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|  | Genetic testing for hereditary mutations in the RET gene |
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|  | August 2013 |
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|  | MSAC application no. 1152  Assessment report |

**Contracted Assessment Report for Application 1152 - Genetic testing for hereditary mutations in the RET gene**

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This report is a contracted technical report for use by the Medical Services Advisory Committee (MSAC) to inform its deliberations. MSAC is an independent committee which has been established to provide advice to the Minister for Health and Ageing on the strength of evidence available on new and existing medical technologies and procedures in terms of their safety, effectiveness and cost effectiveness. This advice will help to inform government decisions about which medical services should attract funding under Medicare.

**MSAC’s advice does not necessarily reflect the views of all individuals who participated in the MSAC evaluation.**

This report was prepared for MSAC by Ms Skye Newton, Ms Camille Schubert, Dr Judy Morona, Mr Paul Fitzgerald, and Associate Professor Tracy Merlin from Adelaide Health Technology Assessment with the assistance of Health Expert Standing Panel member Dr Rory Clifton-Bligh. The report was commissioned by the Department of Health and Ageing on behalf of MSAC. It was edited by Ms Jo Mason of MasonEdit, Adelaide.

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# Executive summary

## Mutation testing of the *RET* gene

### Purpose of application

In October 2010 the Department of Health and Ageing received an application from the Pathology Services Table Committee (PSTC) requesting a Medicare Benefits Schedule (MBS) listing to enable RET (rearranged during transfection) mutation testing for (i) patients with symptoms of multiple endocrine neoplasia type 2 (MEN2) and (ii) unaffected relatives of a patient with a documented RET mutation to determine the risk of disease. It was proposed that two new MBS items cover the use of diagnostic and predictive testing for mutations in the *RET* gene.

A team from Adelaide Health Technology Assessment (AHTA), University of Adelaide, was contracted to conduct a systematic review of the literature and an economic evaluation of RET mutation testing. A decision analytic protocol (DAP) was developed before commencement of the assessment and was approved by the Protocol Advisory Sub-Committee (PASC) of the Medical Services Advisory Committee (MSAC).

The *RET* proto-oncogene encodes a receptor tyrosine kinase, which is involved in processes such as neural crest differentiation, cell migration and proliferation (Burzynski et al. 2005). Hereditary mutations in this gene cause MEN2 and hereditary Hirschsprung’s disease (colonic aganglionosis). Genetic testing may be indicated in a patient with one or more features of the syndrome (diagnostic testing) to make a diagnosis; or it can be used in unaffected relatives of a patient with a documented RET mutation (presymptomatic testing) in order to determine their risk of disease and reduce morbidity and mortality through early intervention.

### Multiple endocrine neoplasia type 2 (MEN2)

MEN2 is a group of disorders associated with tumours of the endocrine system (generally the thyroid, parathyroid and adrenals). Nearly all patients develop a medullary thyroid carcinoma (MTC), and half of patients with MEN2A or MEN2B develop phaeochromocytomas (Margraf et al. 2009). Of those patients with MEN2A, 15–30% may also develop hyperparathyroidism, whereas patients with MEN2B are not at risk of parathyroid disease but will show other abnormalities such as ganglioneuromas, medullated corneal nerves and marfanoid body habitus (Eng 1999). Familial medullary thyroid cancer (FMTC) comprises families who only have MTC. However, some RET mutations are associated with both MEN2A and FMTC, so a clinical history is required to distinguish between the two conditions (Margraf et al. 2009).

RET mutation testing is currently performed in order to triage further investigations. If patients are found to have pathological RET mutations, they are investigated for further MEN2 features before receiving a total thyroidectomy. However, if patients are found to have no pathological RET mutations, they are either assumed to have a sporadic MTC or hyperparathyroidism, or are investigated for other hereditary disorders associated with phaeochromocytoma.

### Proposal for public funding

The proposed MBS items are summarised in Table 1. The suggested fees are based on updated information on the current pricing of RET mutation testing in Australia and differ from the fees proposed in the final DAP.

It is a requirement that all individuals undergoing predictive testing should first receive genetic counselling and give informed consent (or assent in the case of children). It is also recommended that patients undergoing diagnostic RET mutation testing should undergo genetic counselling. As a consequence, it is suggested that the ordering of the genetic test for RET mutations should be limited to specialised genetic services that can provide accredited genetic counselling to patients and their family members.

Table 1 Proposed MBS item descriptors for RET mutation testing

| Category 6 – Pathology services |
| --- |
| MBS [item number]  Detection of germline mutations in the *RET* gene in patients with:   1. medullary thyroid carcinoma 2. adrenal phaeochromocytoma under the age of 50 years 3. hyperparathyroidism plus a diagnosis of medullary thyroid cancer or phaeochromocytoma in a close relativea   1 or more tests  Fee: $400  Prior to ordering these tests the ordering practitioner should ensure that the patient (or their parent/guardian in the case of children) has given informed consent. Testing can only be performed after genetic counselling. Appropriate genetic counselling should be provided to the patient either by the treating practitioner, a genetic counselling service or a clinical geneticist on referral. Further counselling may be necessary upon receipt of the test results. |
| MBS [item number]  Detection of a known mutation in the *RET* gene in:   1. asymptomatic first- or second-degree relatives, at genetic risk, of a patient with a documented pathogenic RET mutation   1 or more tests  Fee: $200  Prior to ordering these tests the ordering practitioner should ensure that the patient (or their parent/guardian in the case of children) has given informed consent. Testing can only be performed after genetic counselling. Appropriate genetic counselling should be provided to the patient either by the treating practitioner, a genetic counselling service or a clinical geneticist on referral. Further counselling may be necessary upon receipt of the test results. |

### Current arrangements for public reimbursement

Currently, RET mutation testing is standard practice, although there is no MBS listing for any test that detects mutations of the *RET* gene. Patients are therefore encouraged to have their blood sample collected through a public hospital so that genetic testing is conducted and billed under state and territory public hospital arrangements. When patients are referred by a private facility, they are billed directly. Private health insurance generally provides a subsidy for testing only if the MBS also provides a rebate for the test (ALRC 2003; PaLMS 2011).

Three accredited pathology laboratories in Australia offer RET mutation testing (RCPA 2012). All offer polymerase chain reaction (PCR) amplification and DNA sequencing of the RET gene, with a 4-week to 3-month turnaround for results. The costs of RET mutation testing in Australia are summarised in Table 6 (page xxxiv).

There have been no previous MSAC considerations of RET mutation testing.

### Prerequisites to implementation of any funding advice

RET mutation testing is currently classified as a Class 3 *in-vitro* diagnostic (IVD) by the Therapeutic Goods Administration (TGA). Laboratories offering the test in house must have National Association of Testing Authorities (NATA) accreditation, with demonstrated compliance with the suite of standards on the validation of in-house IVDs, as published by the National Pathology Accreditation Advisory Council, for each test manufactured.

### Consumer impact statement

The public was invited to provide feedback on the draft protocol for undertaking this evaluation of RET mutation testing during October 2011. No public consultation responses were received from any relevant craft groups or consumer groups.

### Clinical need

It is estimated that the prevalence of MEN2 is 2.5 per 100,000 in the general population (Raue & Frank-Raue 2010). In a population of 22.6 million people (ABS 2011), it is therefore estimated that approximately 500–600 Australians have this rare disorder. The best estimate of the population *suspected* of having MEN2 comprises those who are diagnosed with MTC. In 2007 there were a total of 456 males and 1,331 females newly diagnosed with thyroid carcinomas (AIHW 2010). Approximately 5–10% of thyroid carcinomas are medullary (Keatts & Itano 2006), so it is estimated that, of the 1,787 thyroid carcinomas diagnosed, 89–179 of them would be medullary.

In 2007 there were 150 diagnostic tests performed on the *RET* gene in Australia (Suthers 2008b). This is within the range of what would be expected given the estimated rate of MTCs diagnosed. It is expected that having item numbers on the MBS to allow reimbursement for RET mutation testing would not significantly impact on the number of genetic tests being performed on patients, given that it is already considered standard practice in Australia[[1]](#footnote-1) and ‘best practice’ world-wide (Brandi et al. 2001).

Only 25–30% of MTCs are hereditary (Raue & Frank-Raue 2010), so the use of the genetic test in the proband would rule out the need for further familial genetic testing or MTC surveillance in 65–70% of cases. It is therefore expected that only 22–54 Australian patients per year would have MTC caused by MEN2, resulting in their first-degree relatives requiring genetic screening. Based on data from the Familial Cancer Unit in South Australia, there are approximately 11.5 unaffected first- or second-degree relatives per proband (Suthers et al. 2006). In a study assessing uptake of genetic screening, when family members were contacted both by the proband and directly by letter from the Familial Cancer Unit, 40% of relatives undertook genetic screening within 2 years (Suthers et al. 2006). It is therefore estimated that, on average, 4.6 family members per proband would agree to predictive genetic testing. In 2007 there were 49 presymptomatic tests performed on the *RET* gene in Australia (Suthers 2008b), which is below the rate of what would be expected, assuming that more than one relative per proband would be tested. It is therefore estimated that having an item number on the MBS for detection of a known mutation in the *RET* gene in first- or second-degree relatives at genetic risk would increase the number of presymptomatic tests to approximately 101–248 per year.

## Clinical place for proposed intervention

For the diagnosis of MEN2, RET mutation testing is used to triage (or replace, in the case of pentagastrin-stimulated calcitonin) biochemical screening and imaging in those patients with clinical features suggestive of MEN2.

Two clinical management algorithms have been provided for RET mutation testing in index cases *with* an MTC (Figure 1) and *without* an MTC (Figure 2), and for their close family members. The clinical scenario in Figure 1 is more common than in Figure 2, as an MTC is the first symptom in most MEN2 families due to its earlier and higher penetrance (Brandi et al. 2001). The left side of each management algorithm outlines the approach to the diagnosis and prediction of MEN2 in a setting without genetic testing (as the historical comparator), while the right side shows current clinical practice, which includes the use of genetic testing. The white text boxes and solid arrows relate to the diagnosis and treatment of people with clinical features suggestive of MEN2, while the black boxes and dashed arrows correspond to the management of their close family members.

Special emphasis should be given to material differences between the algorithms outlining the ‘historical’ and ‘current’ clinical management strategies for MEN2 in the type of healthcare resources and the frequencies of their use. Figure 1 shows that, in the absence of RET mutation testing (the historical setting), all patients with an MTC at presentation or detected through initial investigations would be monitored for further clinical features of MEN2, despite there being a 75% chance of the MTC being sporadic. It is also assumed that, in the absence of genetic testing, their first-degree family members would receive annual surveillance for MEN2 features. Family members would undergo a total thyroidectomy once early signs of MTC are detected by elevated calcitonin levels. In comparison, the main differences between this historical setting and the current setting (with RET mutation testing available) are: i) the *targeted* use of lifelong surveillance in patients and family members who have a definitive diagnosis of MEN2 or RET mutation, or the *avoidance* of this requirement in those patients and family members without a RET mutation; and ii) the use of prophylactic total thyroidectomy in family members with a confirmed RET mutation.

In Figure 2 it is shown that, in the absence of RET mutation testing, all those who present with an early onset adrenal phaeochromocytoma or hyperparathyroidism (plus a diagnosis of MTC or phaeochromocytoma in a close relative) who are found *not* to have an MTC would be assumed not to have MEN2. Therefore, the index case and their family members would not be screened or undergo surveillance. However, in the current setting where genetic testing is available, patients with this clinical profile who have a RET mutation would be diagnosed with MEN2 and therefore undergo prophylactic total thyroidectomy and lifelong surveillance. Their family members would also undergo cascade screening and those who also carry the RET mutation would undergo prophylactic thyroidectomy and lifelong surveillance.

Flow chart showing that under the historical pathway, all patients with an MTC would be treated as if they had MEN2, with the index case and their relatives all receiving lifelong surveillance for MEN2 features. 
Under the current pathway, with RET testing available, those index cases without RET mutations can avoid lifelong surveillance, and their family members don't need surveillance. Those with the mutations, are treated the same as in the historical scenario.

Figure Management algorithm for use of a RET mutation test to diagnose and predict MEN2 (MTC identified in index case prior to genetic testing)

a Biochemical screening and imaging for further features of MEN2: plasma or urine catecholamine (and adrenal imaging e.g. adrenal CT scan or MRI and/or MIBG scan if these are elevated) to assess for phaeochromocytoma, serum calcium (and parathyroid hormone if elevated) to assess for hyperparathyroidism.

b Surveillance in those who have had a total thyroidectomy: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+) and calcitonin and carcinoembryonic antigen to detect persistence or recurrence of MTC.

c Historical surveillance in those at risk of MEN2 who have not had a total thyroidectomy: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+); pentagastrin-stimulated serum calcitonin and neck ultrasound to assess for an MTC.

d Second-degree relatives would only be considered to be at genetic risk if first-degree relatives have a RET mutation, clinical features of MEN2, or if information regarding first-degree relatives is unavailable.

Algorithm showing clinical pathway for patients suspected of having MEN2, who do not have an MTC at presentation. In the historical scenario, if an MTC is found, they would be assumed to have MEN2, and their family would receive surveillance. If the index case does not have an MTC, they would be assumed not to have MEN2, and the family would not receive surveillance. In the current scenario, decisions about surveillance and prophylactic thyroidectomies would be based on RET mutation status. 

Figure Management algorithm for use of a RET mutation test to diagnose and predict MEN2 (no MTC in index case prior to genetic testing)

a Screening for other hereditary disorders: genetic testing of the VHL gene for von Hippel Lindau disease, genetic testing for SDHB, SDHC and SDHD mutations; if serum calcium and parathyroid hormone are elevated then additional testing for features of MEN1 (serum gastrin, serum insulin, serum glucagon, serum pancreatic polypeptide, serum vasoactive intestinal peptide, serum prolactin, growth hormone and adrenocorticotrophic hormone).

b Historical biochemical screening and imaging for further features of MEN2: pentagastrin-stimulated serum calcitonin and neck ultrasound to assess for an MTC; plasma or urine catecholamine (and adrenal imaging e.g. adrenal CT scan or MRI and/or MIBG scan if these are elevated) to assess for phaeochromocytoma if not the presenting clinical feature, or serum calcium (and parathyroid hormone if elevated) to assess for hyperparathyroidism if not the presenting clinical features.

c Current biochemical screening and imaging for further features of MEN2: plasma or urine catecholamine (and adrenal imaging e.g. adrenal CT scan or MRI and/or MIBG scan if these are elevated) to assess for phaeochromocytoma if not the presenting clinical features, or serum calcium (and parathyroid hormone if elevated) to assess for hyperparathyroidism if not the presenting feature.

d Historical surveillance: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+); plus pentagastrin-stimulated serum calcitonin and neck ultrasound to assess for an MTC or calcitonin and carcinoembryonic antigen after surgery for MTC.

Current surveillance: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+) ± calcitonin and carcinoembryonic antigen after surgery for MTC.

e Second-degreerelatives would only be considered to be at genetic risk if first-degree relatives have a RET mutation, or if information regarding first-degree relatives is unavailable.

## Comparator

Comparators are usually selected by determining the technology (including testing strategy) most likely to be replaced, or added to, by the technology submitted for a new MBS item number. However, in the situation of RET mutation testing of patients suspected of having MEN2 or of their close family members, genetic testing is already standard practice. As a consequence, when determining the financial implications of RET mutation testing in this report, the comparator is considered to be genetic testing paid for either by the patient or by the states and territories through the public hospital system.

The comparator for financial implications therefore differs from the comparator used to benchmark the safety, effectiveness and cost-effectiveness of RET mutation testing. As RET mutation testing is a means of triagingbiochemical screening and imaging (and a replacement for pentagastrin-stimulated calcitonin measurements) in patients suspected of having MEN2 and their close relatives, the comparator selected was biochemical screening and imaging *alone* for the diagnosis of MEN2. The screening and imaging investigations that patients receive depend on their presenting feature. For index patients, the comparator is outlined in the second column of Table 7 (page xxxvi) and Table 87, in Appendix I outlines the MBS items that correspond to these investigations.

There is no specific alternative test to determine individual *susceptibility* to MEN2. Without genetic testing the diagnosis of MEN2 would rely on tumour type and location, which is not possible to assess prospectively. However, close family members of someone with MEN2 would have lifelong surveillance to ensure early detection of disease. The surveillance regimen for those with MEN2 or at risk of MEN2 is outlined in Table 2 and the MBS items corresponding to these surveillance measures are given in Table 88 n Appendix I. The comparison for first-degree relatives (and second-degree relatives in a cascade fashion) is therefore between genetic counselling and RET mutation testing in addition to a prophylactic thyroidectomy, lifelong thyroxine and lifelong surveillance in those who carry a RET mutation, versus genetic counselling and lifelong surveillance (with a total thyroidectomy and lifelong thyroxine after a rise in calcitonin levels) for all at-risk relatives.

Table 2 Lifelong surveillance regimen for MEN2

| **Age** | **Surveillance** |
| --- | --- |
| From 1–5 years | *Annual*  General clinical examination  Examination of thyroid (or thyroid bed if post-thyroidectomy) by neck ultrasound (and biopsy of any suspicious masses)  Biochemical screen for phaeochromocytoma  Screen for hyperparathyroidism (total and ionised serum calcium)  ± calcitonin and carcinoembryonic antigen after surgery for MTC |

Source: (Genetics Sub-committee of PSTC 2010)

### Scientific basis of comparison

There were no studies identified, that met the inclusion criteria, on which to judge the safety of genetic testing for RET mutations. One historical controlled study (level III-3 interventional evidence) and 8 uncontrolled case series (level IV interventional evidence) reported on the safety of prophylactic thyroidectomies.

The diagnostic effectiveness of RET mutation testing was primarily determined by 9 historical controlled studies with a moderate to high risk of bias. These studies were supported by 171 uncontrolled case series (level IV interventional evidence and/or level IV diagnostic evidence) that provided uncontrolled data on the impact of RET mutation testing on health outcomes and/or diagnostic yield data.

Test performance could not be determined as no studies were identified that provided long-term clinical data that could be used as the reference standard for a MEN2 diagnosis.

One cohort study (level III-2 interventional evidence) was identified that assessed whether the expected change in management from RET mutation testing was associated with better health outcomes for individuals who underwent a prophylactic thyroidectomy.

### Comparative safety

No studies were available that specifically reported on the safety of RET mutation testing or surveillance for MEN2. However, given the availability of RET mutation testing results in asymptomatic gene carriers being recommended to undergo prophylactic total thyroidectomy, rather than waiting for clinical signs of an MTC, the safety of prophylactic total thyroidectomy was assessed.

One historical controlled study (level III-3 interventional evidence) showed similar rates of mortality due to surgical complications in those who underwent surgery prior to knowledge of the link between RET mutation status and MEN2, versus those who underwent surgery knowing their RET mutation status (one death in each cohort). Twelve case series (level IV interventional evidence) reported on the rate of adverse events following total thyroidectomy. Transient hypoparathyroidism was reported in five patients (36.4%) in 4 of the 12 case series. Permanent hypoparathyroidism occurred in between 7.7% and 13.6% of patients from 4 of the 12 studies that reported adverse events after total thyroidectomy. Transient laryngeal nerve palsy was reported in between 4.5% and 5.9% of patients in 4 studies, and one case of permanent laryngeal nerve palsy was reported. Other complications included one case of arterial bleeding, one case of fluctuating thyroid hormone (at 1 year post-surgery) despite adequate compliance with thyroxine replacement, and one case of permanent unilateral Horner’s syndrome[[2]](#footnote-2). It is expected that the rate of surgical complications would be higher in those patients who undergo surgery at a later stage of disease, due to the more invasive surgery required to remove an MTC once the tumour has extended beyond the thyroid, although direct evidence was not available comparing the safety of prophylactic thyroid surgery against curative surgery.

### Key results

There were no safety concerns (either physical or psychological) raised in any of the articles identified regarding RET mutation testing.

### Overall conclusion with respect to comparative safety

RET mutation testing is a safe procedure for patients, involving a simple blood test. In those who are found to be RET mutation carriers, the treatment recommended is a prophylactic thyroidectomy to avoid the risk of developing an MTC. This procedure is associated with a risk of hypoparathyroidism and laryngeal nerve palsy, which is usually transient. The risk of adverse events with prophylactic surgery is likely to be lower than when patients are treated at a later disease stage.

### Comparative effectiveness

Nine historical controlled studies (level III-3 interventional evidence) provided evidence showing that health outcomes are likely to be better for patients diagnosed with the addition of RET mutation testing.

Seven historical controlled studies reported on the incidence and severity of MTC in patients who underwent total thyroidectomy in the era prior to RET mutation testing compared with the era subsequent to the introduction of RET mutation testing. Those diagnosed and treated since RET mutation testing became available had almost half the risk of having an MTC at the time of surgery, compared with those whose treatment decisions were based on biochemical screening in the pre-RET mutation testing era (RR=0.53, 95% CI 0.32, 0.90). It is unknown whether any clinical benefit has occurred in index patients, or whether all the benefits found have been due to more effective management of family members.

One historical controlled study reported that age at diagnosis reduced for patients with MEN2A and FMTC between two surveys in Japan, one performed in 1996 (capturing data prior to the availability of RET mutation testing) and the other in 2002. Age at diagnosis in patients with MEN2B increased marginally, likely just through chance given the small sample; however, the MEN2B phenotype is more clearly diagnosed than the MEN2A, so genetic testing has probably had less impact on patients and their family members with or suspected of having MEN2B than MEN2A. Five additional historical controlled studies reported that the introduction of RET mutation testing allowed the age at time of total thyroidectomy to significantly reduce. One Australian study reported that the mean age decreased from 32 years to 16 years (Learoyd et al. 1997).

Both age at time of total thyroidectomy and severity of MTC are significant predictors of the risk of residual or recurrent disease (Schreinemakers et al. 2010). Six historical controlled studies reported a greatly reduced risk of persistence, recurrence or mortality in those who underwent total thyroidectomy with knowledge of their RET mutation status, compared with total thyroidectomy without this knowledge (RR=0.28, 95% CI 0.17, 0.45). However, this evidence is highly biased, as those in the historical cohort were followed up for longer time periods, allowing a greater chance of disease recurrence simply as a matter of time.

Assessment of individual components in an evidence linkage supported the conclusions based on direct evidence of the impact of testing on patient health outcomes. One historical controlled study and 3 case series reported instances of false positive results based on calcitonin levels, which led to patients either undergoing total thyroidectomy or being scheduled for surgery that was subsequently cancelled after a negative RET mutation status was identified. One single case of an individual free from RET mutations, in a family with known mutations, who had an MTC was noted (Halling et al. 1997). It is unknown whether this could be considered a false negative RET mutation test or a coincidental finding of a spontaneous MTC in a RET-mutation-negative family member of an FMTC kindred. Although a true comparison of accuracy was not able to be performed given the lack of long-term clinical follow-up data to use as a reference standard for MEN2 diagnosis, the limited evidence available would suggest that diagnoses made with the addition of RET mutation testing are likely to be more accurate than those made on the basis of biochemical screening. As the treatment option (thyroidectomy) is the same, irrespective of early or late identification of MEN2, and has proven effectiveness, it is unlikely that studies assessing the comparative effectiveness of thyroidectomy in an ‘earlier (RET-mutation-tested)’ versus ‘later (non-RET-mutation-tested)’ MEN2 diagnosed population are necessary or will be conducted.

Patients who are asymptomatic gene carriers are likely to undergo prophylactic total thyroidectomy on the basis of this knowledge. Prophylactic surgery is associated with having a lower stage of MTC disease at time of surgery, compared with surgery performed on the basis of calcitonin levels.

Overall, clinical management with the addition of RET mutation testing would appear to have superior effectiveness and at least non-inferior safety, compared with diagnosis and treatment of MEN2 without knowledge of RET mutation status.

### Key results

There is evidence that RET mutation testing has allowed patients to undergo total thyroidectomy at an earlier age, and at an earlier stage of MTC disease, than before the introduction of RET mutation testing.

### Key uncertainties

Both age and stage of disease at the time of surgery may be considered surrogate outcomes for survival. Longer term patient-relevant outcomes such as rates of mortality and disease recurrence were reported and were highly in favour of RET mutation testing; however, these results were confounded by different lengths of follow-up in the testing and non-testing study arms.

There is also a high risk of bias in the results due to the comparison against historical cohorts. This type of comparison means that it is unknown to what extent other factors might have influenced the results; for example, if significant advances in surgical methods or surveillance for features of MEN2 have occurred over the same time period as the introduction of RET mutation testing, it would be difficult to correctly attribute the clinical benefits.

### Overall conclusion with respect to comparative clinical effectiveness

All the evidence regarding the comparative clinical effectiveness of RET mutation testing was at high risk of bias. This evidence suggests that the addition of RET mutation testing allows identification of patients at risk of MEN2 at a younger age, allowing prophylactic surgery to occur at a younger age and at a less advanced stage of MTC disease. As age and disease stage are predictors of MTC disease recurrence, it is probable that earlier identification will reduce the risk of disease recurrence in MEN2 patients. Assuming that the findings from the evidence base remain consistently in the same direction, even if the size of this effect is confounded by longer lengths of follow-up in the control arm and differences in patient care over time, the comparative clinical effectiveness of the addition of RET mutation testing would be superior to biochemical screening and imaging.

## Economic evaluation

An economic evaluation was conducted for both (i) RET mutation testing in potential index cases—MTC or phaeochromocytoma under 50 years of age—and (ii) RET mutation testing in index cases and additional familial genetic testing in first- or second-degree relatives of identified RET-mutation-positive index cases.

As genetic testing is currently funded through state hospital budgets, an analysis of the comparison between existing clinical practice and funding arrangements against the proposed MBS listings would only identify a shift in the funding provider, but would not identify whether or not the practice of RET mutation testing has economic merit. Therefore, the economic analysis undertaken for this assessment compares the proposed MBS listings for RET mutation testing against a hypothetical analysis of the historical scenario of medical surveillance before RET mutation testing was available.

In each case the model runs over 30 years and shows accumulated healthcare costs from a societal perspective, with discounting applied to both costs and outcomes (where applicable) at a rate of 5% per year.

With respect to the economic evaluation of genetic testing in potential index cases alone, a cost analysis (cost-minimisation) approach was used, as there is no evidence to suggest that health outcomes within the index case will be affected by genetic testing. The inputs into this model relate to the costs of genetic testing and monitoring (consultation, biochemical tests and imaging) for additional MEN2 symptoms. Resources used are based on the surveillance regimen described by the Genetics Subcommittee of the PSTC and current MBS fees (website, March 2013).

With respect to familial testing, a cost-utility analysis was undertaken, as the ability to identify RET-mutation-positive family members via testing allows for prophylactic thyroidectomy treatment and therefore both health costs and outcomes are affected. The inputs into this model relate to the costs of genetic testing, monitoring (biochemical/imaging etc.) and thyroidectomy (surgical, hospital and pharmaceutical). The health states, which are applicable to family members only, include: healthy (no surgery/surveillance); healthy (pre-surgery, with surveillance); healthy (no MTC) post-thyroidectomy (incorporating adverse effects of surgery); symptomatic MTC; and death. Health outcomes are measured as accumulated quality-adjusted life-years (QALYs). While a decrease in the rate of symptomatic MTC following thyroidectomy in patients receiving medical surveillance is associated with early identification of RET-mutation-positive patients through genetic testing, the quantification of this effect is uncertain and represents a major source of uncertainty in the model. Patient uptake rates with respect to both annual medical surveillance and genetic testing are also uncertain.

The cost-minimisation analysis of genetic testing in potential index cases demonstrates that cost savings occur within 5 years of testing. Over the course of 30 years, savings of approximately $535 per MTC patient tested, or $1,458 per phaeochromocytoma patient under 50 years of age tested, would be expected compared with a scenario where testing was not available.

With respect to the cost-utility analysis of genetic testing of potential index cases and family members of patients identified as RET-mutation-positive, the results indicate that availability of genetic testing ‘dominates’ (i.e. it results in both improved health outcomes and cost-savings), compared with the alternative scenario where testing is not available.

Sensitivity analyses suggest that the base-case economic conclusions are relatively robust.

With respect to diagnostic RET mutation testing in suspected index cases presenting with MTC, a net cost might be expected if i) high test costs ($1,150) are applied or ii) diagnostic yield increases substantially (i.e. testing only occurred in patients with suspected familial disease). With respect to diagnostic testing in suspected index cases presenting with phaeochromytoma, the costs of testing are most sensitive to test price.

The cost-utility model incorporating both diagnostic testing and familial screening is highly robust where the index cohort present with MTC. Adoption of RET mutation testing remained the dominant economic strategy (vs historical biochemical screening) across all analyses of alternative test price, diagnostic yield, uptake rates and relative risk (RR) below 0.97.

The cost-utility model incorporating both diagnostic testing of index cases presenting with phaeochromocytoma and predictive testing of their family members, was also relatively robust. Genetic testing remained the dominant economic strategy across alternative values of test price, and diagnostic yield. When uptake rates of testing or screening are reduced to 15% a relatively low ICER ($485/QALY) is obtained.

The base case estimate of RR is 0.25, however this is highly uncertain. In either model if the RR of MTC recurrence is increased to 1.0, then genetic testing has negative outcomes and is either dominated (resulting in neither health benefits nor savings) in the model where index patients present with MTC, or associated with a cost-saving of $4,721/QALY lost in the model where index patients present with phaeochromocytoma. However the assumption of zero clinical benefit may be considered unreasonable and not consistent with the available evidence. Where the index cohort present with MTC, any RR less than 0.97 results in genetic testing remaining dominant (gaining QALYs and saving money), and this applies to any RR less than 0.43 in the model where index cases present with phaeochromocytoma.

### Key uncertainties

The lack of direct comparative evidence and the hypothetical nature of the economic comparisons mean that the actual quantification of both incremental costs and outcomes in the economic models are not expected to be particularly accurate. Furthermore, the model structure is simplistic and incorporates generalised assumptions that do not capture the distribution of patient age or risk profiles. For this reason the assumptions and inputs in the base case have been selected to be conservative with respect to the cost-effectiveness of RET. However, broad-ranging sensitivity analyses to nevertheless demonstrate that cost-effectiveness is maintained across a range of clinical scenarios.

### Overall conclusion with respect to comparative cost-effectiveness

Despite the shortcomings of the model, the robust nature of the findings—that RET mutation testing results in cost savings and health outcome benefits when model inputs are varied over a wide range of possibilities—is reassuring. On this basis the conclusion—that RET mutation testing and subsequent targeted surveillance (in comparison with broader and increased reliance on imaging/biochemical surveillance) *is cost-effective*—is reasonably certain.

### Financial/budgetary impacts

Diagnostic RET mutation testing is estimated to occur in 130–260 patients in 2013, increasing to 147–294 in 2015. The estimate of the population suspected of having MEN2 is based on those diagnosed with MTC (approximately 5–10% of all thyroid cancers) (Keatts & Itano 2006). An annual increase in thyroid cancers (and MTCs) of 6.3% has been projected based on the average annual increase in thyroid cancer in Australia 2005-09. One diagnostic RET test is required per patient.

The likely number of eligible family members who elect to have RET screening tests is estimated to be 150–359 in 2013, increasing to 169–406 in 2015. One predictive RET mutation test would be required per eligible family member. These estimations are based on the following assumptions:

* Between 25% and 30% of diagnostic RET mutation tests identify a patient with a positive hereditary mutation (Raue & Frank-Raue 2010).
* Each index patient has 11.5 first- or second-degree relatives eligible for predictive RET mutation testing (Suthers et al. 2006).
* Of eligible relatives, 40% accept familial testing (Suthers et al. 2006); i.e., uptake of the test occurs in 4.6 family members per index case.

The extent of uptake in eligible family members is uncertain, with lower uptake (of one or two relatives per index patient) previously reported in the Australian context (Suthers 2008b). The effect of this uncertainty on the financial and budgetary impact is explored in sensitivity analyses.

The cost of diagnostic RET mutation testing used in the base-case estimates is $400, and $200 for familial RET screening. These costs are based on the median quote for RET mutation testing of the 6 exons most commonly examined (exons 10, 11 and 13–16) provided from the pathology laboratories currently providing this service, and are substantially lower than the price previously estimated in the DAP. The financial and budgetary impacts using the DAP-based costs are provided in Appendix K. It is assumed that all testing is provided in an outpatient setting and, as such, the MBS will cover 85% of the cost of the test. A patient contribution of 15% is applied.

The total estimated cost to the MBS, based on an estimated number of 130–260 diagnostic and 150–359 predictive RET mutation tests performed in 2013, is $109,654, increasing to $123,906 in 2015 based on 147–294 diagnostic and 169–406 screening RET mutation tests performed (Table 3). However, an unknown proportion of patients may qualify for the Medicare Safety Net, in which case 100% of the scheduled fee is paid by the MBS. Allowing for application of the Medicare Safety Net, the overall true costs to the Commonwealth health budget would lie between the total costs to the MBS and the total combined costs of RET mutation testing, i.e. up to $129,005 in 2013 and $145,772 in 2015.

A cost saving would be observed in the state and territory systems due to transfer of testing services to the MBS; however, the costs of genetic counselling services provided in hospitals would continue as per current arrangements.

Under the current arrangements some patients who are referred through the public system receive genetic counselling services and testing at no direct cost[[3]](#footnote-3). With the listing of RET mutation testing on the MBS, assuming that most patients would receive testing as outpatients, Medicare would pay 85% of the scheduled fee and a patient contribution of 15% would apply (in addition to any ‘gap’ charges or out-of-pocket expenses). Patients who may be eligible for the Medicare Safety Net, and those whose pathology service bulk-bills tests listed on the MBS, may not be required to contribute a co-payment.

Table Total costs of RET mutation testing

| **Year** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- |
| *Diagnostic RET mutation testing* |  |  |  |
| Number of diagnostic RET mutation testsb | 130–260 | 138–277 | 147–294 |
| Estimated expenditure on diagnostic RET mutation testingc | $52,071–$104,141 | $55,351–$110,702 | $58,838–$117,676 |
| Patient co-paymentd | $7,811–$15,621 | $8,303–$16,605 | $8,826–$17,651 |
| Estimated MBS expendituree | $44,260–$88,520 | $47,048–$94,097 | $50,012–$100,025 |
| *Familial (predictive) RET mutation testing* |  |  |  |
| Relatives eligible for testing | 374–898 | 398–955 | 423–1,015 |
| Number of relatives tested | 150–359 | 159–382 | 169–406 |
| Estimated expenditure on predictive RET mutation testingh | $29,941–$71,857 | $31,827–$76,384 | $33,832–$81,197 |
| Patient co-paymentd | $4,491–$10,779 | $4,774– $11,458 | $5,075–$12,179 |
| Estimated MBS expendituree | $25,450–$61,079 | $27,053–$64,927 | $28,757–$69,017 |
| *Total combined cost of RET mutation testingi* | $129,005 | $137,132 | $145,772 |
| Lower limit | $82,011 | $87,178 | $92,670 |
| Upper limit | $175,999 | $187,087 | $198,873 |
| *Total patient co-paymentd* | $19,351 | $20,570 | $21,866 |
| Lower limit | $12,302 | $13,077 | $13,901 |
| Upper limit | $26,400 | $28,063 | $29,831 |
| ***Total cost to the MBSe*** | **$109,654** | **$116,562** | **$123,906** |
| Lower limit | $69,710 | $74,101 | $78,770 |
| Upper limit | $149,599 | $159,024 | $169,042 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b estimated based on a 5–10% incidence of medullary thyroid cancer in all thyroid cancers

c assuming that the cost of the diagnostic RET mutation test is $400 (see Table 6 on page xxxiv)

d assuming that most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

e assuming that all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees, with no allowance for additional MBS if some patients qualify for the Medicare Safety Net

f estimated based on the identification of a positive hereditary mutation in the *RET* gene in 25–30% of tests performed; each patient was assumed to have, on average, 11.5 first- or second-degree relatives eligible for familial screening

g assuming an uptake rate of 40% in eligible family members

h assuming that the cost of the predictive RET mutation test is $200 (see Table 6 on page xxxiv)

I assuming that all patients qualify for the Medicare Safety Net, then the total cost to the MBS would equate to the total combined cost of RET mutation testing

Sensitivity analyses assuming upper estimates around disease incidence and a 100% uptake rate of familial screening were undertaken to provide an extreme upper limit of the predictable financial costs. The estimated cost of RET mutation testing to the MBS under these limits increases to $272,568 in 2015.

The proposed MBS item descriptors require that appropriate genetic counselling be provided to the patient prior to diagnostic testing or familial screening; further counselling may be required upon receipt of the test results. Genetic counselling services have not been accounted for in the financial and budgetary estimates, as the current distribution of counselling services is unlikely to change, with little impact expected to the overall health budget, MBS, and state and territory systems.

Listing RET mutation testing on the MBS is not expected to have any impact on the costs of the overall Australian healthcare system considered in its entirety. The practice of genetic testing and counselling is routine in diagnostic and familial screening of patients in a manner unchanged by the proposed listing and at a similar cost, which is currently borne by state government hospital budgets.

### Other relevant factors

Clinical trials comparing the health outcomes of patients diagnosed with the addition of RET mutation testing, versus diagnosis without RET mutation testing, would now be considered unethical, as RET mutation testing has become standard clinical practice for patients suspected of having MEN2. Although the evidence identified is at risk of bias, studies controlling for confounding factors are highly unlikely to now be performed.

# Glossary and abbreviations

AHTA Adelaide Health Technology Assessment

DAP decision analytic protocol

DTC direct-to-consumer

FMTC familial medullary thyroid cancer

HESP Health Expert Standing Panel

IMVS Institute of Medical and Veterinary Science

IVD *in-vitro* diagnostic (medical device)

MBS Medicare Benefits Schedule

MEN2 multiple endocrine neoplasia type 2

MSAC Medical Services Advisory Committee

MTC medullary thyroid carcinoma

NATA National Association of Testing Authorities

NHMRC National Health and Medical Research Council

PaLMS Pacific Laboratory Medicine Services

PASC Protocol Advisory Sub-Committee

PCR polymerase chain reaction

PICO population, intervention, comparator and outcomes

PSTC Pathology Services Table Committee

QALYs quality-adjusted life-years

RCPA Royal College of Pathologists of Australasia

RET rearranged during transfection

RET M+ RET-mutation-positive

RET M– RET-mutation-negative

RR relative risk

TGA Therapeutic Goods Administration

# Introduction

A rigorous assessment of evidence is the basis of decision-making when funding is sought under Medicare.

The Medical Services Advisory Committee (MSAC) evaluates new and existing health technologies and procedures for which funding is sought under the Medicare Benefits Schedule (MBS), in terms of their safety, effectiveness and cost-effectiveness, while taking into account other issues such as access and equity. The MSAC adopts an evidence-based approach to its assessments, based on reviews of the scientific literature and other information sources, including clinical expertise.

A team from Adelaide Health Technology Assessment (AHTA), School of Population Health, University of Adelaide, as part of its contract with the Department of Health and Ageing, was engaged to conduct a systematic review of the literature on RET (rearranged during transfection) mutation testing and to conduct economic modelling in order to inform the MSAC’s decision-making regarding public funding of the intervention. A decision analytic protocol (DAP) was developed before commencement of the assessment and was approved by the Protocol Advisory Sub-Committee (PASC) of MSAC. AHTA sought input and advice from members of a Health Expert Standing Panel (HESP; see Appendix A), who are clinicians with expertise in the field.

This report summarises and critically appraises current evidence on RET mutation testing in (i) patients with symptoms of multiple endocrine neoplasia type 2 (MEN2) and (ii) unaffected relatives of a patient with a documented RET mutation to determine the risk of disease, in order to draw conclusions on the likely safety, effectiveness and cost-effectiveness of this testing in the event it is funded by Medicare.

## Rationale for assessment

In October 2010 an application was received from the Pathology Services Table Committee (PSTC) by the Department of Health and Ageing requesting an MBS listing of genetic testing for mutations in the *RET* gene for (i) patients with symptoms of MEN2 and (ii) unaffected relatives of a patient with a documented RET mutation to determine the risk of disease. It was proposed that two new MBS items are created to cover the use of diagnostic and predictive testing for mutations in the *RET* gene.

# Background

## Mutation testing of the *RET* gene

The *RET* proto-onco-gene encodes a transmembrane receptor tyrosine kinase involved in processes such as neural crest differentiation, and cell migration and proliferation (Burzynski et al. 2005). Mutations in the *RET* gene are associated with MEN2 (MEN2A and B), familial medullary thyroid cancer (FMTC) and the seemingly unrelated syndrome concerning the congenital absence of the enteric ganglia (Hirschsprung’s disease[[4]](#footnote-4)). RET mutations that are causative of MEN2 are called *gain in function* mutations as they cause ligand-independent RET activation and constitutive cell signalling (Margraf et al. 2009).

MEN2 comprises a group of disorders that are associated with tumours of the endocrine system (generally the thyroid, parathyroid and adrenals) (Table 4). It includes three distinct phenotypes, the features of which are also outlined in Table 4. Nearly all patients develop a medullary thyroid carcinoma (MTC) and half of patients with MEN2A or MEN2B develop phaeochromocytomas (Margraf et al. 2009). Of patients with MEN2A, 15–30% may also develop hyperparathyroidism, whereas patients with MEN2B are not at risk of parathyroid disease but will show other abnormalities such as ganglioneuromas, medullated corneal nerves and marfanoid body habitus (Eng 1999). FMTC comprises families who only have MTC. However, some RET mutations are associated with both MEN2A and FMTC, so a clinical history is required to distinguish between the two conditions (Margraf et al. 2009).

As can be seen in Table 4, a clinical diagnosis of MEN2 would only be given once a minimum of two features are identified in a family, as a means of distinguishing this inheritable disease from sporadic MTC. Seventy-five per cent of cases of MTC are sporadic and the remainder are hereditary (i.e. MEN2A, MEN2B or FMTC) (Wells Jr & Santoro 2009). In the absence of genetic testing, patients with an MTC would be considered to *potentially* have MEN2 and would consequently, along with their first-degree family members, be recommended to undergo annual surveillance for additional clinical features of MEN2[[5]](#footnote-5).

Table 4 MEN2 phenotype definitions

| **Gene** | **Phenotype** | **Codon** | **Clinical characteristics** | **Risk** | **Timing of thyroidectomy** |
| --- | --- | --- | --- | --- | --- |
| *RET* | MEN2A (55% of all cases) | 634 611 618 620 630 631 | Family (or individual) with MTC, and at least one individual developing hyperparathyroidism, phaeochromocytoma, or both. | 2 (higher aggressiveness) | 5 years of age |
| MEN2B  (5–10% of all cases) | 918 883 | MTC (with or without phaeochromocytoma) and characteristic clinical features: mucosal ganglioneuromas, gastrointestinal ganglioneuromas, eye abnormalities including corneal nerve thickening, and skeletal abnormalities including marfanoid body habitus. | 3 (highest aggressiveness) | 1st year of life |
| FMTC  (35–40% of all cases) | 609 791 790 804 649 891 768 | Four or more family members with MTC only. No clinical evidence of phaeochromocytoma, hyperparathyroidism, or any MEN2B-specific clinical features in affected or at-risk family members. | 1 (high aggressiveness) | When calcitonin rises / 5–10 years of age |

Source: (International *RET* Mutation Consortium 2006; Raue & Frank-Raue 2009, 2010); MTC = medullary thyroid carcinoma; FMTC = familial MTC

Most cases of MEN2 are caused by mutations on the *RET* proto-oncogene (over 98% of MEN2 families have known RET mutations) (Margraf et al. 2009). Furthermore, over 90% of people who have a RET mutation will develop MEN2 (Toledo et al. 2006). MEN2 is autosomal dominant, which means that offspring with one affected parent have a 50% chance of having MEN2 themselves. RET mutation testing is used as a means of *diagnosing* MEN2 in those with symptoms (distinguishing between those who have MEN2 and those who have the more common sporadic form of MTC), and as a way of *predicting* which family members will develop MEN2 based on whether they carry the pathogenic mutation of the *RET* gene. Given that specific genotype–phenotype relationships have become evident, the type of specific mutation found may also be used to determine the age at which a prophylactic thyroidectomy should be performed (Raue & Frank-Raue 2009).

## Intended purpose

### Proposed MBS listing

Based on the populations expected to benefit from RET mutation testing (those with clinical features of MEN2 or their family members), the proposed MBS items are suggested as:

1. A diagnostic test to detect germline mutations in the *RET* gene
2. A predictive test to detect mutations in the *RET* gene of family members of a proband.

In a diagnostic setting, the use of RET mutation testing would constitute Level 1 testing as defined by the National Pathology Accreditation Advisory Council, and would therefore not require formal pre-test genetic counselling or written consent (NPAAC 2007). However, expert opinion suggests that *all* patients undergoing RET mutation testing should participate in genetic counselling[[6]](#footnote-6). Predictive testing in unaffected relatives would constitute Level 2 testing, and therefore would need to be restricted to services which can provide accredited genetic counselling (NPAAC 2007).

Diagnosis of a pathogenic RET mutation in first-degree relatives of a proband allows for cascade testing of *their* first-degree relatives (i.e. second-degree relatives of the proband). Rather than restrict the proposed MBS item to first-degree relatives, the item has been worded to allow for situations where first-degree relatives are unavailable (e.g. have died), so second-degree relatives may be tested.

It is a requirement that all patients undergoing predictive testing should first receive genetic counselling and give informed consent (or assent in the case of children). It is also recommended that patients undergoing diagnostic genetic testing should undergo genetic counselling. It is therefore suggested that the ordering of the genetic test for RET mutations should be limited to specialised genetic services that can provide accredited genetic counselling to patients and their family members.

The proposed MBS items are summarised in Table 5. The suggested fees are only indicative and are substantially different from those originally proposed in the final DAP, which were $1150 for the diagnostic test and $480 for the predictive test. The propsed fees listed in Table 5 are based on updated information on the current pricing of RET mutation testing in Australia (Table 6).

Table 5 Proposed MBS item descriptor for RET mutation testing

| Category 6 – Pathology services |
| --- |
| MBS [item number]  Detection of germline mutations in the *RET* gene in patients with:   1. medullary thyroid carcinoma 2. adrenal phaeochromocytoma under the age of 50 years 3. hyperparathyroidism plus a diagnosis of medullary thyroid cancer or phaeochromocytoma in a close relativea   1 or more tests  Fee: $400  Prior to ordering these tests the ordering practitioner should ensure that the patient (or their parent/guardian in the case of children) has given informed consent. Testing can only be performed after genetic counselling. Appropriate genetic counselling should be provided to the patient either by the treating practitioner, a genetic counselling service or a clinical geneticist on referral. Further counselling may be necessary upon receipt of the test results. |
| MBS [item number]  Detection of a known mutation in the *RET* gene in:   1. asymptomatic first- or second-degree relatives, at genetic risk, of a patient with a documented pathogenic RET mutation   1 or more tests  Fee: $200  Prior to ordering these tests the ordering practitioner should ensure that the patient (or their parent/guardian in the case of children) has given informed consent. Testing can only be performed after genetic counselling. Appropriate genetic counselling should be provided to the patient either by the treating practitioner, a genetic counselling service or a clinical geneticist on referral. Further counselling may be necessary upon receipt of the test results. |

a It has been suggested by the Protocol Advisory Sub-Committee (PASC) that the definition of a ‘close relative’ be limited to first- or second-degree family members.

Screening of RET exons 10, 11 and 13–16 was initially considered as likely to identify all common known MEN2-associated mutations; however, increasing reports of mutations on exons 5 and 8 have led to a suggestion that these exons should be included as ‘common’ potential mutation sites and included in the routine screening of MTC patients (Elisei et al. 2012). When seeking test descriptions and costs from Australian pathology providers, it became apparent that broader screening is not common practice in Australia, although no data are available to confirm this. A full RET gene screen is only suggested where there is a strong suggestion of familial disease but no mutation is identified on any of the previously mentioned exons. Only one Australian laboratory indicated that they were readily able to undertake such testing.

MSAC may wish to consider whether the proposed listing of RET mutation testing for diagnostic purposes in potential index cases is intended to screen for the common mutations currently routinely tested for in Australia, or whether an increased—or even complete—gene screen is intended to be covered by the listing. Clearly, the number of exons tested will affect the proposed fee. While 6-exon (exons 10, 11 and 13–16) RET mutation testing is assumed in the base-case analysis in the economic model, sensitivity analysis of a higher cost for more extensive testing is also included.

Table 6 Current fees for RET mutation testing in Australia (descriptions and prices from varying individual laboratories)

| **Test description (from source)** | **Price** |
| --- | --- |
| **Single mutation predictive testing** |  |
| *RET* gene predictive testing (one exon) | $200 |
| MEN2A specific mutation (predictive) | $200 |
| RET known mutation screen | $250 |
| **Diagnostic testing** |  |
| RET (common mutations; exons 10, 11 and 13–16) | $400 |
| MEN2A full screen (exons 10, 11 and 13–16) | $400 |
| RET (common mutations) URGENT (20% surcharge) | $480 |
| *RET* gene: full screen (exons 10, 11 and 13–16) | $600 |
| *RET* gene screen (complete) | $1,500.00 |

Source: Prices provided by the Institute of Medical and Veterinary Science (03/2013); Pacific Laboratory Medicine Services (published on internet and confirmed 02/2013); Peter MacCallum Cancer Centre (02/2013).

## Clinical need

It is estimated that the prevalence of MEN2 is 2.5 per 100,000 in the general population (Raue & Frank-Raue 2010). In a population of 22.6 million people (ABS 2011), it is therefore estimated that 500–600 Australians would have this rare disorder.

The best estimate of the population suspected of having MEN2 are those who are diagnosed with MTC. In 2007 there were 456 males and 1,331 females newly diagnosed with thyroid carcinomas (AIHW 2010). Approximately 5–10% of thyroid carcinomas are medullary (Keatts & Itano 2006), so it is estimated that, of the 1,787 thyroid carcinomas diagnosed, 89–179 of them would be medullary. In 2007 there were 150 diagnostic tests performed on the *RET* gene in Australia (Suthers 2008b). This is within the range of what would be expected given the estimated rate of MTCs diagnosed. It is not expected that having item numbers on the MBS to allow reimbursement for RET mutation testing would significantly impact on the number of genetic tests being performed, given that testing is already considered standard practice in Australia[[7]](#footnote-7) and ‘best practice’ internationally (Brandi et al. 2001).

Only 25–30% of MTCs are hereditary (Raue & Frank-Raue 2010), so the use of the genetic test in the proband would rule out the need for further familial genetic testing in 65–70% of cases. It is therefore expected that 22–54 Australian patients per year would have MTC caused by MEN2, resulting in their first-degree relatives requiring genetic screening. Based on data from the Familial Cancer Unit in South Australia, there are approximately 11.5 unaffected first- or second-degree relatives per proband (Suthers et al. 2006). In a study assessing uptake of genetic screening, when family members were contacted both by the proband and directly by letter from the Familial Cancer Unit, 40% undertook genetic screening within 2 years (Suthers et al. 2006). It is therefore estimated that, on average, 4.6 family members per proband would agree to predictive genetic testing. In 2007 there were 49 presymptomatic tests performed on the *RET* gene in Australia (Suthers 2008b), which is below the rate of what would be expected, assuming that more than one relative per proband would be tested. It is therefore estimated that having an item number on the MBS for detection of a known mutation in the *RET* gene in first- or second-degree relatives at genetic risk would likely increase the number of presymptomatic tests to approximately 101–248 per year.

## Existing procedures for the diagnosis of MEN2 and screening for associated neoplasms

In the absence of genetic testing, patients with clinical features suggestive of MEN2 were investigated for other features of MEN2 as well as for hereditary disorders that may cause the features detected (Genetics Sub-committee of PSTC 2010). Those with early onset adrenal phaeochromocytoma or hyperparathyroidism (in combination with FMTC or phaeochromocytoma) would have been investigated for an MTC by pentagastrin-stimulated serum calcitonin and neck ultrasound.

In the current clinical setting with RET mutation testing as standard practice, it occurs as a triage to further investigations. If patients are found to have pathological RET mutations, they would be investigated for further MEN2 features before receiving a total thyroidectomy. However, if patients are found to have no pathological RET mutations, they would either be assumed to have a spontaneous MTC or hyperparathyroidism, or would be investigated for other hereditary disorders associated with phaeochromocytoma.

Table 7 summarises the investigations and management for MEN2 and other hereditary disorders that would be in use, for both patients presenting with disease and their close family members, in the settings with or without RET mutation testing.

Table 7 Co-administered and associated investigations and management

| **Clinical feature** | **Historical setting (RET mutation testing not available)a** | **Current setting (RET mutation testing available)** |
| --- | --- | --- |
| Medullary thyroid carcinoma | * Investigate for phaeochromocytoma: plasma or urine catecholamines (and adrenal imaging – e.g. adrenal CT scan or MRI and/or MIBG scan – if these are elevated) * Investigate for hyperparathyroidism: serum calcium (and parathyroid hormone if calcium is elevated) to assess for hyperparathyroidism. * Total thyroidectomy, lifelong thyroxine and surveillance for all | * Investigate for MEN2 with RET mutation testing   *Use RET mutation testing to triage further investigations.*  Those with RET mutations:   * Investigate for phaeochromocytoma: plasma or urine catecholamines (and adrenal imaging – e.g. adrenal CT scan or MRI and/or MIBG scan – if these are elevated) * Investigate for hyperparathyroidism: serum calcium (and parathyroid hormone if calcium is elevated) to assess for hyperparathyroidism.   Those without RET mutations:   * No further investigations required * Total thyroidectomy and lifelong thyroxine |
| Adrenal phaeochromocytoma (under 50 years of age) | * Investigate for MTC: pentagastrin-stimulated calcitonin and neck ultrasound * Investigate for hyperparathyroidism: serum calcium (and parathyroid hormone if elevated)   Those with an MTC:   * Total thyroidectomy, lifelong thyroxine and surveillance   Those without an MTC:   * Investigate for other hereditary disorders: genetic testing of the VHL gene for von Hippel Lindau disease; genetic testing for SDHB, SDHC and SDHD mutations; if serum calcium and parathyroid hormone are elevated, then assess for MEN (serum gastrin, insulin, glucagon, pancreatic polypeptide, vasoactive intestinal peptide, calcium, prolactin, adrenocorticotrophic hormone, growth hormone and somatomedin C) | * Investigate for MEN2 with RET mutation testing   *Use RET mutation testing to triage further investigations*  Those with RET mutations:   * Investigate for hyperparathyroidism: serum calcium (and parathyroid hormone if elevated) * Prophylactic total thyroidectomy, lifelong thyroxine and surveillance   Those without RET mutations:   * Investigate for other hereditary disorders: genetic testing of the VHL gene for von Hippel Lindau disease; genetic testing for SDHB, SDHC and SDHD mutations; if serum calcium and parathyroid hormone are elevated, then assess for MEN (serum gastrin, insulin, glucagon, pancreatic polypeptide, vasoactive intestinal peptide, calcium, prolactin, adrenocorticotrophic hormone, growth hormone, and somatomedin C) |
| Hyperparathyroidism (plus a diagnosis of MTC or phaeochromocytoma in a close relative) | * Investigate for MTC: pentagastrin-stimulated calcitonin and neck ultrasound * Investigate for phaeochromocytoma: plasma or urine catecholamines (and adrenal imaging, e.g. adrenal CT scan or MRI and/or MIBG scan if elevated)   Those with an MTC:   * Total thyroidectomy, lifelong thyroxine and surveillance   Those without an MTC:   * No further investigations required | * Investigate for MEN2 with RET mutation testing   *Use RET mutation testing to triage further investigations*  Those with RET mutations:   * Investigate for phaeochromocytoma: plasma or urine catecholamines (and adrenal imaging, e.g. adrenal CT scan or MRI and/or MIBG scan if elevated) * Prophylactic total thyroidectomy, lifelong thyroxine and surveillance   Those without RET mutations:   * No further investigations required |
| First-degree relatives of patients with a known pathogenic RET mutation | Genetic counselling plus lifelong surveillance  Total thyroidectomy once calcitonin levels rise, or on the basis of biopsy of a thyroid nodule detected clinically or on ultrasound, followed by lifelong thyroxine | Genetic counselling plus genetic testing for known RET mutation  In those with RET mutation:   * Prophylactic total thyroidectomy, lifelong thyroxine and surveillance   In those without RET mutation:   * No further follow-up |

a The comparative situation is termed ‘historical’ to illustrate a scenario that existed without the use of RET mutation testing. However, of interest is what tests *would be used currently in the absence of RET mutation testing*. Therefore, genetic testing for other diseases has been included in the comparative setting, despite not being available prior to the introduction of RET mutation testing.

CT = computed tomography; MIBG = meta-iodobenzylguanidine scintigraphy; MRI = magnetic resonance imaging; MTC = medullary thyroid carcinoma; SDHB = succinate dehydrogenase complex subunit B; SDHC = succinate dehydrogenase complex subunit C; SDHD = succinate dehydrogenase complex subunit D.

Patients with MEN2, those thought to have MEN2, and people at risk of developing MEN2 require lifelong surveillance. The surveillance tests offered are listed in Table 8.

In Australian practice the risk assessment of family members is already predominantly done by genetic testing of the *RET* gene (Fleming 2011). The listing of RET mutation testing on the MBS is therefore unlikely to have much impact on the use of current surveillance regimens. However, for the purposes of evaluating the cost-effectiveness of RET mutation testing, the historic clinical setting will be assessed—whereby genetic testing is not available and there is a reliance on annual surveillance for patients with an MTC and all close family members.

Prior to the introduction of RET mutation testing, decisions regarding the requirement for lifelong screening in asymptomatic family members were made based on family history. All first-degree relatives of a patient with an MTC would have been recommended to undergo lifelong surveillance. If a first-degree family member developed clinical features, their first-degree relatives (ie second-degree relatives of the proband) would then undergo annual surveillance using the principles of cascade testing.

With the introduction of RET mutation testing, the frequency of initial biochemical screening and imaging and surveillance within family members is likely to have changed for a variety of reasons:

1. a better distinction between sporadic and hereditary cases of MTC (i.e. a more accurate diagnosis of the index case), such that fewer index cases require lifelong surveillance for MEN2
2. greater certainty regarding an individual’s risk of developing MEN2, due to knowledge of the presence/absence of the RET mutation and understanding of the biomarker, facilitating better compliance with surveillance recommendations in those with a mutation
3. a better distinction between spontaneous and hereditary cases of MTC in the index case, and therefore that fewer families require screening and surveillance
4. the ability to distinguish between first-degree relatives who are, and who are not, at risk of developing MEN2 on the basis of RET mutations. Only those first-degree family members who have an RET mutation would undergo surveillance for clinical features of MEN2, while those who are free from pathogenic RET mutations would avoid the need for lifelong surveillance. Thus, the genetic test would replace (and has replaced to date) routine lifelong screening of family members without RET mutations, resulting in fewer people undergoing annual surveillance
5. earlier screening in second-degree relatives (and possibly broader) on the basis of genetic mutations in first-degree relatives, rather than waiting for the emergence of clinical features. Once a first-degree family member is found to have a RET mutation, their first-degree relatives may be genetically tested (using cascade testing). Although similar in principle to clinical practice before the introduction of genetic testing, identification of a RET mutation may occur years before the presentation of clinical features. It is therefore likely that a proportion of additional second-degree (and more distant) relatives are currently undergoing annual surveillance much earlier than they would have been before the introduction of RET mutation testing.

Points (i), (iii) and (iv) above are likely to have greatly reduced the number of index cases and family members recommended to undergo annual surveillance, while points (ii) and (v) are likely to have resulted in a slight increase in surveillance utilisation in specific populations.

Table Lifelong surveillance regimen for MEN2

| **Age** | **Surveillance** |
| --- | --- |
| From 1–5 years | *Annual*  General clinical examination  Examination of thyroid (or thyroid bed if post-thyroidectomy) by neck ultrasound (and biopsy of any suspicious masses)  Biochemical screen for phaeochromocytoma  Screen for hyperparathyroidism (total and ionised serum calcium)  ± calcitonin and carcinoembryonic antigen after surgery for MTC |

Source: (Genetics Sub-committee of PSTC 2010); MTC = medullary thyroid carcinoma

Treatment for MEN2 will depend on the presentation. Standard chemotherapy regimens and radiation therapy have been found to be ineffective methods of treating MTC, so treatment depends largely on the adequacy of surgical removal of the thyroid (Brandi et al. 2001). MTCs are treated with total thyroidectomy and central lymph node dissection at a minimum (Brandi et al. 2001; Lundgren et al. 2007). Following thyroidectomy, patients are required to take thyroxine over the remainder of their life. The primary treatment for phaeochromocytoma is resection, most often performed laparoscopically (Alderazi et al. 2005). The current treatment for hyperparathyroidism is parathyroidectomy, with the timing based on the degree of hypercalcaemia and/or associated clinical features (e.g. cognitive symptoms, renal stones, osteoporosis)[[8]](#footnote-8). Bisphosphonate treatment may be used in milder degrees of hypercalcaemia when patients show osteoporosis on bone densitometry testing8. It is not expected that treatment for those diagnosed with MEN2 who have an MTC at presentation will change with greater use of genetic testing. However, those who have an MTC who are found not to have a RET mutation (and therefore do not have MEN2) can avoid the need for lifelong surveillance for additional features of MEN2.

It is expected that those who are diagnosed with MEN2 prior to the development of an MTC (i.e. through initial presentation with early onset phaeochromocytoma) would receive a prophylactic thyroidectomy upon confirmation of having a RET mutation. As the penetrance of RET mutations in MEN2 patients is close to 100% (i.e. nearly all who have a mutation on the *RET* gene will eventually develop an MTC), the ideal treatment for MEN2 is a total thyroidectomy before clinical evidence of an MTC, to reduce disease-related morbidity and death (Margraf et al. 2009). MEN2A was the first disease in history where total removal of an organ was performed prior to development of a carcinoma solely on the basis of genetic testing (Lundgren et al. 2007). Since testing for RET mutations has been introduced, fewer patients have been diagnosed with metastatic MTC. RET mutation testing has resulted in fewer patients needing to undergo more extensive surgery such as a lymphadenectomy, and has replaced the need for palliation in some patients.

Treatment of family members who are found to have a RET mutation also involves a prophylactic total thyroidectomy (Brandi et al. 2001). Historically, pentagastrin-stimulated serum calcitonin measurements were used to identify those who had developed an MTC. In the absence of genetic testing, removal of the thyroid would only occur once calcitonin levels had risen, indicating the presence of a carcinoma. The present strategy of performing prophylactic thyroidectomy would therefore have resulted in an increased need for thyroxine in the years between when a prophylactic total thyroidectomy is performed and when that individual would have developed an MTC had prophylactic surgery not been performed.

## Marketing status of technology

### Regulatory status

*In-vitro* diagnostic medical devices (IVDs) are, in general, pathology tests and related instrumentation used to carry out testing on human samples, where the results are intended to assist in clinical diagnosis or in making decisions concerning clinical management (Therapeutic Goods Administration 2009).

The Therapeutic Goods Administration (TGA) regulatory framework for IVDs changed in July 2010, such that in-house laboratory tests now receive the same level of regulatory scrutiny as commercial kits. As RET mutation testing is currently only provided in-house, it would be classified as a Class 3 in-house IVD (Box 1). There is a transition period of 4 years to allow currently supplied goods to be included under the new regulation (IVD Australia 2010).

Laboratories that manufacture in-house Class 3 IVDs are required to notify the TGA of the types of IVDs manufactured in each laboratory for inclusion on a register. These laboratories must have National Association of Testing Authorities (NATA) accreditation, with demonstrated compliance with the suite of standards on the validation of in-house IVDs, as published by the National Pathology Accreditation Advisory Council, for each test manufactured. Manufacturers of Class 2, Class 3 and Class 4 IVDs must hold certification from a regulatory body to show compliance with a suitable conformity assessment procedure (Therapeutic Goods Administration 2009). NATA accreditation includes (but is not limited to) the use of direct sequencing on both DNA strands in all exons in which pathogenic mutations have been identified; and confirmation of the mutation on an independent polymerase chain reaction (PCR)[[9]](#footnote-9).

As the national demand for RET mutation testing is likely to be low, it is probable that it would be undertaken by a small number of laboratories to ensure that they have sufficient throughput to maintain training and procedural quality. The staffing required will depend on the caseload, throughput and infrastructure of the laboratories that provide testing.

Box 1 Classification of Class 3 *in-vitro* diagnostic medical devices

Source: [Commonwealth Consolidated Regulations](http://www.austlii.edu.au/au/legis/cth/consol_reg/tgdr2002400/sch2a.html) [accessed March 2013]

**Therapeutic Goods (Medical Devices) Regulations 2002 –Schedule 2A**

1.3 Detection of transmissible agents or biological characteristics posing a moderate public health risk or high personal risk

1. **An IVD is classified as a Class 3 IVD medical device or a Class 3 in-house IVD if it is intended for any of the following uses**:
   1. detecting the presence of, or exposure to, a sexually transmitted agent;
   2. detecting the presence in cerebrospinal fluid or blood of an infectious agent with a risk of limited propagation;
   3. detecting the presence of an infectious agent where there is a significant risk that an erroneous result would cause death or severe disability to the individual or foetus being tested;
   4. pre-natal screening of women in order to determine their immune status towards transmissible agents;
   5. determining infective disease status or immune status where there is a risk that an erroneous result will lead to a patient management decision resulting in an imminent life-threatening situation for the patient;
   6. the selection of patients for selective therapy and management, or for disease staging, or in the diagnosis of cancer;
   7. **human genetic testing;**
   8. to monitor levels of medicines, substances or biological components, when there is a risk that an erroneous result will lead to a patient management decision resulting in an immediate life-threatening situation for the patient;
   9. the management of patients suffering from a life-threatening infectious disease;
   10. screening for congenital disorders in the foetus.

Note: For paragraph (f) An IVD medical device would fall into Class 2 under clause 1.5 if:

* 1. therapy decisions would usually be made only after further investigation; or
  2. the device is used for monitoring.

1. Despite subsection (1) an IVD is classified as a Class 3 IVD medical device or a Class 3 in-house IVD if it is used to test for transmissible agents included in the Australian National Notifiable Diseases Surveillance System (NNDSS) list as published from time to time by the Australian Government.

## Current reimbursement arrangements

Currently, there is no MBS listing for any test that detects mutations on the *RET* gene. No previous applications have been made to MSAC for public funding of RET mutation testing.

Patients are therefore encouraged to have their blood sample collected through a public hospital so that the state health system covers the cost of genetic testing. In those instances when patients are referred by a private facility, they will be billed directly, as private health insurance generally provides a subsidy for testing only if the MBS also provides a rebate for the test (ALRC 2003; PaLMS 2011).

The genetic testing website of the Royal College of Pathologists of Australasia (RCPA) lists three pathology laboratories that offer RET mutation testing (RCPA 2012). The Cancer Genetics Diagnostic Laboratory, Pacific Laboratory Medicine Services (PaLMS), Royal North Shore Hospital, Sydney, New South Wales, and the Peter MacCallum Cancer Centre in Victoria offer PCR amplification and DNA sequencing of RET exons 10, 11 and 13–16, with a 4-week to 3-month turnaround for receipt of results. The Molecular Pathology Division of the Institute of Medical and Veterinary Science (IMVS), Adelaide, South Australia, offers a gene screen for all exons and associated splice junctions by direct sequencing, with a turnaround of 3 months (RCPA 2012).

The costs of RET mutation testing in Australia are summarised in Table 6 (page xxxiv).

## Consumer impact statement

No responses were made during the public consultation period for the DAP.

# Approach to assessment

A DAP was developed prior to the commencement of the assessment, was informed by clinical experts (Appendix A) and public consultation, and was approved by the PASC of MSAC. The purpose of a DAP is to describe in detail a limited set of decision options associated with the possible public funding of a proposed medical service. The DAP also accurately captures current clinical practice, reflects the likely future practice with the proposed medical service, and describes all potentially affected healthcare resources. The guiding framework of the DAP was used throughout this assessment.

## Objective

The objective of this assessment was to determine whether there is sufficient evidence of clinical need, safety, effectiveness and cost-effectiveness to recommend the public funding of genetic testing for hereditary mutations in the *RET* gene for (i) patients with symptoms of MEN2, and (ii) a family member of a patient with a known pathogenic RET mutation.

## Clinical pathway

RET mutation testing is current standard practice, and is funded by either the states and territories or by patients directly. For the purposes of the financial analysis, the comparison is RET mutation testing funded by the states and territories versus testing funded through the MBS. However, for the purposes of assessing the safety, effectiveness and cost-effectiveness of RET mutation testing, the comparison is the historical setting where RET mutation testing was not available. RET mutation testing has been used as a replacement for pentagastrin-stimulated calcitonin testing and as a triage for biochemical screening and imaging.

Two management algorithms are presented, one for patients presenting with an MTC plus their first degree relatives (Figure 3) and the second for patients presenting with phaeochromocytoma or hyperparathyroidism, and their first degree relatives (Figure 4). On the left side of both algorithms is the historical setting, showing which investigations would be used in the absence of RET mutation testing (i.e. although it is classified historical, it is a hypothetical situation including the use of other genetic tests that would be used in the absence of RET mutation testing today, but that were not used before the introduction of RET mutation testing). On the right side of both algorithms is the current scenario, with RET mutation testing being standard clinical practice. Historically, without RET mutation testing, all patients with MTC were treated as if they had MEN2, and all patients and first-degree relatives would require ongoing surveillance for features of MEN2. Conversely, in the absence of RET mutation testing, all patients presenting with phaeochromocytoma or hyperparathyroidism would have been assumed *not* to have MEN2 unless an MTC was present. Figure 3 is the more common algorithm, given the higher penetrance of MTC than phaeochromocytoma or hyperparathyroidism.

RET mutation testing is used to triage patients to further biochemical investigations and surveillance, and is a replacement for pentagastrin-stimulatedcalcitonin. Those who have pathological RET mutations associated with MEN2 would undergo a total thyroidectomy and receive lifelong thyroxine supplementation.

Flow chart showing that under the historical pathway, all patients with an MTC would be treated as if they had MEN2, with the index case and their relatives all receiving lifelong surveillance for MEN2 features. 
Under the current pathway, with RET testing available, those index cases without RET mutations can avoid lifelong surveillance, and their family members don't need surveillance. Those with the mutations, are treated the same as in the historical scenario.

Figure Management algorithm for use of a RET mutation test to diagnose and predict MEN2 (MTC identified in index case prior to genetic testing)

a Biochemical screening and imaging for further features of MEN2: plasma or urine catecholamine (and adrenal imaging e.g. adrenal CT scan or MRI and/or MIBG scan if these are elevated) to assess for phaeochromocytoma, serum calcium (and parathyroid hormone if elevated) to assess for hyperparathyroidism.

b Surveillance in those who have had a total thyroidectomy: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+) and calcitonin and carcinoembryonic antigen to detect persistence or recurrence of MTC.

c Historical surveillance in those at risk of MEN2 who have not had a total thyroidectomy: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+); pentagastrin-stimulated serum calcitonin and neck ultrasound to assess for an MTC.

d Second-degree relatives would only be considered to be at genetic risk if first-degree relatives have a RET mutation, clinical features of MEN2, or if information regarding first-degree relatives is unavailable.

Algorithm showing clinical pathway for patients suspected of having MEN2, who do not have an MTC at presentation. In the historical scenario, if an MTC is found, they would be assumed to have MEN2, and their family would receive surveillance. If the index case does not have an MTC, they would be assumed not to have MEN2, and the family would not receive surveillance. In the current scenario, decisions about surveillance and prophylactic thyroidectomies would be based on RET mutation status. 

Figure Management algorithm for use of a RET mutation test to diagnose and predict MEN2 (no MTC in index case prior to genetic testing)

a Screening for other hereditary disorders: genetic testing of the VHL gene for von Hippel Lindau disease, genetic testing for SDHB, SDHC and SDHD mutations; if serum calcium and parathyroid hormone are elevated then additional testing for features of MEN1 (serum gastrin, serum insulin, serum glucagon, serum pancreatic polypeptide, serum vasoactive intestinal peptide, serum prolactin, growth hormone and adrenocorticotrophic hormone).

b Historical biochemical screening and imaging for further features of MEN2: pentagastrin-stimulated serum calcitonin and neck ultrasound to assess for an MTC; plasma or urine catecholamine (and adrenal imaging e.g. adrenal CT scan or MRI and/or MIBG scan if these are elevated) to assess for phaeochromocytoma if not the presenting clinical feature, or serum calcium (and parathyroid hormone if elevated) to assess for hyperparathyroidism if not the presenting clinical features.

c Current biochemical screening and imaging for further features of MEN2: plasma or urine catecholamine (and adrenal imaging e.g. adrenal CT scan or MRI and/or MIBG scan if these are elevated) to assess for phaeochromocytoma if not the presenting clinical features, or serum calcium (and parathyroid hormone if elevated) to assess for hyperparathyroidism if not the presenting feature.

d Historical surveillance: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+); plus pentagastrin-stimulated serum calcitonin and neck ultrasound to assess for an MTC or calcitonin and carcinoembryonic antigen after surgery for MTC.

Current surveillance: annual general clinical examination, examination of thyroid (or thyroid bed if post-thyroidectomy), biochemical screen for phaeochromocytoma, screen for hyperparathyroidism (total and ionised serum Ca2+) ± calcitonin and carcinoembryonic antigen after surgery for MTC.

e Second-degreerelatives would only be considered to be at genetic risk if first-degree relatives have a RET mutation, or if information regarding first-degree relatives is unavailable.

## Comparator

For the sake of determining the safety, effectiveness and cost-effectiveness of RET mutation testing and subsequent investigations and treatments, a comparison was made against investigations and timing of treatments that would occur without the use of RET mutation testing. The investigations would vary depending on the presenting feature leading to patients being suspected of having MEN2. Table 7 (page xxxvi) outlines the investigations and management strategies that are used with RET mutation testing and without RET mutation testing in index cases and asymptomatic close family members.

## Questions for public funding

In order to clearly articulate the policy question to publicly fund RET mutation testing, details on the appropriate population, intervention, comparator and outcomes (PICO) of interest were sought from the PASC. The PICO approach outlines aspects of the following clinical questions that the assessment is intended to answer:

* Population – specification of the characteristics of the population or patients in whom the intervention is intended to be used;
* Intervention – specification of the proposed intervention;
* Comparator – specification of the therapy most likely to be replaced by, or added to, the proposed intervention; and
* Outcomes – specification of the health outcomes and the healthcare resources likely to be affected by the introduction of the proposed intervention.

Table 9 describes the PICO that were used to develop the clinical questions addressed by this evaluation. The questions were:

1. Is RET mutation testing safe, effective and cost-effective, in the diagnosis of patients suspected of having MEN2, when used to replace or triage the investigation of further features of MEN2 or other hereditary disorders, and to determine who should receive a prophylactic total thyroidectomy and/or lifelong surveillance?

2. Is RET mutation testing safe, effective and cost-effective, in first- and second-degree family members of patients diagnosed with MEN2, when used to determine who should receive prophylactic total thyroidectomy and/or lifelong surveillance?

Table 9 Summary of PICO to define clinical questions

| **Patients** | **Intervention** | **Comparator** | **Reference standard** | **Outcomes to be assessed** |
| --- | --- | --- | --- | --- |
| Patients presenting with clinical features of MEN2:   * MTC (any age) | RET mutation testing  *In those with RET mutation*: clinical investigations for phaeochromoctyoma and hyperparathyroidism, plus total thyroidectomy, lifelong thyroxine and surveillance  *In those without RET mutation*: total thyroidectomy and lifelong thyroxine | Clinical investigations for phaeochromoctyoma and hyperparathyroidism plus total thyroidectomy, lifelong thyroxine and surveillance for all | Long-term clinical assessment | **Safety**  Psychological and physical harms from testing  **Effectiveness** *Direct evidence* **Primary outcomes:**  Mortality  Progression-free survival  Quality of life  Incidence and severity (TNM stage) of MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism  **Secondary outcomes:**  Incidence of symptoms arising from MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism  Timing of thyroidectomy  Age at diagnosis  Rates and implications (physical and psychological) of surveillance  **Cost-effectiveness**  Cost, cost per event avoided, cost per life-year gained, cost per quality-adjusted life-year or disability-adjusted life-year, incremental cost-effectiveness ratio |
| Patients presenting with clinical features of MEN2:   * adrenal phaeochromocytoma (under 50 years of age) | RET mutation testing  *In those with RET mutation*: clinical investigations for hyperparathyroidism, plus prophylactic total thyroidectomy, lifelong thyroxine and surveillance  *In those without RET mutation*: investigations for other hereditary disorders | Clinical investigations for MTC and hyperparathyroidism  *In those with an MTC:* total thyroidectomy, lifelong thyroxine and surveillance  *In those without an MTC:* investigations for other hereditary disorders |
| Patients presenting with clinical features of MEN2:   * hyperparathyroidism plus a diagnosis of MTC or phaeochromoctyoma in a close relative | RET mutation testing  *In those with RET mutation*: clinical investigations for phaeochromoctyoma, plus prophylactic total thyroidectomy, lifelong thyroxine and surveillance  *In those without RET mutation*: no further investigations | Clinical investigations for MTC and phaeochromoctyoma  *In those with an MTC:* total thyroidectomy, lifelong thyroxine and surveillance  *In those without an MTC:* no further investigations |
| First-degree relatives of patients with a diagnosis of MEN2 or a known pathogenic RET mutation | Genetic counselling plus genetic testing for known RET mutation  *In those with RET mutation*: prophylactic total thyroidectomy, lifelong thyroxine and surveillance  *In those without RET mutation:* no further follow-up | Genetic counselling plus lifelong surveillance  Total thyroidectomy once calcitonin levels rise, or on the basis of biopsy of a thyroid nodule detected clinically or on ultrasound, followed by lifelong thyroxine |

MTC = medullary thyroid cancer; TNM Classification of Malignant Tumours is a cancer staging system whereby T describes the size of the primary tumour, N the involvement of regional lymph nodes, and M the presence of distant metastasis.

## Diagnostic assessment framework

This assessment of RET mutation testing is based on the framework outlined in the MSAC *Guidelines for the Assessment of Diagnostic Technologies* (MSAC 2005).

The effectiveness of a diagnostic or predictive test depends on whether it improves patient health outcomes. The clinical benefit can be assessed by studies that directly investigate the impact of the test on health outcomes or, alternatively, in some situations by linking evidence from different studies within the diagnostic or predictive pathway.

### Direct evidence

In a very simplified manner, comparative **direct evidence** would present data on patients suspected (due to signs/symptoms) or at risk (due to family history) of having MEN2. These people would receive either clinical screening alone or RET mutation testing. Both study arms would have patients undergo further investigations and treatments on the basis of the test results (Figure 5). If one study arm was better at identifying MEN2 and appropriately targeting treatment than the other study arm, this would be reflected in a difference in the health outcomes between the patient groups.

A diagram showing that the ideal study design to test the benefit of RET testing is to randomise patients to undergo clinical screening in one arm, and RET testing in another arm, and follow the patients through until health outcomes (all those tested, both positive and negative for MEN2 or mutations).

Figure Ideal structure of comparative direct diagnostic evidence

### Linked evidence

Scoping literature searches indicated that the available direct evidence was limited, so a **linked evidence** approach was used to supplement the evidence base. This included assessing evidence on test performance/diagnostic accuracy, identifying studies addressing changes in clinical management as a consequence of RET mutation testing, and evaluating the likely treatment effectiveness in people found to have a *RET* gene mutation.

The questions addressing each linkage are given as follows:

#### Linkage 1 – Test accuracy

1. Is genetic testing for mutations in the *RET* gene, to triage further clinical investigations, as accurate as, or more accurate than, usual clinical diagnosis to identify patients with MEN2?

2. Is genetic testing for mutations in the *RET* gene, plus annual screening, as accurate as, or more accurate than, annual screening to diagnose relatives of patients with a known RET mutation?

The reference standard

The reference standard is long-term clinical assessment (ideally over the life-time of the patient). However, if a person is identified as having a pathological RET mutation associated with MEN2 prior to the development of an MTC, they would be strongly encouraged to have a prophylactic thyroidectomy to remove the possibility of developing an MTC. After removing the thyroid it is not possible to determine whether the individual would have developed an MTC or not (unless histopathological evidence shows evidence of microscopic disease), so the reference standard is imperfect and would potentially classify many people as receiving a false positive diagnosis by RET mutation testing.

Furthermore, the distinction between a spontaneous MTC and a familial MTC (as occurs in MEN2) is not always able to be made on the basis of clinical information when assessing the individual alone (Table 4, page xxxii). An individual with an MTC may have a *de novo* germline RET mutation but still not meet the classification of MEN2 or FMTC, as they will not have developed hyperparathyroidism or phaeochromocytoma, nor have sufficient family members with clinical signs of disease, to be classified as having MEN2.

#### Linkage 2 – Change in patient management

It was assumed that there is a change in patient management based on results of RET mutation testing, as those patients and relatives who have a RET mutation, but do not yet have an MTC, are likely to undergo a prophylactic thyroidectomy (and receive lifelong thyroxine). In the absence of RET mutation testing, this would only occur after clinical signs of disease are found (i.e. raised calcitonin levels). All patients who have an MTC would be assumed to potentially have MEN2 and receive lifelong screening accordingly. This would also occur when a RET mutation is confirmed. However, in patients where a pathological RET mutation is not found, screening for additional features of MEN2 would not be required.

In the absence of RET mutation testing, family members of index cases with an MTC would undergo annual screening for features of MEN2. With the addition of RET mutation testing, only those family members who are RET-mutation-positive would require screening.

Although these changes to clinical management are clear from expert opinion, relevant literature would also be collated to answer the following questions:

3. Does genetic testing for mutations in the *RET* gene, in addition to usual diagnostic assessment, change the management of patients suspected of MEN2 when compared with usual diagnostic assessment alone?

4. Does genetic testing for mutations in the *RET* gene, to triage annual screening and prophylactic thyroidectomy, change the management of relatives of patients with a known RET mutation when compared with usual clinical surveillance?

#### Linkage 3 – Likely impact of change in patient management from RET mutation testing on patient health outcomes

The key difference in clinical management that may impact on patient health outcomes related to RET mutation testing is the use of prophylactic surgery to remove the thyroid in those at risk of developing an MTC due to a RET mutation. Therefore, the key question regarding patient health outcomes concerns the benefit of prophylactic surgery versus surgery upon clinical confirmation of development of an MTC. The research question was therefore:

5. In patients or family members with a RET mutation, is a prophylactic thyroidectomy and replacement thyroxine as safe and effective as a thryoidectomy and replacement thyroxine performed after calcitonin levels rise, or on the basis of biopsy of a thyroid nodule detected either clinically or on ultrasound?

## Review of literature

### Literature sources and search strategies

The medical literature was searched to identify relevant studies and reviews for the period between 1993 and July–August 2012. Searches were conducted via the electronic databases listed in Table 10.

Table Electronic databases searched

| **Database** | **Period covered** |
| --- | --- |
| Cochrane Library – including, Cochrane Database of Systematic Reviews, Database of Abstracts of Reviews of Effects, the Cochrane Central Register of Controlled Trials (CENTRAL), the Health Technology Assessment Database, the NHS Economic Evaluation Database | 1993 – August 2012 |
| Web of Science – Science Citation Index Expanded | 1993 – July 2012 |
| Current Contents | 1993 – July 2012 |
| Embase.com (including Embase and Medline) | 1993 – July 2012 |
| PubMed | 1993 – July 2012 |
| CINAHL | 1993 – August 2012 |
| EconLit | 1993 – July 2012 |

The search strategy used on the Medline platform through PubMed was as follows:

(("Proto-Oncogene Proteins c-ret/analysis"[Mesh] OR "Proto-Oncogene Proteins c-ret/genetics"[Mesh] OR "Proto-Oncogene Proteins c-ret/physiology"[Mesh]) OR "ret"[All Fields]) AND (MEN2[All Fields] OR "Multiple Endocrine Neoplasia"[All Fields])

Other databases were searched with similar text words and medical subject headings that were relevant to the database.

### Selection criteria

#### Direct evidence

Criteria were pre-specified to determine eligible studies to address the main research questions concerning the diagnostic safety and effectiveness of RET mutation testing. These are outlined in Table 11 and Table 12.

Table 11 Inclusion criteria for identification of studies relevant to assess the safety and effectiveness of RET mutation testing in those suspected of having MEN2

| Characteristic | Criteria | | |
| --- | --- | --- | --- |
| **Study design** | All study designs in the ‘Intervention’ column of Table 19 were included. | | |
| **Population** | Patients suspected of having MEN2 due to: | | |
| An MTC | Adrenal phaeochromocytoma | Hyperparathyroidism plus a diagnosis of MTC or phaeochromocytoma in a close relative |
| **Intervention/test** | RET mutation testing plus clinical investigations for phaeochromocytoma and hyperparathyroidism in those RET M+ | RET mutation testing plus clinical investigations for MTC and hyperparathyroidism in those RET M+, or investigations for other hereditary disorders in those RET M– | RET mutation testing plus clinical investigations for phaeochromocytoma |
| **Comparator** | Clinical investigations for phaeochromocytoma and hyperparathyroidism | Clinical investigations for MTC and hyperparathyroidism, and investigations for other hereditary disorders in those without an MTC | Clinical investigations for MTC and phaeochromocytoma |
| **Outcome** | Safety—psychological and physical harms from genetic testing and clinical screening  Effectiveness—  Primary outcomes: mortality, progression-free survival, quality of life, incidence and severity (TNM stage) of MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism  Secondary outcomes: incidence of symptoms arising from MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism, timing of thyroidectomy, age at diagnosis, rates and implications (physical and psychological) of surveillance | | |
| **Search period** | 1993 – July 2012 | | |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified | | |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

Table 12 Inclusion criteria for identification of studies relevant to assess the safety and effectiveness of RET mutation testing in close relatives

| Characteristic | Criteria | | |
| --- | --- | --- | --- |
| **Study design** | All study designs in the ‘Intervention’ column of Table 19 were included. | | |
| **Population** | Close family members of: | | |
| Those with a pathological *RET* gene mutation | Those suspected of having MEN2 due to MTC | Those suspected of having MEN2 due to phaeochromocytoma or hyperparathyroidism |
| **Intervention/test** | RET mutation testing and lifelong surveillance for those who RET M+ | RET mutation testing | RET mutation testing |
| **Comparator** | Clinical investigations and lifelong surveillance | Clinical investigations and lifelong surveillance | No surveillance for family members of index cases without MTC or RET mutation |
| **Outcome** | Safety—psychological and physical harms from genetic testing and clinical screening  Effectiveness —  Primary outcomes: mortality, progression-free survival, quality of life, incidence and severity (TNM stage) of MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism  Secondary outcomes: incidence of symptoms arising from MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism, timing of thyroidectomy, age at diagnosis, rates and implications (physical and psychological) of surveillance | | |
| **Search period** | 1993 – July 2012 | | |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified | | |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive

#### Linked evidence

In the absence of strong direct evidence, a linked evidence approach was also attempted, where evidence of diagnostic accuracy and change in clinical management and treatment effectiveness are linked to provide an assessment of the effectiveness of RET mutation testing in those suspected of having, or at risk of having, MEN2 disease.

The inclusion criteria for selecting studies for such an assessment are outlined in Table 13 to Table 17.

Table 13 Inclusion criteria for identification of studies relevant to assess the diagnostic accuracy of RET mutation testing in those suspected of having MEN2

| Characteristic | Criteria | | |
| --- | --- | --- | --- |
| **Study design** | All study designs in the ‘Diagnostic accuracy’ column of Table 19 were included. | | |
| **Population** | Patients suspected of having MEN2 due to: | | |
| An MTC | Adrenal phaeochromocytoma | Hyperparathyroidism plus a diagnosis of MTC or phaeochromocytoma in a close relative |
| **Intervention/test** | RET mutation testing plus clinical investigations for phaeochromocytoma and hyperparathyroidism in those RET M+ | RET mutation testing plus clinical investigations for MTC and hyperparathyroidism in those RET M+, or investigations for other hereditary disorders in those RET M– | RET mutation testing plus clinical investigations for phaeochromocytoma |
| **Comparator** | Clinical investigations for phaeochromocytoma and hyperparathyroidism | Clinical investigations for MTC and hyperparathyroidism, and investigations for other hereditary disorders in those without an MTC | Clinical investigations for MTC and phaeochromocytoma |
| **Reference standard** | Lifelong clinical assessment | | |
| **Outcome** | Diagnostic accuracy outcomes: Sensitivity and specificity (and therefore rates of false positives and negatives), positive and negative likelihood ratios, positive and negative predictive values, diagnostic odds ratios, receiver–operator characteristic curves, area under the curve, accuracy | | |
| **Search period** | 1993 – July 2012 | | |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified. | | |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

Table 14 Inclusion criteria for identification of studies relevant to assess the diagnostic accuracy of RET mutation testing in family members of those with a RET mutation or suspected of having MEN2

| Characteristic | Criteria | | |
| --- | --- | --- | --- |
| **Study design** | All study designs in the ‘Diagnostic accuracy’ column of Table 19 were included. | | |
| **Population** | Close family members of: | | |
| Those with a pathological *RET* gene mutation | Those suspected of having MEN2 due to MTC | Those suspected of having MEN2 due to phaeochromocytoma or hyperparathyroidism |
| **Intervention/test** | RET mutation testing and lifelong surveillance for those who are mutation + | RET mutation testing | RET mutation testing |
| **Comparator** | Clinical investigations and lifelong surveillance | Clinical investigations and lifelong surveillance | No surveillance for family members of index cases without MTC or RET mutation |
| **Reference standard** | Lifelong clinical assessment | | |
| **Outcome** | Diagnostic accuracy outcomes: Sensitivity and specificity (and therefore rates of false positives and negatives), positive and negative likelihood ratios, positive and negative predictive values, diagnostic odds ratios, receiver–operator characteristic curves, area under the curve, accuracy | | |
| **Search period** | 1993 – July 2012 | | |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified. | | |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive

Table 15 Inclusion criteria for identification of studies relevant to assess a change in patient management as a result of RET mutation testing in those suspected of having MEN2

| Characteristic | Criteria | | |
| --- | --- | --- | --- |
| **Study design** | All study designs in the ‘Intervention’ column of Table 19 were included. In the event that large numbers of pre-test/post-test case series were identified, all would be reviewed but only those that were large case series and/or with long-term follow-up would have data extracted. | | |
| **Population** | Patients suspected of having MEN2 due to: | | |
|  | An MTC | Adrenal phaeochromocytoma | Hyperparathyroidism plus a diagnosis of MTC or phaeochromocytoma in a close relative |
| **Intervention/test** | RET mutation testing plus clinical investigations for phaeochromocytoma and hyperparathyroidism in those RET M+ | RET mutation testing plus clinical investigations for MTC and hyperparathyroidism in those RET M+, or investigations for other hereditary disorders in those RET M– | RET mutation testing plus clinical investigations for phaeochromocytoma |
| **Comparator** | Clinical investigations for phaeochromocytoma and hyperparathyroidism | Clinical investigations for MTC and hyperparathyroidism, and investigations for other hereditary disorders in those without an MTC | Clinical investigations for MTC and phaeochromocytoma |
| **Outcome** | Rates of treatment, method of treatment, rates of referral, type of referral, hospital separations and re-admissions, hospital length of stay | | |
| **Search period** | 1993 – July 2012 | | |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified. | | |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

Table 16 Inclusion criteria for identification of studies relevant to assess a change in patient management as a result of RET mutation testing in close family members of those with a known RET mutation or suspected of having MEN2

| Characteristic | Criteria | | |
| --- | --- | --- | --- |
| **Study design** | All study designs in the ‘Intervention’ column of Table 19 were included. In the event that large numbers of pre-test/post-test case series were identified, all would be reviewed but only those that were large case series and/or with long-term follow-up would have data extracted. | | |
| **Population** | Close family members of: | | |
|  | Those with a pathological *RET* gene mutation | Those suspected of MEN2 with MTC | Those suspected of MEN2 due to phaeochromocytoma or hyperparathyroidism (without MTC) |
| **Intervention/test** | RET mutation testing and lifelong surveillance for those who are RET M+ | RET mutation testing | RET mutation testing |
| **Comparator** | Clinical investigations and lifelong surveillance | Clinical investigations and lifelong surveillance | Assuming not MEN2, therefore no surveillance for family members of index cases without MTC or RET mutation |
| **Outcome** | Rates of treatment, method of treatment, rates of referral, type of referral, hospital separations and re-admissions, hospital length of stay | | |
| **Search period** | 1993 – July 2012 | | |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified. | | |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive

Although RET mutation testing impacts on the management of a large proportion of both the index cases and their relatives, many of the changes are in relation to ruling out the need for surveillance strategies, rather than concerning different treatment strategies, and so would not alter patient health outcomes. The only change in clinical management that could impact on the health of patients is the effect of prophylactic total thyroidectomy (either in the index case presenting with features of MEN2 without MTC) and ongoing surveillance for an MTC, compared with the previous management involving a total thyroidectomy in response to rising levels of calcitonin or on the basis of biopsy results from a thyroid nodule detected clinically or through ultrasound.

Table 17 Inclusion criteria for identification of studies relevant to assess treatment effectiveness following a change in patient management as a result of RET mutation testing

| Characteristic | Criteria |
| --- | --- |
| **Study design** | Level I, II and III-1 evidence listed in the Invention column of Table 19 were included. Should there be sufficient good-quality evidence available from higher level evidence for each type of treatment intervention, then lower level evidence would be reviewed but data would not be extracted. |
| **Population** | Index case: Patients with a pathogenic RET mutation who present with adrenal phaeochromocytoma or hyperparathyroidism, plus a diagnosis of MTC or phaeochromocytoma in a close relative, who do not have an MTC upon presentation  Relatives: Family members with a pathological *RET* gene mutation |
| **Intervention/test** | Index case: Treatment for presenting symptom plus a prophylactic total thyroidectomy, lifelong thyroxine and surveillance  Relatives: A prophylactic total thyroidectomy, lifelong thyroxine and surveillance |
| **Comparator** | Index case: Treatment for presenting symptom plus lifelong surveillance, and a total thyroidectomy once calcitonin levels rise, or on basis of biopsy of a thyroid nodule detected clinically or through ultrasound, plus lifelong thyroxine and surveillance  Relatives: A total thyroidectomy once calcitonin levels rise, or on basis of biopsy of a thyroid nodule detected clinically or through ultrasound, plus lifelong thyroxine and surveillance |
| **Outcome** | *Effectiveness:*  **Primary outcomes:** mortality, progression free survival, quality of life, incidence and severity (TNM stage) of MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism  **Secondary outcomes:** incidence of symptoms arising from MTC, phaeochromocytoma, parathyroid hyperplasia/neoplasia and hyperparathyroidism, timing of thyroidectomy, age at diagnosis, rates and implications (physical and psychological) of surveillance  *Safety:* Psychological and physical harms from testing |
| **Search period** | 1993 – July 2012 |
| **Language** | Non-English language articles were excluded unless they provided a higher level of evidence than the English language articles identified. |

MTC = medullary thyroid carcinoma; TNM Classification of Malignant Tumours is a cancer staging system, whereby T describes the size of the primary tumour, N the involvement of regional lymph nodes, and M the presence of distant metastasis

### Search results

Figure 6 details the steps undertaken to select studies eligible for this assessment, following screening against the inclusion criteria. Studies were selected by one assessor and reviewed by another assessor when inclusion was in doubt. Final selection was a consensus decision.

#### Prisma flowchart

Flowchart oulining how many potential articles there were at different points of the systematic review. Initially 8008 articles were identified, of which 2376 remained after removing duplicates. 493 were throught relevant on title/abstract and had the full text retrieved. 135 were finally included. 

Figure Summary of the process used to identify and select studies for the review

Source: adapted from (Liberati et al. 2009)

The study profiles of all included studies are shown in Table 97, to Table 100 in Appendix L. Full text articles that did not meet the inclusion criteria are listed in Appendix M according to the reason for exclusion.

## Data extraction and analysis

### Appraisal of the evidence

Studies included in the evidence base were appraised in three stages:

Stage 1: Appraisal of the applicability and quality (strength of the evidence) of individual studies included in the assessment.

Stage 2: Appraisal of the precision, size of effect and clinical importance of the results for primary outcomes in individual studies—used to determine the safety and effectiveness of RET mutation testing.

Stage 3: Integration of this evidence for conclusions about the net clinical benefit of RET mutation testing in the context of Australian clinical practice.

#### Stage 1: Strength of the evidence

The evidence presented in the selected studies was assessed and classified using the dimensions of evidence defined by the National Health and Medical Research Council (NHMRC) (NHMRC, 2000).

These dimensions (Table 18) consider important aspects of the evidence supporting a particular intervention and include three main domains: strength of the evidence, size of the effect and relevance of the evidence. The first domain is derived directly from the literature identified as informing a particular intervention. The last two require expert clinical input as part of the determination.

Table Evidence dimensions

| Type of evidence | Definition |
| --- | --- |
| Strength of the evidence:  Level  Quality  Statistical precision | The study design used, as an indicator of the degree to which bias has been eliminated by designa  The methods used by investigators to minimise bias within a study design  The *p*-value or, alternatively, the precision of the estimate of the effect. It reflects the degree of certainty about the existence of a true effect |
| Size of effect | The distance of the study estimate from the ‘null’ value and the inclusion of only clinically important effects in the confidence interval |
| Relevance of evidence | The usefulness of the evidence in clinical practice, particularly the appropriateness of the outcome measures used |

a See Table 19

The three sub-domains (level, quality and statistical precision) are collectively a measure of the strength of the evidence.

The ‘level of evidence’ reflects the effectiveness of a study design to answer a particular research question. Effectiveness is based on the probability that the design of the study has reduced or eliminated the impact of bias on the results. The NHMRC evidence hierarchy provides a ranking of various study designs (‘levels of evidence’) by the type of research question being addressed (Table 19).

Table Designations of levels of evidence for interventions and studies of diagnostic accuracy (including table notes) (NHMRC 2008, 2009a)

| **Level** | **Interventiona** | **Diagnostic accuracyb** |
| --- | --- | --- |
| **I**c | A systematic review of level II studies | A systematic review of level II studies |
| **II** | A randomised controlled trial | A study of test accuracy with: an independent, blinded comparison with a valid reference standardd among consecutive persons with a defined clinical presentation6 |
| **III-1** | A pseudorandomised controlled trial (i.e. alternate allocation or some other method) | A study of test accuracy with: an independent, blinded comparison with a valid reference standardd among non-consecutive persons with a defined clinical presentatione |
| **III-2** | A comparative study with concurrent controls:  ▪ non-randomised, experimental trialf  ▪ cohort study  ▪ case-control study  ▪ interrupted time series with a control group | A comparison with reference standard that does not meet the criteria required for Level II and III-1 evidence |
| **III-3** | A comparative study without concurrent controls:  ▪ historical controlled study  ▪ two or more single arm studyg  ▪ interrupted time series without a parallel control group | A diagnostic case-control studye |
| **IV** | A case series with either post-test or pre-test/post-test outcomes | A study of diagnostic yield (no reference standard)h |

Sources: (NHMRC 2008, 2009a)

### Explanatory notes

a Definitions of these study designs are provided in (NHMRC 2000; pp. 7–8).

b These levels of evidence apply only to studies of assessing the accuracy of diagnostic or screening tests. To assess the overall effectiveness of a diagnostic test, there also needs to be a consideration of the impact of the test on patient management and health outcomes (MSAC 2005; Sackett and Haynes 2002). The evidence hierarchy given in the ‘Intervention’ column should be used when assessing the impact of a diagnostic test on health outcomes relative to an existing method of diagnosis/comparator test(s). The evidence hierarchy given in the ‘Screening’ column should be used when assessing the impact of a screening test on health outcomes relative to no screening or alternative screening methods.

c A systematic review will only be assigned a level of evidence as high as the studies it contains, excepting where those studies are of level II evidence. Systematic reviews of level II evidence provide more data than the individual studies and any meta-analyses will increase the precision of the overall results, reducing the likelihood that the results are affected by chance. Systematic reviews of lower level evidence present results of likely poor internal validity and thus are rated on the likelihood that the results have been affected by bias, rather than whether the systematic review itself is of good quality. Systematic review *quality* should be assessed separately. A systematic review should consist of at least 2 studies. In systematic reviews that include different study designs, the overall level of evidence should relate to each individual outcome/result, as different studies (and study designs) might contribute to each different outcome.

d The validity of the reference standard should be determined in the context of the disease under review. Criteria for determining the validity of the reference standard should be pre-specified. This can include the choice of the reference standard(s) and its timing in relation to the index test. The validity of the reference standard can be determined through quality appraisal of the study (Whiting et al. 2003).

e Well-designed population-based case-control studies (e.g. population-based screening studies where test accuracy is assessed on all cases, with a random sample of controls) do capture a population with a representative spectrum of disease and thus fulfil the requirements for a valid assembly of patients. However, in some cases the population assembled is not representative of the use of the test in practice. In diagnostic case-control studies a selected sample of patients already known to have the disease is compared with a separate group of normal/healthy people known to be free of the disease. In this situation patients with borderline or mild expressions of the disease, and conditions mimicking the disease, are excluded, which can lead to exaggeration of both sensitivity and specificity. This is called spectrum bias or spectrum effect because the spectrum of study participants will not be representative of patients seen in practice (Mulherin and Miller 2002).

f This also includes controlled before-and-after (pre-test/post-test) studies, as well as adjusted indirect comparisons (i.e. using A vs. B and B vs. C to determine A vs. C with statistical adjustment for B).

g Comparing single arm studies, i.e. case series from 2 studies. This would also include unadjusted indirect comparisons (i.e. using A vs. B and B vs. C to determine A vs. C but where there is no statistical adjustment for B).

h Studies of diagnostic yield provide the yield of diagnosed patients, as determined by an index test, without confirmation of the accuracy of this diagnosis by a reference standard. These may be the only alternative when there is no reliable reference standard.

Note A: Assessment of comparative harms/safety should occur according to the hierarchy presented for each of the research questions, with the proviso that this assessment occurs within the context of the topic being assessed. Some harms (and other outcomes) are rare and cannot feasibly be captured within randomised controlled trials, in which case lower levels of evidence may be the only type of evidence that is practically achievable; physical harms and psychological harms may need to be addressed by different study designs; harms from diagnostic testing include the likelihood of false positive and false negative results; harms from screening include the likelihood of false alarm and false reassurance results.

Note B: When a level of evidence is attributed in the text of a document, it should also be framed according to its corresponding research question, e.g. level II intervention evidence; level IV diagnostic evidence; level III-2 prognostic evidence.

Note C: Each individual study that is attributed a ‘level of evidence’ should be rigorously appraised using validated or commonly used checklists or appraisal tools to ensure that factors other than study design have not affected the validity of the results.

Source:Hierarchies adapted and modified from: (Bandolier 1999; Lijmer et al. 1999; NHMRC 2009a; Philips et al. 2001)

In terms of assessing the quality of the identified studies, it was planned that studies assessing test performance (diagnostic accuracy) would be graded according to pre-specified quality and applicability criteria using the QUADAS-2 tool (Whiting et al. 2011). However, no test performance studies were identified that compared RET mutation testing accuracy against the reference standard of lifelong clinical assessment. Many diagnostic yield studies were found, but these do not allow independent confirmation of a RET mutation that has been identified. Given the lack of comparator, these studies do not provide useful information on test performance and would universally rate poorly using the QUADAS-2 tool, and so their quality was not formally evaluated.

The appraisal of historical controlled studies and cohort studies was conducted using the modified checklist by Downs and Black (Downs & Black 1998). Studies were considered to have a high risk of bias if they scored ≤17, a moderate risk if they scored >18 and ≤21, and a low risk if they scored >22, out of 26.

The appraisal of uncontrolled before-and-after case series was assessed according to a checklist developed by the UK National Health Service (NHS) Centre for Reviews and Dissemination (Khan 2001). The six questions were scored 0–1 and summed to give an estimate of study quality: ≤2 = poor quality; > 2 & ≤ 4 = moderate quality; > 4 = high quality.

#### Stage 2: Precision, size of effect and clinical importance

Statistical precision was determined using statistical principles. Small confidence intervals and p-values give an indication as to the probability that the reported effect is real and not attributable to chance (NHMRC 2000). Studies needed to be appropriately powered to ensure that a real difference between groups would be detected in the statistical analysis. In the available direct evidence that compared RET mutation testing versus no testing, the size of the difference in effect (relative or absolute) and corresponding confidence intervals were inspected to determine whether the observed differences were clinically important.

Relevance of the evidence

The outcomes being measured in this report are clinically relevant, particularly for the direct evidence. However, the use of the supporting linked evidence approach means that clinically relevant outcomes could not be provided for some of the linkages (notably test performance). Inadequately validated (predictive) surrogate measures of a clinically relevant outcome should be avoided (NHMRC 2000).

#### Stage 3: Assessment of the body of evidence

Appraisal of the body of evidence (i.e. all the individual studies identified to address each clinical question) was conducted along the lines suggested by the NHMRC in their guidance on clinical practice guideline development (NHMRC 2008). Five components are considered essential by the NHMRC when judging the body of evidence:

1. the evidence base—which includes the number of studies sorted by their methodological quality
2. the consistency of the study results—whether the better quality studies had results of a similar magnitude and in the same direction, i.e. homogeneous or heterogeneous findings
3. the potential clinical impact—appraisal of the precision, size and clinical importance or relevance of the primary outcomes used to determine the safety and effectiveness of the test
4. the generalisability of the evidence to the target Medicare population
5. the applicability of the evidence—whether the studies delivered RET mutation testing in a similar manner to how it will be delivered in Australian clinical practice.

A matrix for assessing the body of evidence for each research question, according to the components above, was used for this assessment (Table 20) (NHMRC 2008).

Table Body of evidence matrix

| **Component** | **A** | **B** | **C** | **D** |
| --- | --- | --- | --- | --- |
| **Excellent** | **Good** | **Satisfactory** | **Poor** |
| **Evidence basea** | One or more level I studies with a low risk of bias or several level II studies with a low risk of bias | One or two level II studies with a low risk of bias or an SR / several level III studies with a low risk of bias | One or two level III studies with a low risk of bias, or level I or II studies with a moderate risk of bias | Level IV studies, or level I to III studies / SRs with a high risk of bias |
| **Consistencyb** | All studies consistent | Most studies consistent and inconsistency may be explained | Some inconsistency reflecting genuine uncertainty around clinical question | Evidence is inconsistent |
| **Clinical impact** | Very large | Substantial | Moderate | Slight or restricted |
| **Generalisability** | Population(s) studied in body of evidence are the same as the target population | Population(s) studied in the body of evidence are similar to the target population | Population(s) studied in the body of evidence differ to target population for guideline but it is clinically sensible to apply this evidence to target populationc | Population(s) studied in the body of evidence differ to target population and it is hard to judge whether it is sensible to generalise to target population |
| **Applicability** | Directly applicable to Australian healthcare context | Applicable to Australian healthcare context with few caveats | Probably applicable to Australian healthcare context with some caveats | Not applicable to Australian healthcare context |

SR = systematic review; several = more than 2 studies

a Level of evidence determined from the NHMRC evidence hierarchy—Table 19

b If there is only 1 study, rank this component as ‘not applicable’.

cFor example, results in adults that are clinically sensible to apply to children OR psychosocial outcomes for one cancer that may be applicable to patients with another cancer

Source: adapted from (NHMRC 2008)

After each of the five components in the matrix are rated, the overall assessment of the body of evidence is integrated and conclusions are drawn regarding the net clinical benefit of RET mutation testing in the context of Australian clinical practice.

## Expert advice

The HESP has been established as a panel of MSAC and is a pool of experts collated from various medical fields who are nominated by their associated professional body or by applicants. HESP members are engaged to provide practical, professional advice to evaluators that directly relates to each application and the service being proposed for the MBS. HESP members are not members of either MSAC or its subcommittees—the Evaluation Sub-Committee (ESC) and the PASC. Their role is limited to providing input and guidance to the assessment groups to ensure that the pathway is clinically relevant and takes into account consumer interests. HESP members’ advice is to inform the deliberations that MSAC presents to the Minister.

# Results of assessment

## Is it safe?

| Summary of safety |
| --- |
| No studies mentioned any safety concerns regarding RET mutation testing or surveillance for features of MEN2.  One historical controlled study (level III-3 interventional evidence) reported one death from surgical complications in the pre-RET mutation testing era, compared with one death from surgical complications after diagnosis by RET mutation testing, with similar numbers of patients treated in both scenarios (n=29 in pre-RET mutation testing era; n=31 in RET mutation testing era). No further details on the nature of these deaths or information on confounding factors were reported.  Twelve case series (level IV interventional evidence) reported on rates of adverse events due to total thyroidectomy, performed after RET mutation testing identified the patients as having (k=2), or being at risk of having (k=10), MEN2. Transient hypoparathyroidism was the most commonly reported adverse event, mentioned in 8 studies, with rates between 5.0% and 36.4%. Permanent hypoparathyroidism occurred in up to 13.6% of patients. Temporary laryngeal nerve palsy occurred in 4.5–5.9% of patients, and one case of permanent laryngeal nerve palsy was reported (1.3% of 1 study).  Other complications following total thyroidectomy were one case of arterial bleeding requiring re-operation, one case of permanent unilateral Horner’s syndrome, and one paediatric case with fluctuating thyroid function test results despite good thyroxine replacement compliance at 1-year follow-up. |

There were no studies identified that provided data on any psychological or physical harms from RET mutation testing plus clinical screening, as per the inclusion criteria outlined in Table 11 and Table 12. However, one of the consequences of the introduction of RET mutation testing was that more patients had prophylactic total thyroidectomies, prior to clinical evidence of disease, and this could result in harms. One historical controlled study (level III-3 interventional evidence) and 12 case series (level IV evidence) provided evidence on the rates of adverse events following total thyroidectomies. These studies are outlined in Table 21 and Table 25.

The most serious complication following total thyroidectomy was death, which was reported in two patients, one of whom was diagnosed with RET mutation testing, and the other was based on clinical testing (Table 21) (Diaz & Wohllk 2012). The specific surgical complications leading to these deaths were not reported. The surgical techniques employed with those who died from surgical complications, and the stage of disease of these patients, was not mentioned. It is therefore unclear whether RET mutation testing affects the likelihood of surgical complications.

Table Deaths following total thyroidectomy

| Study and location | Level of evidence and quality assessment | Study population | Pre-RET mutation testing era | RET mutation testing era |
| --- | --- | --- | --- | --- |
| (Diaz & Wohllk 2012)  Chile | III-3 interventional evidence  High risk of bias (9/26) | N=60 MEN2 patients who underwent total thyroidectomy | n=29 | n=31 |
| 13/29 (44.8%):  3 from hypertensive crisis  1 from surgical complications  9 from distant metastases | 1/31 (3.2%):  Died from surgical complications |

Twelve case series reported on the rates of adverse events following RET mutation testing and subsequent total thyroidectomies (Table 22). The most common adverse event related to total thyroidectomy surgery was transient hypoparathyroidism or hypocalcaemia, requiring calcium substitution postoperatively. Eight studies mentioned this outcome, which occurred in 5.0–36.4% of patients (Table 22). Permanent hypoparathyroidism was identified in 4 studies, with between 5.9% and 13.6% of patients requiring ongoing calcium substitution (Dralle et al. 1998; Lau et al. 2009; Schellhaas et al. 2009; Spinelli et al. 2010).

Laryngeal nerve palsy was reported in 5 studies, with transient laryngeal nerve palsy reported in 4.5–5.9% of patients, and permanent laryngeal nerve palsy reported in one patient (1.3%) (Dralle et al. 1998; Gimm et al. 2002; Lau et al. 2009; Rodriguez Gonzalez et al. 2002; Schellhaas et al. 2009).

Three other complications were reported in individual patients. One patient had arterial bleeding that required re-operation the day after the thyroidectomy (Schellhaas et al. 2009), one patient developed permanent unilateral Horner’s syndrome (Heizmann et al. 2006), and one paediatric patient had fluctuating thyroid function test results despite good thyroxine replacement compliance at 1-year follow-up (Lau et al. 2009).

It is unclear how these complication rates would compare with surgery performed after clinical signs of MTC are detected, and this cannot be determined given the non-comparative nature of the studies identified.

**Table 22 Adverse events following total thyroidectomy**

| Study | Study design and quality appraisal | Population | Adverse events |
| --- | --- | --- | --- |
| In patients with MTC | | | |
| (Spinelli et al. 2010)  Italy | IV interventional evidence  Moderate quality (4/6) | N=13 children with MEN2 who underwent surgery for MTC:  7 (54%) MEN2A  4 (31%) FMTC  2 (15%) MEN2B | Permanent  1/13 (7.7%) developed postoperative permanent hypocalcaemia  All other patients showed no sequelae related to surgery |
| (Bihan et al. 2012)  France | IV interventional evidence  Poor quality (2/6) | N=5 members of an MTC family with a RET L790F mutation who had abnormal pentagastrin-stimulated calcitonin levels and a thyroidectomy (including index patient)  3 had clinical signs of disease | Transient  1/5 (20%) developed temporary hypoparathyroidism |
| In asymptomatic family members | | | |
| (Lau et al. 2009)  Hong Kong | IV interventional evidence  High quality (5/6) | N=22 asymptomatic relatives from 8 MEN2A families, who underwent prophylactic total thyroidectomy based on RET mutation status | Transient  8/22 (36.4%) temporary hypoparathyroidism requiring short-term calcium or vitamin D supplementation  1/17 (5.9%) transient recurrent nerve palsy (at follow-up, laryngeal nerve function described as normal)  1/17(5.9%) had arterial bleeding, requiring re-operation the day after thyroidectomy  Permanent  3/22 (13.6%) had permanent hypoparathyroidism requiring oral calcium and vitamin D supplementation (median 49 months follow-up)  1/22 (4.5%) paediatric patient had fluctuating thyroid function test results despite good thyroxine replacement compliance (at 1-year follow-up) |
| (Wells Jr & Skinner 1998)  USA | IV interventional evidence  High quality (5/6) | N=18 RET M+ first-degree relatives from 7 MEN2A kindreds with no clinical symptoms of disease, who had a thyroidectomy | 0/18 (0%) had surgical complications from total thyroidectomy |
| (Schellhaas et al. 2009)  Germany | IV interventional evidence  High quality (5/6) | N=17 patients with mutation in codon 634 operated on prophylactically  (14 from MEN2A  3 with apparent familial MTC) | 9/17 (52.9%) no perioperative complications  Transient  5/17 (29.4%) temporary hypoparathyroidism (none required calcium or vitamin D supplementation at long-term follow-up)  1/17 (5.9%) transient recurrent nerve palsy (at follow-up, laryngeal nerve function described as normal)  1/17(5.9%) had arterial bleeding, requiring re-operation the day after thyroidectomy  Permanent  1/17 (5.9%) had permanent hypoparathyroidism requiring oral calcium supplementation (15 years follow-up) |
| (Dralle et al. 1998)  Germany | IV interventional evidence  Moderate quality (4/6) | N=75 RET M+ patients <20 years of age who have undergone a prophylactic total thyroidectomy  57 underwent additional lymph node dissections | Transient  20/75 (26.7%) transient hypoparathyroidism  4/75 (5.3%) laryngeal nerve palsy  Permanent  5/75 (6.7%) hypoparathyroidism  3/75 (4%) persistent hypercalcitoninaemia  1/75 (1.3%) laryngeal nerve palsy |
| (Gimm et al. 2002)  Germany, Austria | IV Interventional evidence  Moderate quality (4/6) | N=40 patients with RET codon 790/791 mutations who underwent thyroid operations  22 thyroidectomy with cervicocentral lymph node dissection  8 thyroidectomy with lymph node dissection extending beyond cervicocentral compartment | Transient  2/40 (5%) transient hypoparathyroidism  2/40 (5%) laryngeal nerve palsy  Permanent  No permanent complications for any patients |
| (Heizmann et al. 2006)  Switzerland | IV interventional evidence  Moderate quality (4/6) | N=14 RET M+ patients who were presymptomatic, from 2 MEN2A kindreds | Transient  All patients received oral calcium substitution for several days postoperatively  2/14 (14.3%) required intravenous calcium substitution for 2 days  Permanent  1/14 (7.1%) had permanent unilateral Horner’s syndrome (63-year-old)  No recurrent nerve palsy or persistent hypocalcaemia |
| (Decker et al. 1996)  USA | IV Interventional evidence  Moderate quality (4/6) | N=11 RET M+ children who underwent prophylactic thyroidectomy | Transient  1/11(9.1%) children required transient calcium and vitamin D replacement postoperatively  Permanent  No wound or recurrent nerve complications |
| (Frank-Raue et al. 1996)  Germany | IV Interventional evidence  Moderate quality (4/6) | N=9 patients identified as RET M+ undergoing a prophylactic thyroidectomy | Permanent  No recurrent laryngeal nerve damage or hypoparathyroidism |
| (Rodriguez Gonzalez et al. 2002)  Spain | IV interventional evidence  Moderate quality (3/6) | N=22 RET M+ asymptomatic MEN2A patients with normal basal and pentagastrin-stimulated calcitonin levels who received a prophylactic thyroidectomy | Transient  2/22 (9.1%) transitory hypoparathyroidism, treated with calcium for 1 and 3 months (both had central neck dissection)  1/22 (4.5%) with transitory unilateral recurrent nerve injury, which improved 1 month after operation with normal laryngoscopy |
| (Yoshida et al. 2009)  Japan | IV interventional evidence  Moderate quality (3/6) | N=12 adults who underwent total thyroidectomy for MTC and had MEN2 | 0/12 (0%) patients experienced morbidity after surgery |

FMTC = familial medullary thyroid carcinoma; MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive

## Is it effective?

### Direct evidence

Summary of effectiveness

Nine historical controlled studies (level III-3 interventional studies), with a high risk of bias and confounding, reported on health outcomes in cohorts of patients who were diagnosed either prior to the introduction of RET mutation testing, or after the addition of RET mutation testing.

The studies consistently showed lower rates of disease persistence or recurrence after total thyroidectomy in RET-mutation-positive patients whose mutation status was known prior to surgery, compared with those whose mutation status was unknown when surgical decisions were being made. However, this outcome is confounded by the fact that the historical cohorts had longer follow-up periods than the recent cohorts, and the rates of persistence/recurrence were not adjusted for length of follow-up.

Those who were diagnosed with the addition of RET mutation testing had a lower risk of MTC at the time of undergoing total thyroidectomy than those who were treated before the availability of RET mutation testing. Lower severity of disease at the time of surgery allows less invasive surgical procedures to be performed. It is therefore expected that patient outcomes would be better since the introduction of RET mutation testing, although this could not be confirmed as unconfounded long-term data are not available.

Age at diagnosis was found to be lower in one historical controlled study (level III-3 interventional study) for patients with MEN2A and FMTC since the introduction of RET mutation testing.

Over 50 uncontrolled studies, many of small sample size, provided additional data on the outcomes mentioned above. Results were often heterogeneous because of the different populations and circumstances studied, but were largely consistent with the findings observed in patients receiving RET mutation testing in the historical controlled studies. The only exception was that mortality rates following RET mutation testing were higher in the uncontrolled studies than in the controlled study, but this was likely due to their longer lengths of follow-up. The mortality rates following RET mutation testing in the uncontrolled studies were still lower than the rates reported for the pre-RET mutation testing era in the historical controlled study.

The diagnostic effectiveness of RET mutation testing was assessed using studies meeting the inclusion criteria given in Table 11 and Table 12. Nine historical controlled studies were identified. The majority (8/9) were considered to be at high risk of bias, with 1 considered to have a moderate risk of bias. As those patients who were included in the comparator arms (pre-RET mutation testing) were from a different time period than those in the intervention arms (with RET mutation testing available), it is possible that the observed results were due to confounding factors, for example changes in surgical techniques or screening procedures over time. Another flaw in the included studies was that the health outcomes of those who were determined not to have MEN2 were often not reported. It is therefore possible that cases of MEN2 were missed through the diagnostic and screening process, but no discussion of the clinically negative patients or those with wildtype RET were generally included.

#### Mortality

Seven studies reported on the rate of death following RET mutation testing and subsequent treatments, compared with a pre-RET mutation testing scenario (k=1 historical controlled study) or without a comparator arm (k=10 case series). The historical controlled study was in MEN2 patients who had undergone total thyroidectomy (Table 23). Of those patients diagnosed and treated without knowledge of their RET mutation status, 31% died from distant metastases, compared with no patients diagnosed since the use of RET mutation testing. This study had a high risk of bias, as the use of historical controls meant that length of follow-up was inevitably different between the two study arms. The length of follow-up was not stated but, as it was reported that RET mutation testing was initiated in 1997, the maximum follow-up in the study for those diagnosed with RET mutation testing would be 5 years. It is unknown at what stage of disease patients commonly had a total thyroidectomy in either scenario. While this study may be hypothesis-generating, the lack of details in the article and the high risk of bias inherent in the study design means that no strong conclusions can be made regarding the risk of mortality following RET mutation testing, compared with diagnosis without RET mutation testing.

Table 23 Comparative mortality in pre-RET mutation testing era and RET mutation testing era

| Study and location | Level of evidence and quality of assessment | Study population | Pre-RET mutation testing era | RET mutation testing era |
| --- | --- | --- | --- | --- |
| (Diaz & Wohllk 2012)  Chile | III-3 interventional evidence  High risk of bias (9/26) | N=60 MEN2 patients who underwent total thyroidectomy | n=29 | n=31 |
| 13/29 (44.8%):  3 from hypertensive crisis 1 from surgical complications 9 from distant metastases | 1/31 (3.2%):  Died from surgical complications |

Ten case series reported on deaths after RET mutation testing (level IV interventional evidence) (Table 24). The studies are ordered by risk of bias, then by sample size. These studies were very heterogeneous in regards to their populations and description of outcomes. Most studies reported that deaths were related to MTCs, whereas 1 study reported that half of all deaths were due to phaeochromocytomas.

Table 24 Mortality following RET mutation testing and subsequent treatments

| Study and location | Level of evidence | Study population | Intervention | Mortality |
| --- | --- | --- | --- | --- |
| (Gonzalez et al. 2003)  Mexico | IV Interventional evidence  High quality (5/6) | N=17 RET M+ patients who had MTC or CCH  14 MEN2A  3 MEN2B (all 3 symptomatic)  11 had thyroidectomy | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16, and direct DNA sequencing of exons 13–15  Clinical screening  Thyroidectomy | 2/17 (9.5%) died (mean follow-up 6.7 years, range 1–24 years):  1 patient had increased calcitonin prior to death at age 20 years, suggesting tumour activity/metastases  1 patient died from skeletal metastases detected |
| (Milos et al. 2008)  Worldwide (Romania, Germany, Chile, Brazil, Argentina, Hungary, Spain, The Netherlands, Czech Republic, Poland, USA) | IV interventional evidence  Moderate quality (4/6) | N=92 carriers of RET C634W mutation from 20 unrelated MEN2A families  81 had thyroidectomy  68 had MTC  7 had CCH | RET mutation testing (method not stated)  Thyroidectomy | 18/92 (19.6%) died (mean follow-up 12 years, range 1–29 years)  Mean age of death 41 years  Cause of death unknown in 2 cases  PCC dominant cause in 8/16 (mean age 42 years, range 18–67 years)  Metastatic MTC cause of death in 4/16 (ages 21, 29, 69 years):  2 died from myocardial infarction  1 died from lung cancer  1 died from an accident |
| (Frohnauer et al. 2000)  USA | IV Interventional evidence  Moderate quality (4/6) | N=23 members from 5 MEN2A kindreds who had a RET codon 804 mutation  14 had a thyroidectomy | RET mutation testing by denaturing gradient gel electrophoresis analysis, confirmed by direct DNA sequencing of exon 14  Thyroidectomy | 1/23 (4.3%) died from widespread metastases at age 12 years, 6 years after diagnosis and thyroidectomy |
| (Punales et al. 2003)  Brazil | IV interventional evidence  Moderate quality (3/6) | N=50 index cases and family members with RET codon 634 mutation, who underwent surgery  43 had clinical disease  7 were clinically asymptomatic gene carriers | RET mutation testing by single-strand conformational polymorphism analysis, restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15  Total thyroidectomy, with a central cervical lymph node dissection in those with increased calcitonin levels | 7/50 (14%) died of MTC, all of whom had disseminated disease at diagnosis Length of follow-up not stated |
| (Patocs et al. 2006)  Hungary | IV interventional evidence  Moderate quality (3/6) | N=40 patients from 18 families who had had a thyroidectomy due to hereditary MTC or CCH:  33 MEN2A  1 MEN2B  6 MTC families without PCC or hyperparathyroidism | RET mutation testing by single-strand conformation polymorphism analysis, restriction enzyme digestion, and direct DNA sequencing of exons 10, 11, 13, 14 and, in MEN2B phenotype, exons 15 and 16 | 2/40 (5%) deaths from MTC  C609S: 0/3 C609Y: 0/2 C634F: 1/9 C634Y: 0/9 C634S: 0/4 C634R: 0/2 C634W: 0/2 V804M: 1/4 V804L: 0/4 M918T: 0/1 |
| (Quayle et al. 2004)  USA | IV interventional evidence  Moderate quality (3/6) | N=39 patients with MEN2 or FMTC diagnosed when over 50 years of age:  36 MEN2A  3 FMTC  Mutations:  5 RET codon 608  15 RET codon 618  6 RET codon 620  12 RET codon 634  1 unknown | RET mutation testing (method not stated) | 5-year MTC specific survival: 87%  10-year MTC specific survival: 83%  Overall 5-year survival: 86%  Overall 15-year survival: 74%  When older patients were compared with younger patients, the differences in MTC-specific survival (p=0.04) and overall survival (p>0.001) were statistically significant  Stage-specific survival was similar in both groups  3/39 (7.7%) patients had distant metastases occurring in the liver and bone (n=1), liver, lung and kidney (n=1), and liver and skin (n=1)  All 3 died MTC-specific deaths |
| (Lips et al. 1994)  The Netherlands | IV interventional evidence  Moderate quality (3/6) | N=14 symptomatic RET M+ members from 4 large MEN2A families | MEN2 diagnosed by linkage analysis until June 1993  RET mutation testing by direct DNA sequencing of exons 10 and 11 | 3/14 (21.4%) died from MTC  2/14 (14.3%) died from PCC  1/14 (7.1%) died from an unrelated myocardial infarction |
| (Yoshida et al. 2009)  Japan | IV interventional evidence  Moderate quality (3/6) | N=12 adults who underwent total thyroidectomy for MTC and had MEN2 | RET mutation testing (method not stated)  Total thyroidectomy (unclear whether treatment decisions influenced by RET mutation) | 1/12 (8.3%) patients had surgery that was not considered curative, and died of advanced metastatic MTC at 1 year postoperatively |
| (Jung et al. 2010)  Korea | IV interventional evidence  Moderate quality (3/6) | N=8 RET M+ members of a 3-generation FMTC family (including index case) underwent total thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 of the index case  Analysis of exon 10 in family members  Total thyroidectomy with either central neck dissection or modified radical neck dissection | 0/8 (0%) patients died during median 10-year follow-up period |
| (Vaclavikova et al. 2009)  Czech Republic | IV interventional evidence  Low quality (2/6) | N=10 index cases with a RET Y791F mutation:  3 with apparently sporadic MTC 3 with FMTC/MEN2A/MEN2B 1 with PCC 3 with HSCR  N=21 RET M+ family members | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 3/31 (9.7%) deaths during 15-year follow-up period |

CCH = C-cell hyperplasia; FMTC = familial MTC; HSCR = Hirschsprung’s disease; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

#### Progression-free survival

Six historical controlled studies (level III-3 interventional evidence) reported on the percentage of patients who were disease free or who had residual or recurrent disease following total thyroidectomy (Table 25). The majority of these studies were considered to have a high risk of bias, as confounding factors (such as surgical or screening techniques and lengths of follow-up varying between study arms) could not be ruled out. These studies are presented in Figure 7. The 6 studies were consistent in the direction of effect, indicating that fewer patients who had been diagnosed with RET mutation testing subsequently had residual disease, recurrent disease or died, compared with those who were diagnosed without knowledge of RET mutations. However, as all these studies were historical controlled studies, it is possible that a component of these findings is due to longer follow-up in those who were diagnosed in the earlier time period (before RET mutation testing). The more time that has passed since diagnosis, the greater the proportion of patients who cumulatively have further signs of disease. Skinner et al. (1996) reported that the four patients who had recurrence had it detected at a mean of 10.5 years (range 5–18 years) follow-up. Assuming that recurrence would occur at a similar time-point for those who had undergone diagnosis including RET mutation testing, the mean follow-up of 1.3 years in the RET mutation testing era was too short to conclude if patients were likely to develop recurrence or not. Likewise, Lallier et al. (1998) reported a case of recurrence at 5 years post-thyroidectomy.

Table Persistence or recurrence after total thyroidectomy in the pre-RET mutation testing era and RET mutation testing era

| Study and location | Level of evidence and quality assessment | Study population | Outcome measure | Pre-RET mutation testing era | RET mutation testing era | **Follow-up** |
| --- | --- | --- | --- | --- | --- | --- |
| (Rohmer et al. 2011)  France | III-3 interventional evidence  Moderate risk of bias (18/26) | N=170 patients with a RET mutation who underwent a total thyroidectomy younger than 21 years of age:  109 MEN2A  24 MEN2B  37 FMTC | RET M+ | n=38 | n=132 |  |
| Disease-free patients | 23/37 (62.1%) | 117/129 (90.6%) | Median 5.8 years  (range 0.01–28.7 years) |
| (Schreinemakers et al. 2010)  Sweden | III-3 interventional evidence  High risk of bias (17/26) | N=93 patients with a RET mutation who underwent a total thyroidectomy younger than 20 years of age | RET M+ | n=25 | n=68 |  |
| Residual or recurrent disease at follow-up | 7/18 (38.8%) | 9/42 (21.4%) | Median 7 years (IQR 3, 11 years) |
| (Lallier et al. 1998)  Canada | III-3 interventional evidence  High risk of bias (15/26) | N=13 MEN2 patients (children) who underwent total thyroidectomy between 1981 and 1997  with RET mutation testing: 5 had codon 620 mutation 1 had codon 643 mutation  without RET mutation testing: (codons unknown) | Clinically positive / RET M+ | n=7 | n=6 |  |
| Persistence or recurrence | 1/7 recurrence 5 years postoperatively | 0/6 (0%) | Pre-RET 2–14 years  RET 1–2 years |
| Time free of disease (time to disease, or time to follow-up, for individual patients) | 3 years 8 years 5 years 4 years 5 years 2 years 14 years | 2 years 1 year 1 year 1 year 1 year 1 year |  |
| (Skinner et al. 1996)  USA | III-3 interventional evidence  High risk of bias (13/26) | N=38 children who underwent thyroidectomy prior to 16 years of age for MEN2A or presence of a RET mutation | RET M+ | n=24 | n=14 |  |
| Persistence or recurrence | 1/24 (4.2%) persistence  4/24 (16.7%) recurrence | 0/14 (0%) | Mean: Pre-RET 9.3 years  RET 1.3 years |
| (Sanchez Sobrino et al. 2011)  Spain | III-3 interventional evidence  High risk of bias (10/26) | N=8 members of a family with MEN2A due to RET C634Y mutation | RET M+ | n=5 | n=3 |  |
| Free of disease | 0/5 (0%) | 3/3 (100%) | Total >20 years |
| Time free of disease | 17 years 9 years 6 years 16 years 11 years | 9 years  7 years 7 years |  |
| (Diaz & Wohllk 2012)  Chile | III-3 interventional evidence  High risk of bias (9/26) | N=60 MEN2 patients who underwent total thyroidectomy |  | n=29 | n=31 |  |
| Biochemically cured | 8/29 (27.6%) | 28/31(90.3%) | Not stated |
| Persistent | 8/29 (27.6%) | 2/31 (6.5%) |  |
| Dead | 13/29 (44.8%):  3 hypertensive crisis  1 surgical complications  9 distant metastases | 1/31 (3.2%):  Died from surgical complications |  |

FMTC = familial MTC; IQR = interquartile range; RET M+ = RET-mutation-positive

A forest plot combining the results of Rohmer et al, Schreinemakers et al, Skinner et al, Lallier et al, Sanchez Sobrino et al, and Diaz & Wohllk. All point estimates favour the RET era, with smaller studies having confidence intervals that cross the point of no effect. The combined estimate was clearly in support of RET testing, at reducing the risk of persistence of diseaes. 

Figure Risk of persistence of disease or mortality, RET mutation testing era versus pre-RET mutation testing era

m = mean; med = median; NS = not stated; RET era = RET mutation testing era.

References for studies: Rohmer (Rohmer et al. 2011); Schreinemakers (Schreinemakers et al. 2010); Skinner (Skinner et al. 1996); Lallier (Lallier et al. 1998); Sanchez Sobrino (Sanchez Sobrino et al. 2011); Diaz (Diaz & Wohllk 2012)

Twenty-three uncontrolled studies reported on the rates of persistence or recurrence of disease after total thyroidectomy in RET-mutation-positive patients with MTC (n = 8) and/or in asymptomatic relatives who had inherited a RET mutation (n =16) (see Table 73, Appendix B). The patients included adults and children with clinical signs of MEN2A, MEN2B and FMTC, with raised calcitonin levels or other early signs of disease at baseline, and those with no signs of disease receiving prophylactic treatment. Given the different phenotypes and disease stages included in each study, it is not surprising that results were highly variable. It was not always explicit in the articles whether treatment was initiated before or after, identification of a RET mutation.

In those patients who developed clinical signs of disease prior to total thyroidectomy, the rate of persistence or recurrence was between 4.0% (Dralle et al. 1998), and 56.7% (Quayle et al. 2004). In patients where surgery was considered prophylactic, rates of persistence or recurrence varied between zero and 58.3%, with a median of 9%. Skinner et al. (2005) performed *post hoc* analyses comparing those patients who were operated on before 8 years of age with those operated on after they turned 8 years of age. In the 22 patients who received surgery before 8 years of age, there were no instances of metastasis to cervical lymph nodes and no instances of persistence or recurrence. However, in the 28 patients operated on after the age of 8 years (range 8–19 years), 6 patients (21.4%) suffered from persistence or recurrence.

#### Incidence and severity of MTC

Seven historical controlled studies (level III-3 interventional evidence) compared the incidence and severity of MTC in patients who had been diagnosed and treated prior to RET mutation testing becoming available, against those who were diagnosed and treated with the addition of RET mutation testing (Table 26). These studies were based in Australia, Canada, the United States of America, France, Sweden, the Netherlands and Spain. All the studies were at risk of bias and confounding (although 1 study was rated as moderate quality as the reporting was very good). Overall, data on the incidence and severity of MTC in these studies were more informative than for the other longer term outcomes, where the difference in length of follow-up between arms became a problem in interpretation of the data. Incidence and severity of MTC were determined by histopathology in those who underwent total thyroidectomy, and immediately after surgery in both the intervention and control arms.

The results of these 7 studies were heterogeneous in size of effect but consistent in their direction of effect, showing that the risk of having an MTC detected by histopathology was significantly greater in those patients who were diagnosed in the pre-RET mutation testing era (Table 26). On average, those diagnosed and treated since RET mutation testing became available had almost half the risk of having an MTC at time of treatment (RR=0.53, 95% CI 0.32, 0.90).

Patients are likely to be identified earlier through genetic screening than through clinical screening. Those undergoing surgery at a particular age are therefore more likely to have a lower risk profile if they have been identified by RET mutation testing.

The majority of the studies restricted the comparison of outcomes to those who were RET-mutation-positive. However, Learoyd et al. (1997) also discussed the management and outcomes of family members in MEN2 families who were found to be negative for RET mutations. Four family members had undergone a total thyroidectomy based on an elevated calcitonin response to pentagastrin-stimulation testing. Two of these were performed without knowledge of mutation status and showed normal thyroid histological characteristics. In another family two members underwent a total thyroidectomy despite being found to not have a RET mutation. These two patients were found to have C-cell hyperplasia. Three members from one family had elevated calcitonin responses but refused thyroidectomy on religious grounds. These people were later found to be RET-mutation-negative. One additional person from a MEN2 family was indicated for surgery based on elevated calcitonin, but surgery was not performed as genetic screening became available and this family member was found not to have a RET mutation.

Table Incidence and severity of MTC at time of total thyroidectomy

| Study and location | Level of evidence and quality assessment | Study population | Histology results | Pre-RET mutation testing era | RET mutation testing era | **Significance (p-value)** |
| --- | --- | --- | --- | --- | --- | --- |
| (Rohmer et al. 2011)  France | III-3 interventional evidence  Moderate risk of bias (18/26) | N=170 patients with a RET mutation who underwent a total thyroidectomy younger than 21 years of age:  109 MEN2A  24 MEN2B  37 FMTC | RET M+ | n=38 | n=132 |  |
| Normal or CCH or microscopic MTC | 26/37(70.3%) | 120/129 (93%) | p=0.001 |
| MTC | 11/37(29.7%) | 9/129 (7.0%) |
| Lymph node metastases:  N0  N1  Nx | 20/37 (54.1%)  6/37 (16.2%)  11/37 (29.7%) | 80/129 (62.0%)  9/129 (7.0%)  9/129 (7.0%) | NS |
| (Schreinemakers et al. 2010)  Sweden | III-3 interventional evidence  High risk of bias (17/26) | N=93 patients with a RET mutation who underwent a total thyroidectomy younger than 20 years of age | RET M+ | n=25 | n=68 |  |
| Median age | 10.6 years | 7.8 years | p=0.02 |
| CCH | 7/25 (28%) | 22/67 (32.8%) | NS |
| MTC | 18/25 (72%) | 43/67 (64.2%) |
| Normal histology | 0/25 (0%) | 2/67 (3.0%) |
| Lymph node metastases | 5/25 (20%) | 3/68 (4.4%) | p=0.02 |
| (Learoyd et al. 1997)  Australia | III-3 interventional evidence  High risk of bias (16/27) | N=164 individuals from families with MEN2 and known RET mutations:  56 were RET M+  108 were RET M– | RET M+ | n=45 | n=7 |  |
| Mean age (range) | 32 years (6–65 years) | 16 years (7–28 years) |  |
| CCH | 1/45 (2%) | 4/7 (57%) | p<0.001 |
| MTC | 44/45 (98%) | 3/7 (43%) |  |
| RET M– | n=2 | n=2 |  |
| CCH | 0/2 (0%) | 2/2 (100%) |  |
| MTC | 0/2 (0%) | 0/2 (0%) |  |
| (Lallier et al. 1998)  Canada | III-3 interventional evidence  High risk of bias (15/26) | N=13 MEN2 patients (children) who underwent total thyroidectomy between 1981 and 1997  with RET mutation testing: 5 codon 620 and 1 codon 643 mutations | Clinically positive / RET M+ | n=7 | n=6 |  |
| Mean age (range) | 11.8 years (1.5–16 years) | 9.1 years (6–14 years) |  |
| CCH without MTC | 0/7 (0%) | 2/6 (33.3%) |  |
| MTC | 7/7 (100%) | 1/6 (16.7%) |  |
| Disease-free | 0/7 (0%) | 3/6 (50%) |  |
| (Skinner et al. 1996)  USA | III-3 interventional evidence  High risk of bias (13/26) | N=38 children who underwent thyroidectomy prior to 16 years of age for MEN2A or presence of a RET mutation | RET M+ | n=24 | n=14 |  |
| Mean age (range) | 10.6 years (5–15 years) | 10.5 years (5–15 years) |  |
| CCH without MTC | 4/24 (16.7%) | 3/14 (21.4%) |  |
| MTC without nodal metastases | 12/24 (50%) | 11/14 (78.6%) |  |
| MTC with nodal metastases | 1/24 (4.2%)  Spread to cervical lymph nodes | 0/14 (0%) |  |
| MTC, unsampled lymph nodes | 7/24 (29.2%) | 0/14 (0%) |  |
| (Sanchez Sobrino et al. 2011)  Spain | III-3 interventional evidence  High risk of bias (10/26) | N=8 individuals from a family with MEN2A due to RET C634Y mutation | RET M+ | n=5 | n=3 |  |
| Age range | 23–58 years | 6–34 years |  |
| CCH | 0/5 (0%) | 2/3 (66.7%) |  |
| MTC | 5/5 (100%) | 1/3 (33.3%) |  |
| (Lips et al. 1994)  The Netherlands | III-3 interventional evidence  High risk of bias (7/26) | N=14 members of 4 large MEN2A families who had a thyroidectomy:  8 were based on RET mutation carrier status  6 were based on raised pentagastrin-stimulated calcitonin levels who were later found to be RET M– | RET M+ | Not stated | n=8 |  |
| C-cell clusters or nodules and small irregular foci of MTC |  | 8/8 (100%) |  |
| RET M– | n=6 | Not stated |  |
| C-cell clusters or nodules | 2/6 (33.3%) |  |  |

CCH = C-cell hyperplasia; FMTC = familial medullary thyroid carcinoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

A forest plot showing seven studies, all of which have point estimates favouring the RET testing era, as reducing the risk of having an MTC at time of total thyroidectomy. The combined estimate clearly favours RET testing. 

Figure Risk of having an MTC at time of total thyroidectomy, RET mutation testing era versus pre-RET mutation testing era

m = mean; med = median; RET era = RET mutation testing era

References for studies: Rohmer (Rohmer et al. 2011); Schreinemakers (Schreinemakers et al. 2010); Learoyd (Learoyd et al. 1997); Skinner (Skinner et al. 1996); Lallier (Lallier et al. 1998); Sanchez Sobrino (Sanchez Sobrino et al. 2011).

Twelve uncontrolled studies (level IV interventional evidence) reported on the incidence and severity of MTC in patients who developed clinical signs of disease prior to total thyroidectomy (Table 74, Appendix C). The populations included a mix of index patients (probands) and family members who also showed clinical signs or symptoms. Given the starting population, it is not surprising that the vast majority of these patients had MTC at the time of total thyroidectomy (median 100%, range 64.3–100%), rather than C-cell hyperplasia or no disease.

Fifty-one uncontrolled studies (level IV interventional evidence) provided information on the rate of MTCs in RET-mutation-positive family members who had undergone total thyroidectomy (Table 75, Appendix C). In family members a total thyroidectomy is often considered ‘prophylactic’ if the surgery is based on a genetic diagnosis, in the absence of clinical manifestations (i.e. the patient is asymptomatic and there is no evidence of palpable cervical masses) (Rodriguez Gonzalez et al. 2002). However, MTCs or C-cell hyperplasia are often evident on histopathological examination of the thyroid specimen, with MTCs evident in 0–100% of RET-mutation-positive family members. The median rate of MTCs was 67.6% (interquartile range (IQR) = 50%, 85.7%). Given that, in many of the studies, the populations received prophylactic surgery, this is still very high and is indicative of the high penetrance of MTCs in those with a RET mutation.

Machens et al. (2005) stratified patients who underwent a thyroidectomy for C-cell hyperplasia or MTC into three risk categories according to which RET codon was mutated. They found that patients with RET codon 918 mutations (responsible for MEN2B) had the highest risk (100%) of MTC at the time of surgery, while patients with RET codon 609–634 mutations had a high risk (72.6%) of MTC at the time of surgery, and those with RET codon 786–891 mutations had the lowest risk (45.1%).

#### Incidence of phaeochromocytoma and hyperparathyroidism

One very small historical controlled study (level III-3 interventional evidence) provided the incidence of phaeochromocytoma and hyperparathyroidism in patients who were diagnosed clinically or with the addition of RET mutation testing (Table 27). This study was too small to draw any conclusions, but it is not expected that there would be any difference in the incidence of identified phaeochromocytoma and hyperparathyroidism between the two diagnostic modalities. In both scenarios (diagnosed prior to RET mutation testing availability or diagnosed after) treatment for phaeochromocytoma would only occur after clinical signs of disease, and patients in both arms would continue to undergo surveillance. The article describing the Australian historical controlled study (level III-3 interventional evidence) supported this, reporting that knowledge of having a RET mutation did not impact on the frequency of post-thyroidectomy pentagastrin testing or screening for additional features of phaeochromocytoma or hyperparathyroidism (Learoyd et al. 1997).

The only exception to this would be that availability of RET mutation testing could allow earlier distinction between patients with MEN2A and those from FMTC families. If, based on the genotype, patients are classified as having FMTC rather than MEN2A, screening for other features of MEN2A (phaeochromocytoma or hyperparathyroidism) would not be required. However, no differences in health outcomes are expected.

Table Incidence of phaeochromocytoma and hyperparathyroidism

| Study and location | Level of evidence and quality assessment | Study population | Outcome measure | Pre-RET mutation testing era | RET mutation testing era |
| --- | --- | --- | --- | --- | --- |
| (Sanchez Sobrino et al. 2011)  Spain | III-3 interventional evidence  High risk of bias (10/26) | N=8 individuals from a family with MEN2A due to C634Y mutation | RET M+ | n=5 | n=3 |
| Phaeochromocytoma | 2/5 (40%)  2 x bilateral | 1/3 (33.3%)  Bilateral |
| Hyperparathyroidism | 0/5 | 0/3 |

RET M+ = RET-mutation-positive

Fifteen uncontrolled studies (level IV interventional evidence) reported on the penetrance of phaeochromocytoma and hyperparathyroidism over varying lengths of time (Table 76, **Error! Reference source not found.**). Phaeochromocytoma was observed in 0–76.4% of patients, with a median of 25%. These studies were often limited by short-term follow-up, so the figures presented are not representative of lifetime penetrance of phaeochromocytoma.

It is well known that the rate of phaeochromocytoma and hyperparathyroidism differ by phenotype of MEN2 and by age. The greatest detail on the rate of phaeochromocytoma in those with RET mutations was provided by Machens et al. (2006). This case series was rated as poor quality, as few details were provided on the inclusion criteria for the study or the method of Ret mutation testing. However, they reported that, in those with the highest risk phenotype (mutation in codon 918, corresponding to MEN2B), the penetrance of phaeochromocytoma was 43% by age 30 years and 100% by age 35 years. In those with a high-risk phenotype (mutation in codon 609–634, corresponding to some types of FMTC and MEN2A), the penetrance by age 30 years was 8%, which increased to 18% by age 35 years and 54% by age 50 years. In those with the lowest risk (mutation in codon 768–891, corresponding to FMTC), the penetrance of phaeochromocytoma was zero until age 50 years, when it was 4%. In the largest case series (Frank-Raue et al. 2011) over half of the phaeochromocytoma cases were identified after clinical symptoms were evident rather than being identified through screening.

Twelve uncontrolled studies reported on the rate of hyperparathyroidism in RET-mutation-positive index cases (Table 76, **Error! Reference source not found.**). Up to 15.4% of patients with MEN2A, MEN2B and FMTC combined were diagnosed with hyperparathyroidism. In 1 study 38.5% of patients showed evidence of hypercellular parathyroid pathology on exploration, but this was not correlated to signs of clinical disease (Etit et al. 2008). The largest study reported that the median age at which hyperparathyroidism was diagnosed was 46 years but the range (28–82 years) indicated wide variability (Frank-Raue et al. 2011). The vast majority (87.5%) of cases of hyperparathyroidism were identified through screening (Frank-Raue et al. 2011).

Thirty uncontrolled studies (level IV interventional evidence) provided rates of phaeochromocytoma and hyperparathyroidism in RET-mutation-positive family members (Appendix D). The studies included a mix of patients with MEN2A, MEN2B and FMTC. The overall penetrance of phaeochromocytoma varied between zero and 50%.

The 23 case series reporting on the rate of hyperparathyroidism in patients with MEN2 in RET-mutation-positive family members showed rates of up to 27.3%. Schuffenecker et al. (1998) reported that the mean age of hyperparathyroidism diagnosis was 33.7 years (range 12–70 years), with a penetrance of 14% by age 30 years, 26% by age 40 years and 48% by age 60 years. These rates are unlikely to have been altered at all by RET mutation testing.

### Secondary effectiveness outcomes

#### Age at diagnosis

One article was identified that compared the age at time of diagnosis in patients with an MTC between a survey performed in 1996, and a survey performed in 2002 (Kameyama & Takami 2004). Age at diagnosis in 2002 was lower for patients with MEN2A, FMTC and sporadic MTC, but not with MEN2B. The reduction in age in the survey in 2002 was attributed to RET mutation testing allowing earlier diagnosis of symptomatic patients (rather than requiring more than one disease feature in the family), earlier distinction between MEN2A and FMTC, and pre-clinical diagnosis within family members. Given the smaller numbers of people with MEN2B, the lack of reduction in age may either be due to chance or as a consequence of MEN2B having a clearer phenotype. There is also a higher rate of *de novo* mutations in MEN2B than in MEN2A, such that the proportion of familial cases detected through screening will be much lower than with MEN2A.

Table Mean age (years) at diagnosis

| Study and location | Level of evidence and quality assessment | Study population | Population | Pre-RET mutation testing era | RET mutation testing era |
| --- | --- | --- | --- | --- | --- |
| (Kameyama & Takami 2004)  Japan | III-3 interventional evidence  High risk of bias (10/26) | N=905 MTC patients:  634 patients in 1996:  175 MEN2A  49 FMTC  20 MEN2B  390 sporadic MTC  271 patients in 2002:  83 MEN2A  14 FMTC  11 MEN2B  163 sporadic MTC | MEN2A  FMTC  MEN2B  Sporadic MTC | 40.3±15.3  43.6±15.6  26.5 ±8.8  48.5±13.9 | 35.6±14.4  34.6±12.3  30.5±10.1  47.6±14.0 |

FMTC = familial medullary thyroid carcinoma

Twelve additional uncontrolled studies reported the age at diagnosis for index cases (Table 78, Appendix E). Given that these patients were detected due to symptoms, the age at diagnosis varies largely on genotype and phenotype. The age ranged from a mean of 13.5±2.1 years for patients with MEN2B to a median of 62 years for patients with a RET codon 804 mutation.

Table 79 (Appendix E) outlines the age at diagnosis for family members of someone with a confirmed RET mutation in 12 uncontrolled studies. Punales et al. (2003) reported that those with clinical disease were, on average, 8 years older than those without clinical disease. Historically, asymptomatic gene carriers would not have been able to be diagnosed until they showed clinical or biochemical signs of disease.

Machens et al. (2005) reported that the time to diagnosis of MTC in patients with the highest risk (mutation in RET codon 918, corresponding to MEN2B) was 14.3 years (95% CI 10.3, 18.4). In those with a high risk (mutation in RET codons 609–634, corresponding to some types of FMTC and MEN2A), the time to diagnosis of MTC was 30.1 years (95% CI 26.6, 33.5). In those with the least high risk (mutation in RET codons 768–891, corresponding to FMTC), the time to diagnosis of MTC was 51.6 years (95% CI 46.5, 56.6). However, the authors did not report if diagnosis was based on clinical and/or biochemical signs or on RET mutation status.

#### Mean age at time of thyroidectomy

Five historical controlled studies (level III-3 interventional evidence) reported on the age at time of total thyroidectomy in a cohort of patients treated in the era prior to RET mutation testing, and a cohort of patients treated after RET mutation testing was available (Table 29). Since the introduction of RET mutation testing, patients have undergone total thyroidectomy at a much younger age. Schreinemakers et al. (2010) reported that the mean age at surgery was significantly different between those who were disease free after surgery (mean 8.6 years) compared with those who had residual or recurrent disease (mean 12.1 years, p=0.002).

Table Age at time of total thyroidectomy

| Study and location | Level of evidence and quality assessment | Study population | Outcome measure | Pre-RET mutation testing era | RET mutation testing era | **Significance (p-value)** |
| --- | --- | --- | --- | --- | --- | --- |
| (Rohmer et al. 2011)  France | III-3 interventional evidence  Moderate risk of bias (18/26) | N=170 RET M+ patients who underwent a total thyroidectomy younger than 21 years of age:  109 MEN2A  24 MEN2B  37 FMTC | RET M+ | n=38 | n=132 |  |
| Mean age at thyroidectomy | 10.7± 6.6 years | 8.3± 4.4 years | p=0.003 |
| (Schreinemakers et al. 2010)  Sweden | III-3 interventional evidence  High risk of bias (17/26) | N=93 RET M+ patients who had undergone total thyroidectomy younger than 20 years of age | RET M+ | n=25 | n=68 |  |
| Median age at thyroidectomy (IQR) | 10.6 years (7.4–13.2) | 7.8 years (5.3–12.2) | p=0.022 |
| (Learoyd et al. 1997)  Australia | III-3 interventional evidence  High risk of bias (16/27) | N=164 people from 26 families with MEN2 RET mutations:  56 were RET M+  108 were RET M– | RET M+ | n=45 | n=7 |  |
| Mean age at surgery (range) | 32 years  (6–65) | 16 years  (7–28) | Not stated |
| (Lallier et al. 1998)  Canada | III-3 interventional evidence  High risk of bias (15/26) | N=13 patients with MEN2 who underwent total thyroidectomy between 1981 and 1997 | Clinically positive / RET M+ | n=7 | n=6 |  |
| Mean age at surgery (range) | 11.8± 4.9 years  (1.5–15) | 9.0± 3.3 years  (5–14) | Not stated |
| (Sanchez Sobrino et al. 2011)  Spain | III-3 interventional evidence  High risk of bias (10/26) | N=8 family members with MEN2A due to a RET C634Y mutation | RET M+ | n=5 | n=3 |  |
| Mean age at surgery (range) | 37.6± 14.8 years  (23–58) | 16.7± 15.1 years  (6–34) | Not stated |

N/A = Not applicable; RET M+=RET mutation positive; RET M–=RET mutation negative.

Two of the historical controlled studies further compared whether patients with RET mutations were treated appropriately according to the 1999 consensus statement from the Seventh International Workshop on Multiple Endocrine Neoplasia and the 2009 French guidelines (Rohmer et al. 2011; Schreinemakers et al. 2010). For the highest risk group (mutations in codon 918, 883 and 922) surgery was recommended before 6 months of age, for the higher risk group (mutations in codon 630, 634, 609, 611, 618 and 620) before age 5 years, and for the least high risk group (mutations in codons 768, 790, 791, 804 and 891) prior to age 10 years (Rohmer et al. 2011; Schreinemakers et al. 2010). Across both these studies, only 3 out of 62 (4.7%) patients were treated according to the age-appropriate guidelines in the historical cohort, compared with 42 out of 197 (21.3%) in the cohorts diagnosed since the introduction of RET mutation testing (Table 30). The difference in the rates between cohorts was significant for both studies. Schreinemakers et al. (2010) reported that age at time of thyroidectomy was an independent prognostic variable for persistent or recurrent disease in multivariate analyses.

Both the Rohmer et al. (2011) and Schreinemakers et al. (2010) studies noted that, of those with residual or recurrent disease, none had been operated on at an age recommended by the guidelines. This finding should not be surprising, given that the guidelines were developed with reference to the age at which treatment may be considered curative.

Table Age-appropriate surgery

| Study and location | Level of evidence and quality assessment | Study population | Outcome measure | Pre-RET mutation testing era | RET mutation testing era | **Significance (p-value)** |
| --- | --- | --- | --- | --- | --- | --- |
| (Rohmer et al. 2011)  France | III-3 interventional evidence  Moderate risk of bias (18/26) | N=170 RET M+ patients who underwent a total thyroidectomy younger than 21 years of age:  109 MEN2A  24 MEN2B  37 FMTC | RET M+ | n=38 | n=132 |  |
| Age-appropriate surgery | 2/37 (5.4%) | 21/129 (16.3%) | p=0.091 |
| (Schreinemakers et al. 2010)  Sweden | III-3 interventional evidence  High risk of bias (17/26) | N=93 RET M+ patients who had undergone total thyroidectomy younger than 20 years of age | RET M+ | n=25 | n=68 |  |
| Age-appropriate surgery | 1/25 (4%) | 21/68 (30.9%) | p=0.004 |

FMTC = familial medullary thyroid carcinoma; RET M+ = RET-mutation-positive

Two uncontrolled studies reported a mean age at time of total thyroidectomy in index cases of 24.6±12.2 years and 43.2±22.2 years (Table 31). Due to the lack of a comparator, it is unknown based on this data whether age at time of thyroidectomy has changed for index cases; however, it is hypothesised that little change would be found in those whose MTC is identified before RET mutation testing, as the treatment for MTC would be the same regardless of mutation.

Table 31 Mean age at total thyroidectomy (index cases)

| Study and location | Level of evidence | Study population | Intervention | Mean age at thyroidectomy |
| --- | --- | --- | --- | --- |
| (Chiefari et al. 1998)  Italy | IV Interventional evidence  Moderate quality (4/6) | N=16 RET M+ patients from 8 families with hereditary MTC, who underwent thyroidectomy | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14  Clinical screening  Total thyroidectomy | Mean = 24.6±12.2 years  (range = 10–45 years) |
| (Vaclavikova et al. 2009)  Czech Republic | IV interventional evidence  Poor quality (2/6) | N=6 index cases with a RET Y791F mutation who underwent total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | Mean = 43.2±22.2 years  (range = 14–69 years) |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive

Table 80 (Appendix F) outlines the mean age at total thyroidectomy in family members who were screened and found to have RET mutations as reported in 19 uncontrolled studies. Family members often had a total thyroidectomy at a younger age than index patients, prior to any clinical signs of disease. Mean age at time of thyroidectomy in the studies varied between 7.5 years and 52.8 years, with a median of 25.1 years.

#### Rates of surveillance

Nine direct studies of the impact of RET mutation testing on health outcomes mentioned rates of surveillance in those who were RET-mutation-positive or -negative (Table 32). Prior to RET mutation testing being available, all family members would have been recommended to undergo surveillance for features of MEN2. However, after the introduction of RET mutation testing, those who had no pathological RET mutations were able to cease surveillance and were told that their descendants would not require evaluation for MEN2 either (see also ‘Change in Management’ in linked evidence section).

Those who were RET-mutation-positive were recommended to continue surveillance for features of MEN2 (Learoyd et al. 1997); however, a small proportion of RET-mutation-positive patients refuse further clinical and/or biochemical examinations (Romei et al. 2011).

Table Rates of surveillance

| Study and location | Level of evidence and quality of assessment | Study population | RET M+ | RET M– |
| --- | --- | --- | --- | --- |
| ([Learoyd et al. 1997](#_ENREF_117))  Australia | III-3 interventional evidence  High risk of bias (16/27) | N=164 patients from 26 MEN2 families:  56 were RET M+  108 were RET M– | Unchanged from pre-RET mutation testing era | All discontinued screening |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV Interventional evidence  Moderate quality (4/6) | N=229 members spanning 6 generations of a large extended FMTC family with a RET G533C mutation  76 members were RET M+ | 10/76 (13.2%) RET M+ family members presented with low pentagastrin-stimulated calcitonin levels and surgery was delayed  3/76 (3.9%) refused further clinical investigation | 153/153 (100%) RET M– family members had normal pentagastrin-stimulated calcitonin levels and were excluded from further clinical investigation |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | N=60 RET M+ family members of patients with MTC re-classified from sporadic MTC to FMTC or MEN2A due to a RET mutation:  35 showed clinical and/or biochemical signs of disease on screening  30 (29 FMTC, 1 MEN2A) underwent total thyroidectomy  5 refused treatment | 5/60 (8.3%) RET M+ refused clinical and/or biochemical examinations  20/20 (100%) RET M+ clinically unaffected underwent yearly clinical and biochemical assessment | Not stated |
| ([Lindskog et al. 2004](#_ENREF_124))  Sweden | IV interventional evidence  Moderate quality (3/6) | N=33 RET M– family members of a MEN2A family with a RET codon 618 mutation | Not stated | 33 RET M– family members who had previously undergone biochemical surveillance, ceased surveillance and were told that their descendants would not need to be evaluated for disease |
| ([Shimotake et al. 1996](#_ENREF_186))  Japan | IV interventional evidence  Moderate quality (3/6) | N=6 children who had an affected parent from a MEN2 family and were without clinical signs of disease underwent RET mutation testing  3 were RET M+ | Not stated | 3/6 were found not to have RET mutation  These family members were released from endocrine screening |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV interventional evidence  Poor quality (2/6) | N=25 asymptomatic first-degree relatives from 12 unrelated Greek families  9 MEN2A  1 FMTC  3 probable FMTC (only 3 members diagnosed with MTC) | Not stated | 20 RET M– family members excluded from further screening |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV interventional evidence  Poor quality (2/6) | N=15 RET M+ family members from an MTC index patient with a RET L790F mutation  8 had abnormal pentagastrin-stimulated calcitonin levels | 7/15 (46.7%) had normal calcitonin levels and were advised to have annual follow-ups to check calcitonin levels  3/7 (42.9%) refused follow-up | Not stated |
| ([Kinlaw et al. 2005](#_ENREF_107))  USA | IV interventional evidence  Poor quality (2/6) | N=15 RET M+ family members (including index case) of a MEN2A family with a RET C609S mutation | 11/15 (73.3%) underwent further biochemical and clinical screening  4/15 (26.7%) refused further evaluation | Not stated |
| ([Uchino et al. 1999](#_ENREF_204))  Japan | IV interventional evidence  Low quality (2/6) | N=6 clinically unaffected members from MEN2A families with mutations on RET codon 634  All had raised calcitonin levels. | 2 adult patients refused treatment  1 patient, aged 7 years, is being monitored | Not stated |

FMTC = familial medullary thyroid carcinoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

### Linked evidence

Summary of effectiveness

The accuracy of RET genetic testing could not be determined as no long-term clinical data were available to use as the reference standard. There were 6 uncontrolled studies that reported cases where total thyroidectomy had been recommended, based on calcitonin testing, in patients who were later found to have no RET mutations. Biochemical screening as a means of diagnosis is therefore known to have false positive results. From the data available it is impossible to determine whether mutation testing of the *RET* gene is associated with false positive test results.

There were uncontrolled data suggesting that many patients undergo prophylactic surgery based on knowledge of a RET mutation, and comparative data suggesting that prophylactic surgery is associated with much better health outcomes than curative surgery (surgery performed after clinical signs of disease).

As it was not clear when the systematic review was initiated that direct evidence was available for the assessment of RET mutation testing, a linkage of evidence was also undertaken.

### Is it accurate?

It was initially decided that the accuracy of RET mutation testing would be determined using the reference standard of long-term clinical health outcomes (i.e. whether RET mutation status corresponds to MEN2 features over the patient’s lifetime). However, given the age of the technology, long-term clinical data were not available in those who were RET-mutation-positive, and very little clinical data were available for those patients or family members who tested negative for RET mutations.

Histological data were available for those who had RET mutations and subsequently underwent total thyroidectomy. These data are shown in the section on the incidence and severity of MTC (Table 26; Table 74 and Table 75, Appendix C). Although this short-term data could be used as an imperfect reference standard to calculate the positive predictive value of RET mutation testing, compared with the positive predictive value of pentagastrin-stimulated calcitonin levels, this comparison may be misleading. RET mutation testing allows earlier detection of those at risk of developing an MTC, allowing surgery to be performed at an earlier stage of disease development. A different spectrum of patients would therefore be operated on. In patients with a RET mutation who undergo a total thyroidectomy, a histopathology result that shows no MTC may be thought of as a clinical success due to early intervention, rather than as a lack of accuracy of RET mutation testing. The positive predictive value of RET mutation testing, compared with pentagastrin-stimulated calcitonin testing, has therefore not been presented.

A further difficulty in determining the accuracy of RET mutation testing is that treatment bias may occur. Between RET mutation testing and long-term clinical outcomes, treatment (i.e. total thyroidectomy) occurs in those who are RET-mutation-positive. In those patients who have RET mutations corresponding to FMTC, the only clinical characteristic that patients are at risk of developing is an MTC. If patients have a prophylactic total thyroidectomy, they are unlikely to develop an MTC, so there is no way of confirming whether the results of RET mutation testing correspond to clinical outcomes in these patients.

#### Diagnostic yield

Diagnostic yield informs the question of what proportion of patients who are tested are found to have pathology RET mutations. As the results of this will be largely determined by the population tested, the results for diagnostic yield have been separated into the following different populations: those with an MTC; those with an apparently sporadic MTC; those presenting with phaeochromocytoma; and family members of someone with a confirmed RET mutation.

Due to the volume of small case series reporting diagnostic yield, which do not provide much additional information, studies on diagnostic yield were only included if they tested a minimum of 20 patients. Some results are available for numbers smaller than this, if the article includes several subpopulations.

##### Medullary thyroid carcinoma (MTC)

The diagnostic yield of RET mutation testing among patients presenting with an MTC is presented in Table 81 (hereditary MTC), Table 82 (apparently sporadic MTC) and Table 83 (unspecified MTC) in Appendix G. In the 17 uncontrolled studies that reported the diagnostic yield from patients with hereditary MTC, a median of 95.5% of index cases had a detectable RET mutation, ranging between 25% and 100% in individual studies. Thus, only 4.5% of families with inherited MTC did not have a detectable RET mutation in these studies. In some cases the study design precluded the detection of certain known RET mutations, such as the RET mutations on exons 15 and 16 responsible for MEN2B ([Hedayati et al. 2006](#_ENREF_88); [Kimura et al. 1995](#_ENREF_105)). Additionally, the number of index cases with undetectable RET mutations would be expected to continue to decrease as RET mutation testing methodologies continue to improve.

From the 25 studies that reported the diagnostic yield of RET mutation testing of patients with apparently sporadic MTC, without any family history or additional features of MEN2, the prevalence of germline RET mutations ranged from zero to 55.6% with a median of 6.5% (Table 81, Appendix G). In this population RET mutation testing would be able to rule out 93.5% of patients from additional biochemical and surveillance for MEN2 features. In the 6 studies that presented diagnostic yield from unspecified cases of MTC, the diagnostic yield of RET mutations ranged from 21.5% to 45.7% with a median of 30.6% (Table 82, Appendix G).

##### Phaeochromocytoma

In patients presenting with phaeochromocytoma, the diagnostic yield depended on whether the patients had other indicators of MEN2 or not. In 14 uncontrolled studies that reported diagnostic yield for patients who had apparently sporadic phaeochromocytoma, the proportion of patients who had germline RET mutations ranged from zero to 18.3% with a median of 0.45% (Table 84, Appendix G). Thus, up to 200 patients with apparently sporadic phaeochromocytoma would need to be tested in order to identify one MEN2 case.

In 4 studies with familial phaeochromocytoma, a median of 24.4% of patients had a RET mutation (Table 84, Appendix G), such that four patients would be tested for every case detected. In the 5 studies that reported the diagnostic yield for patients with unspecified phaeochromocytoma, a median of 7.8% of patients had a germline RET mutation.

##### Hyperparathyroidism

There was no evidence on the diagnostic yield of RET mutation testing in patients presenting with hyperparathyroidism, plus a diagnosis of MTC or phaeochromocytoma in a close relative. It is therefore unknown what proportion of patients with these characteristics would be positive for RET mutations.

##### Relatives of patients with a known RET mutation

Biologically, half of all first-degree relatives of someone with a confirmed RET mutation can be expected to also have a RET mutation, given the pattern of inheritance. However, the rates differ largely between studies (Table 85, Appendix G). This is likely due to different proportions of first-degree, versus more distant, relatives and whether symptomatic relatives are included. In families with a higher risk phenotype, those with RET mutations may be likely to already have clinical features of disease, so if the figures shown are only those who are still asymptomatic, the chances of having a RET mutation are much lower.

In the 11 studies that tested only first-degree relatives of someone with a RET mutation, the diagnostic yield ranged between 20% and 57.1% with a median of 37.5% (Table 85, Appendix G). This is lower than the 50% expected biologically, as several studies excluded family members with clinical signs of disease. In fact, 1 study found that, while 57.1% (36/63) of all first-degree relatives from 9 MEN2A families were RET-mutation-positive, only 33.3% (10/30) of those with no clinical signs of disease carried the RET mutation ([McMahon et al. 1994](#_ENREF_133)). The 48 studies that included second-degree relatives or did not specify the relationship to the index patient had a similar median diagnostic yield of 39.4% (range 15.9–65.5%).

### Does it change patient management?

A diagnostic test is only useful if the results then influence clinical management. In the case of RET mutation testing, knowledge of the *RET* gene is useful in determining whether patients require ongoing surveillance for features of MEN2 and would benefit from a total thyroidectomy (if they don’t already have clinical signs of MTC, in which case they would have a thyroidectomy regardless of RET status).

In 1996 Learoyd et al. (1997) performed a survey of clinicians managing MEN2 families in Australia, and reported that 27/28 clinicians surveyed used RET mutation test results to assist in the management of MEN2. Given that mutation testing had only been available for this condition for 3 years, the authors concluded that there was a high level of acceptance by clinicians.

Table 86 (Appendix H) outlines the results of 28 uncontrolled studies that provide information on treatments received by patients. In total, 24 studies reported on the treatment of patients who were RET-mutation-positive. A median of 60.9% (range 0–100%) of patients underwent a total thyroidectomy, many of which were prophylactic. Additionally, a median of 3.9% (range 0–60%) of patients were scheduled for surgery and 17.1% (range 0–100%) were being monitored. However, a median of 8.5% (range 0–33.3%) of patients refused a thyroidectomy and/or further monitoring. Treatment decisions for patients who did not undergo a thyroidectomy were not reported in 10 studies (median 8.3% of patients, range 0–49.2%).

Ten studies listed in Table 86 (Appendix H) included anecdotal evidence that RET mutation testing influenced management of RET-mutation-negative family members. Three studies reported that RET-mutation-negative family members were released from further clinical and biochemical screening ([Alvares Da Silva et al. 2003](#_ENREF_9); [Karga et al. 1998](#_ENREF_102); [Lindskog et al. 2004](#_ENREF_124)). Six studies reported that before RET mutation testing was available, some patients were classified as being clinically affected based on raised pentagastrin-stimulated calcitonin levels, warranting a total thyroidectomy, and were subsequently found to be free of RET mutations ([Decker et al. 1995](#_ENREF_40); [Frank-Raue et al. 1996](#_ENREF_69); [Gagel et al. 1995](#_ENREF_75); [Halling et al. 1997](#_ENREF_87); [Hernandez et al. 1997](#_ENREF_90); [Lips et al. 1994](#_ENREF_125); [Marsh et al. 1996](#_ENREF_133)). In these patients it is clear that RET mutation testing would have avoided unnecessary surgery. This is illustrated in 3 studies in which patients with raised pentagastrin-stimulated calcitonin levels were spared from having a thyroidectomy when found to be RET-mutation-negative ([Decker et al. 1995](#_ENREF_40); [Gagel et al. 1995](#_ENREF_75); [Hernandez et al. 1997](#_ENREF_90)).

Halling et al. (1997) reported on individuals from one FMTC kindred who underwent total thyroidectomy, and in whom both RET mutation status and pre-operative pentagastrin-stimulated calcitonin levels were recorded. This study shows that C-cell hyperplasia is common in both those with and without RET mutations regardless of whether or not pentagastrin-stimulated calcitonin levels were elevated. It was present in 5/7 RET-mutation-negative patients with raised pentagastrin-stimulated calcitonin levels, and in 3/3 with normal levels. Of note is that one RET-mutation-negative patient with raised pentagastrin-stimulated calcitonin levels showed clinical signs of disease on biochemical screening, and was subsequently found on histopathology to have an MTC. From this family it is also clear that some patients had a prophylactic total thyroidectomy prior to RET mutation testing being available, even when pentagastrin-stimulated calcitonin levels were not raised.

#### Does change in management improve patient outcomes?

One key change in management that has occurred in patients with RET mutations is that those at risk of MTC are more likely to be operated on before clinical signs of disease develop.

One small cohort study of patients who underwent total thyroidectomies for MEN2 between 1995 and 2007 in Italy compared the outcomes between those who had a thyroidectomy after clinical signs developed (i.e. a curative thyroidectomy) and those who had a thyroidectomy before clinical signs developed, on the basis of a *RET* gene point mutation (i.e. a prophylactic thyroidectomy) (Table 33). This study is considered to have a high risk of bias as there were confounding factors (such as risk level) that were not evenly distributed between the treatment groups. The health outcomes resulting from these treatments (either curative or prophylactic thyroidectomies) may potentially be the result of risk level of their particular RET codonic mutations rather than due to the management strategy.

The patients who already showed clinical signs of an MTC or MEN2 at the time of thyroidectomy showed more-developed MTC on histology. Lymph node metastases were found in two patients with MEN2B. These patients died 6 and 7 years after diagnosis due to disease progression. Tumour stage at presentation is a key prognostic factor for outcomes.

It is expected that RET mutation testing has increased the proportion of patients having prophylactic thyroidectomies rather than curative thyroidectomies, and it is expected that patient health outcomes are improved.

Table 33 Prophylactic thyroidectomy versus thyroidectomy based on clinical signs/symptoms

| Study and location | Level of evidence | Study population | Histology results | Prophylactic thyroidectomy | Curative thyroidectomy |
| --- | --- | --- | --- | --- | --- |
| ([Spinelli et al. 2010](#_ENREF_193))  Italy | III-2 interventional evidence  High risk of bias (16/26) | N=13 juvenile patients (8–17 years of age) with MEN2 who underwent surgery for MTC:  7 (54%) MEN2A  4 (31%) FMTC  2 (15%) MEN2B |  | n=6 | n=7 |
| C-cell hyperplasia | 6/6 (100%) | 7/7 (100%) |
| Monolateral MTC | 4/6 (66.7%) | 2/7 (28.6%) |
| Bilateral MTC | 0/6 (0%) | 5/7 (71.4%) |
| Extrathyroid invasion | 0/6 (0%) | 2/7 (28.6%) |
| Central lymph node involvement | 0/6 (0%) | 2/7 (28.6%) |
| No. of organs affected by distal metastases | 0 | 2 |
| Stage | 4 x T1N0M0  2 x no MTC | 5 x T1N0M0  2 x T4N1M1 (2 MEN2B) |

FMTC = familial medullary thyroid carcinoma

# Other relevant considerations

## RET mutation testing considered best practice

Despite the limitations in the direct evidence identified in this systematic review, there is unlikely to be any improvement in the evidence base in the future, with the possible exception of longer term follow-up being reported for the historical controlled studies. This is because RET mutation testing and prophylactic surgery are now considered the ‘gold standard’ in the diagnosis of patients with MEN2 and prediction of MEN2 risk in family members ([Learoyd & Robinson 2005](#_ENREF_118)). It would therefore be considered unethical to perform a controlled trial examining the direct impact of RET mutation testing on the health outcomes of these people because the control arm would not, therefore, have access to the ‘gold standard’ method of determining MEN2.

## Implications to the consumer

Public comment was sought during the development of the final DAP, which was released for public comment on 7 October 2011 and closed for comments on 4 November 2011. No comments from the public were received.

## Ethical considerations

### Introduction

Genetic testing of the *RET* gene in children and siblings of individuals with MEN2 is the only effective route to prevention and treatment for hereditary MTC, and is now considered standard care ([Burke, Pinsky & Press 2001](#_ENREF_24)). Early identification permits the use of prophylactic thyroidectomy, which has been demonstrated to improve life expectancy and quality of life ([Raue & Frank-Raue 2012](#_ENREF_167)). Yet a significant number of individuals choose not to undergo Ret mutation testing, indicating that there are barriers to understanding and appreciating its benefits for hereditary MTC ([Rosenthal & Diekema 2011](#_ENREF_174)).

The aim of this assessment report is to synthesise the available evidence in order to inform a public funding decision. In the case of ethical issues, such synthesis equates to reviewing the relevant literature and assessing the balance of the arguments. The synthesis is descriptive but it is also normative insofar as it seeks to identify ethical ideals for framing policy on how medical professionals should conduct themselves.

### Methods of evidence synthesis

Seven core papers ([Burke & Press 2006](#_ENREF_25); [Giarelli 2001](#_ENREF_77); [Green & Botkin 2003](#_ENREF_84); [Kinder 1998](#_ENREF_106); [Korf 1999](#_ENREF_111); [Offit & Thom 2007](#_ENREF_150); [Winslow, Kodner & Dietz 2005](#_ENREF_212)) were selected from the 249 articles identified as potentially relevant in a literature search that obtained papers addressing the linking of ethical theory to genetic testing. These constituted the main body of evidence. Where possible they were supported by additional articles that presented (i) material from an Australian perspective and (ii) issues relating specifically to RET mutation testing. Some of these additional articles were identified in the ‘ethics’ literature search, while others were identified in the systematic literature review searches conducted to assess the clinical safety and effectiveness of RET mutation testing. Additional key texts in medical ethics ([Beauchamp & Childress 2001](#_ENREF_14); [Munson 2000](#_ENREF_139); [Rogers & Braunack-Mayer 2004](#_ENREF_171)) and web resources ([ALRC 2003](#_ENREF_7); [HGSA 2008](#_ENREF_91)) were also sourced.

### Genetic exceptionalism

The ‘exceptionalism’ debate began in 1991, with respect to HIV infection, during the evolution of HIV/AIDS policy ([Bayer 1991](#_ENREF_13)). Genetic exceptionalism, which propounds that genetic information is special and distinct from other forms of information, resulted in development of gene-specific privacy and discrimination policies in many countries ([Gostin & Hodge 1999](#_ENREF_83)). Several features of genetic testing support the concept of genetic exceptionalism:

1. Genetic testing for heritable conditions provides information that is private and personal but is also relevant to individuals other than just the tested individual. Test results have implications for family members, who may or may not wish to know their risk of suffering from a given disease.
2. Many tests are used to predict disease development that may occur many years into the future. Because of this, the psychological ramifications can be very different from the situation where the test is being used for a symptomatic patient.
3. There is concern that genetic information can be used to discriminate against individuals, as illustrated by a few incidences of discrimination by insurers and employers. Under Australian law an insurance company is unable to require that a genetic test is undertaken as a condition of insurance; however if a person has been genetically tested, they are obliged to inform the insurer of the outcomes of that test. Additionally, Australian law does not prevent an employer requiring a new or potential employee to provide a DNA sample.
4. Most patients have only a limited knowledge of genetics. Because of this, an informed consent process requires adequate counselling on an extensive array of issues. These include both standard considerations and those particular to individual and/or family circumstances ([Kinder 1998](#_ENREF_106)).

Genetic exceptionalism is not universally accepted. Some argue that genetic and non-genetic diagnostic and predictive testing feature more similarities than differences ([Diergaarde et al. 2007](#_ENREF_44); [Green & Botkin 2003](#_ENREF_84)), and that the introduction of genetic testing into medical practice does not fundamentally alter the ethical obligations of physicians to their patients. Results from non-genetic tests such as cholesterol levels, HIV status, alcohol or narcotic addiction, blood pressure and a family history of inheritable disease can be used in the same way as genetic test results to discriminate against and/or stigmatise individuals. Likewise, some of these results have the potential to either affect family members or cause psychological harm in the same way as genetic test results can. Thus, they raise the same ethical dilemmas concerning the preservation of autonomous choice, privacy and confidentiality as genetic testing, and should all be handled in the same way ([Green & Botkin 2003](#_ENREF_84); [Lazzarini 2001](#_ENREF_115); [Suthers 2008a](#_ENREF_194)). Some contend that the clinical integration of genetic risk assessment for common malignancies such as colon and breast cancer has negated the need for treating genetic information as special. This belief incorporates the view that many medical interventions are now reliant on genetic information in order to offer the best possible clinical care ([Offit & Thom 2007](#_ENREF_150)).

Despite these counterarguments to genetic exceptionalism, there is no broad acceptance that genetic information is the same as other kinds of medical information. The United Nations Educational, Scientific and Cultural Organization, which is concerned with moral issues in relation to science, developed normative international standards for the use of biomedical applications using the genetic exceptionalism approach ([Soini 2012](#_ENREF_192)). It also forms the basis of genetic-specific legislation governing privacy and discrimination in various countries. As a consequence, for this review we have adopted the conservative view of genetic exceptionalism—that genetic information is indeed unique and particularly vulnerable to misuse.[[10]](#footnote-10)

### Ethical framework

The philosophical approach adopted by this assessment is principlism because it is predominant within the field of biomedical ethics ([Beauchamp & Childress 2001](#_ENREF_14); [Munson 2000](#_ENREF_139); [Rogers & Braunack-Mayer 2004](#_ENREF_171)). Furthermore, no alternative approach was used in any of the papers included in this assessment. Recently it has been recommended that health technology assessments should incorporate a comprehensive ethical analysis ([Duthie & Bond 2011](#_ENREF_50)). Although a philosophical defence of principlism has not been possible within the confines of this assessment, it does not unduly undermine the assessment’s capacity to report on the main ethical issues as identified in the literature search. Nor does it preclude a reasonable understanding of the main issues identified.

### The ‘four principles’ approach

Principlism outlines four main principles—autonomy, non-maleficence, beneficence and justice—which are used to assess the ethical issues associated with genetic testing, as briefly described below.

#### Autonomy

Autonomy refers to self-rule. Individual autonomy is the governing of oneself and the directing of one’s own life, free from coercive interference on the part of others and from limitations that might prevent one from making meaningful choices. A respect for individuals’ autonomy entails that they have a right to self-determination or to act freely in accordance with a self-chosen plan. Such respect underpins the process of informed consent and education in medical care and research, and provides the basis for privacy of medical records ([Burke & Press 2006](#_ENREF_25); [Giarelli 2001](#_ENREF_77)). Most theories of autonomy agree that two conditions are required for autonomous choice—liberty, or independence from controlling influences; and agency, or the capacity for intentional action ([Beauchamp & Childress 2001](#_ENREF_14); [Winslow, Kodner & Dietz 2005](#_ENREF_212)).

#### Non-maleficence

Non-maleficence refers to not inflicting harm or injury to others, and is associated with the dictum *Primum non nocere*: ‘Above all (first) do no harm’. The principle also finds expression in the modern Hippocratic oath: ‘I will use treatment to help the sick according to my ability and judgement, but I will never use it to injure or wrong them’. In clinical practice the principle of non-maleficence is often combined with, and sometimes balanced against, the principle of beneficence, a version of which is expressed in the first half of the above Hippocratic oath ([Beauchamp & Childress 2001](#_ENREF_14); [Giarelli 2001](#_ENREF_77)). For instance, even the best diagnostic tests and treatments can carry certain risks of harm, and it is practically impossible for medical professionals to act without ever causing harm. Indeed, causing some harms may be warranted in the light of greater potential benefits. Hence, the avoidance of unwarranted or unnecessary harm, even if unintentional, is paramount to the non-maleficent conduct of health professionals. Inextricably linked to the concept of non-maleficence is the obligation to exercise ‘due care’, which is not always explicitly defined but rather implied in many professional codes of clinical practice. Aspects of non-maleficent practice that are implied in the clinician’s duty of care are neither more nor less important than those explicitly defined ([Munson 2000](#_ENREF_139)).

#### Beneficence

The principle of beneficence asserts that it is not enough to respect the autonomy of patients and to avoid causing them harm; in addition, clinicians and providers of health services should act in ways that actively promote the welfare of patients ([Kinder 1998](#_ENREF_106)). Just as there are standards of due care that explicitly and implicitly define appropriate conduct in the protection of patients from harm, so too are there explicit and implicit standards of beneficence. For example, an obvious expectation in medical care is the physician’s duty to help patients by providing appropriate treatment. More implicit is the wider societal expectation that physicians should make reasonable sacrifices for the sake of their patients. In the absence of a reasonable cause to act otherwise, a physician’s neglect of a patient requiring medical intervention understandably warrants the disapproval of that patient and of the physician’s colleagues, placing the ethical conduct of the physician in serious question even before potential legal ramifications are considered.

Practical constraints must be applied in acting beneficently. There are countless ways to promote the welfare of a patient, but the majority of people will distinguish between expectations that are reasonable and those that are not. In this way, whether or not clinicians fulfil their duty of beneficence relies on judgement and is constrained by various practical considerations. It is also constrained by the duty to act in accordance with other, sometimes conflicting, ethical principles ([Munson 2000](#_ENREF_139)). Ethical dilemmas arise precisely when one is torn when acting in accordance with two or more ethical principles that commend different courses of action.

#### Justice

Justice refers to treating individuals equally. In medical ethics the principle of justice finds expression in the belief that everyone deserves equal access to advances in medicine, and in the importance of fairness in the treatment of patients, particularly in the distribution of scarce resources. Different theories of justice focus on conditions of entitlement, fair and equal treatment, and concerns that the distribution of social goods such as healthcare occurs on the basis of relevant factors, for example degree of need, capacity to benefit and/or particular rights. Distributive justice concerns how resources are distributed, to whom and for what reasons. For instance, difficult choices are sometimes made between greatly benefiting the few (those with rare diseases) and benefiting to a lesser degree the many ([Giarelli 2001](#_ENREF_77); [Winslow, Kodner & Dietz 2005](#_ENREF_212)).

### The main ethical issues raised by RET mutation testing for MEN2

Questions relevant to ethical inquiry when assessing a health technology have been listed previously ([Hofmann 2005](#_ENREF_92)) and have provided valuable guidance for this assessment. However, the questions proposed by Hofmann have not been used as a ‘checklist’ on a question-by-question basis, as individual concepts cannot be logically separated. The emergent themes or issues are most comprehensibly captured when discussed in a collective manner.

The main ethical issues associated with genetic testing and their most relevant ethical principles are listed in Table 34 and discussed below.

Table 34 Main ethical issues for RET mutation testing and their most relevant principles

| Issue | Most relevant principle(s) |
| --- | --- |
| Informed consent | Autonomy, non-maleficence, beneficence |
| Privacy and confidentiality | Autonomy |
| Balancing risks and benefits | Non-maleficence, beneficence |
| Potential for discrimination | Justice |
| Access | Justice |
| Direct-to-consumer genetic testing | Non-maleficence, beneficence, autonomy |

#### Informed consent

Many people do not have a good understanding of genetics, and seeking informed consent for genetic testing poses particular challenges for clinicians and counsellors. Emphasis is placed on the need for an explicit agreement between the health provider and the patient. The basic elements of informed consent, adapted from guidelines of the American Society of Clinical Oncology ([Kinder 1998](#_ENREF_106)), are listed in Table 35. In line with the principle of respect for autonomy, clinicians and counsellors should stress that testing for a genetic mutation is completely voluntary and optional. The competence of the individual to be tested would need to be assessed, with information provided in a format that the patient can understand. Particular emphasis should be on the likely accuracy of the diagnosis or prediction and the fact that test results will not always provide definitive information about whether the development of disease will ensue. The limits of other methods for predictive testing, if applicable, would also need to be discussed.

Table 35 Elements to be considered when obtaining informed consent for genetic testing

| **Element** |
| --- |
| Autonomy provisions   * information on the specific test being performed * implications of a positive and a negative test result * possibility that the test will be inconclusive or not informative * options for risk estimation without genetic testing * risk of passing mutation to children * options to withdraw from study (in the case of genetic tests conducted for research) |
| Beneficence provisions   * options for medical surveillance, risk reduction and screening following testing |
| Non-maleficence provisions |
| * technical accuracy of the test * risks of psychological distress * risk of insurance or employer discrimination |
| Paternity provisions   * procedures if relatedness (i.e. paternity/maternity) is not as expected * procedures governing notification of family |
| Privacy—professional responsibilities   * confidentiality issues * fees involved in testing, counselling and follow-up care |
| Special considerations   * ownership and research uses of DNA remaining after testing * reproductive uses of genetic information |

Source: adapted from ([Offit & Thom 2007](#_ENREF_150))

In the context of hereditary MTC, information regarding an individual’s disease subtype (MEN2A, MEN2B or FMTC) and the tumours to which they are predisposed, along with the clinical implications of a specific RET mutation, should form an integral part of the pre-test counselling. Individuals should also be given information on non-genetic periodic screening or surveillance methods and their limitations and benefits. Although genetic testing of close relatives of individuals with RET mutations is now considered part of standard clinical care ([Burke, Pinsky & Press 2001](#_ENREF_24)), these individuals still need to be informed of the likelihood of tumour development, the mean age of onset of disease, available treatments and the long-term prognosis (both with and without genetic testing) associated with a familial RET mutation, so that they may make an informed decision.

Special concerns have arisen with regard to the situation where the intended recipients of genetic tests are unable to give informed consent, specifically children and embryos ([Offit et al. 2004](#_ENREF_149)). In Australia the genetic testing of children for clinical purposes is not regulated by legislation. However, the World Health Organization, the Nuffield Council on Bioethics and the American Society of Human Genetics have developed guidelines on the genetic testing of children ([ALRC 2003](#_ENREF_7)), and the Human Genetics Society of Australasia (HGSA) has published a position statement, *Pre-symptomatic and predictive testing in children and young people* ([HGSA 2008](#_ENREF_91)). In essence these guidelines and statements affirm that the predictive genetic testing of minors should only be conducted when there is an availability of treatment options that directly benefit the child. A study investigating the attitudes of clinical geneticists to predictive genetic testing in minors showed support for testing young children when it provides a clear medical benefit, such as in the case of MEN2 ([Borry et al. 2008](#_ENREF_20)). The elevated potential for developing MTC in early childhood and the curative nature of a thyroidectomy suggests that the issue of informed consent cannot safely be deferred until the child is of age. This means that it is important that parents with children at risk of having a RET mutation are well informed about the nature of the disease, the screening procedures that may be avoided if mutations are ruled out by the testing of family members, and disease treatment regimes, with an unbiased presentation of the risks and benefits.

There has been some discussion about the ethical justification for RET mutation screening to be added to existing mandated newborn screening programs, which allow parents to opt out but does not require consent ([Rosenthal & Diekema 2011](#_ENREF_174); [Shuman et al. 2012](#_ENREF_188)). RET mutation screening meets the ethical criteria for newborn screening proposed by the President’s Council on Bioethics and the traditional Wilson-Jungner criteria ([The President's Council on Bioethics 2008](#_ENREF_199); [Wilson & Jungner 1968](#_ENREF_211)). It was estimated that approximately 1,000 children born each year will develop hereditary MTC in their lifetime, and that 90% of at-risk newborns could be identified for early treatment with a newborn screening program (5% of individuals with MEN2A or MEN2B and 12% of those with FMTC have no identifiable RET mutation). The principles of beneficence support mandatory newborn screening for RET mutations as it removes decision-making from the parent. However, parental consent for a prophylactic thyroidectomy to prevent MTC would still be required. There have been many well-documented cases of parents refusing consent to medical procedures or treatment for their children ([Rosenthal & Diekema 2011](#_ENREF_174)).

An ethical dilemma arises when parents refuse to provide consent for genetic testing for children at risk of hereditary MTC. Parents have the right to make decisions with regard to their children’s health on the basis of their individual autonomy and beneficence ([Shuman et al. 2012](#_ENREF_188)). However, the non-maleficence principle only allows an individual complete autonomy over his/her own beliefs and actions as long as they do not cause harm to others. If a parent’s or guardian’s actions or decisions places a child in harm’s way, intervention is potentially justified ([Rosenthal & Diekema 2011](#_ENREF_174)). Theoretically, in cases where a delay in genetic testing could result in a delay in undergoing a prophylactic thyroidectomy to prevent disseminated cancer, legal intervention to overturn the parental decision could be sought. However, as the risk of harm is not immediate and does not constitute a medical emergency, repeated discussions and mediation should be attempted before overriding parental decision-making rights with regard to genetic testing for RET mutations. It should be noted that there is no legal precedent for overriding parental rights in hereditary MTC even though this may violate the ethical principles of beneficence and justice in certain cases ([Shuman et al. 2012](#_ENREF_188)).

The issue of prenatal testing introduces particular ethical considerations insofar as definitions of personhood are contentious. Ethical guidelines for Australian practice in the area of genetic testing at the embryonic stage of human development appear to be lacking; however, various medical associations in the USA and Europe have developed similar positions. The main message is that, while prenatal testing is usually considered acceptable in instances of increased risk of foetal genetic disorders, embryo selection to avoid genetic disease is not appropriate in all circumstances. It depends on the gestational period at which selection would occur, as well as other factors including the disease’s severity, probability of occurrence and age of onset ([Offit & Thom 2007](#_ENREF_150)). Testing for RET mutations in embryos would need to be considered relative to the best available guidelines. Information about the nature of the disease, screening procedures and disease treatment regimens should be provided to RET mutation carriers who are contemplating prenatal testing. Out of continuing respect for autonomy, these individuals should also be informed of the risk of conceiving affected offspring.

#### Privacy and confidentiality

The principle of autonomy affirms the right to voluntary genetic testing, entailing access to the best available evidence of risks and benefits. Furthermore, it affirms the individual’s right to privacy. While a patient may choose to reveal information, genetic test results must usually be kept confidential by medical personnel. In the case of inherited genetic conditions, keeping the results of a genetic test confidential protects an individual’s right to privacy; however, it also limits the ability of other family members to make informed choices with respect to their own health. Given that inheritable genetic disorders are both an individual and a family matter, ethical dilemmas can arise when a clinician is torn between maintaining the confidentiality of a patient’s test results and informing family members of their own corollary predisposition to disease ([Giarelli 2001](#_ENREF_77)).

Although healthcare professionals recognise the need to maintain confidentiality in most clinical scenarios, some circumstances exist in which disclosure may be permissible, even required. From a legal perspective, the courts have ruled that the duty to protect confidentiality is not absolute when a threat to a third party is considered ‘imminent’ and ‘serious’ ([Rosenthal & Pierce 2005](#_ENREF_175)). Judging which specific clinical situations warrant a breach of confidentiality remains one of the most difficult ethical issues raised by genetic testing.

Some authors have identified that the ‘duty to warn’ family members of genetic risk may be justified on the grounds that the clinician regards the entire family as the patient and, in this sense, revealing genetic information among family members does not represent a breach of confidentiality ([Rogers & Braunack-Mayer 2004](#_ENREF_171)). However, as per considerations of informed consent, counselling is required before test results can be disclosed. Counselling helps the initial test recipient understand and deal with information, but also to consider and state how much information they are prepared to share. Counselling the initial test recipient on the benefits of sharing information with close relatives, and in turn providing counselling to those relatives whether they are directly affected or not, is likely to require considerable skill and sensitivity.

Professional societies differ in their positions on confidentiality ([Shuman et al. 2012](#_ENREF_188)). The American Medical Association and the American Society of Clinical Oncology do not support violating privacy in any manner in order to notify family members of genetic risks. In contrast, the World Health Organization, the National Human Genome Research Institute, the American Society of Human Genetics and the US Institute of Medicine’s Committee on Assessing Genetic Risks, in addition to other national and international groups, support the ethical justification for disclosure in selected cases. It has been proposed that the following criteria must be met before a clinician contemplates any disclosure of genetic information ([Winslow, Kodner & Dietz 2005](#_ENREF_212)):

1. All attempts to bring about voluntary disclosure must be exhausted.
2. The seriousness of the harms posed by the genetic mutation must be imminent and certain.
3. Effective means of preventive or therapeutic intervention must be available.

These criteria would likely apply in instances where index cases with a RET mutation resist disclosure to their relatives. The harms risked by maintaining confidentiality are substantial and certain, as close relatives carrying the familial RET mutation will develop MTC if left untreated, and there is an effective treatment available by undergoing a prophylactic thyroidectomy. Consequently, in the case of hereditary MTC, a breach in confidentiality could be considered ethically justifiable.

While best practice is represented by striving to avoid breaking confidentiality, the Australian Government has enabled genetic counsellors to legally do so in serious cases through its 2006 amendment to the *Privacy Act 1988*[[11]](#footnote-11). This amendment allows disclosure of genetic information, without consent, to relatives provided such disclosure is ‘necessary to lessen or prevent a serious threat to the life, health or safety whether or not the threat is imminent’ ([Suthers, McCusker & Wake 2011](#_ENREF_194)). In 2009 the NHMRC developed guidelines that provide the formal mechanism for implementation of the new provisions under section 95AA of the Act, which practitioners must comply with ([NHMRC 2009b](#_ENREF_145)).

Individuals who undergo genetic testing are also likely to have concerns about who will have access to their test results, and how the information will be used and for what purposes. They may be particularly concerned about disclosure to third parties—the potential for health and life insurance companies, employers and financial institutions to use genetic information in order to discriminate against them. Confidentiality and privacy are of particular importance in this respect ([Beauchamp & Childress 2001](#_ENREF_14)). Some commentators have argued that the underwriting of health insurance premiums on the basis of genetic test results should not be an issue for Australian patients. Community rating dictates that all people pay the same rates for the same level of health cover, regardless of their health status and family history ([Delatycki 2008](#_ENREF_41)). Life insurance, on the other hand, is not afforded the same level of protection against genetic discrimination. This may be defensible if healthcare is considered a basic or fundamental good or right, whereas death benefits are considered a commodity. The reduction of life insurance to a commodity, and thus the perspective that it is very different from health insurance, is contestable.

Legislation has sought to deal with this problem—under Australian law, no applicant for insurance is required to undergo genetic testing. However, results of tests already taken must be disclosed and employers can request a DNA sample from new or potential employees. This has practical implications and the fear of insurance or employer actions does deter some people from being tested ([Wilcken 2011](#_ENREF_210)).

#### Weighing risks and benefits

Incorporating the principles of beneficence and non-maleficence means that risks should be minimised and benefits maximised before a genetic test is accepted into clinical practice. Thus, factors pertaining to the predictive value of the test, the benefit provided by interventions that are associated with a positive test result, the availability and acceptability of the interventions, and the possible harms posed by the knowledge of risk or by the interventions used to reduce risk must be evaluated ([Burke & Press 2006](#_ENREF_25)).

Risks associated with genetic testing are generally psychological and social, but by no means should risks to physical health be neglected. For example, tests that erroneously present an individual as a non-carrier of a mutationin the *RET* gene may result in substantial physical harms to that individual, especially in the case of pre-symptomatic testing. Pre-symptomatic individuals will be afforded a false sense of security about their risk and will almost certainly miss the opportunity for screening or surveillance procedures that offer the potential for early detection and intervention against disease.

On the other hand the likely ramifications of a false *negative* test in a symptomatic individual seeking to confirm a clinical diagnosis are that the patient will experience some level of anxiety about symptoms that are not supported by a genetic diagnosis. In this case the potential for harm could be extended to include close relatives who are at risk of carrying an inherited mutation but will not be offered predictive testing due to the false negative test result of the index case.

When it comes to the psychologically harmful effects of a positive genetic test result, the risk must be weighed against the potential benefit of information that can lead to targeted surveillance, preventive measures and/or more-specific and -effective treatment ([Offit & Thom 2007](#_ENREF_150)). Respecting a patient’s wish to keep genetic information private minimises the risk, however small to begin with, that disclosed information will lead to the patient being discriminated against and to attendant psychological, social and economic harms.

Historically, genetic information has been used by civilizations to discriminate against individuals and groups, such as Jewish people, African Americans and other ethnic minorities. Hence, there is considerable concern that individuals with positive genetic test results, and therefore known to suffer from conditions of a hereditary nature, may be stigmatised by society, as seen for AIDS patients in the 1980s ([Green & Botkin 2003](#_ENREF_84)). Societal acceptance of genetic disorders cannot be regulated by law; it can only be ameliorated through education of the public ([Winslow, Kodner & Dietz 2005](#_ENREF_212)).

#### Access issues

Individuals who undergo genetic testing deserve justice in resource allocation. Also, access to treatment should be provided in the event that results provide evidence of a RET mutation. In Australia we are lucky that there is fairly equitable access to medical services, with perhaps the exception of rural and remote communities ([Wilcken 2011](#_ENREF_207)). Nevertheless, the allocation of resources and the resolution of other access issues may create ethical problems for policy makers. They must consider issues such as how cost-effective the treatment should be before publicly funding the Ret mutation testing, and to what extent genetic counselling should be offered as part of the testing process ([Wilcken 2011](#_ENREF_207)). Currently, most molecular tests are expensive and are not included in the MBS. For this reason suspected MEN2 syndrome patients from both rural and remote areas and metropolitan centres would be encouraged to have their blood sample collected through a public hospital so that this facility is charged for the testing. When patients are referred by a private facility they are billed directly and must cover the entire cost themselves. Moreover, no subsidies are offered for pre-implantation genetic testing, which is often preferred by families to avoid the risk of having to abort an affected foetus and which is prohibitively expensive for most people ([Wilcken 2011](#_ENREF_210)).

Five accredited laboratories offer RET mutation testing in Australia ([Suthers 2008b](#_ENREF_195)), although the Royal College of Pathologists of Australasia’s genetic testing website currently lists only three of these[[12]](#footnote-12). It is expected that referral overseas would not be a common occurrence given the relatively low demand for this service. With current funding for RET mutation testing being provided either by the state/territory governments, where testing may be limited by budgetary constraints, or at a personal cost to the patient, it is probable that not all symptomatic patients or at-risk relatives are being tested, and this raises questions of justice. Listing RET mutation testing on the MBS should increase access to the test for all individuals who require it.

It should be noted that the quality of genetic testing varies significantly ([Offit & Thom 2007](#_ENREF_150)) and that RET mutation testing is no exception. The test available from the Cancer Genetics Diagnostic Laboratory of PaLMS, Royal North Shore Hospital in Sydney, New South Wales, offers PCR and sequencing of RET exons 10, 11 and 13–16; the Molecular Pathology Division of the Peter MacCallum Cancer Centre, Victoria, offers targeted RET mutation analysis; and the Molecular Pathology Division of the IMVS in Adelaide, South Australia, offers a gene screen for all exons and associated splice junctions by direct sequencing. Thus, the range of RET mutations that can be detected may differ between laboratories due to differences in detection methods. This may result in a number of patients with rarer types of RET mutations that cannot be detected by some laboratories being misdiagnosed and receiving a false negative test result.

#### Direct-to-consumer testing

There is no clear requirement for direct-to-consumer (DTC) genetic testing companies to offer proper and effective pre-test genetic counselling or psychiatric evaluation, and they lack appropriate regulatory oversight to prevent deceptive practices and ensure a quality product ([Caulfield & McGuire 2012](#_ENREF_30); [Wilcken 2011](#_ENREF_210)). Hence, ethical concerns have been raised about the potential harm of DTC services to consumers, implications for the health system and privacy issues related to the commercial storage of genomic data ([Caulfield & McGuire 2012](#_ENREF_30)).

According to the principle of non-maleficence, the potential for psychological and physical harm caused by DTC genetic testing should be minimised. However, in the absence of appropriate genetic counselling, the results could cause anxiety or lead to an inappropriate behavioural response, either because individuals overinterpret the significance of a positive result or gain a false sense of security from a negative result. There is also potential for psychological and physical harm caused by inappropriate clinical decisions based on an inaccurate performance or interpretation of genetic tests ([Caulfield & McGuire 2012](#_ENREF_30); [Offit & Thom 2007](#_ENREF_150)).

The principle of autonomy advocates that informed consent be obtained from each individual tested and guarantees their right to privacy. It is disconcerting that very few DTC companies have reasonably comprehensive privacy policies for protection of personal information and DNA samples ([Caulfield & McGuire 2012](#_ENREF_30)). There is no guarantee of continued privacy if a DTC company is sold, goes out of business or becomes bankrupt. The new owner of the stored genetic information and/or DNA samples may not feel bound by previous privacy arrangements, raising numerous concerns about what could happen to private information in a commercial setting ([Wilcken 2011](#_ENREF_210)).

In Australia symptomatic patients and their at-risk relatives are unlikely to use DTC genetic testing as most would have testing funded through a public hospital. However, a non-symptomatic individual using such a service may be identified by chance as a RET mutation carrier. These individuals would then require further medical treatment and/or counselling to deal with this finding. The medical practitioner would also need to determine the likely accuracy of the genetic testing that was conducted by the DTC company as well as any further treatment, surveillance or repeat genetic testing that is needed for their ongoing clinical management.

### Summary

The consensus standpoints for a range of ethical issues and, in some cases, dilemmas raised by genetic testing are summarised as follows:

* On balance, RET mutation testing appears ethically acceptable provided that it is both preceded and followed by adequate counselling on, among other things, the limitations and significance of test results, including the possible ramifications for family members and the possible courses of effective treatment should a test result be positive:
  + RET mutations identify MEN2, for which there is an effective prophylactic treatment option.
  + Counselling is necessary in order to ensure informed consent and minimise risks of harm, both psychological and, in the longer term, physical.
* Test results should remain confidential, although the patient should be counselled on the benefit of sharing information with family members who may benefit.
* As always, confidentiality should be broken only if risks to others are serious, imminent, certain and avoidable, and attempts at encouraging voluntary disclosure have been exhausted.
* Testing should be available, and not overly financially burdensome, to all who might benefit from it.
* Direct-to-consumer genetic testing appears to carry substantial risks.

### Conclusion

With respect to RET mutation testing, the above ethical analysis would suggest that the test should only be offered on the MBS if it is performed in conjunction with genetic counselling from accredited counsellors with familiarity in both interpretation of RET mutation test results and management of the implications for the index case and family members.

# What are the economic considerations?

In assessment of a new service MSAC is required to consider not only the comparative effectiveness and safety of the service but also the comparative cost and cost-effectiveness of the service. Thus, an economic evaluation is required, which is based on the clinical evidence for this service when added to or substituted for the main comparator in the relevant setting.

However, because RET mutation testing is *normal practice* for patients who currently present with symptoms that are potentially associated with MEN2 disease or hereditary RET mutations*,* and is currently funded through state hospitals/governments, an economic comparison between MBS-listed RET mutation testing and current clinical practice would not be expected to show incremental effects in either clinical outcomes or resource expenditure. Rather, it would simply highlight the transfer of costs from one healthcare funder to another. This does not provide information to MSAC on the inherent economic value of RET mutation testing.

The purpose of an economic evaluation is to inform MSAC as to the additional costs and additional gains (health or other socially relevant outcomes) of the proposed service over the comparator when used in the Australian healthcare system. In this context, where clinical practice has already adopted the technology irrespective of the funding source, the comparator has been nominated as the ‘historical clinical scenario’ (i.e. when RET mutation testing was not available). The following analysis is therefore intended to assist MSAC to ensure that society’s health resources are allocated to those activities from which it will get the most value.

Two sets of economic analysis were undertaken to consider the two distinct applications of RET mutation testing (diagnostic and familial screening). With consideration of diagnostic testing of a patient presenting as an index case, a cost-minimisation approach is undertaken; and with respect to familial screening, a cost-utility model is undertaken.

Since the publication of the DAP

Updated information on the current pricing of RET mutation testing in Australia has been obtained (see Table 6 on page xxxiv). In this report the base-case economic evaluations have assumed a lower revised fee associated with testing, based on current prices.

The evidence base identified in the assessment provides little directly comparative and/or non-confounded data. The economic modelling is therefore undertaken on the basis of the available data with various assumptions. Where assumptions are made, these will be explicit and, in the base case, tend toward a conservative approach. Given the limitations of the available data, all quantification within the economic evaluation may be considered indicative but uncertain.

## Economic evaluation

### Background literature

A literature search identified four published economic evaluations of RET mutation testing (Table 36).

Table 36 Published economic evaluations of RET mutation testing

| **Publication** | **Setting** | **Model & results** |
| --- | --- | --- |
| Delbridge L and Robinson B. (1998) Genetic and biochemical screening for endocrine disease: III. Costs and logistics ([Delbridge & Robinson 1998](#_ENREF_42)) | Ret mutation testing of family members of known MEN2A families in Australia | Cost-effectiveness model consideration of pentagastrin (biochemical screening) vs. no screening, and RET mutation testing vs. no testing  Results: No testing: $0/lives saved; risk of dying 117/1,000  Pentagastrin testing: $76,315/life saved; risk of dying 22/1,000  RET mutation testing $5,175/life saved; risk of dying 3/1,000 |
| Erlic Z, Rybicki L, Peczkowska M, et al. (2009) Clinical predictors and algorithm for the genetic diagnosis of pheochromocytoma patients ([Erlic et al. 2009](#_ENREF_60)) | Genetic screening of index patients presenting with phaeochromocytoma | The model considers the costs of screening all potential index cases presenting with phaeochromocytoma for genetically relevant germline mutations to diagnose Von Hippel-Lindau Disease (VHL), MEN2 (RET), paraganglioma syndromes 1, 3 and 4 (SDHD) and type 1 neurofibromatosis (NF1). Taking into account expected diagnostic yields, the model determines the most cost-effective order for sequential testing, and also considers further cost reductions obtainable by pre-selection of at-risk patients. |
| Gilchrist DM, Morrish DW, et al. (2004) Cost Analysis of DNA-based testing in a large Candian family with multiple endocrine neoplasia type 2 ([Gilchrist et al. 2004](#_ENREF_79)) | RET mutation testing of a specific family group with MEN2 history, in Canada | The authors conduct a cost analysis of RET mutation testing vs. clinical surveillance in the specific scenario of a family with a history of MTC. In this case the family was large and 58 family members were tested for germline RET mutations, in which only four members were identified as carrying RET mutations. It is concluded that RET mutation testing has resulted in significant cost savings in comparison with biochemical screening in this example. |
| Pigny P, Cardot-Bauters C, Do Cao C, et al. (2009) Should genetic testing be performed in each patient with sporadic pheochromocytoma at presentation? ([Pigny et al. 2009](#_ENREF_158)) | Patients presenting with phaeochromocytoma, with no family history or concomitant disease, in France | This cost analysis of genetic testing for RET, VHL and NF1 (vs. alternative clinical/biological/imaging investigations) in 100 patients presenting with sporadic phaeochromocytoma concludes that cost-savings can be achieved without negative clinical outcomes if routine genetic screening for hereditary phaeochromocytoma is excluded in patients with unilateral adrenal tumour diagnosed after the age of 50 years. |

The Delbridge and Robinson model is based on Australian clinical practice and is relevant to the proposed listing of RET mutation testing for family members, although the resource prices from the 1990s may require updating. This model uses a decision analytic to demonstrate that, where a family history has been established, RET mutation testing appears to be both more effective (i.e. it decreases the risk of dying) and less expensive (per life saved) than biochemical surveillance. However, the model does not consider initial RET mutation testing in potential MEN2 index cases and does not present results in the form of an incremental cost-utility analysis with an estimate of QALYs or life-years gained.

The Gilchrist et al. model is specific to a single, particularly large family and the Canadian healthcare system, and is not generally applicable to Australia at a population level. While the Pigny et al. analysis supports the proposed age restriction on RET mutation testing in phaeochromocytoma, it does not provide cost-effectiveness information on the use of RET mutation testing in the proposed listings.

Although consistent in their conclusion that RET mutation testing is cost-effective in the respective setting of analysis, none of the published models are adequate to confirm the cost-effectiveness of the proposed MBS listings of RET mutation testing in both potential index cases and family members in the Australian population. Therefore, further economic modelling of costs and outcomes based on the proposed listings in the intended Australian population is required.

### Structure

Economic analyses of costs and outcomes associated with RET mutation testing are undertaken for the four following scenarios (as requested in the DAP):

1. index patients presenting with MTC
2. index patients younger than 50 years of age presenting with phaeochromocytoma
3. first- (or second-)degree family members of patients presenting with MTC
4. first- (or second-)degree family members of patients younger than 50 years of age presenting with phaeochromocytoma

#### Scenarios 1 and 2 – Cost-minimisation analysis

In the case of diagnostic RET mutation testing in potential index patients (Scenarios 1 & 2), there is no evidence available to suggest a direct benefit in health outcomes to the index patient who is already presenting with symptoms. Equally, because no harms are associated with testing, it is assumed that there will be no change in clinical outcomes in either direction for the index patient. Therefore, the approach taken for these scenarios is a cost-comparison (cost-minimisation). In these scenarios the costs of RET mutation testing all potential index cases younger than 50 years of age presenting with either MTC or phaeochromocytoma, and subsequently commencing routine annual surveillance for additional MEN2 complications in those patients identified to have a RET mutation, are compared with the costs of providing routine annual surveillance for MEN2 complications in all patients younger than 50 years of age presenting with MTC or a phaeochromocytoma, because MEN2 could not be ruled out.

A diagrammatic representation of the structure of the cost-minimisation analysis of RET mutation testing in potential index patients *presenting with MTC* is shown in Figure 9.

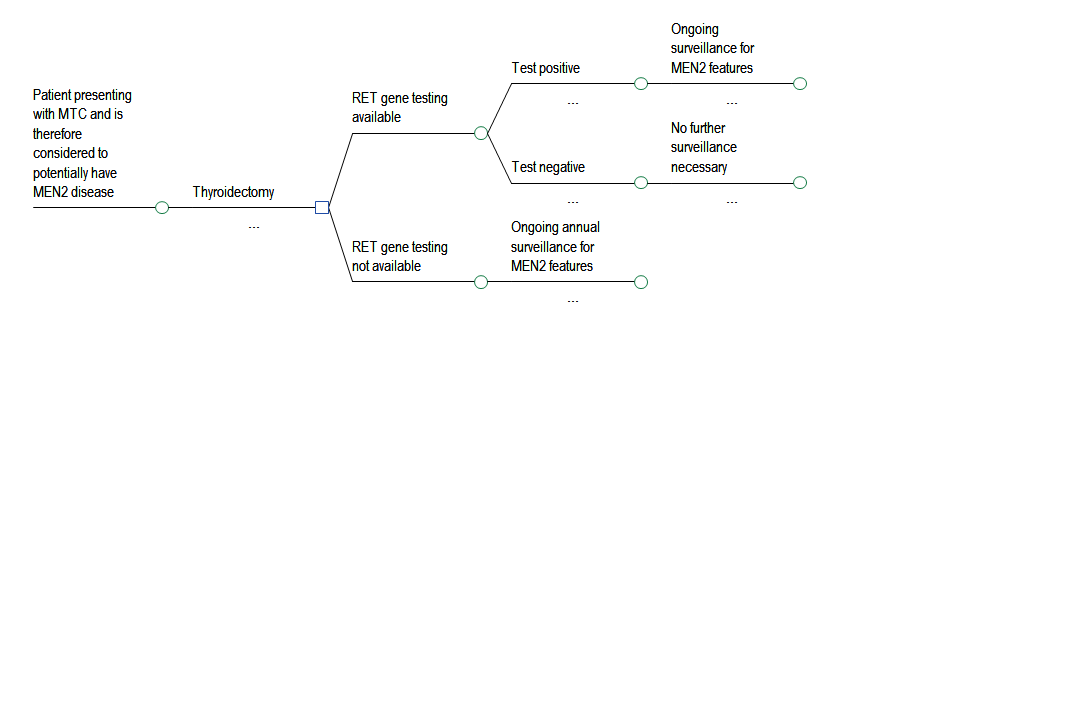


Figure Structure of cost-minimisation comparisons of RET mutation testing and selective surveillance versus non-selective surveillance for MEN2 complications, in patients presenting as potential index cases (Scenarios 1 and 2)

A diagrammatic representation of the structure of the cost-minimisation analysis of RET mutation testing in potential index patients younger than 50 years of age *presenting with phaeochromocytoma* is shown in Figure 10.

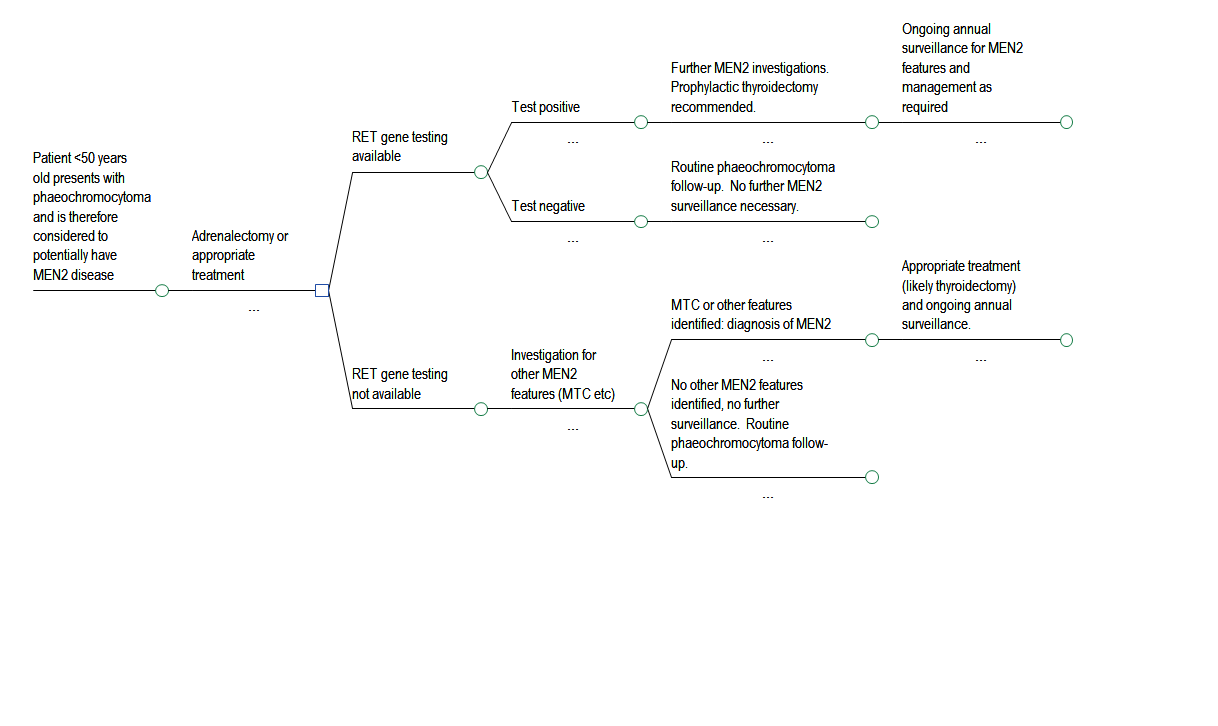


Figure Structure of cost-minimisation comparisons of RET mutation testing and selective surveillance versus non-selective surveillance for MEN2 complications, in patients younger than 50 years of age presenting with phaeochromocytoma (Scenarios 1 and 2)

The cost-minimisation analysis considers only the costs and downstream costs of the proposed diagnostic test (full screen of relevant exons) for RET mutations, and ongoing clinical investigation/surveillance for other MEN2-related conditions in the patient presenting with potential MEN2A.

The incidence of MEN2A and the extent and costs of downstream *treatments* associated with the management of the presenting complaint (MTC or phaeochromocytoma) in the potential index case (confirmed or not as MEN2) will occur identically, irrespective of the availability of RET mutation testing; if these were to be included in the economic evaluation, they would ‘cancel out’ in each arm. Therefore, they have not been included in the cost-minimisation analyses of Scenarios 1 and 2.

The cost-minimisation models for Scenarios 1 and 2 run over 30 years.

#### Scenarios 3 and 4 – Cost-utility analysis of the availability of diagnostic RET mutation testing in suspected index cases and subsequent prognostic familial screening in first- and second-degree relatives of affected cases, versus non-availability of RET mutation testing.

The third and fourth economic analyses are cost-utility models that attempt to measure the incremental difference in clinical outcomes and resource use across the broader population cohort relevant to the proposed listing of the prognostic screen for family members. The modelled cohorts are the suspected index cases *and their potentially at-risk family members* because, given the serious and known hereditary nature of MEN2 diseases, historical clinical practice advised that all family members of a potential MEN2 patient should also receive ongoing medical surveillance for symptoms of disease (MTC, phaeochromocytoma, hyperparathyroidism). The second proposed MBS listing is for RET mutation testing in first- and second-degree family members of patients identified as RET-mutation-positive.

The comparison modelled (with index cases being (i) MTC or (ii) phaeochromocytoma at younger than 50 years of age) is:

RET mutation testing (index and family) available—index cases are tested; RET-mutation-negative index cases and their families require no further surveillance for MEN2/RET associated diseases. RET-mutation-positive index cases maintain ongoing annual surveillance for other MEN2 complications and their first- and second-degree family members are also tested for hereditary RET mutations. RET-mutation-negative family members require no surveillance for MEN2, while RET-mutation-positive family members are recommended prophylactic thyroidectomy where appropriate and ongoing annual surveillance.

versus No RET mutation testing available—all potential index cases and family members of potential index cases are advised to receive ongoing annual surveillance for MEN2 complications, but a MEN2 diagnosis would only be made with the development of additional clinical symptoms or positive biochemical test results or imaging studies. Prophylactic/treatment thyroidectomy occurs as indicated.

The key assumptions that drive the model are:

* + that identification of RET-mutation-negative cases (index and/or family members) will substantially decrease the resource use and patient anxiety associated with what can subsequently be identified as unnecessary patient surveillance.
  + that RET mutation testing enables earlier detection of MEN2 disease and, once identified as RET-mutation-positive, the risk–benefit profile of prophylactic thyroidectomy becomes more favourable. Therefore, prophylactic surgeries can be undertaken more often and sooner. Surgery at a younger age increases the likelihood that the thyroidectomy will more effectively cure or prevent future MTC, thereby increasing both survival and quality of life.

The health states associated with RET mutation testing and MTC included in the model are shown in Figure 11.

Health state transition diagram for potential MEN2 patients (i.e. family members of known MEN2 patient)
Potential patients can be healthy, stay healthy, transition to MTC symptoms, transition to thyroidectomy, to no apparent disease or ongoing MTC diseaes, and death. 

Figure Health state transition diagram for potential MEN2 patients (i.e. family members of known MEN2 patient)

While other health states including phaeochromocytoma, hyperparathyroidism etc. may also be associated with MEN2, there is inadequate evidence to model any effect of RET mutation testing on the prevalence or pattern of these conditions; therefore, they do not need to be included in this model, which is intended to identify incremental effects only.

The cost-utility model runs over a ‘lifetime’ time horizon (70 years); however, results for shorter time horizons (10, 20 and 50 years) are also presented.

Discounting at a rate of 5% *per annum* is applied to costs and outcomes in all models.

Each model is calculated beginning with a cohort of 100 cases presenting with MTC/phaeochromocytoma, and assumes that these patients are an average age of 40 years.

Sensitivity analyses also consider the inclusion of familial screening in the cost-minimisation, although there is an argument that clinical outcomes may change (be improved) with familial screening; therefore, this is more thoroughly investigated with a cost-utility analysis.

**Inputs**

The inputs used in the economic models are from various sources which are detailed in Table 37 to Table 43.

The demographic and diagnostic parameters relating to the patient and family populations are detailed in Table 37. Transition probabilities between health states are detailed in Table 38. With respect to population and disease characteristics the median of results compiled from multiple sources in the systematic review generally informed the base-case inputs, unless an alternative justification is given. Where the data from studies was inconsistent or wide ranging, alternative inputs are tested in the sensitivity analyses. Where possible, inputs based on assumptions are also tested in sensitivity analyses.

Table 37 Inputs used in the economic models relating to population, disease and test characteristics

| **Variable** | **Base case** | **Source and discussion** | **Sensitivity analyses** |
| --- | --- | --- | --- |
| Discount rate | 5% | Convention used by Department of Health and Ageing | Nil |
| Diagnostic yield of RET mutation test in patients with MTC | 30.6% | Median based on 6 studies presenting diagnostic yield from unspecified cases of MTC (Table 83) |  |
| Diagnostic yield of RET mutation test in patients younger than 50 years of age with phaeochromocytoma | 18.3% | The proportion of people with phaeochromocytoma identified as RET M+ in [Krawczyk et al. (2010](#_ENREF_110)). This estimate is highly uncertain. There is no data identifying the prevalence of RET mutations that specifically considers patients younger than 50 years of age as a single risk factor. Alterative estimates based on the findings in this report on the incidence of mutation in phaeochromocytoma include a median of 7.8% in unspecified phaeochromocytoma cases (not defined by age) and 24.4% in cases with familial history (Table 84). These are tested in sensitivity analyses. | 7–25% |
| Diagnostic yield of predictive RET mutation test in first- and second-degree relatives of known RET M+ patients | 39.4% | Median based on 48 studies including second-degree relatives / not specifying relationship (Table 85); similar to median of studies of first-degree relatives only (37.5%, n=11). Biologically, 50% inheritance rate, but expect less due to *de novo* cases and some affected patients already diagnosed/dead | 50% |
| Number of family members per index patient | 11.4 | Familial Cancer Unit of South Australia | 5–15 |
| Compliance rate for family-member surveillance in untested population | 40% | Assumed that surveillance rate would be approximately equal to test uptake rate | 10–90% |
| Familial screening uptake rate | 40% | Suthers et al. (2006) | 10–90% |

RET M+ = RET-mutation-positive

Table Transition probabilities used in cost-utility analysis (applicable to family members only)

| **From health state** | **To health state** | **Probability** | **Discussion** |
| --- | --- | --- | --- |
| Healthy and undertaking either RET mutation test and/or biochemical investigation and surveillance | Post thyroidectomy | Where RET testing available: 100% of RET M+ patients are assumed to receive prophylactic thyroidectomy in the following year.  Where RET testing is not available: 70% of RET M+ patients are assumed to receive prophylactic thyroidectomy.  In the scenario of the index patient presenting with MTC, this is assumed to occur over the following 3 years (30%, 20%, 20% in each year).  In the scenario where the index presentation is phaeochromocytoma, all prophylactic thyroidectomies are assumed to occur in the following year (i.e. 70%). | The assumption that all patients identified as RET M+ receive prophylactic thyroidectomy is based on current recommendations ([Brandi et al. 2001](#_ENREF_21)). However, less than 100% is commonly reported and thyroidectomy uptake rates vary considerably between studies (Table 98), such that modelling 100% thyroidectomy results in a conservative estimate of the maximum likely additional surgical costs associated with testing.  The assumption that only 70% of RET M+ patients receive early thyroidectomy where RET mutation testing is not available is based on the symptomatic penetrance rate of MTC. This is conservative with respect to the allocation of costs of thyroidectomy but does not bias the results with respect to outcomes, as the 30% of people who do not receive the thyroidectomy are assumed to have normal survival. In the MTC index case scenario the assumption that most surgery is performed over the 3 years following initial investigations and monitoring is consistent with the average age of surgery being a few years later in the pre-RET mutation testing era (Table 29). In the model where index patients present with phaeochromocytoma, identification of a RET mutation or signs of MTC in family members is assumed to require management in the year immediately following diagnosis, as (relatively) early manifestation of phaeochromoytoma suggests a patient group with a higher risk mutation ([Moline & Eng 2011](#_ENREF_3)) |
| Healthy with no surveillance | Symptomatic / late-stage disease | Occurs in RET M+ people only: 70% at year 15 | Year 15: (symptoms, thyroidectomy) – Post-thyroidectomy penetrance of symptomatic MTC is ~70%. Undetected MTCs in non-thyroidectomised patients have been assumed to become apparent at year 15. |
| Healthy (no surveillance)  OR post-thyroidectomy | Death | Background mortality rate less people developing symptomatic disease. | Based on average mortality of people (men + women), starting at 40 years of age in year 1, from the Australian Bureau of Statistics ([ABS 2012](#_ENREF_3)). In patients who have had a thyroidectomy: in the RET era 90% are assumed to be successful (i.e. long-term cure); in the pre-RET era 60% are assumed to be successful (based on the findings in Rohmer et al (2011) and the overall RR estimate derived in Results - Figure 7). 'Non-successful' thyroidectomies are not apparent until year 15. |
| Symptomatic / late-stage disease | Death | Kaplan Meir post-thyroidectomy survival curve from Kakudo et al. (1985); with linear extrapolation beyond 30 years. | Kakudo et al. (1985) (stage I, n=19; and stage III, n=27) case series ([Kakudo, Carney & Sizemore 1985](#_ENREF_97)) |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive

Resource costs in the model are described in Table 39 to Table 43. These tables describe the application of the costs as well as their derivation and source. With the exception of pentagastrin ampoules (for which a current local cost could not be obtained), all resources costs used in the economic model are from current published Australian sources.

Table Genetic testing costs applied to index patients and / or family members of known index cases in the ‘RET mutation testing available’ arms (all models)

|  | **Costs** | **Derivation** | **Application** |
| --- | --- | --- | --- |
| RET mutation testing in potential index cases | $436.30 | Comprises of:  RET mutation test (full set of relevant exons) $400  Genetic counselling $36.30 | One-off cost in year 1 for each patient presenting with MTC or phaeochromocytoma. |
| RET mutation testing in first- and second-degree family members of known index cases | $236.30 | Comprises of:  RET mutation test (known mutation) $200  Genetic counselling $36.30 | One-off cost in year 1 for each family member of confirmed index case who undergoes genetic testing |

MTC = medullary thyroid carcinoma

Table Initial investigations and ongoing annual surveillance costs for index patients presenting with MTC, with unknown (untested) or positive RET mutation status.

|  | **Costs** | **Derivation** |
| --- | --- | --- |
| Investigation for phaeochromocytoma | $83.35 | Plasma/urine catecholamines: (described in MBS Item 66695), but combined with ≥5 tests, therefore MBS item 66707 used |
| Investigations for hyperparathyroidism | $9.70 | Serum calcium (MBS Item 66500) +/- parathyroid hormone (described in MBS 66695) but this is combined with ≥5 tests, included in MBS 66707 above |

MBS = Medicare Benefits Schedule

Table Initial investigations and ongoing annual surveillance costs for index patients presenting with phaeochromocytoma, with unknown or positive RET mutation status

|  | **Costs** | **Derivation** |
| --- | --- | --- |
| Investigations for MTC | $152.80 | Calcitonin (described in MBS Item 66695, but combined with parathyroid below), 2 tests, MBS 66698: $43.70  Neck (thyroid) ultrasound (MBS 55032) $109.10. |
| Additional costs for pentagastrin-stimulated calcitonin test (no RET mutation testing available arm) | $302.00 | Pentagastrin ampoule $241, from [Gilchrist et al. (2004](#_ENREF_79)) Calcitonin; baseline (included in sample above) and at 3 minutes and 5 minutes: (MBS 66695) $30.50 x2 |
| Investigate for hyperparathyroidism | $0 | Serum calcium (MBS Item 66500) assumed to be included in patient episode cone and not reimbursed +/- parathyroid hormone (described in MBS 66695) but as it is combined with 2 tests, included in MBS 66698 above |

MBS = Medicare Benefits Schedule; MTC = medullary thyroid carcinoma

Table Initial investigations and ongoing annual surveillance costs for first- or second-degree family members of index patients, with unknown or positive RET mutation status, compliant with testing/surveillance recommendations

|  | **Costs** | **Derivation** |
| --- | --- | --- |
| Consultation | $36.30 | *MBS Item 23* |
| Investigate for phaeochromocytoma | $83.35 | Plasma/urine catecholamines: (described in MBS Item 66695), but combined with ≥5 tests, therefore MBS item 66707 used |
| Investigate for hyperparathyroidism | $9.70 | Serum calcium (MBS Item 66500)  +/- parathyroid hormone (MBS 66695) but as it is combined with ≥5 tests, included in MBS 66707 above |
| Investigate for MTC | $109.10 | Neck (thyroid) ultrasound (MBS 55032) $109.10 Calcitonin (MBS 66707), but ≥5 tests required, therefore included in MBS 66695 above |
| Additional costs for pentagastrin-stimulated calcitonin test (no RET mutation testing available arm only.) | $271.50 | Pentagastrin ampoule $241, from [Gilchrist et al. (2004](#_ENREF_79))  Calcitonin; baseline (included in sample above) and at 3 minutes and 5 minutes: (MBS 66695) $30.50 (additional test assumed to be included in patient episode cone). |

MBS = Medicare Benefits Schedule; MTC = medullary thyroid carcinoma

Table Thyroidectomy and post-thyroidectomy costs in family members of index patients receiving prophylactic thyroidectomy (following positive genetic or biochemical test) or treatment thyroidectomy (after detection of MTC).

|  | **Costs** | **Derivation** |
| --- | --- | --- |
| Thyroidectomy (one-off) | $5,491.59 | Includes:  Thyroidectomy (MBS Item 30296) $1,023.70 Pre-anaesthetic consultation (MBS Item 17615) $85.55 Anaesthesia (MBS Item 20320) $118.80  Age modifier for anaesthesia (MBS item 25015) $19.80  Assistant (MBS item 51303) $204.74 Hospital accommodation for thyroid operation (Private hospital Inpatient NHCDCa cost weights for K06Z: ALOS 2.04 days) $4,039.00 |
| Post-thyroidectomy (annual) | $54.22 | Thyroxine tablets (PBS Item 2173J $27.11, 200mcg x200 tabs), required 2x prescriptions/year (assumed) |
| Ongoing surveillance for other MEN2 symptoms following development of MTC, *in patients previously non-compliant with testing/surveillance recommendations* | $129.35 | Includes:  Consultation (Item 23) $36.30 Investigation for phaeochromocytoma (Plasma/urine catecholamines (≥5 tests described in MBS 66695) (MBS Item 66707): $83.35 Investigation for hyperparathyroidism (Serum calcium, MBS Item 66500, $9.70 +/- parathyroid hormone, included in MBS 66707). |

ALOS = average length of stay; MBS = Medicare Benefits Schedule; MTC = medullary thyroid carcinoma

a National Hospital Cost Data Collection private hospital costs, AR-DRG version 5.1 round 13 (2008–2009) [Round 13 (2008-09) Cost Report](http://www.health.gov.au/internet/main/publishing.nsf/Content/Round_13-cost-reports)

The survival curve following thyroidectomy as presented by Kakudo et al. (1985) is shown in Figure 12.

A survival curve following thyroidectomy, showing 100% at 0 years, 90% at 3-8 years, 50% at 18-25 years, 25% at 24 years. 

Figure Cumulative survival curve for 46 patients following thyroidectomy for MTC by Kaplan-Meier method from Kakudo et al. (1985)

#### Pentagastrin ampoules

The injectable drug pentagastrin used in the calcitonin stimulation tests to identify early MTC prior to availability of RET mutation testing is not routinely available in Australia. No current pricing of this drug was available through routine wholesaler enquiries; therefore, the estimated cost of $241 per biochemical test (including injection-associated consumables) is based on the unadjusted value (in Canadian dollars) used in the published study by Gilchrist et al. (2004). While the currency exchange rates are similar, given inflation, differing healthcare systems and importation costs, this value is uncertain and may be an underestimate. If the cost of pentagastrin is underestimated, this will result in the model underestimating both the costs of biochemical surveillance and the cost savings associated with RET mutation testing.

#### Test accuracy

Test accuracy data is not included in the model and this is a significant shortcoming. Although the RET mutation test is assumed to be relatively accurate, there are reports of false positives, and the negatives do not necessarily rule out MEN2 disease. Given the robustness of the model to identifying both cost savings and health benefits, it is unlikely that the incorporation of sensitivity/specificity data—if available—would change the overall conclusion of economic effectiveness, although it is theoretically possible if the test performance is poor enough. While this assumption of 100% test accuracy does not completely reflect reality, it does not necessarily introduce a bias favouring RET mutation testing into the economic model—equally, the accuracy of biochemical surveillance is excluded from the model, and this too is imperfect. Learoyd et al. (1997) and other publications report on a number of known cases of false positives associated with biochemical monitoring, with estimations that the rate of false positives associated with pentagastrin stimulation may be as high as 10% (Costante et al 2009). False positives associated with biochemical monitoring would increase the costs and decrease the health outcomes associated with the comparator. If, in reality, the rate of false positives is lower with RET mutation testing than with biochemical monitoring, then the omission of this information from the model is a source of bias against RET mutation testing and a further area where the model is conservative with respect to estimates of the incremental benefits associated with RET mutation testing.

#### Palliative care

An accurate estimation of palliative care costs associated with advanced MTC in the Australian setting was not obtained and these costs have not been included in the model. Although palliative care costs, if included, would occur late in the time horizon of the model and be substantially discounted, the omission of these costs results in a likely underestimate of the cost savings associated with RET mutation testing. In the cohort where RET mutation testing is available, relative to where it is not available, the increased prophylactic and early surgery associated with identification of RET mutations would decrease the incidence of advanced MTC requiring palliation.

#### Utility values

A literature search for utility values associated with MEN2A, thyroidectomy (including complication of surgery) and MTC was undertaken. While many sources of published utility values were identified, only one source (Li et al. 2011) was identified that included ‘watchful waiting’—equivalent to surveillance—which has a significant role in the economic model. In general it appeared that the utilities reported by the various sources were generally consistent with each other and not wide-ranging; however, for improved consistency, a single source was selected for the model. The utility values reported by Li et al. (2011) are derived from time-trade-off methodology with a panel of thyroid experts, and also supported by alternative literature where available. The utility values used are described in detail in Table 44, in addition to the weightings applied to them. The actual thyroidectomy procedure has not been allocated a unique utility value; rather, the entire post-thyroidectomy value is assumed for the entire year of the procedure.

Table Health state utilities used in cost-utility analysis

| **Health state** | **Utility** | **Source** | **Discussion** |
| --- | --- | --- | --- |
| Healthy with no symptoms and not undergoing any surveillance | 1 | Assumed | This utility applies to family members of index patients who are RET mutation tested and found to be RET M–, and to family members of index patients who do not participate in surveillance who may be either RET M– or RET M+ but who are not yet symptomatic of MEN2 disease. |
| Healthy and undergoing surveillance | 0.98 | ([Li et al. 2011](#_ENREF_119)) | Described as ‘watchful waiting’, this utility was determined using time-trade-off methodology with a panel of thyroid experts. This appears reasonable at face value and allows for the anxiety of being ‘at risk’ and requiring regular monitoring. |
| Post-thyroidectomy | 0.944 | Weighted combination from ([Li et al. 2011](#_ENREF_119)) | Weighting of utilities from Li et al. (2011) for post-thyroidectomy (no complications): 0.97; and post-thyroidectomy (permanent complications): 0.65. Weighted at 92% and 8%, respectively, based on 8% rate of permanent complications (hypoparathyroidism/hypocalcaemia, 6.7%; and recurrent nerve injury, 1.3%) reported following surgery in asymptomatic family members ([Dralle et al. 1998](#_ENREF_47)). |
| Advanced MTC / recurrence | 0.60 | ([Li et al. 2011](#_ENREF_119)) | Estimated to be a reasonable average utility across all time points (from diagnosis to progression/death) for patients diagnosed with MTC who have not been tested or monitored, and present late due to symptoms. Disease is assumed to be at a later stage (e.g. commonly stage III) compared with cases of MTC identified/predicted through screening/surveillance (generally precancerous CCH/stage I). |

MTC = medullary thyroid carcinoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

### Assumptions

The model does not attempt to replicate all potential health outcomes and pathways that may occur in clinical practice. There is potentially an infinite number of combinations of age, genetic risk, disease stage and treatment outcomes that could exist for both index cases and family members, and there is little reliable data on the distribution of these in RET-mutation-positive populations generally, let alone within Australia. Therefore, the model rather crudely predicts costs and outcomes where times to events, patient age, survival, disease development and treatment outcomes are point estimates. The following assumptions are made:

* *RET mutation testing* *does not impact* *MTC or phaeochromocytoma treatment or outcomes (costs, utilities or survival) in index cases* presenting with symptoms of disease. (These will be identical in each arm of the model and would cancel out of any incremental calculation; *therefore, these parameters are not calculated or included in the model*.) This may be a conservative assumption as patients with an identified RET mutation may be more diligent in routine monitoring for additional symptoms and subsequently have better clinical outcomes. Also, index patients identified as RET-mutation-negative may experience a utility benefit associated with the knowledge that they are not at increased risk of other MEN2-associated conditions.
* *RET mutation testing* *does not impact the incidence or treatment of phaeochromocytoma (costs, utilities or survival) in familial RET-mutation-positive patients*. This is based on the fact that, unlike MTC, no prophylactic measures are taken to prevent development of phaeochromocytoma in RET-mutation-positive patients, and the RET mutation test does not alter the natural pathway of disease. If RET mutation testing is associated with earlier detection of phaeochromocytoma and this is associated with improved outcomes, then this approach is conservative.
* The average age of patients entering the model is 40 years, based on the mean age of diagnosis of index patients in the pre-RET mutation testing era (Table 28). The background mortality rate is then based on ABS data on the average Australian mortality rates from age beginning at 40 years. Although uncertain in reality, for simplicity it is assumed in the model that this is also the average age / annual background mortality rate of family members also. The higher the background mortality rate, the more conservative the model is with respect to RET mutation testing, as the majority of the costs for testing occur in the first few years of the model, whereas costs associated with biochemical screening are spread more evenly over a person’s lifetime.
* Patients identified as RET-mutation-positive through genetic familial screening (non-symptomatic) are all recommended to have prophylactic thyroidectomy. The model assumes 100% of RET-mutation-positive patients receive a prophylactic thyroidectomy in the year immediately after they are tested for a RET mutation. This is a conservative approach with respect to estimating costs associated with RET mutation testing, as in practice some of these thyroidectomies will be delayed a few years (in low risk patients) and therefore costs should be discounted, and some RET-mutation-positive patients refuse prophylactic treatment and/or ongoing surveillance and do not incur these further costs ([Alvares Da Silva et al. 2003](#_ENREF_9); [Kinlaw et al. 2005](#_ENREF_107); [Romei et al. 2011](#_ENREF_173)).
* In untested true RET-mutation-positive patients it is estimated that only 70% would develop a *symptomatic* MTC and subsequent thyroidectomy before the age of 70 years ([Frank-Raue et al. 2011](#_ENREF_70)). This assumption has also been assumed to apply to patients undgoing monitoring in the pre-RET mutation testing era. While actual lifetime penetrance rates of any MTC (including microscopic and those identifiable only through biochemical/pathology testing) are estimated at close to 100%, sometimes the MTC would not develop to be apparent or symptomatic before the patient has died of other conditions or reaches an age where active treatment is inappropriate.
* In the historical comparison arm, of the family members who participate in biochemical screening and are true RET-mutation-positive, the first 5 years of biochemical screening detects all 70% of cases who would develop symptomatic MTC in their lifetime, and these patients then undergo prophylactic/early thyroidectomy. Of these cases, 30% are assumed to be identified in the first year of screening and an additional 20% in each of the second and third years after screening commences.
* The ‘cure rate’ for prophylactic thyroidectomy in RET-mutation-positive family members is assumed to be 90% (i.e. the rate of patients developing symptomatic advanced stage relapse is assumed to be 10%). This assumption approximates the the median estimate of the rate of recurrence in patients for whom surgery is considered prophylactic (9%). Where family members do not have access to RET mutation testing, but have early surgery on the basis of family history and biochemical surveillance, the cure rate is estimated at 60% and the relapse rate at 40%. These estimates are based on the results published by Rohmer et al. (2011) and the meta-analysis presented in Figure 7. These estimates are based on limited data and it is acknowledged that they are highly uncertain and likely to be biased. Alternative RRs, up to 1 are tested in the sensitivity analyses.
* Where prophylactic/early MTC surgery is curative and successful, life-expectancy is equivalent to the normal population. This is supported by literature reports that patients with stage I MTC have a life expectancy similar to the general population ([de Groot et al. 2006](#_ENREF_37)) and that long term mortality in MEN2A patients alive 10-15 years after surgery is similar to the population.([Szinnai et al. 2003](#_ENREF_198))
* Where prophylactic/early MTC surgery is not successful and relapse occurs, this is assumed to present at year 15 of the model (11-14 years after thyroidectomy). This estimate is highly uncertain; but approximates the recurrence time of 10.5 years reported in Skinner et al. (1996) and is consistent with the estimate of symptomatic presentation of MTC in untreated patients.
* Where late stage disease develops (i.e. in true RET-mutation-positive patients *not* undergoing medical surveillance), presentation is assumed to occur at year 15 in the model, at which time these patients require a thyroidectomy and will be subsequently tested and monitored. There is no applicable data available on the time to presentation of symptoms and so this estimate is highly uncertain. This estimate is considered plausible given it represents the development of symptomatic MTC in family members at similar ages to the presentation in index patients, if it is assumed that family members equally comprise people of the same generation and one generation younger than index patients, and a generation approximates 30 years. Furthermore, the estimate has face validity with respect to allowing for the development of late stage disease in patients who may be free of disease initially, given that:
* patients with clinical disease are on average 8 years older than those without clinical disease ([Punales et al. 2003](#_ENREF_161))
* the average interval between primary tumour and metastases is 6.6 years ([Fialkowski & Moley 2006](#_ENREF_64))

Nevertheless, this estimate of the time to the development of symptomatic MTC in non-monitored family members is reasonably arbitrary.

* Equivalent surveillance for MTC, phaeochromocytoma and hyperparathyroidism (and the recommendation for prophylactic thyroidectomy) is undertaken on all patients with a known RET mutation, or suspected of possibly having MEN2, irrespective of the specific codon mutation or predicted risk level (e.g. from low-risk FMTC through to high-risk MEN2B). In reality, increasing research is being undertaken stratifying risk levels on the basis of the specific mutation identified. While some papers suggest that this is too uncertain to yet influence clinical practice, other guidelines suggest that the range of risk profiles justifies varying clinical approaches with respect to prophylactic thyriodectomy etc. There is currently inadequate data to incorporate various mutation specific risk profiles into the economic models. Such a model would be highly complex but, if achievable, such an approach would be likely to improve the accuracy of the model results.

While it can be seen that, to generate quantitative results, the economic models contain numerous assumptions that will not necessarily replicate reality in all circumstances, an attempt has been made to ensure that the base-case assumptions are generally conservative and do not favour economic benefit toward RET mutation testing, such that in reality, RET mutation testing is likely to be of greater economic value than predicted in the economic models presented here.

### Results

#### Scenarios 1 and 2 – Cost-minimisation analysis

The results of the cost-minimisation comparing RET mutation testing (and selective biochemical surveillance) with biochemical investigation and surveillance in patients presenting with MTC or phaeochromocytoma younger than 50 years of age are shown below (Table 45 and Table 46).

Table 45 Results of cost-minimisation analysis or RET mutation testing vs. biochemical screening in a cohort of 100 patients presenting with MTC, over varying time horizons (1, 10, 20 and 30 years)

| **Time horizon** | **Costs associated with RET mutation testing and selective surveillance** | **Costs associated with non-selective biochemical surveillance** | **Increment** |
| --- | --- | --- | --- |
| 1 years | $46,477.33 | $9,305.00 | $37,172.33 |
| 10 years | $66,436.89 | $74,532.32 | –$8,095.43 |
| 20 years | $80,006.73 | $118,878.19 | –$38,871.47 |
| 30 years | $88,036.99 | $145,120.88 | –$57,083.89 |

Table 46 Results of cost-minimisation analysis or RET mutation testing vs biochemical screening in a cohort of 100 patients younger than 50 years of age presenting with phaeochromocytoma, over varying time horizons (1, 10, 20 and 30 years)

| **Time Horizon** | **Costs associated with RET mutation testing and selective surveillance** | **Costs associated with non-selective biochemical surveillance** | **Increment** |
| --- | --- | --- | --- |
| 1 year | $51,953 | $45,480 | $6,473 |

The cost analysis clearly demonstrates that, over the long term, substantial savings would be expected following the use of RET mutation testing to identify RET-mutation-positive patients among those presenting with MTC where target biochemical surveillance for other MEN2 symptoms could subsequently be used.

These results show that, in the case of potential index patients presenting with MTC, while in the year of RET mutation testing there is an increased cost per patient associated with testing, over time the savings in screening costs accumulate and an overall saving is achieved. Savings begin to accrue within 10 years due to the subsequent reduction in the extent of biochemical surveillance required within a genetically tested cohort. Over an extended time period (e.g. 30 years) net savings of around $535 per MTC patient tested would be expected, despite the initial outlay of $400 per RET mutation test per patient.

In the case of phaeochromocytoma patients, assuming no change in patient management on the basis of the test, and on the grounds that ongoing annual surveillance for MEN2 is not undertaken regularly in phaeochromocytoma patients unless the initial investigation shows a further symptom, a marginal increase in costs is observed (approximately $65 per patient), because the diagnostic RET mutation test costs slightly more than biochemical pentagastrin testing and imaging.

#### Scenarios 3 and 4 – Cost-utility analysis of the availability of diagnostic RET mutation testing in suspected index cases and subsequent prognostic familial screening in first- and second-degree relatives of affected cases, versus non-availability of RET mutation testing

##### MTC

The results of the cost utility analysis in a cohort of 100 index cases presenting with MTC (Scenario 3) are presented in Table 47 to Table 50, which show disaggregated costs and outcomes. These are combined in Table 51 to estimate an incremental cost-effectiveness ratio.

Table 47 Results: discounted cumulative societal healthcare costs associated with a cohort of 100 potential index cases presenting with MTC and family members where RET mutation testing is available

| **Time horizon (years)** | **Costs in index patients** | **Costs in family members** | **Total costs** |
| --- | --- | --- | --- |
| 1 | $46,477 | $46,687 | $93,165 |
| 10 | $66,437 | $452,866 | $519,303 |
| 20 | $80,007 | $713,164 | $793,171 |
| 30 | $88,037 | $789,352 | $877,389 |
| 60 | $96,724 | $869,615 | $966,339 |

Table 48 Results: discounted cumulative societal healthcare costs associated with a cohort of 100 potential index cases presenting with MTC and family members where RET mutation testing is not available

| **Time horizon (years)** | **Costs in index patients** | **Costs in family members** | **Total costs** |
| --- | --- | --- | --- |
| 1 | $9,305 | $234,577 | $243,882 |
| 10 | $74,532 | $2,020,587 | $2,095,119 |
| 20 | $118,878 | $3,265,563 | $3,384,441 |
| 30 | $145,121 | $3,923,320 | $4,068,441 |
| 60 | $173,511 | $4,610,136 | $4,783,646 |

Table 49 Results: incremental difference in cumulative societal healthcare costs between where RET mutation testing is available and where RET mutation testing is not available for a cohort of 100 potential index cases presenting with MTC and their family members

| **Time horizon (years)** | **Total costs where RET mutation testing available** | **Total costs where RET mutation testing is not available** | **Incremental cost difference** |
| --- | --- | --- | --- |
| 1 | $93,165 | $243,882 | –$150,717 |
| 10 | $519,303 | $2,095,119 | –$1,575,816 |
| 20 | $793,171 | $3,384,441 | –$2,591,271 |
| 30 | $877,389 | $4,068,441 | –$3,191,052 |
| 60 | $966,339 | $4,783,646 | –$3,817,307 |

Table 50 Results: discounted cumulative quality-adjusted life-years (QALYs) in the family members of 100 potential index cases presenting with MTC

| **Time horizon (years)** | **Where RET mutation testing is available** | **Where RET mutation testing is not available** | **Increment** |
| --- | --- | --- | --- |
| 1 | 1,149 | 1,141 | 8 |
| 10 | 9,188 | 9,129 | 59 |
| 20 | 14,600 | 14,476 | 124 |
| 30 | 17,760 | 17,557 | 203 |
| 60 | 21,171 | 20,822 | 349 |

Table 51 Overall results: incremental costs, quality-adjusted life-years (QALYs) and incremental cost-effectiveness ratio (ICER) of RET mutation testing versus no RET mutation testing, over various time horizons for a cohort of 100 potential index cases presenting with MTC and their family members

| **Time horizon (years)** | **Incremental costs** | **Incremental QALYs** | **ICER** |
| --- | --- | --- | --- |
| 1 | –$150,717 | 8 | Dominant |
| 10 | –$1,575,816 | 59 | Dominant |
| 20 | –$2,591,271 | 124 | Dominant |
| 30 | –$3,191,052 | 203 | Dominant |
| 60 | –$3,817,307 | 349 | Dominant |

The base-case analysis indicates that RET mutation testing is highly cost-effective, both saving costs and improving health outcomes, when made available to index cases presenting with MTC and their family members, who would otherwise be recommended long-term biochemical surveillance. Cost savings and utility gains are made in the first year and continue to increase when longer time horizons are considered.

##### Phaeochromocytoma

Similar analysis is conducted in the alternative scenario (4), a cohort of 100 potential index cases presenting with phaeochromocytoma, and the results of this scenario are shown in Table 52 to Table 56.

Table Results: discounted cumulative societal healthcare costs associated with a cohort of 100 potential index cases younger than 50 years of age presenting with phaeochromocytoma and family members where RET mutation testing is available

| **Time horizon (years)** | **Costs in index patients** | **Costs in family members** | **Total costs** |
| --- | --- | --- | --- |
| 1 | $51,953 | $27,800 | $79,753 |
| 10 | $51,953 | $267,068 | $319,020 |
| 20 | $51,953 | $420,401 | $472,354 |
| 30 | $51,953 | $465,281 | $517,234 |
| 60 | $51,953 | $512,561 | $564,514 |

Table Results: discounted cumulative societal healthcare costs associated with a cohort of 100 potential index cases younger than 50 years of age presenting with phaeochromocytoma and family members where RET mutation testing is not available

| **Time horizon (years)** | **Costs in index patients** | **Costs in family members** | **Total costs** |
| --- | --- | --- | --- |
| 1 | $45,480 | $42,928 | $88,408 |
| 10 | $45,480 | $428,340 | $473,820 |
| 20 | $45,480 | $707,753 | $753,233 |
| 30 | $45,480 | $826,573 | $872,053 |
| 60 | $45,480 | $940,531 | $986,011 |

Table Time horizon (years) Total costs where RET mutation testing available Total costs where RET mutation testing is not available Incremental cost difference

| **Time horizon (years)** | **Total costs where RET mutation testing available** | **Total costs where RET mutation testing is not available** | **Incremental cost difference** |
| --- | --- | --- | --- |
| 1 | $79,753 | $88,408 | -$8,654 |
| 10 | $319,020 | $473,820 | -$154,800 |
| 20 | $472,354 | $753,233 | -$280,879 |
| 30 | $517,234 | $872,053 | -$354,819 |
| 60 | $564,514 | $986,011 | -$421,497 |

Table Results: discounted cumulative quality-adjusted life-years (QALYs) in family members of 100 potential index cases presenting with phaeochromocytoma

| **Time Horizon (years)** | **Where RET mutation testing is available** | **Where RET mutation testing is not available** | **Increment** |
| --- | --- | --- | --- |
| 1 | 1,149 | 1,148 | 1 |
| 10 | 9,198 | 9,200 | –2 |
| 20 | 14,638 | 14,625 | 14 |
| 30 | 17,832 | 17,786 | 46 |
| 60 | 21,283 | 21,165 | 118 |

Table Overall results: Incremental costs, quality-adjusted life-years (QALYs) and incremental cost-effectiveness ratio (ICER) of RET mutation testing vs. no RET mutation testing, over various time horizons, in potential index cases presenting with phaeochromocytoma and their family members

| **Time horizon (years)** | **Incremental costs** | **Incremental QALYs** | **Incremental ICER$/QALYs gained** |
| --- | --- | --- | --- |
| 1 | -$8,654 | 1 | Dominates |
| 10 | -$154,800 | –2 | $72,873 (cost savings per QALY **lost**) |
| 20 | -$280,879 | 14 | Dominates |
| 30 | -$354,819 | 46 | Dominates |
| 60 | -$421,497 | 118 | Dominates |

Overall, the cost-effectiveness of RET mutation testing in potential index cases presenting with phaeochromocytoma and their families is apparent. Cost savings are incurred every year, primarily due to the fact that Ret mutation testing for family members is less expensive than comprehensive biochemical screening and imaging. A negative health outcome is associated with testing in the short term (to 17 years), associated with the increased rate of thyroidectomy (including a complication rate); however, once increased relapse of MTC is included in the model (occurring from 15 years), health outcome benefits associated with RET mutation testing and earlier thyroidectomy are apparent and high cost-effectiveness is demonstrated.

## Sensitivity analyses

Sensitivity analyses are conducted on various parameters in the economic model to identify the importance of uncertainties and key assumptions. The sensitivity tests undertaken and the results of the analyses are presented in Table 57 to Table 60.

The sensitivity analysis of cost-utility of RET mutation testing in the cohort of index patients presenting with MTC and their family members indicates that, despite many uncertainties in the economic model, the cost-effectiveness of RET mutation testing in this cohort appears robust (Table 59). The only parameter that significantly altered cost-effectiveness in the sensitivity analysis was removing all clinical benefit attributed to using the test to achieve quicker, more successful prophylactic thyroidectomies (i.e. such that the RR of recurrence of MTC following thyroidectomy after RET versus thyroidectomy on the basis of biochemical screening was 1.0). However, further testing indicted that, for any RR less than 0.97, RET mutation testing remained dominant.

The sensitivity analysis of inputs used in the phaeochromocytoma model indicates that the conclusion of cost-effectiveness of RET mutation testing in this cohort of index patients and family members is sound, despite many uncertainties in the model (Table 60). The benefit in outcomes is maintained in all circumstances; however, where extreme uptake rates of testing or minimal uptake rates of surveillance are incorporated in the model, an increase in costs becomes apparent such that the ICER increases up to $2,600 per QALY. Where no change in RR of relapse is modelled, unsurprisingly, genetic testing is dominated as it results in both increased costs and quality of life decrements (due to costs and complications in additional thyroidectomies), with no clinical benefit.

Table 57 Sensitivity analyses in the economic model – Scenario 1: 100 index cases only considered, presenting with MTC

| **Parameter** | **Base-case value** | **Alternative values tested** | **Results: net cost at 30 years** |
| --- | --- | --- | --- |
| Base case |  |  | –$57,083 (i.e. savings) |
| Proposed fee for diagnostic RET mutation tests | $400 for index case | $600–$1,150 | –$37,083 (i.e. savings) to $17,916 (costs) |
| Discount rate (costs and outcomes) | 5% | 0% | –$148,097.10 |
| Diagnostic yield in index cases | 30.6% | 5–95% | –$94,234.84 to $36,373.96 |

Table Sensitivity analyses in the economic model – Scenario 2: 100 index cases only considered, patients younger than 50 years of age presenting with phaeochromocytoma

| **Parameter** | **Base-case value** | **Alternative values tested** | **Results: net cost at 30 years** |
| --- | --- | --- | --- |
| Base case |  |  | $6,473 |
| Proposed fee for diagnostic RET mutation tests | $400 for index case | $600–$1,150 | $26,473–$81,473 |
| Discount rate (costs and outcomes) | 5% | 0% | $6,473 |
| Diagnostic yield in index cases | 18.3% | 7–25% | $1,334–$9,520 |

Table Sensitivity analyses in the economic model – Scenario 3: 100 index cases presenting with MTC and their family members

| **Parameter** | **Base-case value** | **Alternative value or range tested** | **Results: ICER at 30 years (across range, respectively)** |
| --- | --- | --- | --- |
| Base case |  |  | Dominates |
| Proposed fee for diagnostic RET mutation test | $400 | $600–$1150 | Dominates (either fee) |
| Proposed fee for predictive RET mutation test | $200 for family screen | $250 | Dominates |
| Discount rate (costs and outcomes) | 5% | 0% | Dominates |
| Health outcome measure | Utility values | Life-years gained | Dominates |
| Diagnostic yield in index cases | 30.6% | 5–95% | Dominates (either yield) |
| Diagnostic yield in family members | 40% | 50% | Dominates |
| Predictive RET mutation testing uptake rate | 40% | 15–100% | Dominates (either uptake) |
| Family surveillance uptake rate (in the absence of testing) | 40% | 15–100% | Dominates (either uptake) |
| RR of relapse (based on likelihood of MTC at time of surgery) in RET mutation testing era vs. no RET mutation testing | Absolute risk of relapse: 10% in RET mutation testing arm and 40% in non-RET mutation testing arm (RR 0.25) | Absolute risk of relapse = 10% in both arms (RR 1.0)  Values of RR ≤ 0.975 (Relapse in no test arm ≥ 10.25%) | Dominated (from year 29)  Dominates |

Table Sensitivity analyses in the economic model – Scenario 4: 100 index cases, patients younger than 50 years of age presenting with phaeochromocytoma, and their family members

| **Parameter** | **Base-case assumption** | **Alternative value or range tested** | **Results: ICER at 30 years (across range, respectively)** |
| --- | --- | --- | --- |
| Base case |  |  | Dominates |
| Proposed fee for diagnostic RET mutation tests | $400 for index case | $600–$1,150 | Dominates (either price) |
| Proposed fee for family screening *RET* gene test | $200 for family screen | $480 | Dominates |
| Discount rate (costs and outcomes) | 5% | 0% | Dominates |
| Health outcome measure | Utility values | Life-years gained | Dominates |
| Diagnostic yield in index cases | 18.3% | 7–25% | Dominates (either yield) |
| Diagnostic yield in family members | 40% | 50% | Dominates |
| Predictive RET mutation testing uptake rate | 40% | 15–100% | Dominates–$485/QALY |
| Family surveillance uptake rate (in the absence of testing) | 40% | 15–100% | $1,123/QALY—dominates |
| Relative risk of relapse (based on likelihood of MTC at time of surgery) in RET mutation testing era vs. no RET mutation testing | Absolute risk of relapse in RET = 10%; risk of relapse in non-RET mutation testing era = 40% (RR 0.25) | Absolute risk of relapse = 10% in both arms (RR 1.0)  RR 0.25-0.43 (Relapse in no test arm 23%-40%) | $4,721 saved/QALY **lost**  Dominates |

The sensitivity analyses show that the assumption of a clinical benefit in terms of a reduced MTC development or relapse in patients who receive genetic testing (and earlier thyroidectomy) is critical to demonstrate cost-effectiveness in each cost-utility model. Although the estimate of the RR of MTC relapse in patients treated prophylactically is uncertain, the evidence consistently indicates that improved outcomes are expected with earlier prophylactic thyroidectomies undertaken on the basis of genetic test results. Therefore it is reasonable to assume that a RR less than one would be realised in practice, such that testing would be cost-effective.

## Cost analysis

### Financial/budgetary impacts

#### Sources of data

Table 61 describes the sources of the data and parameters used in the estimation of the financial and budgetary impact of listing RET mutation testing on the MBS.

Table 61 Sources of data and parameters used to estimate the financial and budgetary impact of listing RET mutation testing on the MBS

| **Data or parameter** | **Value used** | **Source** |
| --- | --- | --- |
| Incidence of thyroid cancer in Australia, 2005–09 | 2005: 1,617 cases  2006: 1,664 cases  2007: 1,789 cases  2008: 1,995 cases  2009: 2,039 cases  *See reference for access to full data set.* | Australian Institute of Health and Welfare website, *Thyroid cancer* workbook ([AIHW 2010](#_ENREF_3)) |
| Incidence of MTC 1982–2009 | 5–10% of total thyroid cancers | [Keatts & Itano (2006](#_ENREF_101)) |
| Projected growth in thyroid cancer and MTC 2010–15 | 6.3% | Calculated based on average annual increase in thyroid cancer 2005–09 |
| Percentage of MTC patients found to have hereditary mutation | 25–30% | Raue & Frank-Raue (2010) |
| Number of first- and second-degree family members to be tested per proband case | 11.5 | [Suthers et al. (2006](#_ENREF_193)) |
| Uptake rate of testing in family members | 40% | [Suthers et al. (2006](#_ENREF_193)) |
| Cost of RET mutation testing (base case) | *RET* gene screen (diagnostic): $500.00  Known RET mutation test (familial): $200.00 | Calculated, based on the quoted price for RET mutation testing of the 6 most common exons (Table 6 on page xxxv) |

In the base-case scenario the costs of RET mutation testing have been based on the average cost of RET mutation testing (see Table 6 on page xxxv). The financial and budgetary impact of testing based on the lowest sourced quote (*RET* gene screen: $400; known RET mutation test: $200) is presented in Table 64 and Table 65; and that based on the costs proposed in the final DAP (diagnostic RET mutation test: $1,150; known RET mutation test: $480) is presented in Appendix K.

Genetic counselling may or may not be covered by the MBS, depending on the setting in which the service is provided. The state and territory systems currently provide services through accredited genetic counsellors and medical specialists (clinical geneticists). Genetic counselling may alternatively be provided by the treating practitioner in consultation and, as such, would be covered by the MBS. The cost of genetic counselling has not been accounted for in the financial and budgetary estimates, as the current distribution of services is unlikely to change, with little impact expected to the overall health budget, MBS, and state and territory systems.

#### Likely volume of diagnostic RET mutation tests per year

RET mutation testing (diagnostic testing) is expected to occur in all patients who are considered to *potentially* have a diagnosis of MEN2. As 95–100% of patients with MEN2 are at risk of developing a thyroid tumour, all patients presenting with MTCs are considered potential cases of MEN2 that require further confirmation ([Margraf et al. 2009](#_ENREF_132)). As described, diagnostic RET mutation testing is standard practice, such that the number of newly diagnosed MTCs each year is a useful proxy for the estimated number of diagnostic RET mutation tests performed annually. In routine practice, patients would only require one diagnostic RET mutation test per lifetime, with repeat testing required only on possibly erroneous results; however, it is uncertain how often these may arise, due to a non-obligatory RCPA recommendation to conduct predictive RET mutation testing in duplicate ([Ravine & Suthers 2012](#_ENREF_168)).

The likely volume of diagnostic RET mutation testing performed per year, presented in Table 62, is based on epidemiological estimates of the proportion of MTC within total thyroid cancer data during 2007–09. Projected numbers of MTC and tests performed beyond 2007 are based on an assumed ongoing annual increase of 6.3% in the incidence of new thyroid cancers. This was the average annual growth rate for the period 2005–09 and was only marginally less than the average growth rate of 6.7% seen over the 27 years of available data.

Table 62 Estimated number of diagnostic RET mutation tests during 2007–15, with or without MBS listing

| **Year** | **2007** | **2008** | **2009** | **2010** | **2011** | **2012** | **2013** | **2014** | **2015** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Incidence of thyroid cancera | 1,789 | 1,995 | 2,039 | 2,168 | 2,304 | 2,449 | 2,604 | 2,768 | 2,942 |
| Estimated number of diagnostic RET mutation testsb | 89–179 | 100–200 | 102–204 | 108–217 | 115–230 | 122–245 | 130–260 | 138–277 | 147–294 |
| Reported number of tests ([Suthers 2008b](#_ENREF_195)) | 150 |  |  |  |  |  |  |  |  |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a incidence of thyroid cancer from 2010 onwards projected based on the average annual incidence during 2005–09 of 6.3%

b estimated number of diagnostic RET mutation tests performed based on the reported incidence of MTC in all thyroid cancers of 5–10% ([Keatts & Itano 2006](#_ENREF_101))

Inaccuracies in the estimates may occur as a result of the following factors:

* Some patients with newly diagnosed MTCs may have had RET mutation testing previously performed as a result of known familial RET mutations and therefore do not require testing upon MTC diagnosis. These patients will be overrepresented in the MTC estimate.
* Some diagnostic RET mutation testing will be initiated in patients suspected of having MEN2 due to other symptoms (e.g. phaeochromocytoma + family history) and will subsequently be underrepresented in the MTC estimate.

However, overall, these estimates are considered reasonable and are consistent with the reported 150 diagnostic RET mutation tests performed in 2007 (and 107 in 2006), the only available quantitative data on the extent of RET mutation testing in Australia ([Suthers 2008b](#_ENREF_192)).

It is considered that*, should listing occur,* these *estimates of potential MBS diagnostic test use and expenditure equate to* estimates of *diagnostic test use and approximate expenditure for state health budgets under the current scenario where, although not MBS-listed*, RET mutation testing in these patients is already considered standard practice in Australia[[13]](#footnote-13).

It is also noted that the incidence of thyroid cancer has increased every year since 1982, except in three individual years (see Appendix J) and therefore, an ongoing annual increase at a similar rate in the numbers of tests ordered, might also be anticipated beyond the years projected in the report.

#### Likely volume of known RET mutation testing per year

Only 25–30% of MTC patients who undergo diagnostic RET mutation testing are expected to have positive hereditary mutations ([Raue & Frank-Raue 2010](#_ENREF_166)) that warrant known RET mutation testing in their families (familial screening). Across index patients with a mutation, it is assumed that there will be an average of 11.5 first- or second-degree relatives eligible for familial screening. Of these, it is expected that approximately 40% will accept an offer of familial screening within 2 years ([Suthers et al. 2006](#_ENREF_196)). Therefore, to estimate the number of familial screens undertaken if MBS listing occurred, it is assumed that an average of 4.6 relatives per index case of MEN2 would be tested. Again, it is assumed that this testing would normally occur only once in a person’s lifetime. The calculations and projections are presented in Table 63.

Table 63 Estimated number of familial screens during 2007–15, with an MBS listing

| **Year** | **2007** | **2008** | **2009** | **2010** | **2011** | **2012** | **2013** | **2014** | **2015** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Estimated number of proband screensa | 89–179 | 100––200 | 102–204 | 108–217 | 115–230 | 122–245 | 130–260 | 138–277 | 147–294 |
| Positive resultsb | 22–54 | 25–60 | 25–61 | 27–65 | 29–69 | 31–73 | 33–78 | 35–83 | 37–88 |
| Number of relatives *eligible* for a familial screenc | 257–617 | 287–688 | 293–703 | 312–748 | 331–795 | 352–845 | 374–898 | 398–955 | 423–1,015 |
| Anticipated number of familial screens performed, assuming 40% uptake rate | 103–247 | 115–275 | 117–281 | 125–299 | 132–318 | 141–338 | 150–359 | 159–382 | 169–406 |

MBS = Medicare Benefits Schedule

a estimated based on a 5–10% incidence of MTC in all thyroid cancers; incidence of thyroid cancer beyond 2010 projected based on the average annual incidence during 2005–09 of 6.3%

b assuming positive hereditary RET mutations identified in 25–30% of MTC patients tested

c assuming 11.5 relatives per proband

In 2007 there were 49 presymptomatic familial RET mutation tests performed in Australia (up from 29 the year before) ([Suthers 2008b](#_ENREF_195)). This represents only one or two relatives screened per index patient identified (assuming a 25% diagnostic yield in the initial test) and is a lower volume of familial screening than would be expected. While the reasons for the low uptake of familial screening seen historically are unknown, potentially increased rates of familial screening would occur following an MBS listing—both directly as a result of MBS funding improving current access/funding-related issues (if they are present); and if patient attitudes toward RET mutation testing become more positive and MBS funding is perceived as government endorsement of the practice.

While the extent to which potentially affected families will uptake familial screening in the future is highly uncertain, it would be prudent to anticipate that predictive RET mutation testing may increase from previously reported levels and retain the estimated 40% uptake rate as a base estimate.

### Total cost to the Australian healthcare system overall

The MBS listing of the diagnostic RET mutation testing in patients with MTC considered to potentially have MEN2 disease is not expected to have any impact on the costs of the overall Australian healthcare system considered in its entirety. The practice of RET mutation testing and counselling already occurs routinely in these patients in a manner unchanged by the proposed listing and at a similar cost, which is currently borne by state government hospital budgets.

Likewise, familial screening and counselling for known RET mutations in family members of identified proband cases is currently available as in the proposed listing and at a similar cost, which is currently borne by the state government. However, the extent of familial screening that has been reported in the past (2006–07) is lower than would have been expected, based on the reported incidence of RET mutations and the number of index cases anticipated.

**Total costs to the MBS**

Table 64 and Table 65, respectively, present the estimated annual costs of listing diagnostic and predictive RET mutation testing on the MBS between 2007 and 2015, assuming that all services are provided in an outpatient setting where the MBS covers 85% of the cost of services. Based on an estimated number of 130–260 diagnostic and 150–359 predictive RET mutation tests performed in 2013, the estimated median cost to the MBS is $109,654. This would increase to $123,906 in 2015 based on 147–294 diagnostic and 169–406 predictive tests performed (Table 66). However, an unknown proportion of patients may qualify for the Medicare Safety Net, in which case 100% of the scheduled fee would be paid by the MBS. Allowing for application of the Medicare Safety Net, the overall true costs to the Commonwealth health budget would lie between the total costs to the MBS and the total combined costs of RET mutation testing, i.e. up to $129,005 in 2013 and $145,772 in 2015.

Sensitivity analyses assuming upper estimates around disease incidence (positive results in 30% of patients with MTCs tested) and a 100% uptake rate of familial screening are presented in Table 67 to provide an extreme upper limit of the predictable financial costs. The cost of RET mutation testing to the MBS under these extreme upper limits increases from $241,217 in 2013 to $272,568 in 2015.

Table 64 Estimated cost of diagnostic RET mutation tests during 2007–15, with or without MBS listing

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Number of diagnostic RET mutation testsb | 89–179 | 100–200 | 102–204 | 108–217 | 115–230 | 122–245 | 130–260 | 138–277 | 147–294 |
| Estimated expenditure on diagnostic RET mutation testingc | $44,725 –$89,450 | $49,875 –$99,750 | $50,976 –$101,953 | $54,188 –$108,376 | $57,602 –$115,204 | $61,231 –$122,461 | $65,088 –$130,176 | $69,189 –$138,378 | $73,548 –$147,095 |
| Patient co–paymentd | $6,709 –$13,418 | $7,481 –$14,963 | $7,646 –$15,293 | $8,128 –$16,256 | $8,640 –$17,281 | $9,185 –$18,369 | $9,763 –$19,526 | $10,378 –$20,757 | $11,032 –$22,064 |
| Estimated MBS expendituree | $30,413 –$60,826 | $33,915 –$67,830 | $34,664 –$69,328 | $36,848 –$73,696 | $39,169 –$78,338 | $41,637 –$83,274 | $44,260 –$88,520 | $47,048 –$94,097 | $50,012 –$100,025 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b estimated based on a 5–10% incidence of medullary thyroid cancer in all thyroid cancers

c assuming the cost of the diagnostic RET mutation test is $400 (see Table 6 on page xxxv)

d assuming most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

e assuming all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

Table 65 Estimated cost of predictive RET mutation tests during 2007–15, with an MBS listing

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Relatives eligible for screeningb | 257–617 | 287–688 | 293–703 | 312–748 | 331–795 | 352–845 | 374–898 | 398–955 | 423–1,015 |
| Number of relatives screenedc | 103–247 | 115–275 | 117–281 | 125–299 | 132–318 | 141–338 | 150–359 | 159–382 | 169–406 |
| Estimated expenditure on predictive RET mutation testingd | $20,574 –$49,376 | $22,943 –$55,062 | $23,449 –$56,278 | $24,926 –$59,823 | $26,497 –$63,592 | $28,166 – $67,599 | $29,941 –$71,857 | $31,827 –$76,384 | $33,832 –$81,197 |
| Patient co-paymente | $3,086 –$7,406 | $3,441 –$8,259 | $3,517 –$8,442 | $3,739 –$8,974 | $3,975 –$9,539 | $4,225 –$10,140 | $4,491 –$10,779 | $4,774 –$11,458 | $5,075 –$12,179 |
| Estimated MBS expendituref | $17,487 –$41,970 | $19,501 –$46,803 | $19,932 –$47,836 | $21,187 –$50,850 | $22,522 –$54,054 | $23,941 –$57,459 | $25,450 –$61,079 | $27,053 –$64,927 | $28,757 –$69,017 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b estimated based on the identification of a positive hereditary mutation in the *RET* gene in 25–30% of tests performed; each patient was assumed to have, on average, 11.5 first- or second-degree relatives eligible for familial screening

c assuming an uptake rate of 40% in eligible family members

d assuming the cost of the familial predictive RET mutation test is $200 (see Table 6 on page xxxv)

e assuming most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

f assuming all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

Table 66 Total MBS costs associated with predictive RET mutation testing (combined costs of listing for diagnostic purposes and listing for familial screening), with or without the safety net

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| *Total combined cost of RET mutation testingb* | $88,645 | $98,852 | $101,035 | $107,401 | $114,167 | $121,359 | $129,005 | $137,132 | $145,772 |
| Lower limit | $56,354 | $62,843 | $64,230 | $68,277 | $72,578 | $77,151 | $82,011 | $87,178 | $92,670 |
| Upper limit | $120,936 | $134,862 | $137,840 | $146,524 | $155,755 | $165,568 | $175,999 | $187,087 | $198,873 |
| *Total patient co–paymentc* | $13,297 | $14,828 | $15,155 | $16,110 | $17,125 | $18,204 | $19,351 | $20,570 | $21,866 |
| Lower limit | $8,453 | $9,426 | $9,635 | $10,242 | $10,887 | $11,573 | $12,302 | $13,077 | $13,901 |
| Upper limit | $18,140 | $20,229 | $20,676 | $21,979 | $23,363 | $24,835 | $26,400 | $28,063 | $29,831 |
| ***Total cost to the MBSd*** | **$75,348** | **$84,024** | **$85,880** | **$91,290** | **$97,042** | **$103,155** | **$109,654** | **$116,562** | **$123,906** |
| Lower limit | $47,900 | $53,416 | $54,596 | $58,035 | $61,692 | $65,578 | $69,710 | $74,101 | $78,770 |
| Upper limit | $102,796 | $114,633 | $117,164 | $124,546 | $132,392 | $140,733 | $149,599 | $159,024 | $169,042 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b assuming all patients qualify for the Medicare Safety Net, the total cost to the MBS would equate to the total combined cost of RET mutation testing

c assuming most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

d assuming all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

Table Sensitivity analyses

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Number of diagnostic RET mutation testsb | 179 | 200 | 204 | 217 | 230 | 245 | 260 | 277 | 294 |
| Total cost of diagnostic RET mutation testsc | $71,560 | $79,800 | $81,562 | $86,701 | $92,163 | $97,969 | $104,141 | $110,702 | $117,676 |
| Total number of relatives screenedd | 617 | 688 | 703 | 748 | 795 | 845 | 898 | 955 | 1015 |
| Total cost of predictive RET mutation testse | $123,441 | $137,655 | $140,695 | $149,559 | $158,981 | $168,997 | $179,644 | $190,961 | $202,992 |
| Combined cost of RET mutation testingf | $195,001 | $217,455 | $222,257 | $236,259 | $251,144 | $266,966 | $283,785 | $301,663 | $320,668 |
| **Patient contributiong** | **$29,250** | **$32,618** | **$33,339** | **$35,439** | **$37,672** | **$40,045** | **$42,568** | **$45,249** | **$48,100** |
| **Total cost to the MBSh** | **$165,751** | **$184,837** | **$188,919** | **$200,821** | **$213,472** | **$226,921** | **$241,217** | **$256,414** | **$272,568** |

MBS: Medicare Benefits Schedule; RET: rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence 2005–09 of 6.3%

b based on 10% incidence of medullary thyroid cancer in all thyroid cancers

c assuming the cost of the diagnostic RET mutation test is $400 (see Table 6 on page xxxv)

d based on 11.5 relatives per proband and assuming 100% uptake of familial screen

e assuming the cost of the predictive RET mutation test is $200 (see Table 6 on page xxxv)

f assuming all patients qualify for the Medicare Safety Net, then the total cost to the MBS would equate to the combined cost of RET mutation testing

g assuming most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

h assuming all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees. No allowance for additional MBS if some patients qualify for the Medicare Safety Net

### Costs to the State and Territory health systems

The costs of RET mutation testing would shift from the state and territory systems to the MBS; however, the costs of genetic counselling services provided in hospitals would continue as per current arrangements. Thus, the overall financial and budgetary impact to the state and territory systems would be a cost saving.

### Costs to the private health insurer and/or patient

Under current arrangements patients who are referred to RET mutation testing by a private facility are billed directly, as private health insurance generally subsidises testing only on MBS items (PaLMS 2011). Currently, some patients who are referred through the public system receive genetic counselling services and testing at no direct cost ([Centre for Genetics Education 2013](#_ENREF_31)). With the listing of RET mutation testing on the MBS, assuming that most patients would receive testing as outpatients, Medicare would pay 85% of the scheduled fee and a patient contribution of 15% of the scheduled fee would apply ($75 per patient for a diagnostic RET mutation test or $30 per predictive RET mutation test, in addition to any ‘gap’ charges or out-of-pocket expenses). Private health insurance may assist with these patient costs. Patients who may be eligible for the Medicare Safety Net, and those whose pathology service bulk-bills tests listed on the MBS ([SA Health 2013](#_ENREF_176)), may not be required to contribute a co-payment.

# Discussion

## Is it safe?

No studies were identified that reported on adverse events directly related to RET mutation testing or due to surveillance for MEN2 features. The RET mutation test itself is considered to have minimal risk as it only involves a peripheral blood sample being taken. However, with any diagnostic test there may be psychological harms that arise from informed knowledge regarding being at risk of a disease. In the absence of RET mutation testing, psychological distress is likely to still be a problem, as first-degree family members would know that they have a 50% chance of having MEN2. Therefore, any psychological harms from RET mutation testing are offset by the reassurance given to relatives who are found to be free from the mutation.

It is considered that the best strategy to cure an MTC is to prevent it from developing ([Spinelli et al. 2010](#_ENREF_193)). Since the introduction of RET mutation testing, those at risk of developing an MTC have been able to be identified with a high degree of accuracy, and more gene carriers have been undergoing surgery prior to clinical evidence of an MTC. A comparison of the safety of prophylactic total thyroidectomy versus thyroidectomy performed upon clinical evidence of disease was thought relevant.

One historical controlled study with a high risk of bias (level III-3 interventional evidence) and 6 uncontrolled studies (level IV interventional evidence) reported on adverse events related to total thyroidectomies performed after RET mutation testing (both those with MTC and asymptomatic family members).

The historical controlled study reported that two patients died from surgical complications, one who was diagnosed clinically and one genetically. However, no information regarding confounding factors, such as stage of disease or surgical technique, was provided. No conclusions regarding the comparative risk of death from surgery can therefore be made. This comparative study did not report on other forms of adverse events after surgery.

From the 12 case series the two common types of complications were hypoparathyroidism and laryngeal nerve palsy. In the majority of cases these were temporary. However, hypoparathyroidism was permanent in a small percentage of patients (5.9–13.6% in 3 studies), requiring ongoing calcium supplementation. In one case (out of 75 patients in 1 study) laryngeal nerve palsy was also permanent. The only other permanent complication was a case of unilateral Horner’s syndrome in a 63-year-old patient. Two further complications reported were 1 case of arterial bleeding requiring re-operation (5.9% of 1 study) and a paediatric case with fluctuating thyroid function test results, despite good thyroxine replacement compliance at 1-year follow-up.

Due to the uncontrolled nature of the studies identified, it is unknown how these rates of complications compare with those who have surgery at a later stage of disease progression. However, surgery performed prior to the development of MTC allows less aggressive surgical procedures to be performed, which reduces the risks associated with neck surgery in children. When performed prophylactically, the chances of local infiltration or involvement of cervical lymph nodes are reduced, such that wide neck dissections may be avoided. Wide neck dissections are associated with higher rates of permanent hypoparathyroidism than removal of the thyroid by itself; therefore, the rates of this adverse outcome are hypothesised to be lower when surgery is performed prophylactically ([Spinelli et al. 2010](#_ENREF_193)).

One concern about operating at an earlier stage of disease is that patients are operated on at an earlier age, which may be difficult for parents to accept. Although no evidence was identified that directly assessed rates of complication from surgery based on age, some authors mentioned that, in their experience, there is no direct relationship between patient age and the risk of complications ([Schellhaas et al. 2009](#_ENREF_179)).

For those with a RET mutation associated with MEN2, the penetrance of MTCs is over 90% ([Raue & Frank-Raue 2012](#_ENREF_167)). Although high, this does mean that, for every 10 patients for whom prophylactic surgery is performed, up to 1 patient may be having the surgery unnecessarily. Although prophylactic total thyroidectomies may use less aggressive surgical techniques and it is therefore logical that they are safer than total thyroidectomies performed for the purposes of treatment rather than prevention, the risks of operating on gene carriers who potentially do not require it need to be considered. However, this situation is not unique to the scenario in which RET mutation testing is available. Prior to RET mutation testing becoming available, total thyroidectomies were often performed if an MTC was suspected due to raised calcitonin levels. This measure is not specific to those at risk of developing an MTC, and there were cases reported in our systematic review who had undergone a total thyroidectomy based on clinical signs and were later found to have wildtype RET, and thus were not at risk of MTC at all ([Algun et al. 2002](#_ENREF_6); [Decker et al. 1996](#_ENREF_39); [Frank-Raue et al. 1996](#_ENREF_69)).

An overall summary of the body of evidence on the safety of total thyroidectomies after RET mutation testing is shown in Table 68. The evidence is rated as poor overall due to the high risk of bias and the uncontrolled nature of the studies identified.

Table Body of evidence matrix for the safety of surgery performed on asymptomatic RET M+ gene carriers

| **Component** | **A** | **B** | **C** | **D** |
| --- | --- | --- | --- | --- |
| **Excellent** | **Good** | **Satisfactory** | **Poor** |
| **Evidence basea** |  |  |  | Level IV studies, or level I to III studies/SRs with a high risk of bias |
| **Consistencyb** |  | Most studies consistent and inconsistency may be explained |  |  |
| **Clinical impact** |  |  |  | Slight or restricted |
| **Generalisability** |  | Population(s) studied in the body of evidence are similar to the target population |  |  |
| **Applicability** |  |  | Probably applicable to Australian healthcare context with some caveats |  |

SR = systematic review

a Level of evidence determined from the NHMRC evidence hierarchy—Table 19

b If there is only 1 study, rank this component as ‘not applicable’.

Source: adapted from [NHMRC (2008](#_ENREF_143))

## Is it effective?

The use of RET mutation testing in the diagnosis of patients with MEN2 was quickly introduced across the world, with no formal trials to assess whether patients who are diagnosed using the test have better health outcomes than those who do not have access to RET mutation testing.

The evidence on the comparative effectiveness of RET mutation testing was 9 historical controlled studies (level III-3 interventional studies). These studies are inherently at high risk of bias due to potential differences that may have occurred over the same time period, e.g. in the surgical techniques and screening procedures used. There are also large differences in the length of follow-up between the intervention (with RET mutation testing) and comparator (without RET mutation testing) arms, such that longer term follow-up data should be interpreted with caution.

Six historical controlled studies all reported that patients were, on average, having surgery at an earlier stage of disease development since the introduction of RET mutation testing. Overall, patients had almost half the risk of having an MTC (rather than C-cell hyperplasia or no disease) at the time of total thyroidectomy compared with those treated prior to RET mutation testing (RR=0.52, 0.32, 0.90). The full range of plausible outcomes includes only a small difference, so the clinical impact is considered ‘substantial’ rather than ‘very large’ in the body of evidence matrix (Table 69).

The risk of persistence or recurrence was also reported in 6 historical controlled studies; however, the length of follow-up in the cohorts diagnosed with the addition of RET mutation testing was too short for this outcome to be clinically meaningful. There were significantly more cases of persistence or recurrence noted in the historical cohorts, prior to the use of RET mutation testing, than in the RET-mutation-tested cohorts (RR=0.28, 0.17, 0.45); however, it is unknown to what degree these health outcomes differ due to the longer time interval since surgery. It is also possible that other historical effects may have influenced the differential health outcomes over time.

One study reported that, in patients who all underwent RET mutation testing, age at time of surgery was a significant predictor of residual or recurrent disease ([Schreinemakers et al. 2010](#_ENREF_180)). Since the introduction of RET mutation testing, the age of patients at the time of thyroidectomy has dramatically decreased. One Australian historical controlled study reported that the mean age of thyroidectomy based on calcitonin testing was 32 years, whereas the mean age of thyroidectomy after RET mutation testing was 16 years. Five other historical controlled studies also reported a reduction in the age at time of surgery.

Although all the identified studies had a high risk of bias, they were consistent that the introduction of RET mutation testing has allowed treatment of those at risk of MTC to occur at a younger age, when it is more likely to be prophylactic, and to be associated with a greater likelihood of the surgery being considered curative.

Overall findings from the body of evidence on the effectiveness of RET mutation testing in improving health outcomes are summarised in the Table 69.

Table Body of evidence matrix for the effectiveness of RET mutation testing on improving health outcomes

| **Component** | **A** | **B** | **C** | **D** |
| --- | --- | --- | --- | --- |
| **Excellent** | **Good** | **Satisfactory** | **Poor** |
| **Evidence basea** |  |  |  | Level III-3 studies with a high risk of bias |
| **Consistencyb** |  | Most studies consistent and inconsistency may be explained |  |  |
| **Clinical impact** |  | Substantial |  |  |
| **Generalisability** |  | Population(s) studied in the body of evidence are similar to the target population |  |  |
| **Applicability** |  | Applicable to Australian healthcare context with few caveats |  |  |

a Level of evidence determined from the NHMRC evidence hierarchy—Table 19

b If there is only 1 study, rank this component as ‘not applicable’.

Source: adapted from [NHMRC (2008](#_ENREF_143))

### Diagnostic accuracy

No evidence was identified that provided a measure of the true diagnostic accuracy of RET mutation testing against an appropriate reference standard. Diagnostic yield studies were provided. The summary of the body of evidence on accuracy is shown in Table 70. Diagnostic yield studies are level IV diagnostic evidene and they provided very limited information on the accuracy of RET mutation testing.

Table Body of evidence matrix showing the body of evidence on the accuracy of RET mutation testing

| **Component** | **A** | **B** | **C** | **D** |
| --- | --- | --- | --- | --- |
| **Excellent** | **Good** | **Satisfactory** | **Poor** |
| **Evidence basea** |  |  |  | Level IV studies |
| **Consistencyb** |  | Most studies consistent and inconsistency may be explained |  |  |
| **Clinical impact** | N/A | | | |
| **Generalisability** |  | Population(s) studied in the body of evidence are similar to the target population |  |  |
| **Applicability** |  | Applicable to Australian healthcare context with few caveats |  |  |

N/A = not applicable

a Level of evidence determined from the NHMRC evidence hierarchy—Table 19

b If there is only 1 study, rank this component as ‘not applicable’.

Source: adapted from [NHMRC (2008](#_ENREF_143))

### Impact on patient management

Studies assessing whether there has been a change in management since the introduction of RET mutation testing were uncontrolled. There was evidence that asymptomatic gene carriers would often undergo prophylactic total thyroidectomy, and there was evidence that some patients had been inappropriately treated prior to RET mutation testing becoming available. There was evidence that family members who were found to have mutations were recommended to undergo surveillance for MEN2, and that those who were found to be free from mutations were able to cease surveillance and their descendants did not require any testing. These studies had good generalisability and applicability although the level of evidence was poor (Table 71).

Table Body of evidence matrix for the assessment of a change in management due to RET mutation testing

| **Component** | **A** | **B** | **C** | **D** |
| --- | --- | --- | --- | --- |
| **Excellent** | **Good** | **Satisfactory** | **Poor** |
| **Evidence basea** |  |  |  | Level IV studies |
| **Consistencyb** |  | Most studies consistent and inconsistency may be explained |  |  |
| **Clinical impact** |  | Substantial |  |  |
| **Generalisability** |  | Population(s) studied in the body of evidence are similar to the target population |  |  |
| **Applicability** |  | Applicable to Australian healthcare context with few caveats |  |  |

a Level of evidence determined from the NHMRC evidence hierarchy—Table 19

b If there is only 1 study, rank this component as ‘not applicable’.

Source: adapted from [NHMRC (2008](#_ENREF_143))

### Impact on health outcomes

One study assessed the difference in severity of disease at the time of surgery between those who had prophylactic surgery and those who had curative surgery. This cohort study had a high risk of bias. It showed a moderate difference in the severity of MTC (as determined by histopathology results) and was generalisable to the Australian population and applicable to the healthcare system (Table 72).

Table Body of evidence matrix for the assessment of a whether the change in management due to RET mutation testing results in differential health outcomes

| **Component** | **A** | **B** | **C** | **D** |
| --- | --- | --- | --- | --- |
| **Excellent** | **Good** | **Satisfactory** | **Poor** |
| **Evidence basea** |  |  |  | Level II study with a high risk of bias |
| **Consistencyb** | N/A | | | |
| **Clinical impact** |  |  | Moderate |  |
| **Generalisability** |  | Population(s) studied in the body of evidence are similar to the target population |  |  |
| **Applicability** |  | Applicable to Australian healthcare context with few caveats |  |  |

N/A = not applicable

a Level of evidence determined from the NHMRC evidence hierarchy—Table 19

b If there is only 1 study, rank this component as ‘not applicable’.

Source: adapted from [NHMRC (2008](#_ENREF_143))

## What are other relevant considerations?

With any genetic test there are ethical issues that should be considered, such as matters of informed consent, privacy and confidentiality, and the balance of risks and harms from information regarding genetic testing. Given that it has clear treatment and screening implications for either the individual or their family members, RET mutation testing would on balance appear useful, as long as patients are fully informed on the implications of testing and give true informed consent following accredited genetic counselling.

Controlled studies assessing the direct health impact of RET mutation testing are unlikely to be able to be performed in the current clinical environment, where RET mutation testing is considered best practice for those considered at risk of MEN2 ([Learoyd & Robinson 2005](#_ENREF_116)). Withholding mutation testing for the purposes of further research would therefore be considered unethical.

## What are the economic considerations?

The quantitative results derived from the economic model are highly uncertain, as many of the inputs were based on non-comparative data or data with a high risk of bias, including those that quantify the extent of estimated clinical benefit in family members (i.e. the RR of developing advanced MTC; the penetrance of symptomatic MTC; and the expected timeframes of testing, prophylactic thyroidectomy and development of symptomatic MTC in at-risk patients). Nevertheless, while the quantification is uncertain, the directions of the effect of RET mutation testing on clinical practice and expected outcomes are consistently associated with a decrease in overall expenditure and an increase in overall health in the relevant cohort. Cost savings can be expected, although their extent ranges significantly depending on patient uptake rates and adherence to compliance. Likewise, a net benefit in health outcome benefits is consistently shown, although the rate of symptomatic MTC with and without prophylactic thyroidectomy, the accuracy of the translation into QALYs, and patient uptake rates influence the extent of these benefits, and accurate estimations of these are unknown. Therefore, the conclusion—that, in economic terms, the intervention of RET mutation testing dominates the alternative (historical) clinical practice—is robust.

Except where practically no clinical benefit (RR>0.97) is assumed for RET mutation testing, with respect to long-term relapse outcomes following thyroidectomy, all sensitivity analyses demonstrate that RET mutation testing across index patients and first- or second-degree family members is cost-effective. Although an unbiased estimate of the RR of MTC relapse following prophylactic thyroidectomy undertaken on the basis of RET mutation testing rather than biochemical screening is not available, the available data support the RR used in the base-case analysis (RR 0.56), such that the upper limit of the RR as tested in the sensitivity analysis is extreme, and cost-effectiveness is almost certain in clinical practice.

The number of patients likely to be treated requires estimations of both potential new index cases requiring full *RET* gene screening and of family members who may undertake familial screening. MEN2 is a relatively rare condition and the most common first-presenting symptom is MTC.

The estimate of the population suspected of having MEN2 and eligible for diagnostic RET mutation testing is 130–260 patients in 2013, increasing to 147–294 in 2015. This is based on the number of Australians diagnosed with thyroid cancer, and assuming 5–10% of cases are MTC ([AIHW 2010](#_ENREF_3); [Keatts & Itano 2006](#_ENREF_103)). This approach is used as MTC is the most common first presentation of MEN2 (and while this will likely include patients diagnosed after familial screening, it omits patients whose initial presentation is phaeochromocytoma, thus providing a reasonable proxy overall). An annual increase in the incidence of thyroid cancer (and MTC) of 6.3% has been projected based on the average annual increase in thyroid cancer in Australia 2005-09. Uncertainty is likely contained within the estimated range.

The likely number of eligible family members who elect to have predictive RET mutation tests is estimated to be 150–359 in 2013, increasing to 169–406 in 2015. This estimate assumes that 25–30% of diagnostic RET mutation tests performed identify a patient with a hereditary RET mutation ([Raue & Frank-Raue 2010](#_ENREF_164)) and that each index patient has 11.5 first- or second-degree relatives eligible for predictive RET mutation testing ([Suthers et al. 2006](#_ENREF_193)). It is also assumed that only 40% of eligible relatives accept (i.e. uptake) familial screening ([Suthers et al. 2006](#_ENREF_193)). The estimates of predictive RET mutation testing are more uncertain than the estimates of diagnostic testing given they are further dependent on family size and uptake rates and therefore familial screening rates are potentially underestimated, although non-MBS data does not suggest this ([Suthers 2008b](#_ENREF_192)).

It is assumed that each patient will only require one test per lifetime, as the relevant germline RET mutations are stable. Therefore, the number of tests is equivalent to the number of patients. (While there would conceivably be cases of false positives/negatives that are at odds with clinical observations, or non-determinable results that may prompt re-testing, the expected re-test rate would be low and has not been considered).

The base case of the model assumes that uptake rates of RET mutation testing will remain the same as uptakes rates of biochemical/imaging surveillance. This is assumed to occur in all potential index cases presenting with potential symptoms of MEN2. For family members it is estimated that, in each comparison arm (and in each scenario), 40% of the eligible population would receive either testing or biochemical/imaging surveillance. The economic analysis is not highly sensitive to uptake rates, with extreme proportions (in the range 15–100%) being tested in the sensitivity analyses and cost-effectiveness still evident.

# Conclusions

## Safety

There was no evidence regarding the comparative safety of RET mutation testing and subsequent management, versus the historical comparator prior to RET mutation testing. However, it is likely that RET mutation testing has allowed total thyroidectomies to be performed at an earlier stage of disease, or prior to any clinical signs of MTC, which allows a less aggressive form of surgery to be performed. Although there was no direct comparative evidence identified to show this, it is highly likely that more conservative surgical techniques are safer for patients than surgical techniques performed once clinical signs of MTC exist.

## Effectiveness

RET mutation testing was found to be associated with treatment at a younger age and lower severity of disease (less chance of having MTC or nodal metastases at time of surgery). These two factors have been found to greatly reduce the risk of having residual or recurrent disease. Although the direct evidence comparing the rates of residual or recurrent disease by era (pre- versus post-RET mutation testing) is at risk of bias due to differences in the length of follow-up between the cohorts, it is logical to expect that health outcomes following diagnosis with RET mutation testing have significantly improved.

An overall summary of the body of evidence is that it may be graded ‘C’ for ‘satisfactory’. The studies themselves were at high risk of bias due to being historical controlled studies. However, they were all consistent both in the direction of effect and between different measures of effectiveness. The clinical impact is expected to be substantial, and the evidence is considered to be generalisable to the Australian population suspected to be at risk of having MEN2 and applicable to the Australian healthcare setting.

### Diagnostic accuracy

The diagnostic accuracy of RET mutation testing was not performed, as no studies provided long-term data to enable true disease status to be known. In addition, the reference standard of ‘long-term clinical data’ is inherently flawed if patients have had a prophylactic total thyroidectomy for MTC—removal of the thyroid should prohibit the development of an MTC, which can be the only clinical presentation of MEN2A for some patients and all patients with FMTC.

### Impact on patient management

Four different articles identified cases where patients who had either undergone or been referred for total thyroidectomy based on calcitonin levels were found not to have MTC (if they had undergone surgery), and were later identified as being RET-mutation-negative. It is expected that RET mutation testing as part of the diagnostic strategy can prevent unnecessary surgery in those not at risk of MEN2.

Uncontrolled studies showed that a substantial proportion of patients who were gene carriers underwent prophylactic total thyroidectomy based on the results of mutation testing.

### Impact on health outcomes

One cohort study (level III-2 interventional evidence) with a high risk of bias showed that a greater proportion of those who underwent total thyroidectomy on the basis of raised calcitonin, compared with those where surgery was considered prophylactic, had disease that had advanced into the lymph nodes or distant metastases. Consistent with the direct evidence, it can be expected that prophylactic surgery based on RET mutation testing allows surgery at an earlier stage of disease, which decreases the chances of residual or recurrent disease.

## Other relevant considerations

RET mutation testing should always be accompanied by accredited genetic counselling to assist patients in understanding the implications of being tested and in the interpretation of test results.

RET mutation testing is already considered best practice, and *not* testing patients suspected of MEN2 for RET mutations would be considered negligent.

## Economic considerations

Overall, RET mutation testing in both potential index cases and their family members, as described in the proposed listing, is almost certainly cost-effective, particularly where the index case presents with MTC but also in phaeochromocytoma where consideration of surveillance of family members is included. The economic model, although lacking in high-quality data, suggests that use of RET mutation testing almost certainly results in improved health outcomes and decreased costs compared with hypothetical scenarios where RET mutation testing is not available.

### Costing

The expected uptake of RET mutation testing in proband patients is estimated at 35–85 tests annually (for 35-85 patients – ie 1 test per patient).

The expected uptake of predictive RET mutation testing in first- or second-degree family members is estimated to be 160–380 tests annually, for the same number of people.

The total cost to the MBS for RET mutation testing is estimated to be $75,000–$160,000 annually.

The total cost to the Australian healthcare system, including the MBS, for the intervention/procedure is not expected to change given that testing is routine clinical practice that is currently funded through state hospital budgets or private sources.

# Appendix Health Expert Standing Panel and Assessment Group

**Health Expert Standing Panel (HESP)**

Member Expertise or affiliation

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Name Position

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**Noted conflicts of interest**

There were no conflicts of interest.

# Appendix Uncontrolled studies reporting persistence or recurrence of disease

Table 73 Persistence or recurrence following RET mutation testing and subsequent treatment

| Study and location | Level of evidence | Study population | Intervention | Incidence of persistence or recurrence | Follow-up |
| --- | --- | --- | --- | --- | --- |
| In patients with MTC or CCH | | | | | |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | N=30 RET M+ family members of patients with MTC reclassified from sporadic MTC to FMTC or MEN2A due to a RET mutation, who showed clinically and/or biochemical signs of disease on screening:  29 FMTC  1 MEN2A | RET mutation testing, method changed over 15 years  Initially used direct DNA sequencing of exons 10,11 and 16; later added exons 13–15; and recently added exons 5 and 8  Total thyroidectomy | 3/30 (10%) persistence or recurrence  27/30 (90%) disease free | Mean = 6.0 years |
| ([Gonzalez et al. 2003](#_ENREF_81))  Mexico | IV Interventional evidence  High quality (5/6) | N=17 RET M+ family members (plus probands) from 5 MEN2 families:  14 with MEN2A (14 x 634 mutations)  3 with MEN2B (3 x 918 mutations) | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16, and direct DNA sequencing of exons 13–15  Clinical screening  Thyroidectomy | 14/17 (82.4%) disease free  1/17 (5.9%) alive with disease  2/17 (11.8%) dead from MTC | Mean = 6.7 years  (range 1–24 years) |
| ([Dralle et al. 1998](#_ENREF_49))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=75 RET M+  patients <20 years of age identified retrospectively through a questionnaire | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy | 3/75 (4%) persistent elevated calcitonin postoperatively | Postoperative |
| ([Milos et al. 2008](#_ENREF_136))  Romania, Germany, Chile, Brazil, Argentina, Hungary, Spain, The Netherlands, Czech Republic, Poland, USA | IV interventional evidence  Moderate quality (4/6) | N=34 carriers of RET C634W mutation from 20 unrelated MEN2A families for whom postoperative data were available | RET mutation testing (method not stated)  Thyroidectomy | 9/34 (26.5%) MTC patients with postoperative data showed raised serum calcitonin levels | Postoperative |
| ([Quayle et al. 2004](#_ENREF_162))  USA | IV interventional evidence  Moderate quality (3/6) | N=38 RET M+ patients with MEN2 or FMTC diagnosed when over 50 years of age who had a thyroidectomy | RET mutation testing (method not stated)  Total thyroidectomy | 17/30 (56.7%) patients had increased pentagastrin-stimulated calcitonin levels  5/38 (13.2%) patients had repeat neck exploration procedures for recurrent or persistent disease  3/38 (7.9%) patients had distant metastases detected during follow-up period | Median = 6.4 years |
| ([Yoshida et al. 2009](#_ENREF_215))  Japan | IV interventional evidence  Moderate quality (3/6) | N=12 adults who underwent total thyroidectomy for MTC and had MEN2 | RET mutation testing (method not stated) and total thyroidectomy (unclear whether treatment decisions influenced by RET mutation) | 1/12 (8.3%) patients had surgery that was not considered curative, and died of advanced metastatic MTC at 1 year postoperatively |  |
| 11/12 (91.7%) patients had no recurrence of MTC | Median = 8.8 years |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Low quality (2/6) | N=6 index cases with a RET Y791F mutation who had total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | 1/6 (16.7%) had increased calcitonin levels  4/6 (66.7%) had normal calcitonin levels  1/6 (16.7%) died | Up to 15 years |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV interventional evidence  Poor quality (2/6) | N=5 members of an MTC family with a RET L790F mutation (including index patient), who had a thyroidectomy  All had abnormal pentagastrin-stimulated calcitonin levels  3 had clinical signs of disease | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–15, and restriction site polymorphism analysis of exon 16  Direct DNA sequencing of exon 13 in family members  Clinical screening  Prophylactic thyroidectomy | 0/5 (0%) had recurrent disease  All patients were cured, with a mean follow-up time of 6.6 years | Mean = 6.6 years (range 6–8 years) |
| **In asymptomatic family members** | | | | | |
| ([Skinner et al. 2005](#_ENREF_191))  USA | IV interventional evidence  High quality (5/6) | N=50 RET M+ patients from MEN2A families, who were <20 years of age at time of thyroidectomy | RET mutation testing by restriction site polymorphism analysis and/or direct DNA sequencing of RET exons 10, 11, 13, 14 and 16  Total thyroidectomy | 6/50 (12%) had persistent or recurrent disease detected  *Post hoc analyses:*  Thyroidectomy patients younger than 8 years of age:  0/22 had raised calcitonin levels postoperatively  Thyroidectomy over age 8 years:  6/28 had raised calcitonin levels postoperatively  No evidence that recurrence or persistence was dependent on codon (p=0.92) | Range = 5–10 years |
| ([Lau et al. 2009](#_ENREF_114))  Hong Kong | IV interventional evidence  High quality (5/6) | N=22 asymptomatic patients from 8 MEN2A families, who underwent prophylactic total thyroidectomy based on RET mutation status | RET mutation testing by restriction enzyme analysis and/or direct DNA sequencing (exons not specified)  Prophylactic thyroidectomy with or without a unilateral central compartment neck dissection | 5/22 (22.7%) patients were diagnosed with PCC and had adrenalectomy  2/22 (9.1%) patients had mild HPT  1/22 (4.5%) patient had fluctuating thyroid function test results, despite good thyroxine replacement compliance after 1 year  0/22 (0%) had clinical, biochemical or ultrasonographic evidence of MTC recurrence | Median = 49 months (range 13–128 months) |
| ([Wells Jr & Skinner 1998](#_ENREF_208))  USA | IV interventional evidence  High quality (5/6) | N=18 first-degree relatives aged 21 years or younger, from 7 MEN2A kindreds with no clinical symptoms, who were RET M+ and underwent a thyroidectomy | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 10 and 11  Prophylactic thyroidectomy with bilateral cervicocentral lymphadenectomy | 18/18 (100%) had normal postoperative pentagastrin-stimulated calcitonin levels  13/13 (100%) had normal pentagastrin-stimulated calcitonin levels after 3 years | Range = 0–3 years |
| ([Schellhaas et al. 2009](#_ENREF_179))  Germany | IV interventional evidence  High quality (5/6) | N=17 patients with a RET codon 634 mutation:  14 with MEN2A  3 with apparent FMTC | RET mutation testing (method not stated)  Prophylactic total thyroidectomy with bilateral cervicocentral lymphadenectomy | 2/17 (11.8%) with increased calcitonin levels during follow-up  1 was a 36-year-old at operation who presented with T2N1 neoplasm  1 was a 9-year-old at operation who presented with T1 MTC and lymph node metastases  Lymph node status associated with probable recurrent/persistent MTC (p=0.024) | Median = 13.3 years |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV Interventional evidence  Moderate quality (4/6) | N=35 RET M+ members of an FMTC family with a RET G533C mutation  All underwent thyroidectomy  Data were not available for 7 (recent surgery) | RET mutation testing by direct DNA sequencing of exon 8  Prophylactic thyroidectomy | 20/28 (71.4%) normal postoperative calcitonin levels  (baseline <11.5 pg/mL)  8/28 (28.6%) elevated calcitonin (6 with lymph node metastases and 2 without) | Postoperative |
| ([Franz & Wells Jr 1997](#_ENREF_71))  USA, Germany | IV Interventional evidence  Moderate quality (4/6) | N=20 RET M+ patients:  19 MEN2A patients  1 FMTC patient | RET mutation testing by restriction site polymorphism analysis and/or DNA sequencing (exons not specified)  Clinical screening  Prophylactic thyroidectomy based on RET status | 20/20 (100%) normal postoperative calcitonin levels | Postoperative |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV interventional evidence  Moderate quality (4/6) | N=18 RET M+ patients (aged 17 months – 21 years) had total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, confirmed by restriction site polymorphism analysis  Prophylactic thyroidectomy | 17/18 (94.4%) had normal postoperative calcitonin levels  In 1 juvenile patient with regional lymph node metastases calcitonin levels remained high after surgery | Postoperative |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV Interventional evidence  Moderate quality (4/6) | N= 16 RET M+ members of separate families with hereditary MTC, who had a thyroidectomy | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14  Clinical screening  Total thyroidectomy | 5/16 (31.3%) had elevated postoperative pentagastrin-stimulated calcitonin levels | Postoperative |
| ([Decker et al. 1996](#_ENREF_39))  USA | IV Interventional evidence  Moderate quality (4/6) | N=11 children from confirmed MEN2A patients who underwent prophylactic thyroidectomy | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10 and 11  Clinical screening  Prophylactic thyroidectomy | 11/11 (100%) normal postoperative calcitonin levels | Postoperative |
| ([Frank-Raue et al. 1997](#_ENREF_68))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=11 asymptomatic children with a RET mutation from 8 MEN2A/FMTC families | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing or restriction site polymorphism analysis of exons 10, 11 and 13  Prophylactic thyroidectomy | 11/11 (100%) normal postoperative calcitonin levels | Postoperative |
| ([Frank-Raue et al. 1996](#_ENREF_69))  Germany | IV interventional evidence  Moderate quality (4/6) | N=9 presymptomatic RET M+ patients from families clinically identified with hereditary MTC, who had prophylactic thyroidectomy  Data available for 8 patients | RET mutation testing by single-strand conformation polymorphism analysis, and then either direct DNA sequencing or restriction site polymorphism analysis of exons 10 and 11  Direct DNA sequencing of exons 13 and 16  Clinical screening  Prophylactic thyroidectomy | 8/8 (100%) normal postoperative calcitonin levels | Postoperative |
| ([Lombardo et al. 2002](#_ENREF_126))  France and Italy | IV interventional evidence  Moderate quality (3/6) | N=31 patients with RET V804L mutations from 5 families, who underwent thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15, and restriction site polymorphism analysis of exon 16  Clinical screening  Total thyroidectomy | 3/31 (9.7%) patients had elevated pentagastrin-stimulated calcitonin levels postoperatively  With a median follow-up of 8.5 years, no biochemical or clinical recurrence of MTC was observed | Median = 8.5 years |
| ([Rodriguez Gonzalez et al. 2002](#_ENREF_170))  Spain | IV interventional evidence  Moderate quality (3/6) | N=22 patients at risk of MEN2A who were found to be RET M+ and received prophylactic thyroidectomy  All had RET codon 634 mutations | RET mutation testing by denaturing gradient gel electrophoresis of exons 10, 11, 13, 14 and 16, confirmed by restriction site polymorphism analysis  Clinical screening  Prophylactic total thyroidectomy ± central neck dissection | 1/22 (4.5%) slightly elevated calcitonin after pentagastrin stimulation after 2 years | Mean = 23 months (range 6–57 months) |
| ([Lindskog et al. 2004](#_ENREF_124))  Sweden | IV interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members of a MEN2A family with a RET codon 618 mutation, who had a thyroidectomy  15 were identified by biochemical screening | RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central neck dissection | 7/12 (58.3%) patients tested had elevated pentagastrin-stimulated calcitonin levels | Mean = 19±9 years |
| ([Jung et al. 2010](#_ENREF_98))  Korea | IV interventional evidence  Moderate quality (3/6) | N=8 RET M+ members of a 3-generation FMTC family (including index case), who underwent total thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 of index case  Analysis of exon 10 in family members  Total thyroidectomy with either central neck dissection or modified radical neck dissection | 3/8 (37.5%) had elevated pentagastrin-stimulated calcitonin levels | Median = 10 years |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Low quality (2/6) | N=12 RET M+ family members who had total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | 1/12 (8.3%) patient had recurrent MTC  11/12 (91.7%) patients had normal calcitonin levels | Up to 15 years |

CCH = C-cell hyperplasia; HPT = hyperparathyroidism; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

# Appendix Uncontrolled studies reporting incidence and severity of MTC

Table 74 Incidence and severity of MTC in index cases and clinically affected relatives

| Study and location | Level of evidence | Study population | Intervention | No disease | Medullary micro-carcinoma | C-cell hyperplasia (without MTC) | MTC (+/– C-cell hyperplasia) | Lymph node metastases |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV interventional evidence  High quality (5/6) | N=23 unrelated RET M+ index cases with MTC:  4 FMTC  9 MEN2A  4 MEN2B  6 sporadic MTCs  Note: Data for 1 FMTC index case was not available | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Thyroidectomy  Surgical decisions often made before RET mutation testing with no separation of data based on clinical or genetic diagnosis | 0/22 | Not stated | 0/22 | 22/22 (100%):  2 x T1N0M0  3 x T2N0M0  2 x T2N2M0  3 x T3N0M0  1 x T3N0M1h  1 x T3N0M1hpp  1 x T3N2M0  1 x T3N2M1h  4 x T4N0M0  1 x T4N2M0  1 x T4N2M1h  1 s T4N2M1pp  1 x T4N2Mx | 9/22 (40.9%) |
| ([Neumann et al. 2002](#_ENREF_141))  Germany and Poland | IV interventional evidence  High quality (5/6) | N=13 RET M+ patients with non-syndromic PCC without family history of disease | RET mutation testing by single-strand conformation polymorphisms and direct DNA sequencing of exons 13–16 | Not stated (unclear if 1/13 is disease free or has CCH) | Not stated | Not stated | 12/13 (92.3%) | Not stated |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV Interventional evidence  Moderate quality (4/6) | N=17 index cases with MEN2A  N=5 index cases with MEN2B | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, confirmed by restriction site polymorphism analysis  Prophylactic thyroidectomy | MEN2A: 0/17  MEN2B: 0/5 | Not stated | MEN2A: 0/17  MEN2B: 0/5 | MEN2A: 17/17 (100%)  MEN2B: 5/5 (100%) with aggressive MTC | Not stated |
| ([Boer et al. 2003](#_ENREF_19))  Hungary | IV Interventional evidence  Moderate quality (4/6) | N=14 consecutive unrelated patients with MTC admitted for genetic screening for MEN2A and FMTC, who were found to be RET M+ and underwent a thyroidectomy | RET mutation testing by direct DNA sequencing (exons not specified)  Thyroidectomy | 0/14 (0%) | Not stated | 5/14 (35.7%) | 9/14 (64.3%) | 3/11 (27.3%) of those examined |
| ([Gimm et al. 2002](#_ENREF_80)).  Germany, Austria | IV interventional evidence  Moderate quality (4/6) | N=13 index patients with a RET codon 790/791 mutation who underwent thyroid operations:  7 had RET L790F mutation  6 had RET Y791F mutation | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11, 13 and 14  Total thyroidectomy with or without lymph node dissection | 1/13 (7.7%)  0/7 (0%) L790F mutation  1/6 (16.7%) Y791F mutation | Not stated | 1/13 (7.7%)  0/7 (0%) L790F mutation  1/6 (16.7%) Y791F mutation | 11/13 (84.6%):  7/7 (100%) L790F mutation  4/6 (66.7%) Y791F mutation | 5/13 (38.5%)  4/7 (57.1%) L790F mutation  1/4 with distant metastases  1/6 (16.7%) Y791F mutation |
| ([Abdelhakim et al. 2009](#_ENREF_1)) Morocco | IV Interventional evidence  Moderate quality (4/6) | N=9 index patients with diagnosed MTC:  3 were RET M+:  2 MEN2A  1 unclassified  6 were suspected sporadic MTC (RET M–) | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16  Total thyroidectomy | 0/9 (0%) | Not stated | Not stated | 9/9 (100%)  Severity not stated | Not stated |
| ([Spinelli et al. 2010](#_ENREF_193))  Italy | IV interventional evidence  High quality (4/6) | N=7 patients with MEN2 who underwent curative surgery for MTC | RET mutation testing by direct DNA sequencing (exons not specified)  Curative or prophylactic total thyroidectomy | 0/7 (0%) | Not stated | 0/7 (0%) | 7/7 (100%):  5 T1N0M0  2 T4N1M1 | 2/7 (28.6%) |
| ([Ameur et al. 2009](#_ENREF_11))  France | IV Interventional evidence  Moderate Quality (3/6) | N=21 tissue samples collected from MTC, CCH, MCC or mixed MTC patients that were found to have a germline RET mutation | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 from normal and diseased tissue samples to determine germline and somatic RET status | 0/21 (0%) | 1/21 (4.7%) | 2/21 (9.5%) | 18/21 (85.7%):  7 x T1N0M0  5 x T1N1M0  2 x T1N1Mx  1 x T2N0M0  1 x T2N1M0  2 x T3N1M0 | 10/21 (47.6%) |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV interventional evidence  Moderate quality (3/6) | N=13 patients with sporadic MTC found to be RET M+ | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction site polymorphism analysis of exon affected in index case  Total thyroidectomy | 0/13 (0%) | Not stated | Not stated | 13/13 (100%):  1 x T1bN1aM0  2 x T1bN1bM0  1 x T2aN0M0  3 x T2aN1aM0  3 x T2bN0M0  2 x T2bN1bM0  1 x T3aN1bM0 | 9/13 (69.2%) |
| ([Machens et al. 2001](#_ENREF_130))  Germany | IV interventional evidence  Poor quality (2/6) | N=63 RET M+ patients with MTC who had a thyroidectomy  36 were index patients | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13 and 14  Thyroidectomy |  |  |  | 63/63 (100%):  38/63 T1  18/63 T2  4/63 T3  3/63 T4 | 37/63 (58.7%)  6 with distant metastases |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV Interventional evidence  Poor quality (2/6) | N=6 index cases with a RET Y791F mutation had a total thyroidectomy:  1 MEN2B case  1 MEN2A case  3 apparently sporadic MTCs  1 PCC case | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | 0/6 (0%) | 2/6 (33.3%) | 2/6 (33.3%) | 4/6 (66.7%):  1 T2NxMx  1 T1N1M0  2 T4N1M0 | 3/6 (50.0%) |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV interventional evidence  Poor quality (2/6) | N=5 members of an MTC family with a RET L790F mutation (including index patient), who had a thyroidectomy  All had abnormal pentagastrin-stimulated calcitonin levels  3 had clinical signs of disease | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–15, and restriction site polymorphism analysis for exon 16  Direct DNA sequencing of exon 13 in family members  Clinical screening  Prophylactic thyroidectomy | 0/5 (0%) | Not stated | Not stated | 5/5 (100%):  5 T1N0M0 | 0/5 (0%) |

CCH = C-cell hyperplasia; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

Table Incidence and severity of MTC in family members

| Study and location | Level of evidence | Study population | Intervention | No disease | MCC (medullary microcarcinoma) | C-cell hyperplasia (without MTC) | MTC | Lymph node metastases | |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ([Machens et al. 2005](#_ENREF_129))  Germany | IV interventional evidence  High quality (5/6) | N=206 consecutive RET M+ patients who underwent surgery for CCH, MTC or PCC:  74 index cases  132 non-index cases (criteria for diagnosis and/or surgery not reported) | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16  Clinical screening  Thyroidectomy and/or adrenalectomy | Not stated | Not stated | Not stated | 135/206 (65.5%):  18/18 (100%) highest risk (RET codon 918)  85/117 (72.6%) high risk (RET codons 609–634)  32/71(45.1%) less high risk (RET codons 768–891) | Not stated |
| ([Nguyen et al. 2001](#_ENREF_143))  France | IV interventional evidence  High quality (5/6) | N=87 first-degree relatives of index cases in MEN2 families, who were diagnosed with MTC and found to be RET M+:  84 patients from 52 MEN2A families  3 patients from 3 MEN2B families | MEN2 diagnosed by linkage analysis between 1989 and 1994  RET mutation testing by sequence analysis since 1994 (method not stated)  Total thyroidectomy | 0/87 (0%) | Not stated | 0/87 (0%) | 87/87 (100%) | Not stated | |
| ([Skinner et al. 2005](#_ENREF_191))  USA | IV interventional evidence  High quality (5/6) | N=50 RET M+ patients from MEN2A families, who were <20 years of age at time of thyroidectomy | RET mutation testing by restriction site polymorphism analysis and/or direct DNA sequencing of RET exons 10, 11, 13, 14 and 16  Total thyroidectomy | 4/50 (8%) | Not stated | 13/50 (26%) | 33/50 (66%) | Not stated | |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | N=30 RET M+ family members of patients with MTC reclassified from spontaneous MTC to FMTC or MEN2A due to RET mutation, who showed clinically and/or biochemical signs of disease on screening and underwent a total thyroidectomy:  29 phenotype FMTC  1 phenotype MEN2A | RET mutation testing method changed over 15 years  Initially used direct DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8  Total thyroidectomy | 1/30 (3.3%) | Not stated | 6/30 (20%) | 23/30 (76.6%):  15 x T1N0M0  3 x T1mN0M0  1 x T1bN0M0  1 x T1AN0M0  1 x T1N1aM0  1 x T3mN0M0  1 x T2MN1M0 | 2/30 (6.7%) | |
| ([Lau et al. 2009](#_ENREF_114))  Hong Kong | IV interventional evidence  High quality (5/6) | N=22 asymptomatic patients from 8 MEN2A families, who underwent prophylactic total thyroidectomy  All had RET codon 634 mutations | RET mutation testing by restriction enzyme analysis and/or direct DNA sequencing (exons not specified)  Prophylactic thyroidectomy with or without a unilateral central compartment neck dissection | 1/22 (4.5%)  RET C634W mutation | Not stated | 4/22 (18.2%)  3 RET C634Y  1 RET C634W | 17/22 (77.3%):  9 RET C634Y  4 RET C634R  1 RET C634W  3 RET C634G | 0/22 (0%) | |
| ([Wells Jr & Skinner 1998](#_ENREF_208))  USA | IV interventional evidence  High quality (5/6) | N=18 RET M+ first-degree relatives aged 21 years or younger from 7 MEN2A kindreds with no clinical symptoms, who underwent a thyroidectomy | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 10 and 11  Prophylactic thyroidectomy with bilateral cervicocentral lymphadenectomy | 0/18 (0%) | 13/18 (72.2%) | | 5/18 (27.7%) | 0/18 (0%) | |
| ([Schellhaas et al. 2009](#_ENREF_179))  Germany | IV interventional evidence  High quality (5/6) | N=17 patients with mutation in codon 634:  14 from MEN2A  3 with apparent FMTC | RET mutation testing (method not stated)  Prophylactic thyroidectomy with bilateral cervicocentral lymphadenectomy | 0/17 (0%) | Not stated | 3/17 (17.6%)  Aged 5, 6 and 9 years | 14/17 (82.4%):  12/17 T1 MTC  2/17 T2 MTC (according to 1997 TNM classification, or 14/17 T1 by 2002 classification) | 2/17 (11.7%)  Aged 9 and 36 years | |
| ([Pinna et al. 2007](#_ENREF_159))  Italy | IV interventional evidence  High quality (5/6) | N=14 RET M+ family members who have undergone prophylactic total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 8–16 | 1/14 (7.1%) | Not stated | 1/14 (7.1%) | 12/14 (85.7%) | Not stated | |
| ([Frank-Raue et al. 2011](#_ENREF_70))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=340 patients proven to be carriers of a germline mutation in exon 10 of the *RET* gene  Identified through:  47% symptomatic  53% screening | RET mutation testing method not disclosed  Analysis of clinical and demographic data | Not stated | Not stated | Not stated | 263/340 (77%) | Not stated | |
| ([Milos et al. 2008](#_ENREF_136))  Romania, Germany, Chile, Brazil, Argentina, Hungary, Spain, The Netherlands, Czech Republic, Poland, USA | IV interventional evidence  Moderate quality (4/6) | N=81 carriers of RET C634W mutation from 20 unrelated MEN2A families, who underwent thyroidectomy | RET mutation testing (no methods stated)  Thyroidectomy. | 6/81 (7.4%) | Not stated | 7/81 (8.6%) | 68/81 (88%):  52% by age 30 years  83% by age 50 years  Distant metastases 4/61 cases (aged 28–69 years) | 16/61 (26%) had lymph node metastases (aged 20–72 years) | |
| ([Dralle et al. 1998](#_ENREF_49))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=75 RET M+ patients <20 years of age who underwent prophylactic thyroidectomy | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy  Retrospectively identified through questionnaire | 0/75 (0%) | Not stated | 29/75 (38.6%) | 46/75 (61.3%) | Not stated | |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia and New Zealand | IV interventional evidence  Moderate quality (4/6) | N=57 members of 2 families:  Family 1: RET M+ 22 family members from 4 generations with RET V804L mutation  Family 2: 5 RET M+ family members from 3 generations with RET V804M mutation | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16  From 1998, analysis also of exons 13–15 for probands  Family members screened for family RET mutation  Prophylactic thyroidectomy | Family 1: 2/22 (9.1%)  Family 2: 0/3 (0%) | Not stated | Family 1: 11/22 (50%)  Family 2: 2/3 (66.7%) | Family 1: 9/22 (40.9%)  Family 2: 1/3 (33.3%) | Family 1: 1/22 (4.5%)  (proband)  Family 2: 1/3 (33.3%) | |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV Interventional evidence  Moderate quality (4/6) | N=35 RET M+ members of a large extended FMTC family with a RET G533C mutation, who underwent a total thyroidectomy | RET mutation testing by direct DNA sequencing of exon 8  Prophylactic thyroidectomy | 0/35 (0%) | Not stated | 6/35 (17.1%) | 29/35 (82.8%) | 11/35 (31.4%) | |
| ([Etit et al. 2008](#_ENREF_61))  USA | IV Interventional evidence  Moderate quality (4/6) | N=42 specimens from patients retrospectively identified from hospital records, who underwent a prophylactic thyroidectomy for possible MTC  32 underwent RET mutation testing  31 with family history:  22 MEN2A  1 MEN2B  8 non-MEN  27 were RET M+ | RET mutation testing by direct DNA sequencing analysis of exons 10, 11 and 13–16 | 3/42 (7.1%)  RET M+  3/27 (11.1%) | 29/42 (71.4%) | 9/42 (21.4%)  RET M+  8/27 (29.6%) | 30/42 (71.4%)  RET M+  16/27 (59.3%) | 22/42 (52.4%) | |
| ([Gimm et al. 2002](#_ENREF_80)).  Germany, Austria | IV interventional evidence  Moderate quality (4/6) | N = 27 patients identified during RET mutation screening with a RET codon 790/791 mutation who underwent thyroid operations.  16 had RET L790F mutation  11 had RET Y791F mutation | RET mutation testing by single strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11, 13 and 14.  Total thyroidectomy.with or without lymph node dissection | 5/27 (18.5%)  2/16 (12.5%) RET L790F mutation  3/11 (27.3%) RET Y791F mutation | Not stated | 14/27 (51.9%)  6/16 (37.5%) RET L790F mutation  8/11 (72.7%) RET Y791F mutation | 8/27 (29.6%)  8/16 (50%) RET L790F mutation  0/11 (0%) RET Y791F mutation | 2/27 (7.4%)  2/16 (12.5%) RET L790F mutation  0/11 (0%) RET Y791F mutation | |
| ([Gosnell et al. 2006](#_ENREF_82))  Australia | IV Interventional evidence  Moderate quality (4/6) | N=22 RET M+ members of a single MEN2A kindred with a RET codon 804 mutation, who underwent thyroidectomy | RET mutation testing (method not stated)  Clinical screening  Prophylactic thyroidectomy | 2/22 (9.1%) | 2/22 (9.1%) | 9/22 (40.9%) | 9/22 (40.9%) | Not stated | |
| ([Franz & Wells Jr 1997](#_ENREF_71))  USA, Germany | IV Interventional evidence  Moderate quality (4/6) | N=20 RET M+ patients:  19 MEN2A  1 FMTC | RET mutation testing by restriction site polymorphism analysis and/or DNA sequencing (exons not specified)  Clinical screening  Prophylactic thyroidectomy based on RET status | 0/20 (0%) | Not stated | 20/20 (100%) | | 0/20 (0%) | |
| ([Feldman et al. 2000](#_ENREF_62))  UK, USA, France | IV Interventional evidence  Moderate quality (4/6) | N=20 members from 2 FMTC families who have a RET V804M mutation | RET mutation testing by restriction analysis of exon 14  Clinical screening.  Thyroidectomy | 1/20 (5%) | Not stated | 6/20 (30%) | 13/20 (65%) | Not stated | |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV interventional evidence  Moderate quality (4/6) | N=18 RET M+ juvenile patients from MEN2 families who had a total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, confirmed by restriction site polymorphism analysis  Prophylactic thyroidectomy | 0/18 (0%) | Not stated | 3/18 (16.7%) | 15/18 (83.3%):  7 unilaterally  8 bilaterally | 1/18 (5.6%) | |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV Interventional evidence  Moderate quality (4/6) | N=17 members of 10 MEN2A or FMTC kindreds with known RET mutations who had a thyroidectomy | RET mutation testing by denaturing gradient gel analysis of exons 10 and 11, with confirmatory direct DNA sequencing | 0/17 (0%) | Not stated | 10/17 (58.8%) | 7/17 (41.2%) | Not stated | |
| ([Calva et al. 2009](#_ENREF_27))  USA | IV Interventional evidence  Moderate quality (4/6) | N=16 RET M+ members of a MEN2 family with a RET C609Y mutation, who underwent a thyroidectomy | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy for treatment or prophylaxis | 5/16 (31.3%) | Not stated | 2/16 (12.5%) | 9/16 (56.3%) | 6/16 (37.5%) | |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV interventional evidence  Moderate quality (4/6) | N=16 RET M+ members from 10 families who had available data, and who underwent total thyroidectomy:  11 MEN2A  2 MEN2B  2 FMTC  1 other | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14  Clinical screening  Total thyroidectomy | 0/16 (0%) | Not stated | 1/16 (6.3%) | 15/16 (93.8%) | Not stated | |
| ([Frohnauer et al. 2000](#_ENREF_73))  USA | IV Interventional evidence  Moderate quality (4/6) | N=14 members of 5 MEN2A kindreds who had a RET codon 804 mutation and a thyroidectomy | RET mutation testing by denaturing gradient gel electrophoresis analysis, confirmed by direct DNA sequencing of exon 14  Thyroidectomy | 1/14 (7.1%) | Not stated | 6/14 (42.9%) | 7/14 (50%) | 3/14 (21.4%)  1/14 (7.1%) had distant metastases | |
| ([Guyetant et al. 2003](#_ENREF_86))  France | IV interventional evidence  High quality (5/6) | N=14 potential MEN2 carriers belonging to 9 families who had been operated on for CCH or MTC  3 MEN2A  14 FMTC  7 were children aged <15 years | RET mutation testing of exons 8, 10, 11 and 13–16 (method not stated) | 0/14 (0%) | Not stated | 3/14 (21.4%)  3/7 (42.9%) children | 11/14 (78.6%)  4/7 (57.1%) children | Not stated | |
| ([Heizmann et al. 2006](#_ENREF_89))  Switzerland | IV interventional evidence  Moderate quality (4/6) | N=14 RET M+ patients who were presymptomatic, from 2 MEN2A kindreds:  3 RET C634Y mutations  11 RET C618G mutations | RET mutation testing by single-strand conformation polymorphism analysis, denaturing gradient gel electrophoresis and direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central compartment dissection in those older than 6 years of age | 0/14 (0%) | Not stated | 3/14 (21.4%) | 11/14 (78.5%)  4/14 bilateral MTCs  9 x pT1 pN0  2 x pT1 pN1a | 2/14 (14.3%)  (<2 mm) | |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV Interventional evidence  Moderate quality (4/6) | N=13 RET M+ family members from 7 MEN2A kindreds, who had a thyroidectomy  7 had elevated calcitonin levels | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of affected exon  Thyroidectomy | 0/13 (0%) | Not stated | 13/13 (100%) had evidence of CCH with or without MTC | | Not stated | |
| ([Algun et al. 2002](#_ENREF_6))  Turkey | IV interventional evidence  Moderate quality (4/6) | N=12 RET M+ members from 4 generations of an extended family with MEN2A, who had a thyroidectomy | RET mutation testing by restriction site polymorphism analysis of exon 11  Confirmation with clinical tests  Total thyroidectomy with central lymph node dissection | 3/12 (25%) | 4/12 (33.3%) | 3/12 (25%) | 5/12 (41.7%) | 6/12 (50%)  1 with bone metastases | |
| ([Vestergaard et al. 2007](#_ENREF_206))  Denmark | IV interventional evidence  Moderate quality (4/6) | N=12 first-degree RET M+ family members of index case with a RET Y791F mutation | RET mutation testing by direct DNA sequencing of exon 13  No thyroidectomy | 12/12 had normal pentagastrin-stimulated calcitonin levels | Not stated | Not stated | Not stated | Not stated | |
| ([Frank-Raue et al. 1997](#_ENREF_68))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=11 asymptomatic RET M+ children from 8 MEN2A/FMTC families | RET mutation testing by single-strand conformation polymorphism analysis or restriction site polymorphism analysis, and direct DNA sequencing of exons 10, 11 and 13  Prophylactic thyroidectomy | 0/11 (0%) | Not stated | 5/11 (45.5%) | 6/11 (54.5%) | Not stated | |
| ([Decker et al. 1996](#_ENREF_39))  USA | IV interventional evidence  Moderate quality (4/6) | N=11 RET M+ children of confirmed MEN2A patients from 4 distinct families who underwent prophylactic surgery | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10 and 11  Clinical screening  Prophylactic thyroidectomy | 1/11(9.1%) | Not stated | 9/11 (81.8%) | 1/11 (9.1%) | Not stated | |
| ([Frank-Raue et al. 1996](#_ENREF_69))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=9 presymptomatic RET M+ patients who underwent prophylactic thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis, and then either direct DNA sequencing or restriction site polymorphism analysis of exons 10 and 11  Direct DNA sequencing of exons 13 and 16  Clinical screening  Prophylactic thyroidectomy | 0/9 (0%) | 5/9 (55.6%) | 9/9 (100%) | 0/9 (0%) | Not stated | |
| ([Spinelli et al. 2010](#_ENREF_193))  Italy | IV interventional evidence  High quality (4/6) | N=6 patients with MEN2 who underwent prophylactic surgery for MTC | RET mutation testing by direct DNA sequencing (exons not specified)  Curative or prophylactic total thyroidectomy | 0/6 (0%) | Not stated | 2/6 (33.3%) | 4/6 (66.7%)  T1N0M0 | 0/7 (0%) | |
| ([Marsh et al. 1996](#_ENREF_131))  Australia and New Zealand | IV interventional evidence  Moderate quality (4/6) | N=5 RET M+ asymptomatic members from 2 MEN2A families | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 | 1/5 (20%)had raised pentagastrin-stimulated calcitonin levels | Not stated | 1/5 (20%) | 3/5 (60%) | Not stated | |
| ([Punales et al. 2003](#_ENREF_159))  Brazil | IV interventional evidence  Moderate quality (3/6) | N=50 RET M+ index cases and family members with 634 mutation, who underwent surgery:  43 had clinical disease  7 were clinically asymptomatic gene carriers | RET mutation testing by single-strand conformational polymorphism analysis, restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15  Total thyroidectomy, with a central cervical lymph node dissection in those with increased calcitonin levels | 0/50 (0%) | Not stated | 50/50 (100%) CCH or MTC | | 22/43 (51.2%) with clinical disease  1/7 (14.3%) asymptomatic gene mutation carriers  14/43 (32.6%) with clinical disease had distant metastases | |
| ([Quayle et al. 2004](#_ENREF_160))  USA | IV interventional evidence  Moderate quality (3/6) | N=39 patients with MEN2 or FMTC diagnosed when over 50 years of age  36 patients from MEN2A families  3 from FMTC families  38 with known RET mutation:  5 with codon 609 mutation  15 with codon 618 mutation  6 with codon 620 mutation  12 with codon 634 mutation  1 with unknown mutation | RET mutation testing (method not stated)  Total thyroidectomy | 0/39 (0%) | Not stated | 2/39 (5.1%) | 37/39 (94.9%)  AJCC staging:  12 (34%) stage I  11 (31%) stage II  11 (31%) stage III  1 (3%) stage IV | 3/7 who underwent central node dissection had N1 disease | |
| ([Lombardo et al. 2002](#_ENREF_124))  France and Italy | IV interventional evidence  Moderate quality (3/6) | N=31 patients with RET V804L mutations from 5 families, who underwent thyroidectomy:  3 index cases with MTC  1 with follicular tumour  14 with detectable basal calcitonin levels  13 with significant increase in pentagastrin-stimulated calcitonin levels | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15, and restriction site polymorphism analysis of exon 16  Clinical screening  Total thyroidectomy | 1/31 (3.2%) | Not stated | 12/31 (38.7%) | 18/31 (58.1%):  Before age 40 years: 2/11 (18.2%)  After age 40 years: 16/20 (80%) | 6/31 (19.4%) | |
| ([Erdogan et al. 2007](#_ENREF_55))  Turkey | IV Interventional evidence  Moderate quality (3/6) | N=30 RET M+ patients identified from 15 pedigrees, who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–15, and restriction enzyme analysis of exon 16  Total thyroidectomy | 0/30 (0%) | Not stated | 30/30 (100%) had multifocal MTC and/or CCH | | Not stated | |
| ([Halling et al. 1997](#_ENREF_85))  USA | IV interventional evidence  Moderate quality (3/6) | N=28 RET M+ family members from 1 large FMTC kindred with a RET C609Y mutation, who had thyroidectomy before prior to testing  19 with elevated pentagastrin-stimulated calcitonin levels  N=10 RET M– family members who had thyroidectomy prior to genetic testing, with elevated pentagastrin-stimulated calcitonin levels | RET mutation testing by direct DNA sequencing of exon 10  Clinical screening  Thyroidectomy | RET M+  5/28 (17.9%)  RET M–  1/10 (10%) | Not stated | RET M+  8/28 (28.6%)  RET M–  8/10 (80%) | RET M+  15/28 (53.6%)  RET M–  1/10 (10%) | Not stated | |
| ([Rodriguez Gonzalez et al. 2002](#_ENREF_168))  Spain | IV interventional evidence  Moderate quality (3/6) | N=22 RET M+ patients with normal basal and pentagastrin-stimulated calcitonin levels who received a prophylactic thyroidectomy | RET mutation testing by denaturing gradient gel electrophoresis of exons 10, 11, 13, 14 and 16, confirmed by restriction site polymorphism analysis  Clinical screening  Prophylactic total thyroidectomy ± central neck dissection | 0/22 (0%) | Not stated | 7/22 (31.8%) | 15/22 (68.2%)  14 x T1N0M0  1 x T1N1aM0 | 1/22 (4.5%) | |
| ([Bergant et al. 2006](#_ENREF_15))  Slovenia | IV interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members screened from index patients | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction site polymorphism analysis of exon affected in index case  Total thyroidectomy with central neck dissection | 3/16 (18.8%)  T0N0M0 | Not stated | 2/16 (12.5%) | 13/16 (81.3%):  3 x T1bN0M0  2 x T1bN1bM0  2 x T2bN0M0  4 x T2bN1bM0  1 x T3bN1bM0  1 x T4bN1bM0 | 8/16 (50.0%) | |
| ([Lindskog et al. 2004](#_ENREF_122))  Sweden | IV interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members of a MEN2A family with a RET codon 618 mutation, who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central neck dissection | 0/16 (0%) | Not stated | Not stated | 16/16 (100%):  1 T1NXM0  3 T1N0M0  4 T1N1M0  6 T2N1M0  2 T4N1M1 | 12/16 (75%) | |
| ([Shifrin et al. 2009](#_ENREF_182))  USA | IV interventional evidence  Moderate quality (3/6) | N=15 family members from family with RET V804M mutation, who had total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (not clear if relatives tested for specific mutation or all)  Total thyroidectomy with central and ipsilateral lateral neck dissection | 0/15 (0%) | Not stated | 5/15 (33%) | 10/15 (67%) |  | |
| ([Wu et al. 1998](#_ENREF_211))  Taiwan | IV interventional evidence  Moderate quality (3/6) | N=13 RET M+ first- and second-degree relatives from 2 unrelated MEN2A families | RET mutation testing by direct DNA sequencing of exons 10 and 11 | 5/13 (38.5%) showed no clinical signs of disease | Not stated | Not stated | 8/13 (61.5%) | Not stated | |
| ([Jung et al. 2010](#_ENREF_96))  Korea | IV interventional evidence  Moderate quality (3/6) | N=8 members of a 3-generation FMTC family, who underwent total thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 of index case  Analysis of exon 10 in family members  Total thyroidectomy with either central neck dissection or modified radical neck dissection | 0/8 (0%) | Not stated | 0/8 (0%) | 8/8 (100%):  1 x T1N1M0  1 x T2N0M0  4 x T2N1M0  2 x T3N1M0 | 7/8 (87.5%) | |
| ([Lips et al. 1994](#_ENREF_123))  The Netherlands | IV interventional evidence  Moderate quality (3/6) | N=8 asymptomatic RET M+ members of 4 large MEN2A families, who had a thyroidectomy | MEN2 diagnosed by linkage analysis until June 1993  RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy | 0/8 (0%) | Not stated | 8/8 (100%) | 8/8 (100%)  (scattered, generally small, irregular foci of MTC) | Not stated | |
| ([Gagel et al. 1995](#_ENREF_73))  USA | IV Interventional evidence  Moderate quality (3/6) | N=4 RET M+ patients (children aged 3–12 years) who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16  Thyroidectomy based on genetic screening | 0/4 (0%) | Not stated | 3/4 (75%) | 1/4 (25%)  Unilateral microscopic MTC | Not stated | |
| ([Pacini et al. 1995](#_ENREF_150))  Italy | IV interventional evidence  Moderate quality (3/6) | N=4 clinically unaffected RET M+ family members from 7 MEN2A and 2 MEN2B families, who had a thyroidectomy | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 | 0/4 (0%) | Not stated | 0/4 (0%) | 4/4 (100%) | 1/4 (25%) | |
| ([Vaclavikova et al. 2009](#_ENREF_202))  Czech Republic | IV interventional evidence  Poor quality (2/6) | N=12 family members with a RET Y791F mutation, who underwent a total thyroidectomy  1 MEN2B family  1 MEN2A family  1 FMTC family  1 apparently sporadic MTC family | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | 1/12 (8.3%) | Not stated | 9/12 (75.0%) | 2/12 (16.7%)  T1N1M0 | 2/12 (16.7%) | |
| ([Hernandez et al. 1997](#_ENREF_88))  Spain | IV interventional evidence  Poor quality (2/6) | N=6 RET M+ asymptomatic members of 3 MEN2A families, who had a thyroidectomy  All had raised preoperative pentagastrin-stimulated calcitonin levels | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11  Clinical screening  Total thyroidectomy | 0/6 (0%) | Not stated | 3/6 (50%) | 3/6 (50%) | 0/6 (0%) | |
| ([Kinlaw et al. 2005](#_ENREF_105))  USA | IV interventional evidence  Poor quality (2/6) | N=6 asymptomatic RET M+ members of 3 MEN2A families, who had a thyroidectomy  All 6 had raised pentagastrin-stimulated calcitonin levels and a thyroidectomy | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11  Clinical screening  Total thyroidectomy | 0/6 (0%) | Not stated | 4/6 (66.7%) | 2/6 (33.3%) | Not stated | |
| ([Uchino et al. 1999](#_ENREF_201))  Japan | IV interventional evidence  Low quality (2/6) | N=6 clinically unaffected RET M+ members from 5 MEN2A families with mutations on codon 634 | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11, 13, 14 and 16 | 1/6 (16.7%)  Slightly increased tetragastrin-stimulated calcitonin levels | Not stated | Not stated | 5/6 (83.3%)  Diagnosis:  3 by pathology  3 by ultrasound | 0/3 (0%) of those examined | |

CCH = C-cell hyperplasia; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

# Appendix Uncontrolled studies reporting incidence of phaeochromocytoma and hyperparathyroidism

Table Penetrance of phaeochromocytoma and hyperparathyroidism in RET-mutation-positive index cases

| Study and location | Level of evidence | Study population | Intervention | Incidence | |
| --- | --- | --- | --- | --- | --- |
| Phaeochromocytoma | Hyperparathyroidism |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV interventional evidence  High quality (5/6) | N=23 unrelated index cases with MTC clinically and biochemically characterised as:  82 sporadic MTCs  10 FMTC  10 MEN2A  4 MEN2B  23 were RET M+  Details provided on 22 cases | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 12/22 (54.5%)  confirmed or suspected | Not stated |
| ([Frank-Raue et al. 2011](#_ENREF_70))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=340 patients from 14 different countries who were proven carriers of germline mutation in exon 10 of the *RET* gene | RET mutation testing (method not stated)  Clinical screening | 54/319 (17%)  Identified through:  54% symptomatic  46% screening | 8/299 (2.7%)  Identified through:  12.5% symptomatic  87.5% screening |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV interventional evidence  Moderate quality (4/6) | N=17 index cases with MEN2A (5 men, 12 women, aged 19–60 years)  N=5 index cases with MEN2B (3 men, 2 women, aged 5–22 years) | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, confirmed by restriction site polymorphism analysis | MEN2A: 13/17 (76.4%)  (bilateral in 7 and malignant in 2)  4/17 had parathyroid adenoma  MEN2B: 2/5 (bilateral) | Not stated |
| ([Boer et al. 2003](#_ENREF_19))  Hungary | IV Interventional evidence  Moderate quality (4/6) | N=14 consecutive unrelated patients with MTC admitted for genetic screening for MEN2A and FMTC, who were RET M+ and underwent a thyroidectomy | RET mutation testing by direct DNA sequencing (exons not specified)  Thyroidectomy | Not stated | 1/14 (7.1%) |
| ([Etit et al. 2008](#_ENREF_61))  USA | IV Interventional evidence  Moderate quality (4/6) | N=13 patients retrospectively identified from hospital records who had undergone a prophylactic thyroidectomy for possible MTC | RET mutation testing by direct DNA sequencing analysis of exons 10, 11 and 13–16 | Not stated | 5/13 (38.5%) hypercellular parathyroids were encountered on exploration |
| ([Gimm et al. 2002](#_ENREF_80)).  Germany, Austria | IV interventional evidence  Moderate quality (4/6) | N = 13 index patients with a RET codon 790/791 mutation who underwent thyroid operations | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11, 13 and 14  Thyroidectomy | 2/13 (15.4%) | 0/13 (0%) |
| ([Abdelhakim et al. 2009](#_ENREF_1)) Morocco | IV Interventional evidence  Moderate Quality (4/6) | N=9 patients with confirmed MTC  3 were RET M+:  2 MEN2A  1 unclassified  0/6 suspected sporadic MTC cases had a RET mutation | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16  Total thyroidectomy | 2/9 (22.2%) | 0/9 (0%) |
| ([Paszko et al. 2007](#_ENREF_155))  Poland | IV interventional evidence  Moderate quality (3/6) | N=46 patients with MTC who were RET M+ | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 19/46 (41.3%) | 3/46 (6.5%) with parathyroid pathologies (adenoma and hyperplasia) |
| ([Erdogan et al. 2007](#_ENREF_57))  Turkey | IV Interventional evidence  Moderate quality (3/6) | N=41 RET M+ patients identified from 15 pedigrees:  12 MEN2A  2 MEN2B  1 FMTC  26 were asymptomatic | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–15, and restriction enzyme analysis of exon 16  Total thyroidectomy | 19/41 (46.3%) | 1/41 (2.4%) |
| ([Patocs et al. 2006](#_ENREF_157))  Hungary | IV interventional evidence  Moderate quality (3/6) | N=40 patients from 18 families who had a thyroidectomy due to hereditary MTC or CCH:  33 MEN2A  1 MEN2B  6 from MTC families without PCC or HPT | RET mutation testing by single-strand conformation polymorphism analysis, restriction site polymorphism analysis, and direct DNA sequencing of exons 10, 11, 13, 14 and, in MEN2B phenotype, exons 15 and 16 | Total: 16/40 (40%)  Exon 10: 2/5  Exon 11: 14/26  Exon 14: 0/8  Exon 16: 0/1 | Total: 3/40 (7.5%)  Exon 10: 0/5  Exon 11: 2/26  Exon 14: 1/8  Exon 16: 0/1 |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV interventional evidence  Moderate quality (3/6) | N=13 RET mutation + patients out of 69 with ‘sporadic’ MTC | RET mutation teting by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction enzyme analysis to confirm mutations in FMTC, MEN2A and MEN2B families | 5/13 (38.5%) | 2/13 (15.4%) |
| ([Machens et al. 2006](#_ENREF_128))  Germany | IV interventional evidence  Poor quality (2/6) | N=219 patients with RET mutations divided into 3 categories:  RET codon 918 mutations (highest risk)  RET codons 609–634 (high risk)  RET codons 768–891 (least high risk)  (Machens et al. 2005) previously described 206 patients:  74 index cases  132 non-index cases | RET mutation testing (method not stated) | Highest risk (918):  Penetrance at age:  30 years: 43%  35 years: 100%  50 years: 100%  70 years: 100%  High risk (609–634):  Penetrance at age:  30 years: 8%  35 years: 18%  50 years: 54%  70 years: 73%  Least high risk (768–891):  Penetrance at age:  30 years: 0%  35 years: 0%  50 years: 4%  70 years: 9% | Not stated |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV interventional evidence  Poor quality (2/6) | N=24 clinically affected RET M+ patients who had a thyroidectomy for MTC prior to genetic testing | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and 16 | 10/24 (41.7%) | 1/24 (4.2%) |
| ([Hernandez et al. 1997](#_ENREF_90))  Spain | IV interventional evidence  Poor quality (2/6) | N=17 symptomatic RET M+ members of 3 MEN2A families | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11  Clinical screening  Total thyroidectomy | 6/17 (35.3%) | 0/17 (0%) |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Poor quality (2/6) | N=10 index cases with RET Y791F mutation  1 MEN2B case  1 MEN2A case  1 FMTC case  3 apparently sporadic MTCs  1 with PCC  3 HSCR cases  6 underwent total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | 2/10 (20%):  1 case both sides (aged 31 years)  1 case right (aged 38 years, malignant) | 0/10 (0%) |

CCH = C-cell hyperplasia; HPT = hyperparathyroidism; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

Table Penetrance of phaeochromocytoma and hyperparathyroidism in RET-mutation-positive family members

| Study and location | Level of evidence | Study population | Intervention | Incidence | |
| --- | --- | --- | --- | --- | --- |
| Phaeochromocytoma | Hyperparathyroidism |
| ([Nguyen et al. 2001](#_ENREF_143))  France | IV interventional evidence  High quality (5/6) | N=87 first-degree relatives of index cases in MEN2 families, who were diagnosed with MTC and found to be RET M+:  84 patients from 52 MEN2A families  3 patients from 3 MEN2B families | MEN2 diagnosed by linkage analysis between 1989 and 1994  RET mutation testing by sequence analysis since 1994 (method not stated) | 14/87 (16.1%)  12/84 MEN2A  2/3 MEN2B | 4/87 (4.6%) |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | N=30 RET M+ family members of patients with MTC reclassified from spontaneous MTC to FMTC or MEN2A due to RET mutation, who showed clinical and/or biochemical signs of disease on screening and had a thyroidectomy:  29 phenotype FMTC  1 phenotype MEN2A | RET mutation testing method changed over 15 years  Initially used direct DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8  Total thyroidectomy | 0/30 | 0/30 |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV interventional evidence  High quality (5/6) | N=14 RET M+ relatives of index cases with MTC, who had high calcitonin levels and a thyroidectomy  Index cases:  1 x sporadic MTC  10 x MEN2A  1 x FMTC  2 x MEN2B | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Thyroidectomy  Surgical decisions often made prior to RET mutation testing with no separation of data based on clinical or genetic diagnosis | 7/14 (50%) suspected | Not stated |
| ([Kameyama, Okinaga & Takami 2004](#_ENREF_100))  Japan | IV interventional evidence  Moderate quality (4/6) | N=108 patients with histologically confirmed familial MTC:  83 MEN2A  14 FMTC  11 MEN2B  53 were symptomatic when diagnosed | RET mutation testing (method not stated) | MEN2A: 45%  FMTC: 0%  MEN2B: 0% | MEN2A: 11%  FMTC: 0%  MEN2B: 0% |
| ([Schuffenecker et al. 1994](#_ENREF_181))  France | IV interventional evidence  Moderate quality (4/6) | N=259 affected members from 53 families (including index cases) with RET codon 634 mutations  N=60 affected members from 13 families (including index cases) with RET codon 618 or 620 mutations | RET mutation testing by direct DNA sequencing of exons 10 and 11 | Codon 634: 151/259 (58%, 95% CI 52, 64)  C634R: 70/110 (64%)  C634Y: 53/95 (56%)  Other: 28/54 (52%)  Codon 618 or 620: 5/60 (8%, 95% CI 3, 18) | See below ([Schuffenecker et al. 1998](#_ENREF_182)) |
| ([Schuffenecker et al. 1998](#_ENREF_182))  France | IV interventional evidence  Moderate quality (4/6) | N=188 patients from 30 families with RET codon 634 mutation  10 C634R mutations  11 C634Y mutations  9 other 634 mutations | RET mutation testing by direct DNA sequencing of exons 10 and 11 for index cases, and by restriction site polymorphism analysis for relatives | Not stated | Total: 36/188 (19.1%):  C634R: 15/65 (23.1%)  C634Y: 14/80 (17.5%)  C634F: 6/17 (35.3%)  C634S: 0/11 (0%)  C634G: 1/12 (8/3%)  C634W: 0/3 (0%)  Penetrance at age:  30 years: 14%  40 years: 26%  60 years: 48%  70 years: 81% |
| ([Milos et al. 2008](#_ENREF_136))  Worldwide (Romania, Germany, Chile, Brazil, Argentina, Hungary, Spain, The Netherlands, Czech Republic, Poland, USA) | IV interventional evidence  Moderate quality (4/6) | N=92 carriers of RET C634W mutation from 20 unrelated MEN2A families | RET mutation testing (method not stated) | 41/92 (44.5%)  Age-related penetrance:  20% by age 30 years  67% by age 50 years | 6/64 (9.4%)  Age-related penetrance:  3% by age 30 years  21% by age 50 years |
| ([Dralle et al. 1998](#_ENREF_49))  Germany | IV Interventional evidence  Moderate Quality (4/6) | N=75 RET M+ patients <20 years of age who have undergone a prophylactic total thyroidectomy | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy  Retrospectively identified through questionnaire | 2/75 (2.6%) | 0/75 (0%) |
| ([Algun et al. 2002](#_ENREF_6))  Turkey | IV interventional evidence  Moderate quality (4/6) | N=18 members from a family with MEN2A who were RET M+  N=12 RET M+ family members who underwent total thyroidectomy | RET mutation testing by restriction site polymorphism analysis of exon 11  Clinical screening  Total thyroidectomy with central lymph node dissection | 3/18 (16.7%)  3/12 (25%) | 1/18 (5.6%)  1/12 (8.3%) |
| ([Calva et al. 2009](#_ENREF_27))  USA | IV Interventional evidence  Moderate quality (4/6) | N=16 RET M+ family members who underwent a thyroidectomy  RET C609Y mutation | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy | 0/16 (0%) | 1/16 (6.3%) |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia, New Zealand | IV interventional evidence  Moderate quality (4/6) | N=57 members of 2 families:  Family 1: 22 RET M+ family members from 4 generations with a RET V804L mutation  Family 2: 5 RET M+ family members from 3 generations with a RET V804M mutation | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16  From 1998, analysis also of exons 13–15 for probands  Family members screened for family RET mutation | Family 1: 1/22 (4.5%)  Family 2: 0/5 | Family 1: 6/22 (27.3%)  Family 2: 0/5 |
| ([Gosnell et al. 2006](#_ENREF_82))  Australia | IV Interventional evidence  Moderate quality (4/6) | N=22 RET M+ members from a MEN2A family with RET codon 804 mutation, who underwent total thyroidectomy | RET mutation testing (method not stated)  Clinical screening  Prophylactic thyroidectomy | 1/22 (4.5%)  (index patient) | 3/22 (13.6%) with parathyroid involvement  1 with parathyroid hyperplasia  1 with parathyroid adenoma  1 with parathyroid cyst |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV interventional evidence  Moderate quality (4/6) | N=16 RET M+ patients from 8 families with hereditary MTC, who had a thyroidectomy:  11 x MEN2A  2 x MEN2B  2 x FMTC  1 x other | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14  Clinical screening  Thyroidectomy | 5/16 (31.3%) | 1/16 (6.2%) |
| ([Vestergaard et al. 2007](#_ENREF_206))  Denmark | IV interventional evidence  Moderate quality (4/6) | N=12 first-degree family members who had a RET Y791F mutation | RET mutation testing by direct DNA sequencing of exon 13 | 0/12 | 0/12 |
| ([Marsh et al. 1996](#_ENREF_133))  Australia and New Zealand | IV interventional evidence  Moderate quality (4/6) | N=5 asymptomatic RET M+ members from 2 MEN2A families | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 | 2/5 (40%) | Not stated |
| ([Lips et al. 1994](#_ENREF_125))  The Netherlands | IV interventional evidence  Moderate quality (3/6) | N=80 MEN2A gene carriers (61 diagnosed by DNA sequence analysis)  14 were symptomatic | MEN2 diagnosed by linkage analysis until June 1993  RET mutation testing by direct DNA sequencing of exons 10 and 11 | 39/80 (48.8%) | 3/80 (3.8%) |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV interventional evidence  Moderate quality (3/6) | N=69 RET M+ index cases and family members from 52 MEN2A and 3 MEN2B families with RET codon 634 mutations | RET mutation testing by single-strand conformational polymorphism analysis, restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15 | 18/69 (26.1%) | 9/69 (13.0%) |
| ([Lecube et al. 2002](#_ENREF_119))  Spain | IV interventional evidence  Moderate quality (3/6) | N=25 family members of a FMTC family who had the RET V804M mutation | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–16  Biochemical screening on those who were RET M+ | 0/25 | 0/25 |
| ([Rodriguez Gonzalez et al. 2002](#_ENREF_170))  Spain | IV interventional evidence  Moderate quality (3/6) | N=22 RET M+ patients without clinical signs of disease who received prophylactic thyroidectomy  All had mutations in RET codon 634 | RET mutation testing by denaturing gradient gel electrophoresis, confirmed by restriction analysis  Prophylactic total thyroidectomy +/– central neck dissection | 2/22 (9.1%) had bilateral PCC detected prior to total thyroidectomy | 1/22 (4.5%) had parathyroid hyperplasia diagnosed preoperatively |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members who had a total thyroidectomy | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16 or restriction site polymorphism analysis of exon affected in index case  Total thyroidectomy with central neck dissection | 7/16 (43.8%) | 3/16 (18.8%) |
| ([Lindskog et al. 2004](#_ENREF_124))  Sweden | IV interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members of a MEN2A family with a RET codon 618 mutation, who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central neck dissection | 1/16 (6.3%) | 4/16 (25%) |
| ([Shifrin et al. 2009](#_ENREF_185))  USA | IV interventional evidence  Moderate quality (3/6) | N=15 members from a family with a RET V804M mutation, who had a total thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (not clear if relatives tested only for specific mutation)  Total thyroidectomy with central and ipsilateral lateral neck dissection | 0/15 (0%) (95% CI 0, 22) | 2/15 (13.3%) (95% CI 2, 40) |
| ([Wu et al. 1998](#_ENREF_214))  Taiwan | IV interventional evidence  Moderate quality (3/6) | N=13 RET M+ first- and second-degree relatives from 2 unrelated MEN2A families | RET mutation testing by direct DNA sequencing of exons 10 and 11 | 6/13 (46.2%) | 3/13 (23.1%) |
| ([Yoshida et al. 2009](#_ENREF_215))  Japan | IV interventional evidence  Moderate quality (3/6) | N=12 adults who underwent total thyroidectomy for MTC and had MEN2  5 were symptomatic  All had raised pentagastrin-stimulated calcitonin levels | RET mutation testing (method not stated)  Total thyroidectomy; the parathyroid gland was also removed and autotransplanted (unclear whether treatment decisions influenced by RET mutation) | 5/12 (41.7%):  1 had adrenalectomy before thyroidectomy  3 had adrenalectomy after thyroidectomy  1 had right adrenalectomy before thyroidectomy and left adrenalectomy after thyroidectomy | 2/12 (16.7%) |
| ([Pacini et al. 1995](#_ENREF_152))  Italy | IV interventional evidence  Moderate quality (3/6) | N=5 clinically unaffected RET M+ family members from 7 MEN2A and 2 MEN2B families | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 | 1/5 (20%)  One 23 year old patient has MTC with lymph node metastases, PCC and HPT | |
| ([Quayle et al. 2007](#_ENREF_163))  USA | IV interventional evidence  Poor quality (2/6) | N=323 patients from 65 MEN2A families who were RET M+ | RET mutation testing (method not stated) | 102/323 (31.5%):  C609G: 1/1 (100%)  C609Y: 0/23  C618F: 0/7  C618G: 5/21 (23.8%)  C618R: 11/27 (40.7%)  C618S: 7/41 (17.1%)  C618Y: 0/9  C620F: 0/2  C620R: 2/23 (8.7%)  C620S: 0/4  C620Y: 2/16 (12.5%)  C634G: 0/3  C634R: 49/103 (47.5%)  C634S: 4/4 (100%)  C634W: 4/5 (80%)  C634Y: 17/34 (50%) | Not stated |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Poor quality (2/6) | N=21 family members with RET Y791F mutation  1 MEN2B family  1 MEN2A family  1 FMTC family  3 apparently sporadic MTC families  3 HSCR families | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 1/21 confirmed (4.8%)  (left, at 30 years of age)  1/21 suspected (at 17 years of age) | Not stated |
| ([Kinlaw et al. 2005](#_ENREF_107))  USA | IV interventional evidence  Poor quality (2/6) | N=11 RET M+ family members (including index case) of a MEN2A family with a RET C609S mutation, who were evaluated | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index case  Restriction site polymorphism analysis to detect C609S mutation in family members | 3/11 (27.3%) | Not stated |
| ([Uchino et al. 1999](#_ENREF_204))  Japan | IV interventional evidence  Low quality (2/6) | N=6 clinically unaffected RET M+ members from 5 MEN2A families with mutations on codon 634 | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11, 13, 14 and 16 | 1/6 (16.7%) | Not stated |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV interventional evidence  Poor quality (2/6) | N=5 asymptomatic RET M+ children | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and 16 | 0/5 (0%) | 1/5 (20%) |

HPT = hyperparathyroidism; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

# Appendix Uncontrolled studies reporting age at diagnosis

Table Mean age at diagnosis (index cases)

| Study and location | Level of evidence | Study population | Intervention | Mean age at diagnosis |
| --- | --- | --- | --- | --- |
| ([Neumann et al. 2002](#_ENREF_141))  Germany and Poland | IV interventional evidence  High quality (5/6) | N=271 patients with nonsyndromic PCC without family history of disease  13 were RET M+ | RET mutation testing by single-strand conformation polymorphisms and direct DNA sequencing of exons 13-16.  Also checked for mutations in SDHB, SDHD and VHL. | Mean=36.4 years (range 21 – 50) |
| ([Kameyama, Okinaga & Takami 2004](#_ENREF_100))  Japan | IV interventional evidence  Moderate quality (4/6) | N=271 patients with histologically confirmed MTC:  108 hereditary MTC:  83 MEN2A; 14 FMTC; 11 MEN2B  53 were symptomatic  55 were asymptomatic  163 were sporadic MTC | RET mutation testing (method not stated) | MEN2A: mean=35.6 years  MEN2B: mean=30.5 years  FMTC: mean=34.6 years  Sporadic MTC: mean=47.6 years |
| ([Abdelhakim et al. 2009](#_ENREF_1))  Morocco | IV Interventional evidence  Moderate quality (4/6) | N=9 patients clinically diagnosed with MTC | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16  Total thyroidectomy | Mean = 37.8±15.8 years  Median age for:  MEN2A = 20 years  Sporadic MTC = 38 years |
| ([Patocs et al. 2004](#_ENREF_156))  Hungary | IV interventional evidence  Moderate quality (4/6) | N=7 RET M+ patients with PCCs | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 14 | Mean = 49.4±10.9 years (range 33–63 years) |
| ([Ameur et al. 2009](#_ENREF_11))  France | IV Interventional evidence  Moderate quality (3/6) | N=46 tissue samples from patients diagnosed with MTC  21 had a germline RET mutation | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 from normal and diseased tissue samples to determine germline and somatic RET status | *Germline mutations:* Mean = 27.5 years (range 4–73 years)  *Sporadic:*  Mean = 50.8 years (range 21–74 years) |
| ([Patocs et al. 2006](#_ENREF_157))  Hungary | IV interventional evidence  Moderate quality (3/6) | N=40 patients from 18 families who had had a thyroidectomy due to hereditary MTC or CCH:  33 MEN2A  1 MEN2B  6 from MTC families without PCC or HPT | RET mutation testing by single-strand conformation polymorphism analysis, restriction site polymorphism analysis, and direct DNA sequencing of exons 10, 11, 13, 14 and, in MEN2B phenotype, exons 15 and 16 | C609S: 35 years (range 15–48 years)  C609Y: 41 years (range 27–55 years)  C634F: 43 years (range 27–55 years)  C634Y: 27 years (range 16–51 years)  C634S: 41 years (range 33–51 years)  C634R: 28.5 years (range 22–35 years)  C634W: 35 years (range 16–55 years)  V804M: 38 years (range 34–45 years)  V804L: 38 years (range 33–55 years)  M918Y: 18 years |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV interventional evidence  Moderate quality (3/6) | N=17 MEN2 index cases:  11 MEN2A  1 FMTC  4 MEN2B  1 other (fewer than 4 MTC cases in family) | RET mutation testing by single-strand conformational polymorphism analysis, restriction enzyme analysis and direct sequencing of exons 10, 11 and 13–15 | Mean = 30.6±12.6 years (range 11–55 years)  *MEN2A*  Mean = 32.9±8.4 years (range 19–45 years)  *MEN2B*  Mean = 15.0±4.2 years (range 11–21 years) |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV Interventional evidence  Moderate quality (3/6) | N=13 ‘sporadic’ MTC cases found to be RET M+. | RET mutation testing by single strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11, 13-16. | Median age at diagnosis.  31 ± 20.6 |
| ([Chang et al. 2009](#_ENREF_32))  Taiwan | IV Interventional evidence  Moderate quality (3/6) | N=8 probands from 8 unrelated MTC families:  4 MEN2A  2 MEN2B  1 FMTC  1 sporadic MTC (possibly *de novo* MEN2A) | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 | 27.4±10.2 years (range 12–40 years)  *MEN2A*  32.3±4.5 years (range 27–36 years)  *MEN2B*  13.5±2.1 years (range 12–15 years) |
| ([Machens et al. 2001](#_ENREF_130))  Germany | IV interventional evidence  Poor quality (2/6) | N=63 RET M+ patients with MTC  36 were index patients | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11, 13 and 14 | Medians:  Codon 611: 44 years  Codon 618: 29 years  Codon 620: 36 years  Codon 634: 27 years  Codon 768: 60 years  Codon 790: 39 years  Codon 804: 62 years |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV interventional evidence  Poor quality (2/6) | N=22 clinically affected RET M+ patients who had a thyroidectomy for MTC before genetic testing | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and 16 | 22.8±11.1 years (range 12–54 years) |
| ([Neocleous et al. 2011](#_ENREF_140))  Cyprus | IV interventional evidence  Poor quality (2/6) | N=8 probands from 7 FMTC families and 1 MEN2A family | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index cases, and only exon 10 in family members | 27.9±5.8 years (range 19–34 years) |

CCH = C-cell hyperplasia; FMTC = familial medullary thyroid cancer; HPT = hyperparathyroidism; MTC = medullary thyroid cancer; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

Table Mean age at diagnosis (family members)

| Study and location | Level of evidence | Study population | Intervention | Mean age at diagnosis |
| --- | --- | --- | --- | --- |
| (Machens et al. 2005)  Germany | IV interventional evidence  High quality (5/6) | N=206 consecutive RET+ patients who underwent surgery for CCH, MTC or PCC:  74 index cases  132 nonindex cases (criteria for diagnosis and/or surgery not reported)  Stratified by risk category:  18 highest risk (codon 918)  117 high risk (codons 609-634)  71 less high risk (codons 768-891) | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16  Clinical screening  Thyroidectomy and/or adrenalectomy | Time to diagnosis:  Highest risk  14.3 years (95% CI 10.3, 18.4)  High risk  30.1 years (95% CI 26.6, 33.5)  Least high risk  51.6 years (95% CI 46.5, 56.6) |
| ([Nguyen et al. 2001](#_ENREF_143))  France | IV interventional evidence  High quality (5/6) | N=87 first-degree relatives of index cases in MEN2 families who were RET M+:  84 patients from 52 MEN2A families  3 patients from 3 MEN2B families | MEN2 diagnosed by linkage analysis between 1989 and 1994  RET mutation testing by sequence analysis since 1994 (method not stated) | 14.0±7.0 years (range 0.8–29 years) |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | N=30 RET M+ family members of patients with MTC re-classified from sporadic MTC to FMTC or MEN2A due to a RET mutation, who had a thyroidectomy | RET mutation testing, method changed over 15 years  Initially used DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8  Total thyroidectomy | 38.5±17.8 years (range 5–76 years) |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV interventional evidence  High quality (5/6) | N=23 RET M+ index cases with MTC:  6 with apparently sporadic MTC  4 with FMTC  9 with MEN2A  4 with MEN2B | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Thyroidectomy  Surgical decisions often made before RET mutation testing with no separation of data based on clinical or genetic diagnosis  Note: thyroidectomy outcomes data were not available for 1 FMTC index case | Median (range):  Exon 10 = 28 years (21–35 years)  Exon 11 = 28 years (18–4 years 7)  Exon 13 = 44 years (40–48 years)  Exon 14 = 49 years (46–52 years)  Exon 16 = 20.5 years (14–31 years) |
| ([Frank-Raue et al. 2011](#_ENREF_70))  Germany | IV Interventional evidence  Moderate quality (4/6) | N=340 patients proven to be carriers of germline mutation in exon 10 of the *RET* gene  Identified through:  47% symptomatic  53% screening | RET mutation testing (method not stated)  Clinical screening | Median = 35 years (range 4–86 years)  Codon 609 median = 37 years (range 4–86 years)  Codon 611 median = 42 years (range 14–69 years)  Codon 618 median = 35 years (range 5–72 years)  Codon 620 median = 31 years (range 6–76 years) |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV Interventional evidence  Moderate quality (4/6) | N=15 RET M+ members of 8 separate families with hereditary MTC with available data, who had a prophylactic thyroidectomy | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14  Clinical screening  Total thyroidectomy | 24.7±11.4 years (range 11–44 years) |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV interventional evidence  Moderate quality (3/6) | N=72 RET M+ index cases and family members from 17 MEN2 families:  49 diagnosed on clinical evidence  23 diagnosed through molecular screening  9/23 had elevated serum basal calcitonin | RET mutation testing by single-strand conformational polymorphism analysis, restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15 | With clinical disease (n=49): 29.8±11.6 years  Without clinical disease (n=23): 21.7±21.6 years  (difference p<0.04) |
| ([Shifrin et al. 2009](#_ENREF_185))  USA | IV interventional evidence  Moderate quality (3/6) | N=40 RET M+ family members from a family with a RET V804M mutation  The majority of family members were diagnosed in the same year | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (not clear if relatives tested for specific mutation or all) | Generation I: 71–83 years (median 75 years)  Generation II: 41–64 years (median 45 years)  Generation III: not stated |
| ([Erdogan et al. 2007](#_ENREF_57))  Turkey | IV Interventional evidence  Moderate quality (3/6) | N=38 RET M+ patients identified from 12 MEN2A pedigrees  26 were asymptomatic | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–15, and restriction enzyme analysis of exon 16  Total thyroidectomy | Median = 33 years (range 2 months – 58 years) |
| ([Lindskog et al. 2004](#_ENREF_124))  Sweden | IV interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members of a MEN2A family with a RET codon 618 mutation, who had a thyroidectomy:  15 identified by biochemical screening  1 identified by genetic testing | RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central neck dissection | Mean = 42 years |
| ([Pacini et al. 1995](#_ENREF_152))  Italy | IV interventional evidence  Moderate quality (3/6) | N=5 clinically unaffected RET M+ family members from 7 MEN2A and 2 MEN2B families | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 | Mean = 18.4±6.1 years (range 10–25 years) |

CCH = C-cell hyperplasia; FMTC = familial medullary thyroid cancer; MTC = medullary thyroid cancer; RET M+ = RET-mutation-positive

# Appendix Uncontrolled studies reporting age at thyroidectomy in family members

Table Mean age at total thyroidectomy (family members)

| Study and location | Level of evidence | Study population | Intervention | Mean age at thyroidectomy |
| --- | --- | --- | --- | --- |
| ([Skinner et al. 1996](#_ENREF_190))  USA | IV interventional evidence  High quality (5/6) | N=50 RET M+ patients from MEN2A families, who were <20 years of age at time of thyroidectomy | RET mutation testing by restriction site polymorphism analysis and/or direct DNA sequencing of RET exons 10, 11, 13, 14 and 16  Total thyroidectomy | Mean = 8.6 years (range 3–19 years)  Median = 7 years |
| ([Lau et al. 2009](#_ENREF_114))  Hong Kong | IV interventional evidence  High quality (5/6) | N=22 asymptomatic patients from 8 MEN2A families, who underwent prophylactic total thyroidectomy  All had RET codon 634 mutations | RET mutation testing by restriction enzyme analysis and/or direct DNA sequencing (exons not specified)  Prophylactic thyroidectomy with or without a unilateral central compartment neck dissection | Mean = 25.2±17.5 years (range 6.1–71.9 years) |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV interventional evidence  High quality (5/6) | N=14 RET M+ relatives of MTC cases who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Thyroidectomy  Surgical decisions often made prior to RET mutation testing with no separation of data based on clinical or genetic diagnosis  Note: thyroidectomy outcomes data were not available for 1 FMTC index case | Mean = 18.8±13.8 years (range 5–51 years) |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV Interventional evidence  Moderate quality (4/6) | N=35 RET M+ members of a large extended FMTC family with a RET G533C mutation, who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exon 8  Prophylactic thyroidectomy | Mean = 42 years (range 5–73 years) |
| ([Frohnauer et al. 2000](#_ENREF_73))  USA | IV Interventional evidence  Moderate quality (4/6) | N=13 members (degree not stated) from 5 MEN2A kindreds, who had a RET codon 804 mutation and a thyroidectomy | RET mutation testing by denaturing gradient gel electrophoresis analysis confirmed by direct DNA sequencing of exon 14  Thyroidectomy | Mean = 35.3±15.8 years (range 6–56 years) |
| ([Decker et al. 1996](#_ENREF_39))  USA | IV interventional evidence  Moderate quality (4/6) | N=11 RET M+ children from 4 confirmed MEN2A families, who had a prophylactic thyroidectomy | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10 and 11  Clinical screening  Prophylactic thyroidectomy | Mean = 7.5 years (range 2–12 years) |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia, New Zealand | IV interventional evidence  Moderate quality (4/6) | N=25 RET M+ members of 2 families who had a thyroidectomy:  Family 1: 22 family members with a RET V804L mutation  Family 2: 3 family members with a RET V804M mutation | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16  From 1998, analysis also of exons 13–15 for probands  Family members screened for family RET mutation  Prophylactic thyroidectomy | Family 1:  Mean = 37.1±17.9 years  (range = 9-63 years)  Family 2:  Mean = 34.6±30.5 years  (range = 5-68 years) |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV Interventional evidence  Moderate quality (4/6) | N=17 RET M+ members of MEN2A or FMTC kindreds who had a thyroidectomy based on RET mutational status:  13 MEN2A  4 FMTC | RET mutation testing by denaturing gradient gel analysis of exons 10 and 11, with confirmatory direct DNA sequencing | Mean = 25.1±17.4 years (range = 5-64 years)  *MEN2A* Mean = 28.7±18.4 years (range = 5-64 years)  *MEN2B* Mean = 15.8±8.5 years (range = 10-28 years) |
| ([Heizmann et al. 2006](#_ENREF_89))  Switzerland | IV interventional evidence  Moderate quality (4/6) | N=14 RET M+ patients who were presymptomatic, from 2 MEN2A kindreds | RET mutation testing by single-strand conformation polymorphism analysis, denaturing gradient gel electrophoresis and direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central compartment dissection in those older than 6 years of age | Mean = 25±19 years  (range = 4-63 years) |
| ([Algun et al. 2002](#_ENREF_6))  Turkey | IV Interventional evidence  Moderate quality (4/6) | N=12 RET M+ members of a MEN2A family, who had a prophylactic thyroidectomy | RET mutation testing by restriction site polymorphism analysis of exon 11  Clinical screening  Total thyroidectomy with central lymph node dissection | Mean = 24.2±16.1 years  (range = 1-60 years) |
| ([Rodriguez Gonzalez et al. 2002](#_ENREF_170))  Spain | IV interventional evidence  Moderate quality (3/6) | N=22 RET M+ patients who had normal basal and pentagastrin-stimulated calcitonin levels and received a prophylactic thyroidectomy  All had mutations in RET codon 634 | RET mutation testing by denaturing gradient gel electrophoresis of exons 10, 11, 13, 14 and 16, confirmed by restriction site polymorphism analysis  Clinical screening  Prophylactic total thyroidectomy ± central neck dissection | Mean = 15.2±8.7 years  (range = 5-36 years) |
| ([Lombardo et al. 2002](#_ENREF_126))  France and Italy | IV interventional evidence  Moderate quality (3/6) | N=71 patients with RET V804L mutations from 2 families, who underwent thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15, and restriction site polymorphism analysis of exon 16  Clinical screening  Total thyroidectomy | Mean = 49.9±16.0 years (range 12–75 years) |
| ([Jung et al. 2010](#_ENREF_98))  Korea | IV interventional evidence  Moderate quality (3/6) | N=8 RET M+ members of a 3-generation FMTC family, who underwent a total thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 of index case  Analysis of exon 10 in family members  Total thyroidectomy with either central neck dissection or modified radical neck dissection | Mean = 36.8±17.5 years |
| ([Calva et al. 2009](#_ENREF_27))  USA | IV Interventional evidence  Moderate quality (3/6) | N=16 RET M+ family members who had a thyroidectomy | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy for treatment or prophylaxis | Mean = 39.7 years (range 5–59 years) |
| ([Lips et al. 1994](#_ENREF_125))  The Netherlands | IV interventional evidence  Moderate quality (3/6) | N=8 RET M+ juvenile members from 2 large MEN2A families, who had total thyroidectomy on basis of RET mutation status | MEN2 diagnosed by linkage analysis until June 1993  RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy | Mean = 9.4±4.3 years (range 4–18 years) |
| ([Pacini et al. 1995](#_ENREF_152))  Italy | IV interventional evidence  Moderate quality (3/6) | N=5 clinically unaffected RET M+ family members from 7 MEN2A and 2 MEN2B families | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 | Mean = 18.4±6.1 years (range 10–25 years) |
| ([Gagel et al. 1995](#_ENREF_75))  USA | IV Interventional evidence  Moderate quality (3/6) | N=4 RET M+ patients (children aged 3–12 years) who had a thyroidectomy | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16  Thyroidectomy based on genetic screening | Mean = 9.1±3.8 years (range 3.5–12 years) |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Poor quality (2/6) | N=12 family members from 4 families with a RET Y791F mutation, who underwent total thyroidectomy:  1 MEN2B family  1 MEN2A family  1 FMTC family  1 apparently sporadic MTC family | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | Mean = 20.7±14.1 years (range 5–43 years) |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV interventional evidence  Poor quality (2/6) | N=4 members of an MTC family (including index patient), who had RET L790F mutation and thyroidectomy | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–15, and restriction site polymorphism analysis for exon 16  Direct DNA sequencing of exon 13 in family members  Clinical screening  Prophylactic thyroidectomy | Mean = 52.8±12.0 years (range 45–74 years) |

RET M+ = RET-mutation-positive

# Appendix Studies reporting diagnostic yield

Table 81 Diagnostic yield in patients with a hereditary MTC

| Study and location | Level of evidence | Study population | Intervention | Diagnostic yield |
| --- | --- | --- | --- | --- |
| ([Gagel et al. 1995](#_ENREF_75))  USA | IV diagnostic evidence | N=71 members from 28 families with clinically confirmed MEN2A | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16 | 71/71 (100%) MEN2A RET M+ |
| ([Shirahama et al. 1998](#_ENREF_187))  Japan | IV diagnostic evidence | N=44 patients with MTC:  34 MEN2A  4 MEN2B  6 FMTC | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11; if no mutations found, then restriction site polymorphism analysis of codons 768 and 918 | 42/44 (95.5%) RET M+:  33/34 (97.1%) MEN2A  4/4 (100%) MEN2B  5/6 (83.3%) FMTC |
| ([Elisei et al. 2007](#_ENREF_53))  Italy | IV diagnostic evidence | N=37 patients with inherited who underwent genetic screening for RET mutations during 1993–2006 | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 36/37 (97.3%) RET M+:  31/32 (96.9%) FMTC  5/5 (100%) MEN2B |
| ([Etit et al. 2008](#_ENREF_61))  USA | IV diagnostic evidence | N=32 patients retrospectively identified from hospital records who had undergone a prophylactic thyroidectomy for MTC:  24 MEN2A  8 non-MEN  30 with family history | RET mutation testing by direct DNA sequencing analysis of exons 10, 11 and 13–16 | 29/32 (90.6%) RET M+  27/30 (90%) with family history  24/24 (100%) MEN2A  5/8 (62.5%) non-MEN |
| ([Fernandez et al. 2006](#_ENREF_63))  Spain | IV diagnostic evidence | N=27 patients clinically diagnosed with familial MTC:  16 MEN2A  3 MEN2B  8 FMTC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 23/27 (85.2%) RET M+:  16/16 (100%) MEN2A  3/3 (100%) MEN2B  4/8 (50%) FMTC |
| ([Fink et al. 1996](#_ENREF_65))  Austria | IV diagnostic evidence | N=27 patients clinically diagnosed with MTC from 16 families with FMTC, MEN2A, MEN2B, or suspected of inheritable MTC | RET mutation testing by single-strand conformational analysis or restriction site polymorphism analysis, and confirmatory direct DNA sequencing of exons 10, 11, 13 and 16; if no mutation detected, direct DNA sequencing of exon 14 was conducted | 20/27 (74.1%) RET M+ |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV diagnostic evidence | N=24 unrelated index cases with MTC:  10 MEN2A  4 MEN2  10 FMTC | RET mutation testing by single-strand conformation polymorphism analysis, confirmed by direct DNA sequencing of exons 10, 11 and 13–16 | 17/24 (70.8%) RET M+:  9/10 (90%) MEN2A  4/4 (100%) MEN2B  4/10 (40%) FMTC |
| ([Komminoth et al. 1995](#_ENREF_110))  Switzerland | IV diagnostic evidence | N=22 specimens from patients with MTC suspected of having MEN2 or FMTC | RET mutation testing by single-strand conformation polymorphism and heteroduplex gel electrophoresis analysis of exons 10, 11, 13 and/or 16 | 22/22 (100%) had germline RET mutation  3/22 (13.6%) had exon 10 mutations at codon 618  15/22 (68.2%) had exon 11 mutations at codon 634  3/22 (13.6%) had exon 16 mutations at codon 918 |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV diagnostic evidence | N=17 index cases with MTC from MEN2 families:  11 MEN2A  4 MEN2B  2 FMTC | RET mutation testing by single-strand conformation polymorphism analysis, restriction enzyme analysis and direct DNA sequencing of exons 10, 11, 13, 14 or 15 | 17/17 (100%) RET M+ |
| ([Neumann et al. 1995](#_ENREF_142))  Germany | IV diagnostic evidence | N=10 families with MEN2:  9 MEN2A  1 MEN2B | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 (also checked for mutations in SDHB, SDHD and VHL) | 8/10 (80%) families were RET M+:  7/9 (77.8%) MEN2A  1/1 (100%) MEN2B  2/10 (20%) had VHL mutations, so were reclassified as VHL families |
| ([Chang et al. 2009](#_ENREF_32))  Taiwan | IV diagnostic evidence | N=8 probands from 8 unrelated MTC families:  4 MEN2A  1 suspected MEN2A  2 MEN2B  1 FMTC | RET mutation testing by direct DNA sequencing of exons 1–20 | 8/8 (100%) RET M+ |
| ([Blaugrund et al. 1994](#_ENREF_18))  USA | IV diagnostic evidence | N=7 patients with MTC:  3 MEN2A  1 MEN2B  3 FMTC | RET mutation testing by DNA sequencing of cloned exons 10, 11 and 16, and Southern blot analysis for genomic rearrangements | 7/7 (100%) RET M+ |
| ([Kimura et al. 1995](#_ENREF_105))  Japan | IV diagnostic evidence | N=7 specimens from patients:  1 FMTC  2 MEN2A  4 MEN2B | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11 | 3/7 (42.9%) had germline RET mutations in exons 10 or 11  (test cannot detect MEN2B mutations) |
| ([Gonzalez et al. 2003](#_ENREF_81))  Mexico | IV diagnostic evidence | N=6 probands  3 MEN2B  2 MEN2A  4 sporadic MTC | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16, and direct DNA sequencing of exons 13–15 | 6/9 (66.7%) RET M+ |
| ([Kitamura et al. 1997](#_ENREF_108))  Japan | IV diagnostic evidence | N=6 unrelated patients with inherited MTC | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11, 13, 14 and 16, followed by direct DNA sequencing of exons 10, 11, 13 and 14, and restriction site polymorphism analysis to detect codon 918 mutation | 6/6 (100%) RET M+ |
| ([Hedayati et al. 2006](#_ENREF_88))  Iran | IV diagnostic evidence | N=4 unrelated index cases with MTC:  1 MEN2A,  1 MEN2B  2 FMTC | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 | 1/4 (25%) RET M+:  1/1 MEN2A  0/1 MEN2B  0/2 FMTC |
| ([Abdelhakim et al. 2009](#_ENREF_1))  Morocco | IV diagnostic evidence | N=3 index cases:  2 MEN2A  1 hereditary MTC | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 | 3/3 (100%) RET M+ |

RET M+ = RET-mutation-positive

Table 82 Diagnostic yield in patients with an apparently sporadic MTC

| Study and location | Level of evidence | Study population | Intervention | Diagnostic yield |
| --- | --- | --- | --- | --- |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV diagnostic evidence | N=729 patients with apparently sporadic MTC (no familial history of MTC or other endocrine disease) | RET mutation testing method changed over 15 years  Initially used DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8 | 47/729 (6.5%) RET M+:  32/47 in a non-cysteine-encoding codon in exons 5, 11, 13, 14 or 15  15/47 in a cysteine encoding codon in exons 10, or 11  6/47 MEN2A  41/47 FMTC |
| ([Elisei et al. 2007](#_ENREF_53))  Italy | IV diagnostic evidence | N=481 apparently sporadic MTC patient samples submitted for genetic screening for RET mutations during 1993–2006 | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 35/481 (7.3%) RET M+ |
| ([Bugalho et al. 2007](#_ENREF_22))  Portugal | IV diagnostic evidence | N=77 apparently sporadic cases of MTC | RET mutation testing by direct DNA sequencing of exons 10–16, or restriction site polymorphism analysis of exons 13–16  Exon 8 was screened for gross insertions/deletions (method not stated) | 3/77 (3.9%) RET M+ |
| ([Fernandez et al. 2006](#_ENREF_63))  Spain | IV diagnostic evidence | N=73 patients identified in a hospital through clinical presentation and classified as sporadic MTCs | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 2/73 (2.7%) RET M+ |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV diagnostic evidence | N=69 sporadic MTC patients | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction site polymorphism analysis of exon affected in index case | 13/69 (18.8%) RET M+:  6 x codon 634  4 x codon 790  3 x codon 618 |
| ([Eng, Mulligan, et al. 1995](#_ENREF_56))  UK | IV diagnostic evidence | N=67 sporadic MTC patients  No history of first- or second-degree family MTC or PCC  No multiple tumours  MTC confirmed histopathologically | RET mutation testing by direct DNA sequencing of exons 10, 11, 13 and 16 | 1/67 (1.5%) RET M+ |
| ([Fink et al. 1996](#_ENREF_65))  Austria | IV diagnostic evidence | N=59 sporadic MTC patients | RET mutation testing by single-strand conformational analysis or restriction site polymorphism analysis and confirmatory direct DNA sequencing of exons 10, 11, 13 and 16; if no mutation detected, direct DNA sequencing of exon 14 was conducted | 0/59 (0%) RET M+ |
| ([Erdogan et al. 2005](#_ENREF_58))  Turkey | IV diagnostic evidence | N=56 apparently sporadic MTC, clinically & histopathologically confirmed  Family history negative to PCC, HPT or MTC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 6/56 (10.7%) RET M+ |
| ([Hedayati et al. 2006](#_ENREF_88))  Iran | IV diagnostic evidence | N=53 unrelated index cases with apparently sporadic MTC | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 | 3/53 (5.7%) RET M+ |
| ([Prazeres et al. 2006](#_ENREF_160))  Portugal | IV diagnostic evidence | N=53 patients with apparently sporadic MTC | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16, plus restriction site polymorphism analysis when possible | 2/53 (3.8%) RET M+:  1 C611Y mutation  Other mutation not stated |
| ([Alvandi et al. 2011](#_ENREF_8))  Iran | IV diagnostic evidence | N=49 unrelated index patients diagnosed with MTC and classified as apparently sporadic | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16, and restriction site polymorphism analysis to detect C634R mutation | 7/49 (14.2%) RET M+ |
| ([Lendvai et al. 2012](#_ENREF_120))  Hungary | IV diagnostic evidence | N=47 consecutive patients with apparently sporadic MTCs | RET mutation testing by direct DNA sequencing of exons 10, 11 and 14 | 0/47 (0%) RET M+ |
| ([Fitze et al. 2002](#_ENREF_66))  Germany | IV diagnostic evidence | N=45 patients clinically identified with sporadic MTC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 5/45 (11.1%) RET M+ |
| ([Shan et al. 1998](#_ENREF_183))  Japan, China | IV diagnostic evidence | N=40 patients with apparently sporadic MTCs | RET mutation testing by restriction site polymorphism analysis of codon 918 mutations | 0/40 (0%) had germline mutation on RET codon 918 |
| ([Uchino et al. 1998](#_ENREF_203))  Japan | IV diagnostic evidence | N=40 patients of apparently sporadic MTCs who had surgery between 1965 and 1996. | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 or 16 | 6/40 (15.0%) RET M+:  2 mutations on codon 618  3 mutations on codon 634  1 mutation on codon 804 |
| ([Guerrero et al. 2006](#_ENREF_85))  Brazil | IV diagnostic evidence | N=24 unrelated patients with apparently sporadic MTC) | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10 and 11, and direct DNA sequencing of exons 13–16 | 8/24 (33.3%) RET M+  1/24 (4.2%) at exons 10 or 11  7/24 (29.2%) at exons 13 or 15 |
| ([Komminoth et al. 1995](#_ENREF_110))  Switzerland | IV diagnostic evidence | N=24 specimens from patients with apparently sporadic MTC or PCCs | RET mutation testing by single-strand conformation polymorphism and heteroduplex gel electrophoresis analysis of exons 10, 11, 13 and/or 15 | 1/24 (4.2%) had germline RET mutation on exon 11 |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV diagnostic evidence | N=21 patients diagnosed with apparently sporadic MTC | RET mutation testing by denaturing gradient gel electrophoresis mutational analysis of exons 10 and 11, with confirmatory direct DNA sequencing | 5/21 (23.8%) RET M+ |
| ([Blaugrund et al. 1994](#_ENREF_18))  USA | IV diagnostic evidence | N=15 apparently sporadic MTC | RET mutation testing by DNA sequencing of cloned exons 10, 11 and 16, and Southern blot analysis for genomic rearrangements | 7/15 (46.7%) RET M+ |
| ([Bugalho et al. 1997](#_ENREF_23))  Portugal | IV diagnostic evidence | N=13 sporadic MTC  No family history of MTC, PCC or parathyroid disease | RET mutation testing by direct DNA sequencing of exons 10, 11, 13, 15 and 16, confirmed using restriction site polymorphism analysis where appropriate | 0/13 (0%) RET M+ |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV diagnostic evidence | N=10 with sporadic MTC | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14 | 0/10 (0%) RET M+ |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV diagnostic evidence | N=8 sporadic MTC | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing in exons 1–20 | 5/9 (55.6%) RET M+ |
| ([Abdelhakim et al. 2009](#_ENREF_1))  Morocco | IV diagnostic evidence | N=6 sporadic MTC | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 | 0/6 (0%) RET M+ |
| ([Gonzalez et al. 2003](#_ENREF_81))  Mexico | IV diagnostic evidence | N= 4 sporadic MTC | RET mutation testing by single-strand conformational polymorphism analysis, with direct DNA sequencing of exons 10, 11 and 16 and direct DNA sequencing of exons 13–15 | 1/4 (25%) RET M+ |
| ([Kimura et al. 1995](#_ENREF_105))  Japan | IV diagnostic evidence | N=3 sporadic MTC | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11 | 1/3 (33.3%) had germline RET mutations in exons 10 or 11 |

PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

Table 83 Diagnostic yield in patients with an unspecified MTC

| Study and location | Level of evidence | Study population | Intervention | Diagnostic yield |
| --- | --- | --- | --- | --- |
| ([Boer et al. 2003](#_ENREF_19))  Hungary | IV diagnostic evidence | N=65 consecutive patients during 1992–2000 with MTC undergoing screening | RET mutation testing by direct DNA sequencing (exons not specified) | 25/65 (38.5%) RET M+ |
| ([Klein et al. 2001](#_ENREF_109))  Hungary | IV diagnostic evidence | N=65 unrelated people with MTC (index cases) | RET mutation testing by restriction site polymorphism analysis of exon 11 and/or direct DNA sequencing of exons 10, 13 and/or 14 | 14/65 (21.5%) RET M+:  12 x codon 634 mutations  1 x codon 609 mutation  1 x codon 804 mutation |
| ([Sharma & Saranath 2011](#_ENREF_184))  India | IV diagnostic evidence | N=51 MTC patients | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 for index cases, and specific exons for family members | 15/51 (29.4%) RET M+:  Mutation at exon 11 codon 634:  9/15 (60%) RET M+  Mutation at exon 10 codon 609/618: 3/15 RET M+  Mutation at exon 16 codon 918: 2/15 RET M+  Mutation at exon 14 codon 814 : 1/15 RET M+ |
| ([Ameur et al. 2009](#_ENREF_11))  France | IV diagnostic evidence | N=46 tissue samples collected from MTC, CCH, MCC or mixed MTC patients | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 from normal and diseased tissue samples to determine germline and somatic RET status | 21/46 (45.7%) had a germline RET mutation |
| ([Chung et al. 2004](#_ENREF_36))  Korea | IV diagnostic evidence | N=33 MTC patients who underwent a thyroidectomy (diagnosed clinically and by histopathology) | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (postoperative) | 9/33 (27.3%) RET M+ |
| ([Pinna et al. 2007](#_ENREF_159))  Italy | IV diagnostic evidence | N=22 patients with MTC | RET mutation testing by direct DNA sequencing of exons 8–16 in index case, and appropriate exon in family members | 7/22 (31.8%) RET M+ |

CCH = C-cell hyperplasia; RET M+ = RET-mutation-positive

Table Diagnostic yield in patients presenting with a phaeochromocytoma

| Study and location | Level of evidence | Study population | Intervention | Diagnostic yield |
| --- | --- | --- | --- | --- |
| Apparently sporadic | | | | |
| ([Neumann et al. 2002](#_ENREF_141))  Germany and Poland | IV diagnostic evidence | N=271 patients with non-syndromic PCC and/or paragangliomas without family history of disease  22 had paragangliomas only  8 had both a paraganglioma and a PCC | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 (also checked for mutations in SDHB, SDHD and VHL) | 13/271 (4.8%) RET M+:  4 C634R, 1 C634G, 3 C634Y, 1 C634S,  1 C634F, 2 C634T  1 Y791F (exon 13) |
| ([Amar et al. 2005](#_ENREF_10))  France | IV diagnostic evidence | N=258 patients with apparently sporadic PCC or paraganglioma | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  All the coding exons of SDHB, SDHD, SDHC and VHL were also sequenced | 1/258 (0.4%) RET M+ |
| ([Cascon et al. 2009](#_ENREF_29))  Spain | IV diagnostic evidence | N=192 consecutively enrolled patients with functioning or non-functioning PCC or paraganglioma with no personal or familial history | Complete genetic characterisation of RET, SDHB, SDHC, SDHD and VHL (method not stated) | 1/192 (0.5%) RET M+ |
| ([Radien et al. 1997](#_ENREF_164))  France | IV diagnostic evidence | N=120 patients with apparently sporadic PCC | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10, 11, 13 and 16 | 1/120 (0.8%) RET codon 790 mutation |
| ([Krawczyk et al. 2010](#_ENREF_112))  Poland | IV diagnostic evidence | N=60 patients with diagnosis of apparently sporadic PCC or paraganglioma  53 had PCC  8 had paraganglioma  (1 had both)  41 were benign tumours  11 had malignant lesions | RET mutation testing by direct DNA sequencing of exons 10, 11, 14 and 16 (also checked for mutations in SDHB, SDHD and VHL) | 11/60 (18.3%) RET M+:  6 had mutations at codon 634  5 had mutations at codon 791 |
| ([Iacobone et al. 2011](#_ENREF_93))  Italy | IV diagnostic evidence | N=59 patients with apparently sporadic PCC (without evident hereditary disease and/or syndromic appearance) | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 (also checked for mutations in SDHB, SDHC, SDHD and VHL) | 0/59 (0%) RET M+ |
| ([Lindor et al. 1995](#_ENREF_123))  USA | IV diagnostic evidence | N=29 patients who had undergone an operation for a sporadic PCC | RET mutation testing by direct DNA sequencing of exons 10 and 11, and mutation specific PCR for exon 16 | 0/29 (0%) RET M+ |
| ([Bar et al. 1997](#_ENREF_12))  Israel | IV diagnostic evidence | N=27 patients diagnosed with sporadic PCC | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10, 11 and 16 (also checked for mutations in VHL) | 0/27 (0%) RET M+ |
| ([Beldjord et al. 1995](#_ENREF_15))  France | IV diagnostic evidence | N=28 patients diagnosed clinically with sporadic PCC | RET mutation testing by denaturing gradient gel electrophoresis analysis confirmed by direct DNA sequencing of exons 10, 11 and 16 | 0/28 (0%) RET M+ |
| ([Eng, Crossey, et al. 1995](#_ENREF_55))  UK, USA | IV diagnostic evidence | N=48 patients with apparently sporadic PCC | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 9, 10, 11 and 13–16 (also checked for mutations in VHL) | 5/48 (10.4%) RET M+ |
| ([Lendvai et al. 2012](#_ENREF_120))  Hungary | IV diagnostic evidence | N=48 consecutive patients with apparently sporadic PCC  Mean age of 36±14 years in men and 42±14 years in women | RET mutation testing by direct DNA sequencing of exons 10, 11 and 14 | 0/48 (0%) RET M+ |
| ([Kimura et al. 1995](#_ENREF_105))  Japan | IV diagnostic evidence | N=12 sporadic PCCs | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11 | 0/12 (0%) RET M+ |
| ([Fernandez et al. 2006](#_ENREF_63))  Spain | IV diagnostic evidence | N=12 patients identified in a hospital through clinical presentation of PCC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 1/12 (8.3%) RET M+ |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV diagnostic evidence | N=8 sporadic PCC patients | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing in exons 1–20 | 0/8 (0%) RET M+ |
| Hereditary phaeochromoctyoma | | | | |
| ([Eisenhofer et al. 2011](#_ENREF_51))  Germany | IV diagnostic evidence | N=173 patients with hereditary PCC and paraganglioma patients (retrospective analysis) | Genetic characterisation of RET, VHL, SDHB, SDHC, and SDHD (method not stated) | 38/173 (21.9%) RET M+ |
| ([Cascon et al. 2009](#_ENREF_29))  Spain | IV diagnostic evidence | N=69 consecutively enrolled patients with functioning or non-functioning PCC or paraganglioma with a personal or familial history of disease:  35 had history of MEN2  34 had other familial syndromes:  10 had history of VHL | Complete genetic characterisation of RET, VHL, SDHB, SDHC and SDHD (method not stated) | 54/69 (78.3%) RET M+  35/35 (100%) with history of MEN2  19/34 (55.9%) with history of other familial syndromes  0/10 (0%) with history of VHL |
| ([Amar et al. 2005](#_ENREF_10))  France | IV diagnostic evidence | N=56 patients with a family history of PCC or paraganglioma | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  All the coding exons of SDHB, SDHD, SDHC, and VHL were also sequenced | 15/56 (26.8%) RET M+ |
| ([Woodward et al. 1997](#_ENREF_213))  United Kingdom | IV diagnostic evidence | N=16 kindreds with familial PCC | RET mutation testing of exons 10 and 11 (method not stated), (also checked for mutations in GDNF and VHL) | 0/16 (0%) RET M+ |
| Unspecified phaeochromocytoma | | | | |
| ([Erlic et al. 2010](#_ENREF_59))  USA, Spain, Germany, Poland, Finland | IV diagnostic evidence | N=1,475 patients identified on the European-American Phaeochromocytoma-Paraganglioma Registry | Genetic characterisation of RET exons 10, 11 and 13–16 (method not stated), (also checked for mutations in SDHB, SDHC, SDHD and VHL) | 14/1475 (0.9%) RET M+  13/1475 (Tyr791Phe)  1/1475 (Ser649Leu) |
| ([Mannelli et al. 2009](#_ENREF_131))  Italy | IV diagnostic evidence | N=501 consecutively enrolled patients presenting with PCC or paragangliomas (new or previously identified) | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16, and multiplex ligation-dependent probe amplification assay to detect genomic rearrangements (also checked for mutations in SDHB, SDHC, SDHD and VHL) | 27/501 (5.3%) RET M+ |
| ([Januszewicz et al. 2000](#_ENREF_96))  Poland | IV diagnostic evidence | N=77 unselected patients with PCC surgically treated (who responded to invitation, 85 did not respond) | RET mutation testing by single-strand conformation polymorphism analysis confirmed by direct DNA sequencing of exons 10, 11 and 13–16 | 6/77 (7.8%) RET M+  All 6 had mutations of exon 11, codon 634 (TGC to CGC) |
| ([Patocs et al. 2004](#_ENREF_156))  Hungary | IV diagnostic evidence | N=41 patients with PCCs | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 14 (also checked for mutations in VHL) | 7/41 (17.1%) RET M+:  2 x C609S  3 x C634F  1 x C634Y  1 x C634R |
| ([De Krijger et al. 2006](#_ENREF_38))  Nederlands | IV diagnostic evidence | N=10 PCC tissue samples | RET mutation testing by denaturing gradient gel electrophoresis analysis, confirmed by direct DNA sequencing of exons 10, 11 and 16 (also checked for mutations in SDHB, SDHD and VHL) | 4/10 (40%) RET M+ |

PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

Table Diagnostic yield in relatives of someone with a confirmed RET mutation

| Study and location | Level of evidence | Study population | Intervention | Diagnostic yield |
| --- | --- | --- | --- | --- |
| First-degree family members | | | | |
| ([Elisei et al. 2007](#_ENREF_53))  Italy | IV diagnostic evidence | N=274 first-degree relatives of patients with confirmed RET mutations screened during 1993–2006 | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | 91/274 (33.2%) first-degree family members were RET M+ |
| ([McMahon et al. 1994](#_ENREF_135))  UK | IV diagnostic evidence | N=63 affected or unaffected first-degree relatives from 9 MEN2A families with mutations in RET codon 634:  29 affected  30 unaffected  4 not tested but categorised as non-carriers when parents tested RET M– | RET mutation testing by restriction site polymorphism analysis of codon 634, and confirmatory direct DNA sequencing of exon 11 | 36/63 (57.1%) first-degree family members were RET M+  10/30 (33.3%) asymptomatic first-degree family members were RET M+ |
| ([Wells Jr & Skinner 1998](#_ENREF_208))  USA | IV diagnostic evidence | N=58 first-degree family members from 7 kindreds with MEN2A, showing no clinical signs/symptoms | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 10 and 11 | 21/58 (36.2%) first-degree family members were RET M+ |
| ([Frilling et al. 1995](#_ENREF_72))  Germany | IV diagnostic evidence | N=56 clinically unaffected first-degree relatives from 21 MEN2 and FMTC families | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11, and restriction site polymorphism analysis of exon 16 | 21/56 (37.5%) first-degree family members were RET M+ |
| ([Gagel et al. 1995](#_ENREF_75))  USA | IV diagnostic evidence | N=54 first-degree relatives (affected parent) from 28 families with MEN2A | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16 | 19/54 (35.2%) first-degree family members were RET M+ |
| ([Pinna et al. 2007](#_ENREF_159))  Italy | IV diagnostic evidence | N=43 first-degree relatives of 7 RET M+ index cases with MTC | RET mutation testing by direct DNA sequencing of exons 8–16 in index case, and appropriate exon in family members | 22/43 (51.2%) of first-degree family members were RET M+ |
| ([Shimotake et al. 1996](#_ENREF_186))  Japan | IV diagnostic evidence | N=37 first-degree relatives in a MEN2 family with a RET C634R mutation  6/37 without clinical signs | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exon 11 | 18/37 (48.6%) first-degree family members were RET M+ |
| ([Vestergaard et al. 2007](#_ENREF_206))  Denmark | IV diagnostic evidence | N=27 first-degree family members (children of RET M+ patients) from a large kindred with a RET Y791F mutation | RET mutation testing by direct DNA sequencing of exon 13 | 12/27 (44.4%) first-degree family members were RET M+ |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV diagnostic evidence | N=25 asymptomatic first-degree relatives from 12 unrelated Greek families  9 MEN2A  1 FMTC  3 likely FMTC  Aged 3 months to 86 years | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and/or 16 | 5/25 (20%) asymptomatic first-degree family members were RET M+ |
| ([Sharma & Saranath 2011](#_ENREF_184))  India | IV diagnostic evidence | N=25 first-degree relatives from 7 RET M+ MTC index patients | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 for index cases and specific exons for family members | 13/25 (52%) first-degree family members were RET M+:  11/15 exon 11 codon 634 (C→R)  0/3 exon 11 codon 634 (C→Y)  0/1 exon 10 codon 618 (C→G)  2/2 exon 10 codon 609 (C→R)  0/4 exon 16 codon 918 (M→T) |
| ([Dourisboure et al. 2005](#_ENREF_47))  Argentina | IV diagnostic evidence | N=21 first-degree relatives from a MEN2B family with a RET C630R mutation  5 affected with MTC | RET mutation testing by direct DNA sequencing of exon 11 | 7/21 (33.3%) first-degree family members were RET M+ |
| First- and second-degree, or degree not stated | | | | |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV diagnostic evidence | N=229 extended family members from 6 generations of an MTC index patient with RET G533C mutation | RET mutation testing by direct DNA sequencing of exon 8 | 76/229 (33.2%) extended family members were RET M+ |
| ([Frank-Raue et al. 1996](#_ENREF_69))  Germany | IV diagnostic evidence | N=159 at-risk members (degree not stated) from 35 families with hereditary MTC | RET mutation testing by single-strand conformation polymorphism analysis, and then either direct DNA sequencing or restriction site polymorphism analysis of exons 10 or 11  Direct DNA sequencing of exons 13 or 16 | 84/159 (52.8%) extended family members were RET M+:  64/111(57.7%) MEN2A  16/31(51.6%) FMTC  4/17(23.5%) MEN2B |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV diagnostic evidence | N=146 relatives (degree not stated) of 47 RET M+ index cases who had MTCs without family history of endocrine disorders | RET mutation testing of relatives by direct DNA sequencing of exon affected in index case | 60/146 (41.1%) relatives were RET M+:  35/60 carriers showed clinical or biochemical evidence but were unaware of condition  20/60 showed no signs of disease  5/60 refused clinical/biochemical investigations |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV diagnostic evidence | N=133 family members (degree not stated) from 17 MEN2 families  113 were from families with MEN2A  37 had clinical signs of disease | RET mutation testing by single-strand conformation polymorphism analysis, restriction enzyme analysis and direct DNA sequencing of exons 10, 11, 13, 14 or 15 | 61/133 (45.8%) extended family members were RET M+  57/113 (50.4%) MEN2A family members were RET M+ |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV diagnostic evidence | N=132 relatives (degree not stated) from 7 MEN2A families | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing for family members | 21/132 (15.9%) family members were RET M+ |
| ([Tsai et al. 1994](#_ENREF_202))  USA | IV diagnostic evidence | N=109 members (degree not stated) of 13 kindreds:  9 MEN2A  2 MEN2B  2 FMTC  47 clinically affected  62 non-affected | RET mutation testing by direct DNA sequencing of exons 10 and 11 | 41/109 (37.6%) relatives were RET M+:  41/85 (48.2%) MEN2A family members  0/16 (0%) MEN2B family members  0/8 (0%) FMTC family members  (NB test not appropriate to detect mutations for phenotype MEN2B) |
| ([Shifrin et al. 2009](#_ENREF_185))  USA | IV diagnostic evidence | N=107 family members (degree not stated) from a family with RET V804M mutation (exon 14) | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exon 11 | 40/107 (37.4%) family members were RET M+  Generation I: 7/7 (100%)  Generation II: 17/22 (77%)  Generation III: 15/22 (68%) |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV diagnostic evidence | N=98 relatives (degree not stated) from 17 MEN2A index cases (aged 6 months – 81 years)  N=13 relatives (degree not stated) from 5 MEN2B index cases | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, confirmed by restriction site polymorphism analysis | 42/98 (42.9%) MEN2A relatives were RET M+  All had the RET C634R mutation  0/13 (0%) MEN2B relatives were RET M+ |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV diagnostic evidence | N=93 relatives (degree not stated) from 10 MEN2A or FMTC kindreds | RET mutation testing by denaturing gradient gel electrophoresis mutational analysis of exons 10 and 11, with confirmatory direct DNA sequencing | 29/93 (31.2%) relatives were RET M+ |
| ([Algun et al. 2002](#_ENREF_6))  Turkey | IV diagnostic evidence | N=88 relatives (degree not stated) from 4 generations of an extended MEN2A family with a RET C634G mutation | RET mutation testing by restriction site polymorphism analysis of exon 11 | 18/88 (20.5%) relatives were RET M+ |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV diagnostic evidence | N=77 relatives (degree not stated) of 23 RET M+ index cases with MTC:  6 previously classified as sporadic MTC  4 FMTC families  9 MEN2A families  4 MEN2B families | RET mutation testing by direct DNA sequencing of exons 10, 11, 13, 14 and/or 16 | 24/77 (42.1%) relatives were RET M+:  3/9 (33.3%) sporadic MTC  9/22 (40.9%) FMTC  10/39 (25.6%) MEN2A  2/7 (28.6%) MEN2B |
| ([Chi et al. 1994](#_ENREF_33))  Japan | IV diagnostic evidence | N=74 relatives (degree not stated) from an extended MEN2A pedigree with a RET C634R mutation:  43 clinically affected  31 considered at risk | RET mutation testing by restriction site polymorphism analysis of exon 11 | 45/74 (60.8%) relatives were RET M+  43/43 (100%) clinically affected  2/31 (6.5%) at risk relatives |
| ([Halling et al. 1997](#_ENREF_87))  USA | IV diagnostic evidence | N=72 family members (degree not stated) from one large FMTC kindred with a RET C609Y mutation | RET mutation testing by direct DNA sequencing of exon 10 | 34/72 (47%) first- and second-degree family members were RET M+:  Generation III: 16/23 (70%)  Generation IV: 18/49 (37%) |
| ([Bugalho et al. 2007](#_ENREF_22))  Portugal | IV diagnostic evidence | N=65 relatives (degree not stated) of 8 probands of established FMTC/MEN2 kindreds with a RET mutation  53 were asymptomatic | RET mutation testing by direct DNA sequencing of exons 10–16, or restriction site polymorphism analysis of exons 13–16  Exon 8 was screened for gross insertions/deletions (method not stated) | 32/65 (49.2%) relatives were RET M+  20/53 (37.7%) asymptomatic |
| ([Chang et al. 2009](#_ENREF_32))  Taiwan | IV diagnostic evidence | N=61 relatives (degree not stated) from 8 unrelated MTC families:  45 from 5 MEN2A families  9 from 2 MEN2B families  7 from 1 FMTC family | RET mutation testing by direct DNA sequencing of exons 1–20 | 22/61 (36.1%) RET M+:  18/45 (40%) MEN2A  0/9 (0%) MEN2B  4/7 (57.1%) |
| ([Oriola et al. 1996](#_ENREF_151))  Spain | IV diagnostic evidence | N=59 family members (degree not stated) from 7 MEN2A families:  20 symptomatic  39 at risk of disease | RET mutation testing by direct DNA sequencing of RET exons 10 and 11, and restriction site polymorphism analysis of exons 10 and exon 11 | 28/59 (47.5%) family members were RET M+  8/39 asymptomatic family members |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV diagnostic evidence | N=58 members (degree not stated) of 12 unrelated Greek families  9 MEN2A  1 FMTC  3 likely FMTC  33 clinically affected (aged 12–65 years) | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and/or 16 | 38/58 (65.5%) family members were RET M+  33/33 (100%) were clinically affected |
| ([Pacini et al. 1995](#_ENREF_152))  Italy | IV diagnostic evidence | N=58 family members (degree not stated) from 9 MEN2 families:  16 affected  42 at risk of disease | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 | 21/58 (36.2%) family members were RET M+  5/42 (11.9%) clinically unaffected family members |
| ([Dos Santos et al. 2007](#_ENREF_46))  Brazil | IV diagnostic evidence | N=57 at-risk family members (degree not stated) from 7 index cases:  3 MEN2A  1 MEN2B  3 FMTC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index patients, and restricted to specific exon for family members | 35/57 (61.4%) relatives were RET M+:  19 MEN2A  15 FMTC  1 MEN2B |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia, New Zealand | IV diagnostic evidence | N=54 family members (degree not stated) of 2 probands:  47 family members from 4 generations; proband has RET V804L mutation  7 family members from 3 generations; proband has RET V804M mutation | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16 between 1993 and 1998, then exons 13–15 were included | Family 1: 22/47 (46.8%)  Family 2: 5/7 (71.4%) |
| ([Fink et al. 1996](#_ENREF_65))  Austria | IV diagnostic evidence | N=52 asymptomatic relatives (degree not stated) from 13 families clinically diagnosed with FMTC, MEN2A, MEN2B or suspected of inheritable MTC | RET mutation testing by single-strand conformational analysis or restriction site polymorphism analysis, and confirmatory direct DNA sequencing of exons 10, 11, 13 and 16; if no mutation detected, direct DNA sequencing of exon 14 was conducted | 10/52 (19.2%) relatives were RET M+:  5/19 (26.3%) FMTC family members  5/18 (27.8%) MEN2A family members  0/5 (0%) MEN2B family members |
| ([Lecube et al. 2002](#_ENREF_119))  Spain | IV diagnostic evidence | N=52 family members (degree not stated) of an FMTC family with a RET V804M mutation | RET mutation testing by restriction site polymorphism analysis of exon 14 | 25/52 (48.1%) family members were RET M+ |
| ([Gonzalez et al. 2003](#_ENREF_81))  Mexico | IV diagnostic evidence | N=48 family members (degree not stated) of 6 RET M+ probands:  3 MEN2B  2 MEN2A  1 sporadic MTC | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16, and direct DNA sequencing of exons 13–15 | 15/48 (31.3%) family members were RET M+ |
| ([Gosnell et al. 2006](#_ENREF_82))  Australia | IV diagnostic evidence | N=48 at-risk family members (degree not stated) from a MEN2A kindred with a RET V804L mutation | RET mutation testing (method not stated) | 23/48 (47.9%) family members were RET M+ |
| ([Fugazzola et al. 2002](#_ENREF_74))  Italy | IV diagnostic evidence | N=44 members (degree not stated) of a large FMTC pedigree with a RET A891S mutation | RET mutation testing by direct DNA sequencing of exon 15 | 14/44 (31.8%) family members were RET M+ |
| ([Klein et al. 2001](#_ENREF_109))  Hungary | IV diagnostic evidence | N=43 relatives (degree not stated) of 14 index cases with MTC and RET M+ | RET mutation testing by restriction site polymorphism analysis of exon 11, and/or direct DNA sequencing of exons 10, 13 and/or 14 | 25/43 (58.1%) family members were RET M+ |
| ([Marsh et al. 1996](#_ENREF_133))  Australia and New Zealand | IV diagnostic evidence | N=39 members (degree not stated) of 16 MEN2A and FMTC families at risk of being a gene carrier | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 | 7/39 (17.9%) family members were RET M+  5/21 (23.8%) from 2 MEN2A families were RET M+  1/7 (14.3%) with raised stimulated calcitonin levels were RET M+ |
| ([Frohnauer et al. 2000](#_ENREF_73))  USA | IV diagnostic evidence | N=38 members (degree not stated) from 5 MEN2A kindreds with a RET codon 804 mutation | RET mutation testing by denaturing gradient gel electrophoresis analysis confirmed by direct DNA sequencing of exon 14 | 23/38 (60.5%) family members were RET M+ |
| ([Komminoth et al. 1995](#_ENREF_110))  Switzerland | IV diagnostic evidence | N=38 members (degree not stated) from 3 MEN2A families, 2 MEN2B families and 4 suspected MEN2 families | RET mutation testing by single-strand conformation polymorphism and heteroduplex gel electrophoresis analysis of exons 10, 11, 13 and/or 15 | 11/38 (28.9%) family members were RET M+:  5/21 (23.8%) had exon 10 mutations  4/9 (44.4%) had exon 11 mutations  2/8 (25.0%) had exon 16 mutations |
| ([Hernandez et al. 1997](#_ENREF_90))  Spain | IV diagnostic evidence | N=36 asymptomatic members (degree not stated) of 3 families with MEN2A  8 had raised pentagastrin-stimulated calcitonin levels | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11 | 6/36 (16.7%) family members were RET M+  6/8 (75%) with raised stimulated calcitonin levels were RET M+ |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV diagnostic evidence | N=34 members (degree not stated) of 9 separate families with hereditary MTC:  5 MEN2A  2 MEN2B  1 FMTC  1 with <4 MTC cases | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14 | 22/34 (64.7%) family members were RET M+ |
| ([Siggelkow et al. 2001](#_ENREF_189))  Germany | IV diagnostic evidence | N=34 first- and second-degree relatives of an index case with FMTC and a RET C611F mutation | RET mutation testing restriction site polymorphism analysis of exon 10 in family members | 19/34 (55.9%) first- and second-degree family members were RET M+:  Generation III: 6/8 (75%)  Generation IV: 9/17 (52.9%)  Generation V:4/9 (44.4%) |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV diagnostic evidence | N=31 relatives (degree not stated) of 13 RET M+ sporadic MTC patients | RET mutation testing by restriction site polymorphism analysis of exon affected in index case | 16/31 (17.9%) family members were RET M+:  9 x codon 634  1 x codon 790  6 x codon 618 |
| ([Calva et al. 2009](#_ENREF_27))  USA | IV diagnostic evidence | N=31 first- and second-degree family members from a MEN2A kindred with a RET C609Y mutation | RET mutation testing (method not stated) | 22/31 (70.9%) first- and second-degree family members were RET M+ |
| ([Kinlaw et al. 2005](#_ENREF_107))  USA | IV diagnostic evidence | N=29 first- and second-degree relatives in a family with MEN2A due to RET C609S mutation  6 with manifestations of MEN2A | RET mutation testing by restriction site polymorphism analysis of exon 10 | 14/29 (48.3%) family members |
| ([Neocleous et al. 2011](#_ENREF_140))  Cyprus | IV diagnostic evidence | N=29 family members (degree not stated) from 7 FMTC families and 1 MEN2A family with a RET C618R mutation | RET mutation testing by direct DNA sequencing of exon 10 | 15/29 (51.7%) family members  15/15 RET M+ had Cys618Ser mutations |
| ([Jung et al. 2010](#_ENREF_98))  Korea | IV diagnostic evidence | N=28 first- and second-degree members (excluding the proband) of a 3-generation FMTC family with a RET C618S mutation  8 had MTC | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exon 10 | 11/28 (39.3%) first- and second-degree family members were RET M+ |
| ([Uchino et al. 1999](#_ENREF_204))  Japan | IV diagnostic evidence | N=27 members (degree not stated) from 5 MEN2A families whose clinical status was unknown | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 or 16 | 6/27 (22.2%) family members were RET M+ |
| ([Pasini et al. 2002](#_ENREF_154))  Italy | IV diagnostic evidence | N=26 family members (degree not stated) of a patient with Hirschsprung’s disease and MEN2 (RET C618R mutation) | RET mutation testing restriction site polymorphism analysis and direct DNA sequencing of exon 10 in family members | 12/26 (46.2%) family members were RET M+ |
| ([Wu et al. 1998](#_ENREF_214))  Taiwan | IV diagnostic evidence | N=26 first- and second-degree relatives of 2 probands from 2 unrelated MEN2A families | RET mutation testing by direct DNA sequencing of exons 10 and 11 | 11/26 (42.3%) first- and second-degree family members |
| ([Gil et al. 2002](#_ENREF_78))  Spain | IV diagnostic evidence | N=23 members of 4 independent MEN2A families (degree not stated):  13 clinically affected:  9 MTC only 4 MTC + PCC  10 unaffected | RET mutation testing by single-strand conformation polymorphism analysis and restriction site polymorphism analysis, with confirmatory direct DNA sequencing of exons 10 and 11 | 13/23 (56.6%) family members were RET M+:  13/13 (100%) clinically affected  0/10 (0%) unaffected |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV diagnostic evidence | N=22 extended family members of an MTC patient confirmed to have a RET L790F mutation on exon 13  3 were symptomatic | RET mutation testing by direct DNA sequencing of exon 13 | 14/22 (63.6%) extended family members were RET M+ |
| ([Mastroianno et al. 2011](#_ENREF_134))  Italy | IV diagnostic evidence | N=21 first- and second-degree relatives of a proband with MEN1 (MEN1 IVs4+IG>T mutation) and MEN2 (RET K666M mutation on exon 11) | RET mutation screening of exons 8, 10, 11, 13–16 and 18 (method not stated) | 7/21 (33.3%) first- and second-degree relatives were RET M+  3/21 (14.3%) were both MEN1+ and RET M+ |
| ([Chiefari et al. 2001](#_ENREF_34))  Italy | IV diagnostic evidence | N=20 first- and second-degree relatives of proband with RET C634F mutation  6 were affected with MTC | RET mutation testing by restriction site polymorphism analysis of exon 11, confirmed by direct DNA sequencing | 7/20 (35%) first- and second-degree relatives were RET M+ |
| ([Morita et al. 1996](#_ENREF_137))  Japan | IV diagnostic evidence | N=20 individuals:  1 proband with MEN2A (RET C618S mutation on exon 10)  6 children of the proband  10 grandchildren  3 great-grandchildren | RET testing by PCR amplification and restriction enzyme analysis of exon 10 C618S | 11/19 (55%) family members were RET M+:  4/6 (66.7%) children  4/10 (40%) grandchildren  2/3 (66.7%) great-grandchildren |
| ([Caron et al. 1996](#_ENREF_28))  France | IV diagnostic evidence | N=14 extended family members of a confirmed MEN2A patient with a RET C618R mutation | RET mutation testing by direct DNA sequencing of exons 10 and 11 | 4/14 (28.6%) extended family members were RET M+:  All 4 RET M+ family members were symptomatic |
| ([Abdelhakim et al. 2009](#_ENREF_1))  Morocco | IV diagnostic evidence | N=13 family members (degree not stated) of 3 RET M+ index cases | RET mutation testing by direct DNA sequencing of exons 8, 10, 11, 13–15 and 16 | 2/13 (15.4%) family members were RET M+ |
| ([Neumann et al. 1995](#_ENREF_142))  Germany | IV diagnostic evidence | N=27 family members (degree not stated) from 7 MEN2A families and 1 MEN2B family who had had negative clinical screening (n=19) or unknown phenotype (n=8) | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 (also checked for mutations in SDHB, SDHD and VHL) | 4/27 (14.8%) clinically negative family members were RET M+ |

PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

# Appendix Studies reporting rates of treatment

Table 86 Rates of treatment

| Study and location | Level of evidence | Study population | Intervention | Rates of treatment/surveillance |
| --- | --- | --- | --- | --- |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | N=60 RET M+ family members of patients with MTC reclassified from spontaneous MTC to FMTC or MEN2A due to RET mutation | RET mutation testing method changed over 15 years  Initially used direct DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8  Total thyroidectomy | 35/60 (58.3%) RET M+ family members were found to be affected clinically or biochemically  25/60 (41.7%) RET M+ were clinically unaffected  5/60 (8.3%) RET M+ refused clinical and/or biochemical examinations  30/35 (85.7%) RET M+-affected patients underwent total thyroidectomy  20/20 (100%) RET M+ clinically unaffected underwent yearly clinical and biochemical assessment. |
| ([Pinna et al. 2007](#_ENREF_159))  Italy | IV interventional evidence  High quality (5/6) | N=22 RET M+ family members | RET mutation testing by direct DNA sequencing of exons 8–16  Total thyroidectomy | 14/22 (63.6%) RET M+ family members underwent prophylactic thyroidectomy |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV interventional evidence  Moderate quality (4/6) | N=229 members spanning 6 generations of a large extended FMTC family with a RET G533C mutation  76 members were RET M+ | RET mutation testing by direct DNA sequencing of exon 8  Prophylactic thyroidectomy | 35/76 (46.1%) RET M+ family members had a thyroidectomy  37/76 (53.9%) had not yet undergone surgery:  3/37 had scheduled surgery  10/37 presented with low pentagastrin-stimulated calcitonin levels and surgery was delayed  24/37 had not yet completed clinical evaluation because molecular diagnosis was too recent  3/76 refused further clinical investigation  1/76 refused surgery  153/153 (100%) RET M– family members had normal pentagastrin-stimulated calcitonin levels and were excluded from further clinical investigation |
| ([Frank-Raue et al. 1996](#_ENREF_69))  Germany | IV interventional evidence  Moderate quality (4/6) | N=159 members of 35 hereditary MTC families who had RET mutation testing  84 were RET M+ | RET mutation testing by single-strand conformation polymorphism analysis, and then either direct DNA sequencing or restriction site polymorphism analysis of exons 10 and 11, and direct DNA sequencing of exons 13 and 16  Clinical screening  Prophylactic thyroidectomy | 9/17 (52.9%) asymptomatic patients had prophylactic thyroidectomy  4 patients with elevated calcitonin levels who had thyroidectomy prior to genetic testing were RET M–  Histopathology results revealed only minor CCH or normal results’ |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia, New Zealand | IV interventional evidence  Moderate quality (4/6) | Family 1:  N=48 family members from 4 generations  23 were RET M+  Family 2:  N=9 family members from 3 generations  6 were RET M+ | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16 From 1998, analysis also of exons 13–15 for probands  Family members screened for family RET mutation  Prophylactic thyroidectomy | Family 1:  22/23 (95.7%) (including proband) RET M+ family members underwent prophylactic thyroidectomy  1/23 (3.7%) was awaiting surgery  Family 2:  3/6 (50%) (including proband) RET M+ family members had thyroidectomy  2 had elevated basal calcitonin levels but declined surgery  1 was under further investigation |
| ([Marsh et al. 1996](#_ENREF_133))  Australia and New Zealand | IV interventional evidence  Moderate quality (4/6) | N=39 members of 16 MEN2A and FMTC families at risk of being a gene carrier  7 were RET M+ | Restriction site polymorphism analysis of RET exons 10 and 11  Thyroidectomy | 2/32 (6.3%) RET M– members from 2 MEN2A families had undergone thyroidectomy prior to RET mutation testing based on pentagastrin results:  1 had CCH pathology  1 was normal  5 other RET M– members of these 2 families also had elevated calcitonin levels but their management wasn’t stated |
| ([Boer et al. 2003](#_ENREF_19))  Hungary | IV interventional evidence  Moderate quality (4/6) | N=25 RET M+ consecutive unrelated patients with MTC admitted for genetic screening | RET mutation testing by direct DNA sequencing (exons not specified)  Thyroidectomy | 14/25 (56%) underwent surgery  5/25 (20%) screened postoperatively  6/25 (24%) refused treatment |
| ([Frohnauer et al. 2000](#_ENREF_73))  USA | IV interventional evidence  Moderate quality (4/6) | N=23 members from 5 MEN2A kindreds who had a RET codon 804 mutation | RET mutation testing by denaturing gradient gel electrophoresis analysis, confirmed by direct DNA sequencing of exon 14  Thyroidectomy | 14/23 (60.9%) had a thyroidectomy  2/23 (8.7%) were aged 2 years and awaiting consideration for prophylactic thyroidectomy  2/23 (8.7%) were aged 80 and 85 years with no clinical signs of disease, and refused further testing  1/23 (4.3%) had no further testing (reason not given)  4/23 (17.4%) had calcitonin levels checked  1/4 (aged 84 years) had an elevated basal calcitonin level |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV interventional evidence  Moderate quality (4/6) | N=21 RET M+ family members from 7 MEN2A kindreds  7 had elevated calcitonin levels | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of RET exon affected in index case  Thyroidectomy | 13/21 (61.9%) elected to have prophylactic thyroidectomy based on genetic screening |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV interventional evidence  Moderate quality (4/6) | N=5 RET M+ patients with apparently sporadic MTC  N=10 patients clinically diagnosed as MEN2A from calcitonin levels but dubious results or histopathology | RET mutation testing by denaturing gradient gel analysis of exons 10 and 11, with confirmatory direct DNA sequencing | 5 patients with apparently sporadic MTC (lack of family history) were found to have a germline RET mutation, prompting genetic testing of first-degree relatives  9/10 (90%) patients previously classified as MEN2A (but had questionable results) were reclassified as RET M–  7/8 (87.5%) patients who had thyroidectomy prior to genetic testing were RET M–  2 patients were spared surgery based on RET M– status |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV interventional evidence  Moderate quality (3/6) | N=69 index cases and family members with a RET codon 634 mutation:  47 were clinically diagnosed  22 had no clinical signs of disease | RET mutation testing by single-strand conformational polymorphism analysis, restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15  Total thyroidectomy, with a central cervical lymph node dissection in those with increased calcitonin levels | 43/47 (91.5%) clinically affected patients underwent thyroidectomy  7/22 (31.8%) gene carriers without clinical signs underwent thyroidectomy |
| ([Lombardo et al. 2002](#_ENREF_126))  France and Italy | IV interventional evidence  Moderate quality (3/6) | N=61 patients with RET V804L mutations from 5 families | RET mutation testing by single strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15, and restriction site polymorphism analysis of exon 16  Clinical screening  Total thyroidectomy | 31/61 (50.8%) underwent thyroidectomy:  3 (index case) due to thyroid tumour  14 based on detectable basal calcitonin levels  13 based on increased pentagastrin-stimulated calcitonin levels  1 based on increase in basal calcitonin levels and the willingness of parents  1 based on follicular tumour |
| ([Lindskog et al. 2004](#_ENREF_124))  Sweden | IV interventional evidence  Moderate quality (3/6) | N=49 family members of a MEN2A family with a RET codon 618 mutation:  16 were RET M+  33 were RET M– | RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central neck dissection | 16/49 patients were RET M+:  15 were initially identified by biochemical screening  1 identified by genetic testing  16/16 (100%) had a thyroidectomy  33/33 RET M– patients previously under biochemical surveillance were informed that they had not inherited the mutation and that they and their descendants would no longer need to be evaluated for disease |
| ([Erdogan et al. 2007](#_ENREF_57))  Turkey | IV interventional evidence  Moderate quality (3/6) | N=41 RET M+ patients identified from 12 MEN2A, 2 MEN2B and 1 FMTC pedigrees  26 were asymptomatic | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–15, and restriction enzyme analysis of exon 16  Total thyroidectomy | 30/41 (73.2%) had a thyroidectomy  29/30 had elevated basal and/or stimulated calcitonin levels preoperatively  10/41 (24.4%) RET mutation carriers refused thyroidectomy despite all efforts of the medical team, including a medical student with a RET codon 634 mutation and elevated calcitonin levels |
| ([Quayle et al. 2004](#_ENREF_162))  USA | IV interventional evidence  Moderate quality (3/6) | N=39 RET M+ patients with MEN2 or FMTC diagnosed when over 50 years of age:  36 patients from MEN2A families  3 from FMTC families | RET mutation testing (method not stated)  Total thyroidectomy | 38/39 (97.4%) underwent total thyroidectomy  7/38 also underwent central node dissection  Prior to surgery:  28/38 had abnormal basal and/or pentagastrin-stimulated calcitonin levels  8/38 had palpable nodule on physical examination  2/38 were RET M+ with no physical signs of disease |
| ([Halling et al. 1997](#_ENREF_87))  USA | IV interventional evidence  Moderate quality (3/6) | N=38 family members in 1 large kindred with FMTC who had a thyroidectomy before genetic testing:  28 were RET M+  10 were RET M– | RET mutation testing by direct DNA sequencing of exon 10  Clinical screening  Thyroidectomy | 19/38 were RET M+ with elevated pentagastrin-stimulated calcitonin levels (2 were normal, 4 had CCH, 13 had MTC and CCH)  9/38 were RET M+ with normal pentagastrin-stimulated calcitonin levels  (3 were normal, 4 had CCH, 2 had MTC and CCH)  7/38 were RET M– with elevated pentagastrin-stimulated calcitonin levels (1 was normal, 5 had CCH, 1 had MTC)  3/38 were RET M– with normal pentagastrin-stimulated calcitonin levels (3 had CCH) |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV interventional evidence  Moderate quality (3/6) | N=29 relatives of RET M+ sporadic MTC patients who had the RET mutation and had a total thyroidectomy | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction site polymorphism analysis of exon affected in index case  Total thyroidectomy with central neck dissection | 27/29 (93.1%) of relatives had a thyroidectomy  2/29 (6.9%) refused surgery |
| ([Lecube et al. 2002](#_ENREF_119))  Spain | IV interventional evidence  Moderate quality (3/6) | N=22 family members of a FMTC family who had a RET V804M mutation | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–16 Biochemical screening on those RET M+ | 20/22 had normal pentagastrin-stimulated calcitonin levels (test had not yet been performed in 2 children aged 3 and 5 years)  Consequently, thyroidectomy was not recommended for these patients at that time |
| ([Lips et al. 1994](#_ENREF_125))  The Netherlands | IV interventional evidence  Moderate quality (3/6) | N=20 members from 4 large MEN2A families, who had a RET mutation and/or a thyroidectomy  14 were RET M+ | MEN2 diagnosed by linkage analysis until June 1993  RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy | 14/20 were RET M+ with normal or equivocal pentagastrin-stimulated calcitonin levels:  8/14 (57.1%) had total thyroidectomy on basis of RET mutation status  6/14 (42.9%) were scheduled for surgery  6/20 patients were RET M– but had had a thyroidectomy on the basis of raised pentagastrin-stimulated calcitonin levels:  2/6 (33.3%) had CCH  4/6 (66.7%) had normal thyroid pathology |
| ([Jung et al. 2010](#_ENREF_98))  Korea | IV interventional evidence  Moderate quality (3/6) | N=11 RET M+ members (including index case) of a 3-generation FMTC family who underwent genetic testing  6 diagnosed with MTC before genetic testing  3 diagnosed with MTC after genetic testing | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 of index case  Analysis of exon 10 in family members  Total thyroidectomy with either central neck dissection or modified radical neck dissection | 9/11 (81.8%) RET M+ were clinically affected:  8/9 (88.9%) underwent surgery  1/9 (11.1%) was scheduled for surgery  2/11 (18.2%) were asymptomatic:  1 (a 12-year-old) was being monitored to determine timing of thyroidectomy  1 (a 37-year-old) was recommended prophylactic thyroidectomy and refused |
| ([Gagel et al. 1995](#_ENREF_75))  USA | IV Interventional evidence  Moderate quality (3/6) | N=4 RET M+ children aged 3–12 years identified from genetic screening of 197 MEN2A patients | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16  Thyroidectomy based on genetic screening | 4 RET M+ children underwent thyroidectomies  All had positive histopathology for CCH and 1 also had microscopic MTC  1 mother had thyroidectomy based on raised pentagastrin-stimulated calcitonin levels; her son also had raised levels, and both were found to be RET M–  It was decided the son would be monitored instead of undergoing a thyroidectomy |
| ([Hernandez et al. 1997](#_ENREF_90))  Spain | IV interventional evidence  Poor quality (2/6) | N=36 asymptomatic members of 3 families with MEN2A  6 identified as RET M+ | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11  Clinical screening  Total thyroidectomy | 6/6 (100%) RET M+ also had raised pentagastrin-stimulated calcitonin levels  Treatment decisions the same: total thyroidectomy  2/13 (15.4%) family members tested had false positive pentagastrin-stimulated calcitonin test results  2/30 (6.6%) RET M– had raised pentagastrin-stimulated calcitonin levels  RET M– status has changed treatment decisions and prevented unnecessary thyroidectomy |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Poor quality (2/6) | N=31 family members with a RET Y791F mutation  10 index cases | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | 20/31 (64.5%) had a thyroidectomy  7/31 (22.6%) have had a thyroidectomy recommended  4/31 (12.9%) have refused the surgery |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV interventional evidence  Poor quality (2/6) | N=25 asymptomatic first-degree relatives  5 were RET M+ | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and 16  Thyroidectomy | 5/25 (20%) family members (all children) were RET M+  2/5 children (aged 10 and 14 years) underwent thyroidectomy  3/5 children aged <6 years had not yet been operated on  20/20 (100%) RET M– family members were excluded from further screening |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV interventional evidence  Poor quality (2/6) | N=15 patients from an MTC family with a RET L790F mutation (including index patient)  8 had abnormal pentagastrin-stimulated calcitonin levels | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–15, and restriction site polymorphism analysis for exon 16  Direct DNA sequencing of exon 13 in family members  Clinical screening  Prophylactic thyroidectomy | 8/15 (53.3%) had abnormal pentagastrin-stimulated calcitonin levels:  5 had prophylactic thyroidectomy (including index case)  3 refused recommended surgery due to ‘fear of future discomfort related to L-thyroxin’  7/15 (46.7%) had normal calcitonin levels, and an annual follow-up to check calcitonin levels was recommended:  4/7 had a mean follow-up of 4.5 years with normal calcitonin levels.  3/7 were lost to follow-up |
| ([Kinlaw et al. 2005](#_ENREF_107))  USA | IV interventional evidence  Poor quality (2/6) | N=15 RET M+ family members (including index case) of a MEN2A family with a RET C609S mutation | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index case  Restriction site polymorphism analysis to detect C609S mutation in family members  Thyroidectomy | 11/15 (73.3%) RET M+ underwent further biochemical and clinical screening  3/11 (27.3%) had borderline or slightly elevated pentagastrin-stimulated calcitonin levels  6/15 (40%) had prophylactic thyroidectomy (including 3 with elevated calcitonin levels)  4/15 (26.7%) refused further evaluation |
| ([Neocleous et al. 2011](#_ENREF_140))  Cyprus | IV interventional evidence  Poor quality (2/6) | N=15 RET M+ family members from 7 FMTC families and 1 MEN2A family | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index cases, and only exon 10 in family members  Thyroidectomy | 15/15 (100%) RET M+ underwent prophylactic total thyroidectomy |
| ([Uchino et al. 1999](#_ENREF_204))  Japan | IV interventional evidence  Poor quality (2/6) | N=6 clinically unaffected members from MEN2A families with mutations on RET codon 634  All had raised calcitonin levels | RET mutation testing by single strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11, 13, 14 and 16  Total thyroidectomy | 3/6 (50%) patients, aged 15, 32 and 70 years, had total thyroidectomy (the eldest patient also had a left adrenalectomy)  2/6 (33.3%) patients, aged 54 and 67 years, refused a thyroidectomy  1/6 (16.7%) patient, aged 7 years, was being followed; she had no signs of MTC by ultrasound and only slightly elevated tetragastrin-stimulated calcitonin levels (from basal level of 45 pg/mL to peak level of 110 pg/mL) |

CCH = C-cell hyperplasia; RET M+ = RET-mutation-positive, RET M– = RET-mutation-negative

# Appendix MBS items associated with investigations and treatment of MEN2

Table 87 summarises the investigations for MEN2 and other hereditary disorders. The use of these tests in those suspected of having MEN2 has likely reduced since the introduction of RET mutation testing as a triage test for further investigations. Table 88 summarises the requirements for liflong surveillance of patients with confirmed or suspectred MEN2.

Table 87 MBS items for investigating clinical features of MEN2

| **Type of resource and identifier** | **Description** | **Fees** |
| --- | --- | --- |
| MBS item 66695 | Quantitation in blood or urine of hormones and hormone binding proteins - **ACTH**, aldosterone, androstenedione, C-peptide, **calcitonin**, cortisol, DHEAS, 11-deoxycortisol, dihydrotestosterone, FSH, **gastrin**, **glucagon**, **growth hormone**, hydroxyprogesterone, **insulin**, LH, oestradiol, oestrone, progesterone, **prolactin**, **parathyroid hormone**, renin, sex hormone binding globulin, **somatomedin C(IGF-1),** free or total testosterone, urine steroid fraction or fractions, **vasoactive intestinal peptide** - 1 test | Fee: $30.50  Benefit:  75% = $22.90  85% = $25.95 |
| MBS item 66707 | 5 or more tests described in item 66695 (Item is subject to rule 6) | **Fee:** $83.35  **Benefit:**  75% = $62.55  85% = $70.85 |
| MBS item 12527 | RENAL FUNCTION TEST (with imaging and at least 2 blood samples) | **Fee:** $84.95  **Benefit:**  75% = $63.75  85% = $72.25 |

Source: March 2013 Medicare Benefits Schedule [MBS online](http://www.mbsonline.gov.au/)

MBS = Medicare Benefits Schedule

Table 88 MBS items for lifelong surveillance regimen for MEN2

| **Type of resource and identifier** | **Description** | **Fees** |
| --- | --- | --- |
| MBS item 23 | **Level B Consultation at consultation rooms**  Professional attendance at consulting rooms by a general practitioner (not being a service to which any other item in this table applies) lasting less than 20 minutes, including any of the following that are clinically relevant:   1. Taking a patient history 2. Performing a clinical examination 3. Arranging any necessary investigation 4. Implementing a management plan 5. Providing appropriate preventive health care   In relation to 1 or more health-related issues, with appropriate documentation. | Fee: $36.30  Benefit:  100% = $36.30 |
| MBS item 55032 | NECK, 1 or more structures of, **ultrasound scan** of, where:  (a) the patient is referred by a medical practitioner for ultrasonic examination not being a service associated with a service to which an item in Subgroups 2 or 3 of this Group applies; and  (b) the referring medical practitioner is not a member of a group of practitioners of which the providing practitioner is a member (R) | **Fee:** $109.10  **Benefit:**  75% = $81.85  85% = $92.75 |
| MBS item 66500 | Quantitation in serum, plasma, urine or other body fluid (except amniotic fluid), by any method except reagent tablet or reagent strip (with or without reflectance meter) of: acid phosphatase, alanine aminotransferase, albumin, alkaline phosphatase, ammonia, amylase, aspartate aminotransferase, bicarbonate, bilirubin (total), bilirubin (any fractions), C-reactive protein, **calcium (total or corrected for albumin**), chloride, creatine kinase, creatinine, gamma glutamyl transferase, globulin, glucose, lactate dehydrogenase, lipase, magnesium, phosphate, potassium, sodium, total protein, total cholesterol, triglycerides, urate or urea - 1 test | **Fee:** $9.70 **Benefit:**  75% = $7.30  85% = $8.25 |
| MBS item 66584 | Quantitation of **ionised calcium** (except if performed as part of item 66566) - 1 test | **Fee:** $9.70 **Benefit:**  75% = $7.30  85% = $8.25 |
| MBS item 66695 | Quantitation in blood or urine of hormones and hormone binding proteins - ACTH, aldosterone, androstenedione, C-peptide, **calcitonin**, cortisol, DHEAS, 11-deoxycortisol, dihydrotestosterone, FSH, gastrin, glucagon, growth hormone, hydroxyprogesterone, insulin, LH, oestradiol, oestrone, progesterone, prolactin, **parathyroid hormone**, renin, sex hormone binding globulin, somatomedin C(IGF-1), free or total testosterone, urine steroid fraction or fractions, vasoactive intestinal peptide - 1 test | **Fee:** $30.50  **Benefit:**  75% = $22.90  85% = $25.95 |
| MBS item 66650 | Alpha-fetoprotein, CA-15.3 antigen (CA15.3), CA-125 antigen (CA125), CA-19.9 antigen (CA19.9), cancer associated serum antigen (CASA), **carcinoembryonic antigen** (CEA), human chorionic gonadotrophin (HCG), neuron specific enolase (NSE), thyroglobulin in serum or other body fluid, in the monitoring of malignancy or in the detection or monitoring of hepatic tumours, gestational trophoblastic disease or germ cell tumour - quantitation - 1 test (Item is subject to rule 6) | **Fee:** $24.35 **Benefit:**  75% = $18.30 85% = $20.70 |
| MBS item 66779 | Adrenaline, noradrenaline, dopamine, histamine, hydroxyindoleacetic acid (5HIAA), hydroxymethoxymandelic acid (HMMA), homovanillic acid (HVA), **metanephrines**, methoxyhydroxyphenylethylene glycol (MHPG), phenylacetic acid (PAA) or serotonin quantitation - 1 or more tests | **Fee:** $39.95  **Benefit:**  75% $30.00  80% = $34.00 |

Source: March 2013 Medicare Benefits Schedule [MBS online](http://www.mbsonline.gov.au/)

NB: A maximum of three pathology costs are claimable under MBS arrangements due to coning.

MBS = Medicare Benefits Schedule

Treatment costs associated with the different clinical features of MEN2 are outlined in Table 89, Table 90 and Table 91.

Table 89: Possible treatment costs associated with treatment of, or prophylaxis for, an MTC

| **Type of resource and identifier** | **Description** | **Fees** |
| --- | --- | --- |
| MBS item 30296 | **THYROIDECTOMY, total** (Anaes.) (Assist.) | **Fee:** $1,023.70 **Benefit:**  75% = $796.80 |
| MBS Item 17615 | **Pre-anaesthesia consultation**  - on a patient undergoing advanced surgery or who has complex medical problems, involving a selective history and an extensive examination of multiple systems and the formulation of a written patient management plan documented in the patient notes  *AND of more than 15 minutes but not more than 30 minutes duration,* not being a service associated with a service to which items 2801–3000 applies | **Fee:** $85.55  **Benefit:**  75% = $64.20  85% = $72.75 |
| MBS item 20320 | **INITIATION OF MANAGEMENT OF ANAESTHESIA**  **-** for procedures on oesophagus, thyroid, larynx, trachea, lymphatic system, muscles, nerves or other deep tissues of the neck, not being a service to which another item in this Subgroup applies  (6 basic units) | **Fee:** $118.80 **Benefit:**  75% = $89.10  85% = $101.00 |
| MBS item 51303 | **Assistance** at any operation identified by the word "Assist." for which the fee exceeds $537.15 or at a series of operations identified by the word "Assist." for which the aggregate fee exceeds $537.15  One fifth of the established fee for the operation or combination of operations | 1/5 of $1,023.70 = $204.74 |
| MBS item 25015  *For those with MEN2B or mutations in codons 883, 918, 922a* | **ANAESTHESIA, PERFUSION OR ASSISTANCE AT ANAESTHESIA**  - where the patient is less than 12 months of age or 70 years or greater  (1 basic unit) | **Fee:** $19.80  **Benefit**:  75% = $14.85  85% = $16.85 |
| NHCDC cost weights for K06Z | Accommodation costs for **Thyroid procedure,** average length of stay 2.04 days | **Average total cost:** $4,039 |
| PBS item 2173J | **Thyroxine sodium,** 200 µg | **DPMQ:** $27.11 |
| PBS item 2175L | **Thyroxine sodium,** 100 µg | **DPMQ:** $24.08 |
| PBS item 9287T | **Thyroxine sodium,** 75 µg | **DPMQ:** $24.12 |
| PBS item 2174K | **Thyroxine sodium**, 50 µg | **DPMQ:** $23.47 |

Source: March 2013 Medicare Benefits Schedule [MBS online](http://www.mbsonline.gov.au/), Pharmaceutical Benefits Scheme website update 1 April 2013 [Pharmaceutical Benefits Scheme](http://www.pbs.gov.au/pbs/home), National Hospital Cost Data Collection private hospital costs, AR-DRG version 5.1 round 13 (2008–2009) [Round 13 (2008-09) Cost Report](http://www.health.gov.au/internet/main/publishing.nsf/Content/Round_13-cost-reports)

DPMQ = dispensed price for maximum quantity; MBS = Medicare Benefits Schedule; NHCDC = National Hospital Cost Data Collection; PBS = Pharmaceutical Benefits Schedule

a Patients with MEN2 or mutations in codons 883, 918, 922 are recommended to undergo total thyroidectomies in the first month of life;

Table 90: Possible treatment costs associated with hyperparathyroidism

| **Type of resource and identifier** | **Description** | **Fees** |
| --- | --- | --- |
| MBS item 30315 | **Parathyroid operation for hyperparathyroidism** (Anaes.) (Assist.) | **Fee:** $1,139.90 **Benefit:**  75% = $854.95 |
| MBS Item 17615 | **Pre-anaesthesia consultation**  - on a patient undergoing advanced surgery or who has complex medical problems, involving a selective history and an extensive examination of multiple systems and the formulation of a written patient management plan documented in the patient notes  *AND of more than 15 minutes but not more than 30 minutes duration,* not being a service associated with a service to which items 2801–3000 applies | **Fee:** $85.55  **Benefit:**  75% = $64.20  85% = $72.75 |
| MBS item 20320 | **INITIATION OF MANAGEMENT OF ANAESTHESIA**  **-** for procedures on oesophagus, thyroid, larynx, trachea, lymphatic system, muscles, nerves or other deep tissues of the neck, not being a service to which another item in this Subgroup applies  (6 basic units) | **Fee:** $118.80 **Benefit:**  75% = $89.10  85% = $101.00 |
| MBS item 51303 | **Assistance** at any operation identified by the word "Assist." for which the fee exceeds $537.15 or at a series of operations identified by the word "Assist." for which the aggregate fee exceeds $537.15  One fifth of the established fee for the operation or combination of operations | 1/5 of $1,139.90 = $227.98 |
| NHCDC cost weights for K05Z | Accommodation costs for **Parathyroid procedure,** average length of stay 1.96 days | **Average total cost:** $3,481 |

Source: March 2013 Medicare Benefits Schedule [MBS online](http://www.mbsonline.gov.au/), National Hospital Cost Data Collection private hospital costs, AR-DRG version 5.1 round 13 (2008–2009) [Round 13 (2008-09) Cost Report](http://www.health.gov.au/internet/main/publishing.nsf/Content/Round_13-cost-reports)

MBS = Medicare Benefits Schedule; NHCDC = National Hospital Cost Data Collection; PBS = Pharmaceutical Benefits Schedule; DPMQ = dispensed price for maximum quantity

Table 91: Possible treatment costs associated with adrenal phaeochromocytoma

| **Type of resource and identifier** | **Description** | **Fees** |
| --- | --- | --- |
| MBS item 30324 | **ADRENAL GLAND TUMOUR**, excision of (Anaes.) (Assist.) | **Fee:** $1,364.90 **Benefit:**  75% = $1,023.70 |
| MBS Item 17615 | **Pre-anaesthesia consultation**  - on a patient undergoing advanced surgery or who has complex medical problems, involving a selective history and an extensive examination of multiple systems and the formulation of a written patient management plan documented in the patient notes  *AND of more than 15 minutes but not more than 30 minutes duration,* not being a service associated with a service to which items 2801–3000 applies | **Fee:** $85.55  **Benefit:**  75% = $64.20  85% = $72.75 |
| MBS item 20320 | **INITIATION OF MANAGEMENT OF ANAESTHESIA**  **-** for procedures on oesophagus, thyroid, larynx, trachea, lymphatic system, muscles, nerves or other deep tissues of the neck, not being a service to which another item in this Subgroup applies  (6 basic units) | **Fee:** $118.80 **Benefit:**  75% = $89.10  85% = $101.00 |
| MBS item 51303 | **Assistance** at any operation identified by the word "Assist." for which the fee exceeds $537.15 or at a series of operations identified by the word "Assist." for which the aggregate fee exceeds $537.15  One fifth of the established fee for the operation or combination of operations | 1/5 of $1,364.90 = $272.98 |
| NHCDC cost weights for K03Z | Accommodation costs for **Adrenal procedure,** average length of stay 5.05 days | **Average total cost:** $9,454 |
| PBS item 1499X *(for after bilateral adrenalectomy)* | **Hydrocortisone,** 4 mg | **DPMQ:** $16.88 |
| PBS item 1500Y  *(for after bilateral adrenalectomy)* | **Hydrocortisone,** 20 mg | **DPMQ:** $21.31 |
| PBS item 1433K  *(for after bilateral adrenalectomy)* | **Fludrocortisone acetate,** 100 µg | **DPMQ:** $46.60 |

Source: March 2013 Medicare Benefits Schedule [MBS online](http://www.mbsonline.gov.au/), Pharmaceutical Benefits Scheme Website update 1 April 2013 [Pharmaceutical Benefits Scheme](file:///\\uofa\shared$\HealthSciences\SPHCP\Public%20Health\Projects\AHTA\MSAC\CAs\Completed%20CAs\RET%20gene\Recover\Pharmaceutical%20Benefits%20Schemehttp:\www.pbs.gov.au\pbs\home), National Hospital Cost Data Collection private hospital costs, AR-DRG version 5.1 round 13 (2008–2009) [Round 13 (2008-09) Cost Report](http://www.health.gov.au/internet/main/publishing.nsf/Content/Round_13-cost-reports)

MBS = Medicare Benefits Schedule; NHCDC = National Hospital Cost Data Collection; PBS = Pharmaceutical Benefits Schedule; DPMQ = dispensed price for maximum quantity

# **Appendix J Thyroid cancer incidence**, 1982–2009

The total numbers of thyroid cancer cases observed between 1982 and 2009 are presented in Table 92. The average annual increase in thyroid cancer incidence between 1982 and 2009 was 6.73%, with 6.27% observed between 2005 and 2009. Thyroid cancer incidence has increased each year since 1982 with the exception of 1984, 1993 and 1997.

Table 92 Thyroid cancer incidence, 1982–2009

| **Year** | **Number of thyroid cancer cases** | **Annual change (%)** |
| --- | --- | --- |
| 1982 | 366 |  |
| 1983 | 420 | 14.7541% |
| 1984 | 406 | –3.3333% |
| 1985 | 411 | 1.2315% |
| 1986 | 423 | 2.9197% |
| 1987 | 456 | 7.8014% |
| 1988 | 472 | 3.5088% |
| 1989 | 483 | 2.3305% |
| 1990 | 527 | 9.1097% |
| 1991 | 571 | 8.3491% |
| 1992 | 690 | 20.8406% |
| 1993 | 677 | –1.8841% |
| 1994 | 726 | 7.2378% |
| 1995 | 814 | 12.1212% |
| 1996 | 894 | 9.8280% |
| 1997 | 870 | –2.6846% |
| 1998 | 987 | 13.4483% |
| 1999 | 1,012 | 2.5329% |
| 2000 | 1,062 | 4.9407% |
| 2001 | 1,205 | 13.4652% |
| 2002 | 1,218 | 1.0788% |
| 2003 | 1,414 | 16.0920% |
| 2004 | 1,508 | 6.6478% |
| 2005 | 1,617 | 7.2281% |
| 2006 | 1,664 | 2.9066% |
| 2007 | 1,789 | 7.5120% |
| 2008 | 1,995 | 11.5148% |
| 2009 | 2,039 | 2.2084% |

Source: AIHW thyroid cancer workbook

# Appendix Financial and Budgetary impact, DAP costs

The financial and budgetary impact of the addition of RET gene mutation testing to the MBS using the costs outlined in the Final DAP (*RET* gene screen: $1150, known RET mutation test: $480) are presented in Table 93, Table 94, Table 95 and Table 96.

**Total costs to the MBS**

Table 93 and Table 94, respectively, present the estimated annual costs of listing diagnostic and predictive RET mutation testing on the MBS between 2007 and 2015, assuming that all services are provided in an outpatient setting, where the MBS is responsible for 85% of the service. Based on an estimated number of 130–260 diagnostic and 150–359 predicitve RET mutation test performed in 2013, the estimated cost to the MBS is $294,705. This increases to $333,008 in 2015, based on 147–294 diagnostic and 169–406 predictive RET mutation tests performed (Table 95). However, an unknown proportion of patients may qualify for the Medicare Safety Net, in which case 100% of the scheduled fee is paid by the MBS. Allowing for application of the Medicare Safety Net, the overall true costs to the Commonwealth health budget would lie between the total costs to the MBS and the total combined costs of RET mutation testing, i.e. up to $346,712 in 2013 and $391,774 in 2015.

Table 93 Estimated cost of diagnostic RET mutation tests 2007–2015, with and without MBS listing

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Number of diagnostic RET mutation testsb | 89–179 | 100–200 | 102–204 | 108–217 | 115–230 | 122–245 | 130–260 | 138–277 | 147–294 |
| Estimated expenditure on diagnostic RET mutation testingc | $102,868–$205,735 | $114,713–$229,425 | $117,246–$234,492 | $124,632–$249,265 | $132,484–$264,968 | $140,831–$281,661 | $149,703–$299,406 | $159,134–$318,268 | $169,160–$338,319 |
| Patient co-paymentd | $15,430–$30,860 | $17,207–$34,414 | $17,587–$35,174 | $18,695–$37,390 | $19,873–$39,745 | $21,125–$42,249 | $22,455–$44,911 | $23,870–$47,740 | $25,374–$50,748 |
| Estimated MBS expendituree | $87,437–$174,875 | $97,506–$195,011 | $99,659–$199,318 | $105,937–$211,875 | $112,612–$225,223 | $119,706–$239,412 | $127,248–$254,495 | $135,264–$270,528 | $143,786–$287,571 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b estimated based on a 5–10% incidence of MTC in all thyroid cancers

c assuming that the cost of the diagnostic RET mutation test is $1,150, see final DAP

d assuming that most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

e assuming that all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

Table 94 Estimated cost of predictive RET mutation tests 2007–2015, with an MBS listing

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Relatives eligible for screeningb | 257–617 | 287–688 | 293–703 | 312–748 | 331–795 | 352–845 | 374–898 | 398–955 | 423–1,015 |
| Number of relatives screenedc | 103–247 | 115–275 | 117–281 | 125–299 | 132–318 | 141–338 | 150–359 | 159–382 | 169–406 |
| Estimated expenditure on predictive RET mutation testingd | $49,376–$118,503 | $55,062–$132,149 | $56,278–$135,067 | $59,823–$143,576 | $63,592–$152,622 | $67,599–$162,237 | $71,857–$172,458 | $76,384–$183,323 | $81,197–$194,872 |
| Patient co–paymente | $7,406–$17,776 | $8,259–$19,822 | $8,442–$20,260 | $8,974–$21,536 | $9,539–$22,893 | $10,140–$24,336 | $10,779–$25,869 | $11,458–$27,498 | $12,179–$29,231 |
| Estimated MBS expendituref | $41,970–$100,728 | $46,803–$112,326 | $47,836–$114,807 | $50,850–$122,040 | $54,054–$129,728 | $57,459–$137,901 | $61,079–$146,589 | $64,927–$155,824 | $69,017–$165,641 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b estimated based on the identification of a positive hereditary mutation in the *RET* gene in 25–30% of tests performed; each patient was assumed to have, on average, 11.5 first- or second-degree relatives eligible for familial screening

c assuming an uptake rate of 40% in eligible family members

d assuming that the cost of the predictive RET mutation test is $480, see final DAP

e assuming that most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

f assuming that all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

Table 95 Total MBS costs associated with RET mutation testing (combined costs of listing for diagnostic purposes and listing for familial screening)

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| *Total combined cost of RET mutation testing* | $238,241 | $265,674 | $271,541 | $288,648 | $306,833 | $326,164 | $346,712 | $368,555 | $391,774 |
| Lower limit | $152,244 | $169,775 | $173,524 | $184,456 | $196,076 | $208,429 | $221,560 | $235,519 | $250,356 |
| Upper limit | $324,238 | $361,574 | $369,559 | $392,841 | $417,590 | $443,898 | $471,864 | $501,591 | $533,191 |
| *Total patient co-paymentc* | $35,736 | $39,851 | $40,731 | $43,297 | $46,025 | $48,925 | $52,007 | $55,283 | $58,766 |
| Lower limit | $22,837 | $25,466 | $26,029 | $27,668 | $29,411 | $31,264 | $33,234 | $35,328 | $37,553 |
| Upper limit | $48,636 | $54,236 | $55,434 | $58,926 | $62,638 | $66,585 | $70,780 | $75,239 | $79,979 |
| ***Total cost to the MBSa*** | **$202,505** | **$225,823** | **$230,810** | **$245,351** | **$260,808** | **$277,239** | **$294,705** | **$313,272** | **$333,008** |
| Lower limit | $129,407 | $144,308 | $147,495 | $156,787 | $166,665 | $177,165 | $188,326 | $200,191 | $212,803 |
| Upper limit | $275,603 | $307,338 | $314,125 | $333,915 | $354,951 | $377,313 | $401,084 | $426,352 | $453,213 |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence 2005–09 of 6.3%

b assuming all patients qualify for the Medicare Safety Net, then the total cost to the MBS would equate to the total combined cost of RET mutation testing

c assuming most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

d assuming all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

Sensitivity analyses assuming upper estimates around disease incidence and a 100% uptake rate of familial screening are also presented in Table 96 to provide an extreme upper limit of predictable financial costs. The cost of RET mutation testing to the MBS under these extreme upper limits increases from $620,968 in 2013 to $701,674 in 2015.

Table 96 Sensitivity analyses

| **Year** | **2007** | **2008** | **2009** | **2010a** | **2011a** | **2012a** | **2013a** | **2014a** | **2015a** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Number of diagnostic RET mutation testsb | 179 | 200 | 204 | 217 | 230 | 245 | 260 | 277 | 294 |
| Total cost of diagnostic RET mutation testsc | $205,735 | $229,425 | $234,492 | $249,265 | $264,968 | $281,661 | $299,406 | $318,268 | $338,319 |
| Total number of relatives screenedd | 617 | 688 | 703 | 748 | 795 | 845 | 898 | 955 | 1015 |
| Total cost of predictive RET mutation testse | $296,258 | $330,372 | $337,668 | $358,941 | $381,554 | $405,592 | $431,144 | $458,307 | $487,180 |
| Combined cost of RET mutation testingf | $501,993 | $559,797 | $572,160 | $608,206 | $646,522 | $687,253 | $730,550 | $776,575 | $825,499 |
| **Patient contributiong** | $75,299 | $83,970 | $85,824 | $91,231 | $96,978 | $103,088 | $109,583 | $116,486 | $123,825 |
| **Total cost to the MBSh** | **$426,694** | **$475,827** | **$486,336** | **$516,975** | **$549,544** | **$584,165** | **$620,968** | **$660,089** | **$701,674** |

MBS = Medicare Benefits Schedule; RET = rearranged during transfection (proto-oncogene)

a projected incidence of thyroid cancer based on the average annual incidence during 2005–09 of 6.3%

b based on 10% incidence of MTC in all thyroid cancers

c assuming that the cost of the diagnostic RET mutation test is $1,150, see final DAP

d based on 11.5 relatives per proband and assuming 100% uptake of familial screen

e assuming that the cost of the predictive RET mutation test is $480, see final DAP

f assuming that all patients qualify for the Medicare Safety Net, then the total cost to the MBS would equate to the combined cost of RET mutation testing

g assuming that most patients are outpatients and Medicare pays 85% of the scheduled fees, with no Medicare Safety Net concessions or bulk-billed pathology service

h assuming that all services are provided in an outpatient setting such that Medicare pays 85% of the scheduled fees; no allowance for additional MBS if some patients qualify for the Medicare Safety Net

# Appendix Studies included in the review

Table 97 Study profiles of studies showing direct comparative evidence

| Study and location | Level of evidence and quality assessment | Study design | Study population | Intervention | Comparator | Inclusion/exclusion criteria | Outcomes assessed | Duration of follow-up |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ([Diaz & Wohllk 2012](#_ENREF_43))  Chile | III-3 interventional evidence  High risk of bias (9/26) | Historical controlled study | N=60 MEN2 patients who underwent total thyroidectomy | N=31  Total thyroidectomy after genetic diagnosis | N=29  Total thyroidectomy after clinical diagnosis | Inclusion  MEN2 phenotypes or RET M+  Exclusion  Not stated | Incidence of residual/recurrent disease  Mortality | Not stated |
| ([Kameyama & Takami 2004](#_ENREF_101))  Japan | III-3 interventional evidence  High risk of bias (10/26) | Historical controlled study | N=905 MTC patients:  634 patients in 1996 271 patients in 2002  1996: 175 MEN2A  49 FMTC  20 MEN2B  390 sporadic MTC  2002: 83 MEN2A  14 FMTC  11 MEN2B  163 sporadic MTC | N=271  Diagnosis with RET mutation testing (as well as clinical information, neck mass, serum calcitonin, and other findings) | N=634  Diagnosis based on clinical information (mass in the neck, serum calcitonin level, hypertension and other findings) | Inclusion  MTC patients from institutional members of the Japanese Society of Thyroid Surgery, surveys in 1996 and 2002  Exclusion  Not stated | Age at diagnosis | Not stated |
| ([Lallier et al. 1998](#_ENREF_113))  Canada | III-3 interventional evidence  High risk of bias (15/26) | Historical controlled study | N=13 MEN2 patients (children) who had a total thyroidectomy between 1981 and 1997  *With RET mutation testing:* 5 codon 620 mutations  1 codon 643 mutation  *Without RET mutation testing:*  codons unknown | N=6  Total thyroidectomy when individual identified as gene carrier | N=7  Total thyroidectomy when serum calcitonin was elevated | Inclusion  MEN2 and underwent total thyroidectomy between 1981 and 1997  Exclusion  Not stated | Incidence and severity of MTC  Age at time of thyroidectomy  Incidence of residual/recurrent disease | Pre-RET: 2–14 years  RET: 1–2 years |
| ([Learoyd et al. 1997](#_ENREF_117))  Australia | III-3 interventional evidence  High risk of bias (16/27) | Historical controlled study | N=164 individuals from families with MEN2 and known RET mutations:  56 were RET M+  108 were RET M- | N=7  Total thyroidectomy with knowledge of RET | N=45  Total thyroidectomy without knowledge of RET | Inclusion  Families who had requested RET mutation testing from the one location in Australia performing the test  Exclusion  Not stated | Incidence and severity of MTC  Age at time of thyroidectomy  Rate of surveillance | No long-term outcomes |
| ([Lips et al. 1994](#_ENREF_125))  The Netherlands | III-3 interventional evidence  High risk of bias (7/26) | Historical controlled study | N=14 members of 4 large MEN2A families, who had a thyroidectomy:  8 on the basis of RET mutation carrier status  6 on the basis of raised pentagastrin-stimulated calcitonin levels, who were later found to be RET M– | N=8  Total thyroidectomy based on RET | N=6  Total thyroidectomy based on raised calcitonin, later found to be RET M– | Included  Asymptomatic members of 4 large MEN2A families, who had a thyroidectomy  Excluded  Not stated | Incidence and severity of MTC | Not stated |
| ([Rohmer et al. 2011](#_ENREF_172))  France | III-3 interventional evidence  Moderate risk of bias (18/26) | Historical controlled study | N=170 patients with a RET mutation who underwent a total thyroidectomy younger than 21 years of age:  109 MEN2A  24 MEN2B  37 FMTC | N=38  Total thyroidectomy after 1992 | N=132  Total thyroidectomy before 1993 | Inclusion  RET mutation from families with MEN or familial MTC and aged 21 years at time of surgery  Exclusion  Not stated | Incidence and severity of MTC  Age at time of thyroidectomy  Age-appropriate surgery  Incidence of residual/recurrent disease | Median = 5.8 years  (range 0.01–28.7 years) |
| ([Sanchez Sobrino et al. 2011](#_ENREF_177))  Spain | III-3 interventional evidence  High risk of bias (10/26) | Historical controlled study | N=8 individuals from a family with MEN2A due to RET C634Y mutation | N=3  Total thyroidectomy after genetic diagnosis | N=5  Total thyroidectomy after clinical diagnosis | Inclusion  Family with MEN2A due to C634Y mutation  Exclusion  Not stated | Incidence and severity of MTC  Incidence of PCC  Incidence of HPT  Age at time of thyroidectomy  Progression-free survival | Total over 20 years |
| ([Schreinemakers et al. 2010](#_ENREF_180))  Sweden | III-3 interventional evidence  High risk of bias (17/26) | Historical controlled study | N=93 patients with a RET mutation, who underwent a total thyroidectomy younger than 20 years of age | N=68  Total thyroidectomy with knowledge of RET | N=25  Total thyroidectomy without knowledge of RET | Inclusion  MEN2 syndrome, younger than 20 years of age at the time of surgery, and had undergone a total thyroidectomy  Exclusion  Not stated | Incidence and severity of MTC  Age at time of thyroidectomy  Age-appropriate surgery  Incidence of residual/recurrent disease | Median duration = 7 years (IQR 3, 11) |
| ([Skinner et al. 1996](#_ENREF_190))  USA | III-3 interventional evidence  High risk of bias (13/26) | Historical controlled study | N=38 children who underwent thyroidectomy younger than 16 years of age for MEN2A or presence of a RET mutation | N=14  Prophylactic thyroidectomy based on RET mutation status  4/14 with elevated calcitonin | N=24  Thyroidectomy without knowing of RET mutations (elevated calcitonin levels or strong family history of MTC, or characteristics of phenotype) | Inclusion  Children who underwent thyroidectomy prior to 16 years of age for MEN2A or MEN2B  Exclusion  No RET mutation | Incidence and severity of MTC  Incidence of residual/recurrent disease | Pre-RET: mean = 9.3 years post thyroidectomy  RET mutation testing era: mean = 1.3 years |

IQR = interquartile range; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

Table Study profiles of studies showing direct uncomparative evidence

| Study and location | Level of evidence and quality assessment | Study design | Study population | | Intervention | Inclusion/exclusion criteria | Outcomes assessed | Duration of follow-up |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ([Abdelhakim et al. 2009](#_ENREF_1))  Morocco | IV interventional evidence  Moderate Quality (4/6) | Case series | N=9 index patients with diagnosed MTC  3 were RET M+:  2 MEN2A  1 unclassified  0/6 suspected sporadic MTC cases were RET M+ | | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16  Total thyroidectomy | Inclusion  Patients with diagnosed MTC  Exclusion  Not stated | RET mutation status  Incidence of MTC  Incidence of PCC  Incidence of HPT  Age at diagnosis | Not stated |
| ([Algun et al. 2002](#_ENREF_6))  Turkey | IV interventional evidence  Moderate quality (4/6) | Case series | N=88 members from 4 generations of an extended family with MEN2A  18 were RET M+  12 had a thyroidectomy | RET mutation testing by restriction site polymorphism analysis of exon 11  Clinical screening  Total thyroidectomy with central lymph node dissection | | Inclusion  Members of an extended MEN2A family  Exclusion  Not stated | RET mutation status  Incidence of MMC, CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at time of thyroidectomy | Not stated |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV interventional evidence  Moderate quality (4/6) | Case series | N=229 members spanning 6 generations of a large extended FMTC family with a RET G533C mutation  76 members were RET M+  35 RET M+ members have undergone a total thyroidectomy | | RET mutation testing by direct DNA sequencing of exon 8  Prophylactic thyroidectomy | Inclusion  Patients with RET G533C mutation from 1 FMTC family  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Postoperative calcitonin levels  Age at time of thyroidectomy  Treatment decisions | Not stated |
| ([Ameur et al. 2009](#_ENREF_11))  France | IV interventional evidence  Moderate quality (3/6) | Case series | N=46 tissue samples collected from MTC, CCH, MCC or mixed MTC patients  21 had a germline RET mutation | | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 from normal and diseased tissue samples to determine germline and somatic RET status | Inclusion  Thyroid samples from MTC and CCH patients obtained from Institut Gustave-Roussy  Exclusion  Not stated | RET mutation status  Incidence of MMC, CCH, MTC, and lymph node metastases  Age at diagnosis | N/A |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV interventional evidence  Moderate quality (3/6) | Case series | N=69 sporadic MTC patients  13 found to be RET M+  N=31 relatives of RET M+ sporadic MTC patients  16 were RET M+  27/29 RET M+ patients had total thyroidectomies | | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction site polymorphism analysis of exon affected in index case  Total thyroidectomy with central neck dissection | Inclusion  Sporadic MTC patients who underwent RET mutation testing between 1997 and 2003  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis  Treatment decisions | Not stated |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV interventional evidence  Poor quality (2/6) | Case series | N=22 members of an MTC family (including index patient)  15 had RET L790F mutation  8 had abnormal pentagastrin-stimulated calcitonin levels  5 had a thyroidectomy  3 had clinical signs of disease | | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–15, and restriction site polymorphism analysis for exon 16  Direct DNA sequencing of exon 13 in family members  Clinical screening  Prophylactic thyroidectomy | Inclusion  Family members of an index case with MTC  Exclusion  Not stated | RET mutation status  Incidence of MTC and lymph node metastases  Age at time of thyroidectomy  Safety of thyroidectomy  Rate of surveillance | Mean = 4.2 years (range 1–6 years)  After surgery:  Mean = 6.6 years  (range 6–8 years) |
| ([Boer et al. 2003](#_ENREF_19))  Hungary | IV interventional evidence  Moderate quality (4/6) | Case series | N=65 consecutive unrelated patients with MTC admitted for genetic screening for MEN2A and FMTC  25 were RET M+  5/25 were screened postoperatively  14/25 underwent thyroidectomy | | RET mutation testing by direct DNA sequencing (exons not specified)  Thyroidectomy | Inclusion  Unrelated probands diagnosed with MTC and no signs of MEN2B between 1992 and 2000  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of HPT  Rates of treatment | Mean = 47 months (range 29–78 months) |
| ([Calva et al. 2009](#_ENREF_27))  USA | IV interventional evidence  Moderate quality (4/6) | Case series | N=31 family members who underwent genetic testing  22/31 had a RET C609Y mutation  16/22 underwent a thyroidectomy | | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy for treatment or prophylaxis | Inclusion  Members of a 3-generation MEN2 family with a RET C609Y mutation  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at time of thyroidectomy | Not stated |
| ([Chang et al. 2009](#_ENREF_32))  Taiwan | IV interventional evidence  Moderate quality (3/6) | Case series | N=8 probands from 8 unrelated MTC families:  4 MEN2A  2 MEN2B  1 FMTC  1 sporadic MTC (possibly *de novo* MEN2A) | | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 | Inclusion  Probands from 8 families affected by MTC  Exclusion  Not stated | RET mutation status  Age at diagnosis | Not stated |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV interventional evidence  Moderate quality (4/6) | Case series | N=47 patients:  10 with sporadic MTC:  1/10 had a germline RET mutation  37 members of 10 separate families with hereditary MTC  22/37 were RET M+:  3 were asymptomatic  18/22 had available data  16/18 had a thyroidectomy (from 8 families) | | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14  Clinical screening  Total thyroidectomy | Inclusion  Patients either affected by MTC or belonging to families with hereditary MTC  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis  Age at time of thyroidectomy  Postoperative calcitonin levels | Not stated |
| ([Decker et al. 1996](#_ENREF_39))  USA | IV interventional evidence  Moderate quality (4/6) | Case series | N=36 children (1 month – 12 years) from confirmed MEN2A patients  18 were RET M+  11 underwent prophylactic thyroidectomy | | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10 and 11  Clinical screening  Prophylactic thyroidectomy | Inclusion  Children from 1 of 4 distinct, well-characterised, multigenerational MEN2A kindreds at direct risk for disease  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Postoperative calcitonin levels  Age at time of thyroidectomy  Safety of thyroidectomy | 36 hours post-surgery |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV interventional evidence  Moderate quality (4/6) | Case series | N=93 members of 10 MEN2A or FMTC kindreds with known RET mutations  29 were RET M+  17 had a thyroidectomy  4 are awaiting operation  8 are planned before 5 years of age  N=21 with sporadic MTC  5 were RET M+  N=10 patients clinically diagnosed as MEN2A from calcitonin levels but dubious results or histopathology  1 was RET M+ | | RET mutation testing by denaturing gradient gel analysis of exons 10 and 11 with confirmatory direct DNA sequencing | Inclusion  Consecutive patients at risk of MEN2A/FMTC referred for genetic screening during a 3-month period  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Age at time of thyroidectomy  Treatment decisions | Not stated |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV interventional evidence  Moderate quality (4/6) | Case series | N=21 RET M+ family members from 7 MEN2A kindreds  13 had thyroidectomy  7 had elevated calcitonin levels | | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of affected exon  Thyroidectomy | Inclusion  Member of MEN2A family  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Treatment decisions | Not stated |
| ([Dralle et al. 1998](#_ENREF_49))  Germany | IV interventional evidence  Moderate quality (4/6) | Case series | N=75 RET M+ patients <20 years of age who underwent a prophylactic total thyroidectomy  Identified retrospectively through a questionnaire  57 underwent additional lymph node dissections | | RET mutation testing (method not stated)  Clinical screening  Total thyroidectomy  Retrospectively identified through questionnaire | Inclusion  Patients who:  (1) had a preoperatively proved RET mutation;  (2) age at operation was younger than 20 years  (3) were clinically asymptomatic with regard to thyroid C-cell disease  (4) TNM classification  pT0–1/pNX/pN0–1/M0  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Postoperative calcitonin levels  Safety of thyroidectomy | Not stated |
| ([Erdogan et al. 2007](#_ENREF_57))  Turkey | IV interventional evidence  Moderate quality (3/6) | Case series | N=41 RET M+ patients identified from 15 pedigrees:  12 MEN2A  2 MEN2B  1 FMTC  26 were asymptomatic  30 had a thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–15, and restriction enzyme analysis of exon 16  Total thyroidectomy | Inclusion  Patients had to fulfil the clinical and molecular criteria proposed by the International RET Mutation Consortium  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Incidence of PCC  Incidence of HPT  Age at diagnosis  Treatment decisions | Not stated |
| ([Etit et al. 2008](#_ENREF_61))  USA | IV interventional evidence  Moderate quality (4/6) | Case series | N=42 specimens from patients retrospectively identified from hospital records who had undergone a prophylactic thyroidectomy for possible MTC  32 underwent RET mutation testing:  24 MEN2A  8 non-MEN  30 with family history | | RET mutation testing by direct DNA sequencing analysis of exons 10, 11 and 13–16 | Inclusion  Thyroidectomy must have been performed due to a positive family history of MEN2A, MEN2B or FMTC, an elevated serum calcitonin level, or the presence of a RET mutation, between 1977 and 2007  Exclusion  Not stated | RET mutation status  Incidence of MMC, CCH, MTC and lymph node metastases  Incidence of HPT | Mean = 4.7 years  (range 1 month – 13 years) |
| ([Feldman et al. 2000](#_ENREF_62))  UK, USA, France | IV interventional evidence  Moderate quality (4/6) | Case series | N=20 members from 2 FMTC families who have a RET V804M mutation | | RET mutation testing by restriction analysis of exon 14  Clinical screening  Thyroidectomy | Inclusion  A member of 1 of 2 FMTC families who have a RET exon 14 codon 804 V804M mutation  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC | Not stated |
| ([Frank-Raue et al. 2011](#_ENREF_70))  Germany | IV interventional evidence  Moderate quality (4/6) | Case series | N=340 patients proven to be carriers of germline mutation in exon 10 of the *RET* gene  Identified through:  47% symptomatic  53% screening | | RET mutation testing (method not stated)  Clinical screening | Inclusion  A proven carrier status of a germline mutation in exon 10 of the *RET* gene, or a relative of index registrants if diagnosed with an MTC or PCC  (from 14 different countries)  Exclusion  Not stated | RET mutation status  Incidence of MTC  Incidence of PCC  Incidence of HPT  Age at diagnosis | Not stated |
| ([Frank-Raue et al. 1997](#_ENREF_68))  Germany | IV interventional evidence  Moderate quality (4/6) | Case series | N=11 asymptomatic RET M+ children from 8 MEN2A/FMTC families | | RET mutation testing by single-strand conformation polymorphism analysis or restriction site polymorphism analysis, and direct DNA sequencing of exons 10, 11 and 13  Prophylactic thyroidectomy | Inclusion  Patients with MEN2A or FMTC from 8 families  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Postoperative calcitonin levels | Range = 2–32 months |
| ([Frank-Raue et al. 1996](#_ENREF_69))  Germany | IV interventional evidence  Moderate quality (4/6) | Case series | N=178 members of 35 families clinically identified with hereditary MTC  159 had RET mutation testing  84 were RET M+  67/84 patients were symptomatic  9/17 presymptomatic patients had prophylactic thyroidectomy | | RET mutation testing by single-strand conformation polymorphism analysis, and then either direct DNA sequencing or restriction site polymorphism analysis of exons 10 and 11  Direct DNA sequencing of exons 13 and 16  Clinical screening  Prophylactic thyroidectomy | Inclusion  A member of 1 of 35 families clinically identified as having hereditary MTC since 1993  Exclusion  Not stated | RET mutation status  Incidence of MMC, CCH and MTC  Postoperative calcitonin levels  Safety of thyroidectomy  Treatment decisions | Not stated |
| ([Franz & Wells Jr 1997](#_ENREF_71))  USA, Germany | IV interventional evidence  Moderate quality (4/6) | Case series | N=20 RET M+ patients:  19 MEN2A patients  1 FMTC patient | | RET mutation testing by restriction site polymorphism analysis and/or DNA sequencing (exons not specified)  Clinical screening  Prophylactic thyroidectomy based on RET status | Inclusion  Patients with MEN2A or FMTC from 12 distinct kindreds  Exclusion  Not stated | RET mutation status  Incidence of CCH and lymph node metastases  Postoperative calcitonin levels | Not stated |
| ([Frohnauer et al. 2000](#_ENREF_73))  USA | IV interventional evidence  Moderate quality (4/6) | Case series | N=38 members from 5 MEN2A kindreds with a RET codon 804 mutation  23 were RET M+  14 had a thyroidectomy | | RET mutation testing by denaturing gradient gel electrophoresis analysis, confirmed by direct DNA sequencing of exon 14  Thyroidectomy | Inclusion  At-risk family members of a MEN2A kindred  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Age at time of thyroidectomy  Treatment decisions  Mortality | Not stated |
| ([Gagel et al. 1995](#_ENREF_75))  USA | IV interventional evidence  Moderate quality (3/6) | Case series | N=178 members from 28 families with MEN2A:  71 were clinically confirmed: all were found to be RET M+  53 were clinically negative: all were found to be RET M–  54 were unknown status but at 50% risk of RET mutation: 19 were found to be RET M+  4 RET M+ patients (children aged 3–12 years) had a thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16  Thyroidectomy based on genetic screening | Inclusion  Member of a MEN2A kindred  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Age at time of thyroidectomy  Treatment decisions | Not stated |
| ([Gimm et al. 2002](#_ENREF_80)).  Germany, Austria | IV interventional evidence  Moderate quality (4/6) | Case series | N = 40 patients with a RET codon 790/791 mutation who underwent thyroid operations:  13 were index patients  27 were identified during RET mutation screening  10 had a thyroidectomy  30 had a thyroidectomy and lymph node dissection | | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11, 13 and 14  Thyroidectomy | Inclusion  Patients diagnosed with a codon 790/791 mutation who underwent thyroid operations  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC, and lymph node metastases  Incidence of PCC  Incidence of HPT  Postoperative calcitonin levels  Safety of thyroidectomy | Not stated |
| ([Gonzalez et al. 2003](#_ENREF_81))  Mexico | IV interventional evidence  High quality (5/6) | Case series | N=57 patients  9 Probands: 3 MEN2B  2 MEN2A  4 sporadic MTC  48 Family members  21 had MTC or CCH  17 were RET M+ (5 probands and 12 relatives)  11 had thyroidectomy  4 sporadic MTC patients were RET M– | | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16, and direct DNA sequencing of exons 13–15  Clinical screening  Thyroidectomy | Inclusion  Proband had MTC present, elevated calcitonin and thyroid resection for histopathological confirmation  Exclusion  Patients found to be RET M– were excluded from further study | RET mutation status  Progression to disease over time  Mortality | Mean = 6.7 years (range 1–24 years) |
| ([Gosnell et al. 2006](#_ENREF_82))  Australia | IV interventional evidence  Moderate quality (4/6) | Case series | N=48 at-risk individuals in a single MEN2A kindred with a RET V804L mutation  23 were RET M+  22 RET M+ family members (including proband) underwent thyroidectomy | | RET mutation testing (method not stated)  Clinical screening  Prophylactic thyroidectomy | Inclusion  At-risk individuals in 1 MEN2A kindred  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT | Not stated |
| ([Guyetant et al. 2003](#_ENREF_86))  France | IV interventional evidence  High quality (5/6) | Case series | N=66 patients who had been operated on for CCH or MTC:  43 with diffuse goiter or nodular thyroid disease  18 were potential MEN2 carriers due to prior familial MTC  5 had isolated hypercalcitoninemia  N=46 sporadic cases  27 sporadic MTC  19 sporadic CCH | | RET mutation testing of exons 8, 10, 11 and 13–16 (method not stated) | Inclusion  Consecutive patients with an MTC or CCH diagnosis in pathology department files from 1993 to 2000), representing 2.9% of 3,342 thyroid pathological specimens examined  Exclusion  Consultation cases and patients with non-total thyroidectomy, or with incomplete biological or clinical data | RET mutation status  Incidence of CCH, MTC and lymph node metastases | Not stated |
| ([Halling et al. 1997](#_ENREF_87))  USA | IV interventional evidence  Moderate quality (3/6) | Case series | N=72 family members from 1 large FMTC kindred with a RET C609Y mutation  34 were RET M+  41 had thyroidectomy before genetic testing  28 were RET M+ (19 with elevated pentagastrin-stimulated calcitonin levels)  10 were RET M– (6 with elevated pentagastrin-stimulated calcitonin levels)  I had an unknown RET mutation status with normal pathology  2 had no available pathology | | RET mutation testing by direct DNA sequencing of exon 10  Clinical screening  Thyroidectomy | Inclusion  Member of large FMTC kindred  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Treatment decisions | Not stated |
| ([Heizmann et al. 2006](#_ENREF_89))  Switzerland | IV interventional evidence  Moderate quality (4/6) | Case series | N=14 RET M+ patients who were presymptomatic, from 2 MEN2A kindreds | | RET mutation testing by single-strand conformation polymorphism analysis, denaturing gradient gel electrophoresis and direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central compartment dissection in those older than 6 years of age | Inclusion  Presymptomatic kindreds with MEN2A operated on between 1997 and 2004 by a senior surgeon  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Age at time of thyroidectomy  Safety of thyroidectomy | 6 weeks postoperative |
| ([Hernandez et al. 1997](#_ENREF_90))  Spain | IV interventional evidence  Poor quality (2/6) | Before and after case series | N=53 members of 3 MEN2A families:  17 members affected  36 members asymptomatic  6 identified as RET M+: all 6 had raised pentagastrin-stimulated calcitonin levels and a thyroidectomy  2/13 who tested as RET M– had raised pentagastrin-stimulated calcitonin levels | | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11  Clinical screening  Total thyroidectomy | Inclusion  Family members from 3 families with MEN2A, where both clinical and genetic information was available  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Treatment decisions | Not stated |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV interventional evidence  High quality (5/6) | Case series | N=106 unrelated index cases with MTC  23 index cases were RET M+  N=76 relatives of RET M+ cases:  6 previously sporadic MTC (9 family members tested)  4 FMTC families (21 family members tested)  9 MEN2A families (39 family members tested)  4 MEN2B families (7 family members tested)  24 relatives were RET M+  10 had normal levels with no signs of disease  14 had high calcitonin levels and had thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Thyroidectomy  Surgical decisions often made prior to genetic testing with no separation of data based on clinical or genetic diagnosis  Note: thyroidectomy outcomes data were not available for 1 FMTC index case | Inclusion  Unrelated index cases with MTC, and family members of those with RET mutations  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Age at diagnosis  Age at time of thyroidectomy | 1–20 years |
| ([Jung et al. 2010](#_ENREF_98))  Korea | IV interventional evidence  Moderate quality (3/6) | Case series | N=30 members of a 3-generation FMTC family  29 (index case plus 28 relatives) underwent genetic testing:  11 were RET M+ (including index case)  6 relatives (including index case) diagnosed with MTC before genetic testing  3 relatives diagnosed with MTC after genetic testing  8 relatives (including index case) underwent total thyroidectomy | | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 of index case  Analysis of exon 10 in family members  Total thyroidectomy with either central neck dissection or modified radical neck dissection | Inclusion  Members of a FMTC family  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Age at time of thyroidectomy  Postoperative calcitonin levels  Treatment decisions  Mortality | Median = 10 years |
| ([Kameyama, Okinaga & Takami 2004](#_ENREF_100))  Japan | IV interventional evidence  Moderate quality (4/6) | Case series | N=271 patients with histologically confirmed MTC  108 had hereditary MTC:  83 MEN2A  11 MEN2B 14 FMTC  53 were symptomatic:  39 had a neck mass 14 had other (e.g. adrenal) tumour  55 were asymptomatic:  45 identified as RET M+ 10 had elevated calcitonin levels  163 had sporadic MTC | | RET mutation testing (method not stated). | Inclusion  Histologically proved MTC between 1995 and 2002  Exclusion  Not stated | RET mutation status  Incidence of PCC  Incidence of HPT  Age at diagnosis | Not stated |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV interventional evidence  Poor quality (2/6) | Case series | N=58 individuals from 12 unrelated Greek families  9 MEN2A  1 FMTC  3 probable FMTC (only 3 members diagnosed with MTC)  33 clinically affected patients had a thyroidectomy for MTC prior to genetic testing  25 asymptomatic first-degree relatives of patients  5 children were RET M+ | | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and 16 | Inclusion  Individuals from families with MEN2A or FMTC  Exclusion  Not stated | RET mutation status  Incidence of PCC  Incidence of HPT  Age at diagnosis  Rate of surveillance  Treatment decisions | Not stated |
| ([Kinlaw et al. 2005](#_ENREF_107))  USA | IV interventional evidence  Poor quality (2/6) | Case series | N=30 family members (including index case) of a MEN2A family with a RET C609S mutation  15 were RET M+  6 had thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in the index case  Restriction site polymorphism analysis to detect C609S mutation in family members  Thyroidectomy | Inclusion  Family members in a family with MEN2A due to Cys609Ser mutation  Exclusion  Refusal to be tested | RET mutation status  Incidence of CCH and MTC  Incidence of PCC  Treatment decisions | Not stated |
| ([Lau et al. 2009](#_ENREF_114))  Hong Kong | IV interventional evidence  High quality (5/6) | Case series | N=22 asymptomatic patients from 8 MEN2A families who underwent prophylactic total thyroidectomy based on RET mutation status  All had RET codon 634 mutations:  12 C634Y  4 C634R  3 C634W  3 C634G | | RET mutation testing by restriction enzyme analysis and/or direct DNA sequencing (exons not specified)  Prophylactic thyroidectomy with or without a unilateral central compartment neck dissection | Inclusion  Genetic carriers who were completely asymptomatic and had no clinical evidence of MTC  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Age at time of thyroidectomy  Incidence of residual/recurrent disease  Safety of thyroidectomy | Median = 49 months  (range 13–128 months) |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia, New Zealand | IV interventional evidence  Moderate quality (4/6) | Case series | N=57 members of 2 families  Family 1:  1 proband diagnosed with PCC and MTC had RET V804L mutation  47 family members from 4 generations: 22 were RET M+  22/23 (including proband) had thyroidectomy  Family 2:  1 proband diagnosed with MTC had RET V804M mutation  8 family members from 3 generations: 5 were RET M+  3/6 (including proband) had thyroidectomy | | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16  From 1998, analysis also of exons 13–15 for probands  Family members screened for family RET mutation  Prophylactic thyroidectomy | Included  Families referred to their institute for genetic testing from the early 1990s  Excluded  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at time of thyroidectomy  Treatment decisions | Not stated |
| ([Lecube et al. 2002](#_ENREF_119))  Spain | IV interventional evidence  Moderate quality (3/6) | Case series | N=52 family members of an FMTC family with RET V804M mutation  25 were RET M+ | | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11 and 13–16  Biochemical screening on those RET M+ | Included  Family members of an FMTC family with V804M mutations  Exclusion  Not stated | RET mutation status  Incidence of PCC  Incidence of HPT  Treatment decisions | Up to 2 years |
| ([Lindskog et al. 2004](#_ENREF_124))  Sweden | IV interventional evidence  Moderate quality (3/6) | Before and after case series | N=16 RET M+ family members of a MEN2A family with a RET codon 618 mutation, who had a thyroidectomy:  15 identified by biochemical screening  1 identified by genetic testing  N=33 RET M– family members | | RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy with central neck dissection | Included  Family members from 1 large family | RET mutation status  Incidence of MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis  Age at time of thyroidectomy  Postoperative calcitonin levels  Treatment decisions  Rate of surveillance | Mean = 19±9 years |
| ([Lips et al. 1994](#_ENREF_125))  The Netherlands | IV interventional evidence  Moderate quality (3/6) | Before and after case series | N=148 members from 4 large MEN2A families:  80 MEN2A gene carriers (61 diagnosed by DNA sequence analysis)  14 were symptomatic  14 had normal pentagastrin-stimulated calcitonin results  8 had total thyroidectomy on basis of RET mutation status  6 were scheduled for surgery  68 non-carriers | | MEN2 diagnosed by linkage analysis until June 1993  RET mutation testing by direct DNA sequencing of exons 10 and 11  Total thyroidectomy | Included  Members of 4 large families with MEN2A  Exclusion  Not stated | RET mutations status  Incidence of CCH and MTC  Incidence of PCC  Incidence of HPT  Age at time of thyroidectomy  Treatment decisions  Mortality | Not stated |
| ([Lombardo et al. 2002](#_ENREF_126))  France and Italy | IV interventional evidence  Moderate quality (3/6) | Case series | N=61 patients with RET V804L mutations, from 5 families  31/61 underwent thyroidectomy:  3 index cases with MTC  1 patient with follicular tumour  14 patients with detectable basal calcitonin levels  13 patients with significant increase in pentagastrin-stimulated calcitonin levels | | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15, and restriction site polymorphism analysis of exon 16  Clinical screening  Total thyroidectomy | Included  Patients with V80L RET mutations  Excluded  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Age at time of thyroidectomy  Incidence of residual/recurrent disease  Postoperative calcitonin levels  Treatment decisions | Median = 8.5 years |
| ([Machens et al. 2006](#_ENREF_128))  Germany | IV interventional evidence  Poor quality (2/6) | Case series | N=219 patients with RET mutations divided into three categories  RET codons 918 mutations (highest risk)  RET codons 609–634 (high risk)  RET codons 768–891 (least high risk  206 patients previously described (Machens et al. 2005) | | RET mutation testing (method not stated). | Included  RET mutation carriers recruited between November 1994 and April 2005  Excluded  Not stated | RET mutation status  Incidence of PCC | Not stated |
| (Machens et al. 2005)  Germany | IV interventional evidence  High quality (5/6) | Case series | N=206 consecutive RET M+ patients who underwent surgery for CCH, MTC or PCC:  74 index cases  132 non-index cases (criteria for diagnosis and/or surgery not reported)  Stratified by risk category:  18 highest risk (codon 918)  117 high risk (codons 609–634)  71 less high risk (codons 768–891) | | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16  Clinical screening  Thyroidectomy and/or adrenalectomy | Inclusion  Patients submitted to surgery for MTC, CCH or PCC and/or adrenalectomy  Exclusion  Not stated | RET mutation status  Incidence of MTC  Time to progression stratified by RET risk category | Not stated  Mean time to progression is stated |
| ([Machens et al. 2001](#_ENREF_130))  Germany | IV interventional evidence  Poor quality (2/6) | Case series | N=63 RET M+ patients with MTC who had a thyroidectomy  36 were index patients | | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11, 13 and 14  Thyroidectomy | Inclusion  Patients operated on for MTC between November 1994 and October 1999 and had RET mutations in exons 10, 11, 13 or 14  Exclusion  Carriers of codon 918 mutation (7/198) | RET mutation status  Incidence of MTC and lymph node metastases  Age at diagnosis | Not stated |
| ([Marsh et al. 1996](#_ENREF_133))  Australia, New Zealand | IV interventional evidence  Moderate quality (4/6) | Case series | N=39 asymptomatic members of 16 MEN2A and FMTC families at risk of being a gene carrier  7 were RET M+  21 members were from 2 MEN2A families: 5 were RET M+ | | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11  Thyroidectomy | Inclusion  Family member at risk of carrying RET mutation  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Incidence of PCC  Treatment decisions | Not stated |
| ([Milos et al. 2008](#_ENREF_136))  Worldwide (Romania, Germany, Chile, Brazil, Agentina, Hungary, Spain, The Netherlands, Czech Republic, Poland, USA) | IV interventional evidence  Moderate quality (4/6) | Case series | N=92 carriers of RET C634W mutation from 20 unrelated MEN2A families  81 underwent thyroid operations  49 had available histological data  34 had available postoperative data | | RET mutation testing (method not stated) | Inclusion  Patients with C643W mutation  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Postoperative calcitonin levels  Mortality | Mean = 12 years (range 1–29 years) |
| ([Neocleous et al. 2011](#_ENREF_140))  Cyprus | IV interventional evidence  Poor quality (2/6) | Case series | N=8 probands from 7 FMTC families and 1 MEN2A family  N=29 family members from 7 of the probands: 15 were RET M+ | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index cases, and only exon 10 in family members  Thyroidectomy | Inclusion  FMTC or MEN2A patients and their family members  Exclusion  Not stated | RET mutations status  Age at diagnosis  Treatment decisions | Range = 1–10 years |
| ([Neumann et al. 2002](#_ENREF_141))  Germany and Poland | IV interventional evidence  High quality (5/6) | Case series | N=271 patients with non-syndromic PCC without family history of disease  13 were RET M+ | | RET mutation testing by single-strand conformation polymorphisms and direct DNA sequencing of exons 13–16  Also checked for mutations in SDHB, SDHD and VHL | Inclusion  Patients with PCCs consecutively registered in Freiburg, Germany, and Warsaw, Poland  Exclusion  Cases discovered by clinical or genetic screening without symptoms of illness Excluded 11 with neurofibromatosis type 1 due to clear clinical diagnosis, 14 with family history of VHL or MEN1 | RET mutation status  Incidence of MTC  Age at diagnosis | Not stated |
| ([Nguyen et al. 2001](#_ENREF_143))  France | IV interventional evidence  High quality (5/6) | Case series | N=87 first-degree relatives of index cases in MEN2 families who were diagnosed with MTC and found to be RET M+:  84 patients from 52 MEN2A families  3 patients from 3 MEN2B families | | MEN2 diagnosed by linkage analysis between 1989 and 1994  RET mutation testing by sequence analysis since 1994 (method not stated) | Inclusion  Non-index patients, descendants of index cases  MEN2A or 2B diagnosed by sequence analysis  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Incidence of PCC  Incidence of HPT  Age at diagnosis | Mean = 7.6±2.8 years  (range 1.5–10 years) |
| ([Pacini et al. 1995](#_ENREF_152))  Italy | IV interventional evidence  Moderate quality (3/6) | Case series | N=58 family members from 7 MEN2A and 2 MEN2B families  16 clinically affected patients were RET M+  5/42 clinically unaffected but at risk of disease were RET M+  4/5 had thyroidectomy | | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 | Inclusion  Non-index patients, descendants of index cases  MEN2A or 2B diagnosed by sequence analysis  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Incidence of PCC  Incidence of HPT  Age at diagnosis  Age at time of thryoidectomy | Not stated |
| ([Paszko et al. 2007](#_ENREF_155))  Poland | IV interventional evidence  Moderate quality (3/6) | Case series | N=46 patients with MTC who were RET M+  N=19 RET M+ asymptomatic relatives | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 | Inclusion  Patients with MTC  Exclusion  Not stated | RET mutation status  Incidence of PCC  Incidence of HPT | 2 years |
| ([Patocs et al. 2006](#_ENREF_157))  Hungary | IV interventional evidence  Moderate quality (3/6) | Case series | N=40 patients from 18 families who had had a thyroidectomy due to hereditary MTC or CCH:  33 MEN2A  1 MEN2B  6 from MTC families without PCC or HPT | | RET mutation testing by single-strand conformation polymorphism analysis, restriction site polymorphism analysis, and direct DNA sequencing of exons 10, 11, 13, 14 and, in MEN2B phenotype, exons 15 and 16 | Inclusion  Patients operated on for hereditary MTC or CCH  Exclusion  Patients with unidentified mutations of the *RET* gene | RET mutations status  Incidence of MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis  Mortality | Not stated |
| ([Patocs et al. 2004](#_ENREF_156))  Hungary | IV interventional evidence  Moderate quality (4/6) | Case series | N=41 patients with PCCs  7 were RET M+ | | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 14 | Inclusion  Adrenal tumours (only results for PCC presented)  Exclusion  Not stated | RET mutation status  Age at diagnosis | Not stated (only short-term outcomes) |
| ([Pinna et al. 2007](#_ENREF_159))  Italy | IV interventional evidence  High quality (5/6) | Case series | N=22 patients with MTC who had a total thyroidectomy  7 were RET M+  N=43 relatives of the 7 index cases  22 RET M+ family members  14 have undergone prophylactic total thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 8–16  Total thyroidectomy | Inclusion  Family members who underwent prophylactic total thyroidectomy  Exclusion  Not stated | RET mutation status  Incidence of CCH and MTC  Treatment decisions | Not stated |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV interventional evidence  Moderate quality (3/6) | Case series | N=160 individuals:  150 family members in 17 MEN2 families (54 had clinical signs of thyroid neoplasia or endocrine-related neoplasia)  10 patients with apparently sporadic MTCs  88 patients were RET M+:  17/88 index cases  61/88 family members  10/88 sporadic MTC cases  24/78 patients from MEN2 families had no clinical evidence of disease  69/88 had RET codon 634 mutation  50/69 underwent thyroid surgery:  43/50 had clinical disease  7/50 were clinically asymptomatic gene carriers | | RET mutation testing by single-strand conformational polymorphism analysis, restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–15  Total thyroidectomy, with a central cervical lymph node dissection in those with increased calcitonin levels | Inclusion  Diagnosis of MTC, or family member  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis  Treatment decisions  Mortality | Not stated |
| ([Quayle et al. 2007](#_ENREF_163))  USA | IV interventional evidence  Poor quality (2/6) | Case series | N=323 patients from 65 MEN2A families who were RET M+ | | RET mutation testing (method not stated) | Inclusion  Patients with MEN2A and data on PCC status, with a RET mutation  Exclusion  Patients with mutations in codons 611 and 804 | RET mutation status  Incidence of PCC | Median = 9 years |
| ([Quayle et al. 2004](#_ENREF_162))  USA | IV interventional evidence  Moderate quality (3/6) | Case series | N=39 RET M+ patients with MEN2 or FMTC diagnosed when over the age of 50 years:  36 patients with MEN2A  3 patients with FMTC  5 RET codon 609 mutations  15 RET codon 618 mutations  6 RET codon 620 mutations  12 RET codon 634 mutations  1 unknown mutation  38 had thyroidectomy | | RET mutation testing (method not stated)  Total thyroidectomy | Inclusion  Patients with MEN2A, MEN2B or FMTC who were diagnosed after the age of 50 years  Exclusion  Not stated | RET mutations status  Incidence of CCH, MTC and lymph node metastases  Incidence of residual/recurrent disease  Postoperative calcitonin levels  Treatment decisions  Mortality | Median = 6.4 years |
| ([Rodriguez Gonzalez et al. 2002](#_ENREF_170))  Spain | IV interventional evidence  Moderate quality (3/6) | Case series | N=203 MTC patients  82 patients were RET M+ (RET codon 634 mutation) and diagnosed as MEN2A  60 patients had high calcitonin levels and were not discussed further in this study  22 had normal basal and pentagastrin-stimulated calcitonin levels and received prophylactic thyroidectomy | | RET mutation testing by denaturing gradient gel electrophoresis of exons 10, 11, 13, 14 and 16, and confirmed by restriction site polymorphism analysis  Clinical screening  Prophylactic total thyroidectomy ± central neck dissection | Inclusion  Patients at risk (family) or with confirmed MTC or CCH  Only clinically negative but RET M+ were used for further data analysis  Exclusion  Patients RET M– or clinically confirmed MTC and CCH (not considered to be prophylactic surgery) | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at time of thyroidectomy  Incidence of residual/recurrent disease  Postoperative calcitonin levels  Safety of thyroidectomy | Mean = 23 months (range 6–57 months) |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV interventional evidence  High quality (5/6) | Case series | N=60 RET M+ family members of patients with MTC reclassified from sporadic MTC to FMTC or MEN2A due to a RET mutation:  5 refused treatment  20 had no clinical or biochemical signs of disease and were monitored  35 showed clinical and/or biochemical signs of disease on screening  30 (29 FMTC, 1 MEN2A) underwent total thyroidectomy | | RET mutation testing method changed over 15 years  Initially used direct DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8  Total thyroidectomy | Inclusion  Family members who underwent a total thyroidectomy after RET mutation testing  Exclusion  Did not undergo surgery (n=5) | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis  Incidence of residual/recurrent disease  Progression-free survival  Treatment decisions | Mean = 6.0 years |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV interventional evidence  Moderate quality (4/6) | Case series | N=133 patients:  17 index cases with MEN2A  5 index cases with MEN2B  98 relatives of MEN2A patients  13 relatives from MEN2B families  42 (26 juveniles and 16 adults) had RET mutation  18 carriers (aged 17 months – 21 years) had total thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, and confirmed by restriction site polymorphism analysis  Prophylactic thyroidectomy | Inclusion  Cases with clinical signs of MEN2 and their relatives  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Postoperative calcitonin levels | Not stated |
| ([Schellhaas et al. 2009](#_ENREF_179))  Germany | IV interventional evidence  High quality (5/6) | Case series | N=17 patients with a RET codon 634 mutation  14 with MEN2A  3 with apparent FMTC | | RET mutation testing (method not stated)  Prophylactic total thyroidectomy with bilateral cervicocentral lymphadenectomy | Inclusion  Total thyroidectomy between 1992 and 1999 with mutation in codon 634 in exon 11  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of residual/recurrent disease  Postoperative calcitonin levels  Safety of thyroidectomy | Median = 147 months (range 90–181 months) |
| ([Schuffenecker et al. 1998](#_ENREF_182))  France | IV interventional evidence  Moderate quality (4/6) | Case series | N=188 patients from 30 families with RET 634 mutations:  10 with C634R mutations 11 with C634Y mutations 9 with other 634 mutations | | Exhaustive RET mutation testing (method not stated, data from registry) | Inclusion  Family with 634 mutation and genotyping data from at least 2 generations with comprehensive follow-up of MTC, PCC and HPT  Exclusion  Not stated | RET mutation status  Incidence of HPT | Not stated |
| ([Schuffenecker et al. 1994](#_ENREF_181))  France | IV interventional evidence  Moderate quality (4/6) | Case series | N=86 unrelated MTC patients with RET codon 618, 620 or 634 mutations  54/58 MEN2A were RET M+  6/9 FMTC were RET M+  10/19 with other hereditary MTC were RET M+  N=259 affected members from 53 families with RET codon 634 mutations  N=60 affected members from 13 families with RET codon 618 or 620 mutations | | RET mutation testing by direct DNA sequencing of exons 10 and 11 | Inclusion  Individuals from French families with hereditary MTC  Exclusion  MEN2B | RET mutation status  Incidence of PCC | Not stated |
| ([Shifrin et al. 2009](#_ENREF_185))  USA | IV interventional evidence  Moderate quality (3/6) | Case series | N=107 members of a family with a RET V804M mutation  81 underwent genetic testing  40/81 had RET V804M mutation  15/40 had total thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (not clear if relatives tested for specific mutation or all)  Total thyroidectomy with central and ipsilateral lateral neck dissection | Inclusion  Family members from family with RET V804M mutation  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Age at diagnosis | Not stated |
| ([Shimotake et al. 1996](#_ENREF_186))  Japan | IV interventional evidence  Moderate quality (3/6) | Case series | N=6 children without clinical signs of disease who underwent RET mutation testing  3 were RET M+ | | RET mutation testing by direct DNA sequencing of exons 10 and 11 | Inclusion  First-degree family members in 1 Japanese pedigree  Exclusion  Did not undergo genetic testing | RET mutation status  Rate of surveillance | Not stated |
| ([Skinner et al. 2005](#_ENREF_191))  USA | IV interventional evidence  High quality (5/6) | Case series | N=50 RET M+ patients from MEN2A families who were <20 years of age at time of thyroidectomy | | RET mutation testing by restriction site polymorphism analysis and/or direct DNA sequencing of RET exons 10, 11, 13, 14 and 16  Total thyroidectomy | Inclusion  RET M+ patients, <20 years of age at time of thyroidectomy and were followed up at least 5 years postoperatively  Exclusion  Older than 19 years of age, or not followed up at 5 years | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Incidence of residual/recurrent disease  Age at time of thyroidectomy  Postoperative calcitonin levels | Range = 5–10 years |
| ([Spinelli et al. 2010](#_ENREF_193))  Italy | IV interventional evidence  High quality (4/6) | Case series | N=13 patients (8–17 years of age) with MEN2 who underwent surgery for MTC:  7 (54%) MEN2A  4 (31%) FMTC  2 (15%) MEN2B | | RET mutation testing by direct DNA sequencing (exons not specified)  Curative or prophylactic total thyroidectomy | Inclusion  Patients with MEN2 who underwent surgery for MTC, ≤17 years of age  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Safety of thyroidectomy | N/A |
| ([Uchino et al. 1999](#_ENREF_204))  Japan | IV interventional evidence  Low quality (2/6) | Case series | N=36 members from 5 MEN2A families with mutations on codon 634  15 were RET M+:  9 clinically affected  6 clinically non-affected | | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11, 13, 14 and 16  Total thyroidectomy | Inclusion  Members of 5 MEN2 kindreds  Exclusion  Not stated | RET mutation status  Incidence of MTC and lymph node metastases  Incidence of PCC  Treatment decisions | Not stated |
| ([Vaclavikova et al. 2009](#_ENREF_205))  Czech Republic | IV interventional evidence  Low quality (2/6) | Case series | N=10 index cases with a RET Y791F mutation:  3 with apparently sporadic MTC 3 with FMTC/MEN2A/MEN2B 1 with PCC 3 with HSCR  N=21 RET M+ family members  18 patients (12 relatives from 4 families and 6 index cases) had total thyroidectomy | | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16  Total thyroidectomy | Inclusion  Families with germline Y791F mutations  Exclusion  Not stated | RET mutation status  Incidence of MMC, CCH, MTC and lymph node metastases  Incidence of PCC  Incidence of HPT  Incidence of residual/recurrent disease  Postoperative calcitonin levels  Age at time of thyroidectomy  Treatment decisions  Mortality | Up to 15 years |
| ([Vestergaard et al. 2007](#_ENREF_206))  Denmark | IV interventional evidence  Moderate Quality (4/6) | Case series | N=27 first-degree family members of index case with a RET Y791F mutation  12 had the RET Y791F mutation | | RET mutation testing by direct DNA sequencing of exon 13  No thyroidectomy | Inclusion  First-degree family members of an index case with FMTC and a RET Y791F mutation  Exclusion  Not stated | RET mutation status  Incidence of PCC  Incidence of HPT  Incidence of abnormal calcitonin tests | Not stated |
| ([Wells Jr & Skinner 1998](#_ENREF_208))  USA | IV interventional evidence  High quality (5/6) | Case series | N=58 first-degree relatives, aged 21 years or younger, from 7 MEN2A kindreds with no clinical symptoms  21 were RET M+  18 underwent a thyroidectomy  8 had elevated pentagastrin-stimulated calcitonin levels | | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 10 and 11  Prophylactic thyroidectomy with bilateral cervicocentral lymphadenectomy | Inclusion  First-degree family members from 7 kindreds at risk of MEN2A  Exclusion  Not stated | RET mutation status  Incidence of CCH, MTC and lymph node metastases  Postoperative calcitonin levels  Safety of thyroidectomy | Range = 0–3 years |
| ([Wu et al. 1998](#_ENREF_214))  Taiwan | IV interventional evidence  Moderate quality (3/6) | Case series | N=28 first- and second-degree relatives from 2 unrelated MEN2A families:  15/17 members of family 1: 10 were RET M+  13/15 members of family 2: 3 were RET M+ | | RET mutation testing by direct DNA sequencing of exons 10 and 11 | Inclusion  Members of 2 unrelated Taiwanese MEN2A kindreds  Exclusion  Not stated | RET mutation status  Incidence of MTC  Incidence of PCC  Incidence of HPT | Not stated |
| ([Yoshida et al. 2009](#_ENREF_215))  Japan | IV interventional evidence  Moderate quality (3/6) | Case series | N=12 adults who underwent total thyroidectomy for MTC and had MEN2  5 were symptomatic  All had raised pentagastrin-stimulated calcitonin levels | | RET mutation testing (method not stated)  Total thyroidectomy, the parathyroid gland was also removed and autotransplanted (unclear whether treatment decisions influenced by RET mutation) | Inclusion  MEN2A patients aged over 25 years who underwent surgery for MTC between 1994 and 2006  Exclusion  Not stated | RET mutation status  Incidence of PCC  Incidence of HPT  Incidence of residual/recurrent disease  Safety of thyroidectomy  Mortality | 1–14 years |

CCH = C-cell hyperplasia; HPT = hyperparathyroidism; MCC = medullary microcarcinoma; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive; RET M– = RET-mutation-negative

Table Study profiles of studies showing diagnostic yield

| Study and location | Level of evidence | Study design | Study population | Intervention |
| --- | --- | --- | --- | --- |
| ([Abdelhakim et al. 2009](#_ENREF_1))  Morocco | IV diagnostic evidence | Diagnostic yield | N=9 index cases with MTC:  2 MEN2A, 1 hereditary MTC, 6 sporadic MTC  N=13 family members (degree not stated) of 3 RET M+ index cases | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 |
| ([Algun et al. 2002](#_ENREF_6))  Turkey | IV diagnostic evidence | Diagnostic yield | N=88 relatives (degree not stated) from 4 generations of an extended MEN2A family with a RET C634G mutation | RET mutation testing by restriction site polymorphism analysis of exon 11 |
| ([Alvandi et al. 2011](#_ENREF_8))  Iran | IV diagnostic evidence | Diagnostic yield | N=49 unrelated index patients diagnosed with MTC and classified as apparently sporadic | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16, and restriction site polymorphism analysis to detect C634R mutation |
| ([Alvares Da Silva et al. 2003](#_ENREF_9))  Brazil | IV diagnostic evidence | Diagnostic yield | N=229 extended family members from 6 generations of an MTC index patient with a RET G533C mutation | RET mutation testing by direct DNA sequencing of exon 8 |
| ([Amar et al. 2005](#_ENREF_10))  France | IV diagnostic evidence | Diagnostic yield | N=258 patients with apparently sporadic phaeochromocytoma or paraganglioma  N=56 patients with a family history | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–6  All the coding exons of SDHB, SDHD, SDHC, and VHL were also sequenced |
| ([Ameur et al. 2009](#_ENREF_11))  France | IV diagnostic evidence | Diagnostic yield | N=46 tissue samples collected from MTC, CCH or mixed MTC patients | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 from normal and diseased tissue samples to determine germline and somatic RET status |
| ([Bar et al. 1997](#_ENREF_12))  Israel | IV diagnostic evidence | Diagnostic yield | N=27 patients diagnosed with sporadic PCC | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10, 11 and 16 (also checked for mutations in VHL) |
| ([Beldjord et al. 1995](#_ENREF_15))  France | IV diagnostic evidence | Diagnostic yield | N=28 patients diagnosed clinically with sporadic PCC | RET mutation testing by denaturing gradient gel electrophoresis analysis confirmed by direct DNA sequencing of exons 10, 11 and 16 |
| ([Bergant et al. 2006](#_ENREF_16))  Slovenia | IV diagnostic evidence | Diagnostic yield | N=69 sporadic MTC patients  N=31 relatives (degree not stated) of RET M+ sporadic MTC patients | RET mutation testing by single-strand conformational analysis and confirmatory direct DNA sequencing of exons 10, 11 and 13–16, or restriction site polymorphism analysis of exon affected in index case |
| ([Bihan et al. 2012](#_ENREF_17))  France | IV diagnostic evidence | Diagnostic yield | N=22 extended family members of an MTC patient confirmed to have a RET L790F mutation on exon 13 | RET mutation testing by direct DNA sequencing of exon 13 |
| ([Blaugrund et al. 1994](#_ENREF_18))  USA | IV diagnostic evidence | Diagnostic yield | N=22 specimens from patients with MTC:  15 apparently sporadic MTC; 3 MEN2A; 1 MEN2B; 3 FMTC | RET mutation testing by DNA sequencing of cloned exons 10, 11 and 16, and Southern blot analysis for genomic rearrangements |
| ([Boer et al. 2003](#_ENREF_19))  Hungary | IV diagnostic evidence | Diagnostic yield | N=65 consecutive patients from 1992–2000 with MTC and no signs of MEN2B who were undergoing genetic screening | RET mutation testing by direct DNA sequencing (exons not specified) |
| ([Bugalho et al. 2007](#_ENREF_22))  Portugal | IV diagnostic evidence | Diagnostic yield | N=77 apparently sporadic cases of MTC  N=65 relatives (degree not stated) of 8 probands of established FMTC/MEN2 kindreds with a RET mutation  53 were asymptomatic | RET mutation testing by direct DNA sequencing of exons 10–16 or restriction site polymorphism analysis of exons 13–16  Exon 8 was screened for gross insertions/deletions (method not stated) |
| ([Bugalho et al. 1997](#_ENREF_23))  Portugal | IV diagnostic evidence | Diagnostic yield | N=13 sporadic MTC  No family history of MTC, PCC or parathyroid disease | RET mutation testing by direct DNA sequencing of exons 10, 11, 13, 15 and 16, confirmed using restriction site polymorphism analysis where appropriate |
| ([Calva et al. 2009](#_ENREF_27))  USA | IV diagnostic evidence | Diagnostic yield | N=31 first- and second-degree family members from a MEN2A kindred with a RET C609Y mutation | RET mutation testing (method not stated) |
| ([Caron et al. 1996](#_ENREF_28))  France | IV diagnostic evidence | Diagnostic yield | N=14 extended family members of a confirmed MEN2A patient with a RET C618R mutation  4 were symptomatic | RET mutation testing by direct DNA sequencing of exons 10 and 11 |
| ([Cascon et al. 2009](#_ENREF_29))  Spain | IV diagnostic evidence | Diagnostic yield | N=237 consecutively enrolled patients identified in Spanish hospital with functioning or non-functioning PCCs and/or paragangliomas  35 had personal or familial history of MEN2  10 had personal or familial history of VHL  24 had other familial syndrome | Complete genetic characterisation of RET, SDHB, SDHC, SDHD and VHL (method not stated) |
| ([Chang et al. 2009](#_ENREF_32))  Taiwan | IV diagnostic evidence | Diagnostic yield | N=69 members from 8 unrelated MTC families:  8 probands and 61 relatives (degree not stated)  N=7 sporadic MTC patients | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16  For sporadic cases RET mutation testing by direct DNA sequencing was extended to includes all exons 1–20 |
| ([Chi et al. 1994](#_ENREF_33))  Japan | IV diagnostic evidence | Diagnostic yield | N=74 relatives (degree not stated) from an extended MEN2A pedigree with a RET C634R mutation:  43 clinically affected; 31 considered at risk | RET mutation testing by restriction site polymorphism analysis of exon 11 |
| ([Chiefari et al. 2001](#_ENREF_34))  Italy | IV diagnostic evidence | Diagnostic yield | N=20 first- and second-degree relatives of proband with RET C634F mutation  6 affected with MTC | RET mutation testing by restriction site polymorphism analysis of exon 11, confirmed by direct DNA sequencing |
| ([Chiefari et al. 1998](#_ENREF_35))  Italy | IV diagnostic evidence | Diagnostic yield | N=10 with sporadic MTC  N=37 members (degree not stated) of 10 separate families with hereditary MTC:  6 MEN2A; 2 MEN2B; 1 FMTC; 1 with <4 MTC cases | RET mutation testing by restriction analysis of exons 11, 13, 15 and 16, and DNA sequencing of exons 10 and 14 |
| ([Chung et al. 2004](#_ENREF_36))  Korea | IV diagnostic evidence | Diagnostic yield | N=33 MTC patients who underwent a thyroidectomy (diagnosed clinically and by histopathology) | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (postoperative) |
| ([De Krijger et al. 2006](#_ENREF_38))  Nederlands | IV diagnostic evidence | Diagnostic yield | N=10 PCC tissue samples | RET mutation testing by denaturing gradient gel electrophoresis analysis confirmed by direct DNA sequencing of exons 10, 11 and 16 (also checked for mutations in SDHB, SDHD and VHL) |
| ([Decker et al. 1995](#_ENREF_40))  USA | IV diagnostic evidence | Diagnostic yield | N=103 consecutive patients at risk for MEN2A/FMTC  93 relatives (degree not stated) from 10 MEN2A or FMTC kindreds  21 patients diagnosed with apparently sporadic MTC | RET mutation testing by denaturing gradient gel electrophoresis mutational analysis of exons 10 and 11, with confirmatory direct DNA sequencing |
| ([Donis-Keller 1995](#_ENREF_45))  USA | IV diagnostic evidence | Diagnostic yield | N=9 sporadic MTC patients  N=8 sporadic PCC patients  N=132 relatives (degree not stated) from 7 MEN2A families | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing in exons 1–20 for index cases  Restriction site polymorphism analysis or direct DNA sequencing for family members |
| ([Dos Santos et al. 2007](#_ENREF_46))  Brazil | IV diagnostic evidence | Diagnostic yield | N=57 at-risk family members (degree not stated) from 7 index cases:  3 MEN2A; 1 MEN2B; 3 FMTC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 in index patients, and restricted to specific exon for family members |
| ([Dourisboure et al. 2005](#_ENREF_47))  Argentina | IV diagnostic evidence | Diagnostic yield | N=22 members from a MEN2B family with a RET C630R mutation:  21 first-degree relatives  1 second-degree relative (grandchild with deceased parent) | RET mutation testing by direct DNA sequencing of exon 11 |
| ([Eisenhofer et al. 2011](#_ENREF_51))  Germany and USA | IV diagnostic evidence | Diagnostic yield | N=173 hereditary PCC and paraganglioma patients:  22 from Europe; 151 from USA | Genetic characterisation of RET, VHL, SDHB, SDHC, and SDHD (method not stated) |
| ([Elisei et al. 2007](#_ENREF_53))  Italy | IV diagnostic evidence | Diagnostic yield | N=37 patients with familial MTC  N=481 apparently sporadic MTC patients submitted for RET screening between 1993 and 2006  N=274 first-degree relatives of patients with confirmed RET mutations | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 |
| ([Eng, Crossey, et al. 1995](#_ENREF_55))  UK and USA | IV diagnostic evidence | Diagnostic yield | N=48 patients with apparently sporadic PCC | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 9, 10, 11 and 13–16 (also checked for mutations in VHL) |
| ([Eng, Mulligan, et al. 1995](#_ENREF_56))  UK | IV diagnostic evidence | Diagnostic yield | N=67 apparently sporadic MTC patients  No history of first- or second-degree family MTC or PCC, no multiple tumours, and MTC confirmed histopathologically | RET mutation testing by direct DNA sequencing of exons 10, 11, 13 and 16 |
| ([Erdogan et al. 2005](#_ENREF_58))  Turkey | IV diagnostic evidence | Diagnostic yield | N=56 apparently sporadic MTC patients Histopathologically and clinically confirmed, with a negative family history of MTC, PCC or HPT | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 |
| ([Erlic et al. 2010](#_ENREF_59))  USA, Spain, Germany, Poland, Finland | IV diagnostic evidence | Diagnostic yield | N=1,475 patients identified on the European-American Phaeochromocytoma-Paraganglioma Registry | Genetic characterisation of RET exons 10, 11 and 13–16 (method not stated), (also checked for mutations in SDHB, SDHC, SDHD and VHL) |
| ([Etit et al. 2008](#_ENREF_61))  USA | IV diagnostic evidence | Diagnostic yield | N=32 patients retrospectively identified from hospital records who had undergone a prophylactic thyroidectomy for MTC  30 with family history: 24 MEN2A; 8 non-MEN | RET mutation testing by direct DNA sequencing analysis of exons 10, 11 and 13 –16 |
| ([Fernandez et al. 2006](#_ENREF_63))  Spain | IV diagnostic evidence | Diagnostic yield | N=27 clinically diagnosed patients with hereditary MTC:  16 MEN2A; 3 MEN2B; 8 FMTC  N=73 sporadic MTC  N=14 clinically diagnosed patients who presented with PCC (N=12) and/or HPT (N=4)  N = 238 family members | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 |
| ([Fink et al. 1996](#_ENREF_65))  Austria | IV diagnostic evidence | Diagnostic yield | N=33 patients clinically diagnosed with FMTC, MEN2A, MEN2B or suspected of inheritable MTC from 16 families:  27 had MTC; 6 had CCH  N=52 asymptomatic relatives from the 13 families  N=59 sporadic MTC patients | RET mutation testing by single-strand conformational analysis or restriction site polymorphism analysis, and confirmatory direct DNA sequencing of exons 10, 11, 13 and 16; if no mutation detected, direct DNA sequencing of exon 14 was conducted |
| ([Fitze et al. 2002](#_ENREF_66))  Germany | IV diagnostic evidence | Diagnostic yield | N=45 patients clinically identified with sporadic MTC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 |
| ([Frank-Raue et al. 1996](#_ENREF_69))  Germany | IV diagnostic evidence | Diagnostic yield | N=159 at-risk family members (degree not stated) from 35 families with hereditary MTC:  111 MEN2A; 31 FMTC; 17 MEN2B | RET mutation testing by single-strand conformation polymorphism analysis, and then either direct DNA sequencing or restriction site polymorphism analysis of exons 10 or 11  Direct DNA sequencing of exons 13 or 16 |
| ([Frilling et al. 1995](#_ENREF_72))  Germany | IV diagnostic evidence | Diagnostic yield | N=56 clinically unaffected first-degree relatives (at 50% risk) from 21 hereditary MTC families:  15 MEN2A; 2 MEN2B; 4 FMTC | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11, and restriction site polymorphism analysis of exon 16 |
| ([Frohnauer et al. 2000](#_ENREF_73))  USA | IV diagnostic evidence | Diagnostic yield | N=38 members (degree not stated) from 5 MEN2A kindreds with a RET codon 804 mutation | RET mutation testing by denaturing gradient gel electrophoresis analysis confirmed by direct DNA sequencing of exon 14 |
| ([Fugazzola et al. 2002](#_ENREF_74))  Italy | IV diagnostic evidence | Diagnostic yield | N=44 members (degree not stated) of a large FMTC pedigree with a RET A891S mutation | RET mutation testing by direct DNA sequencing of exon 15 |
| ([Gagel et al. 1995](#_ENREF_75))  USA | IV diagnostic evidence | Diagnostic yield | N=178 members (degree not stated) from 28 families with MEN2A:  54 first-degree relatives (affected parent); 71 clinically affected with MTC, PCC or HPT; 53 clinically negative | RET mutation testing by direct DNA sequencing of exons 10, 11 or 16 |
| ([Gil et al. 2002](#_ENREF_78))  Spain | IV diagnostic evidence | Diagnostic yield | N=23 members of 4 independent MEN2A families (degree not stated):  13 clinically affected (9 MTC only, 4 MTC + PCC); 10 unaffected | RET mutation testing by single-strand conformation polymorphism analysis and restriction site polymorphism analysis, with confirmatory direct DNA sequencing of exons 10 and 11 |
| ([Gonzalez et al. 2003](#_ENREF_81))  Mexico | IV diagnostic evidence | Diagnostic yield | N=9 proband:  3 MEN2B; 2 MEN2A; 4 sporadic MTC  N=48 family members (degree not stated) of 6 RET M+ probands | RET mutation testing by single-strand conformational polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16, and direct DNA sequencing of exons 13–15 |
| ([Gosnell et al. 2006](#_ENREF_82))  Australia | IV diagnostic evidence | Diagnostic yield | N=48 at-risk family members (degree not stated) from a MEN2A kindred. | RET mutation testing (method not stated). |
| ([Guerrero et al. 2006](#_ENREF_85))  Brazil | IV diagnostic evidence | Diagnostic yield | N=24 unrelated patients with apparently sporadic MTC | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10 and 11, and direct DNA sequencing of exons 13–16 |
| ([Halling et al. 1997](#_ENREF_87))  USA | IV diagnostic evidence | Diagnostic yield | N=72 family members (degree not stated) from 1 large FMTC kindred with a RET C609Y mutation | RET mutation testing by direct DNA sequencing of exon 10 |
| ([Hedayati et al. 2006](#_ENREF_88))  Iran | IV diagnostic evidence | Diagnostic yield | N=57 unrelated index cases with MTC:  1 MEN2A; 1 MEN2B; 2 FMTC; 53 apparently sporadic | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 |
| ([Hernandez et al. 1997](#_ENREF_90))  Spain | IV diagnostic evidence | Diagnostic yield | N=36 asymptomatic members of 3 families with MEN2A | RET mutation testing by direct DNA sequencing and/or restriction site polymorphism analysis of exons 10 and 11 |
| ([Iacobone et al. 2011](#_ENREF_93))  Italy | IV diagnostic evidence | Diagnostic yield | N=59 with apparently sporadic PCC (without evident hereditary disease and/or syndromic appearance) | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16 (also checked for mutations in SDHB, SDHC, SDHD and VHL) |
| ([Januszewicz et al. 2000](#_ENREF_96))  Poland | IV diagnostic evidence | Diagnostic yield | N=77 unselected patients with PCC surgically treated (who responded to invitation; 85 did not respond) | RET mutation testing by single-strand conformation polymorphism analysis confirmed by direct DNA sequencing of exons 10, 11 and 13–16 |
| ([Jindrichova et al. 2004](#_ENREF_97))  Czech Republic | IV diagnostic evidence | Diagnostic yield | N=106 unrelated index cases with MTC:  10 MEN2A; 4 MEN2B; 10 FMTC; 82 sporadic MTC  N=77 relatives(degree not stated) of 23 RET M+ index cases with MTC | RET mutation testing by direct DNA sequencing of exons 10, 11, 13, 14 and/or 16 |
| ([Jung et al. 2010](#_ENREF_98))  Korea | IV diagnostic evidence | Diagnostic yield | N=28 first- and second-degree members (excluding the proband) of a 3-generation FMTC family with a RET C618S mutation | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exon 10 |
| ([Karga et al. 1998](#_ENREF_102))  Greece | IV diagnostic evidence | Diagnostic yield | N=58 members (degree not stated) of 12 unrelated Greek families  9 MEN2A families; 1 FMTC families; 3 likely FMTC families  25 asymptomatic first-degree relatives of patients  33 clinically affected family members | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 and/or 16 |
| ([Kimura et al. 1995](#_ENREF_105))  Japan | IV diagnostic evidence | Diagnostic yield | N=25 specimens from patients:  1 with FMTC; 2 with MEN2A; 4 with MEN2B; 3 with neurofibromatosis type 1; 3 with apparently sporadic MTCs; 12 with sporadic PCCs | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11  (NB test not appropriate to detect mutations for phenotype MEN2B) |
| ([Kinlaw et al. 2005](#_ENREF_107))  USA | IV diagnostic evidence | Diagnostic yield | N=29 first- and second-degree relatives in a family with MEN2A due to RET C609S mutation  6 with manifestations of MEN2A | RET mutation testing by restriction site polymorphism analysis of exon 10 |
| ([Kitamura et al. 1997](#_ENREF_108))  Japan | IV diagnostic evidence | Diagnostic yield | N=33 unrelated MTC patients:  27 clinically sporadic; 6 classified as inherited | RET mutation testing by single-strand conformation polymorphism analysis of exons 10, 11, 13, 14 and 16, followed by direct DNA sequencing of exons 10, 11, 13 and 14, and restriction site polymorphism analysis to detect codon 918 mutation |
| ([Klein et al. 2001](#_ENREF_109))  Hungary | IV diagnostic evidence | Diagnostic yield | N=108 individuals:  65 unrelated index cases with MTC; 43 relatives (degree not stated) of RET M+ index cases | RET mutation testing by restriction site polymorphism analysis of exon 11 and/or direct DNA sequencing of exons 10, 13 and/or 14 |
| ([Komminoth et al. 1995](#_ENREF_110))  Switzerland | IV diagnostic evidence | Diagnostic yield | N=46 specimens from patients with MTC:  22 suspected of being MEN2 or FMTC; 24 apparently sporadic MTC or PCCs  N=38 members (degree not stated) from 3 MEN2A families, 2 MEN2B families, and 4 suspected MEN2 families | RET mutation testing by single-strand conformation polymorphism and heteroduplex gel electrophoresis analysis of exons 10, 11, 13 and/or 15 |
| ([Krawczyk et al. 2010](#_ENREF_112))  Poland | IV diagnostic evidence | Diagnostic yield | N=60 patients with diagnosis of apparently sporadic PCC or paraganglioma  53 had PCC; 8 had paraganglioma; 1 had both a PCC and a paraganglioma  41 were benign tumours; and 11 had malignant lesions | RET mutation testing by direct DNA sequencing of exons 10, 11, 14 and 16 (also checked for mutations in SDHB, SDHD and VHL) |
| ([Learoyd et al. 2005](#_ENREF_116))  Australia and New Zealand | IV diagnostic evidence | Diagnostic yield | N=54 family members (degree not stated) of 2 probands:  47 family members from 4 generations, proband had RET V804L mutation  7 family members from 3 generations, proband had RET V804M mutation | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exons 10, 11 and 16 between 1993 and 1998; then exons 13–15 were included |
| ([Lecube et al. 2002](#_ENREF_119))  Spain | IV diagnostic evidence | Diagnostic yield | N=52 family members (degree not stated) of an FMTC family with a RET V804M mutation | RET mutation testing by restriction site polymorphism analysis of exon 14 |
| ([Lendvai et al. 2012](#_ENREF_120))  Hungary | IV diagnostic evidence | Diagnostic yield | N=95 patients:  47 consecutive patients with apparently sporadic MTCs 48 consecutive patients with apparently sporadic PCC | RET mutation testing by direct DNA sequencing of exons 10, 11 and 14 |
| ([Lindor et al. 1995](#_ENREF_123))  USA | IV diagnostic evidence | Diagnostic yield | N=29 patients who had undergone an operation for a sporadic PCC | RET mutation testing by direct DNA sequencing of exons 10 and 11, and mutation specific PCR for exon 16 |
| ([Mannelli et al. 2009](#_ENREF_131))  Italy | IV diagnostic evidence | Diagnostic yield | N=501 consecutively enrolled patients presenting with PCC or paragangliomas (new or previously identified) | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16, and multiplex ligation-dependent probe amplification assay to detect genomic rearrangements (also checked for mutations in SDHB, SDHC, SDHD and VHL) |
| ([Marsh et al. 1996](#_ENREF_133))  Australia and New Zealand | IV diagnostic evidence | Diagnostic yield | N=39 members (degree not stated) of 16 MEN2A and FMTC families at risk of being a gene carrier | RET mutation testing by restriction site polymorphism analysis of exons 10 and 11 |
| ([Mastroianno et al. 2011](#_ENREF_134))  Italy | IV diagnostic evidence | Diagnostic yield | N=21 first- and second-degree relatives of a proband with MEN1 (MEN1 IVs4+IG>T mutation) and MEN2 (RET K666M mutation on exon 11) | RET mutation testing by of exons 8, 10, 11, 13–16 and 18 (method not stated) |
| ([McMahon et al. 1994](#_ENREF_135))  UK | IV diagnostic evidence | Diagnostic yield | N=63 affected or unaffected first- degree relatives from 9 MEN2A families with mutations in RET codon 634:  29 affected; 30 unaffected; 4 not tested but categorised as non-carriers when parents tested RET M– | RET mutation testing by restriction site polymorphism analysis of codon 634, and confirmatory direct DNA sequencing of exon 11 |
| ([Morita et al. 1996](#_ENREF_137))  Japan | IV diagnostic evidence | Diagnostic yield | N=20 members of one MEN2A family:  6 children of the proband; 10 grandchildren of the proband; 3 great-grandchildren of the proband | RET mutation testing by PCR amplification and restriction enzyme analysis of exon 10 C618S |
| ([Neocleous et al. 2011](#_ENREF_140))  Cyprus | IV diagnostic evidence | Diagnostic yield | N=29 family members from 7 FMTC families and 1 MEN2A family | RET mutation testing by direct DNA sequencing of exon 10 |
| ([Neumann et al. 2002](#_ENREF_141))  Germany and Poland | IV diagnostic evidence | Diagnostic yield | N=271 patients with non-syndromic PCC and/or paragangliomas without family history of disease:  241 had PCCs only; 22 had paragangliomas only; 8 had both a PCC and a paraganglioma | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 13–16 (also checked for mutations in SDHB, SDHD and VHL) |
| ([Neumann et al. 1995](#_ENREF_142))  Germany | IV diagnostic evidence | Diagnostic yield | N=10 families with MEN2A or MEN2B  N=27 family members (degree not stated) from 7 MEN2A families and 1 MEN2B family who had had negative clinical screening (N=19) or unknown phenotype (N=8) | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16; if no RET mutation identified, screened for VHL mutation |
| ([Oriola et al. 1996](#_ENREF_151))  Spain | IV diagnostic evidence | Diagnostic yield | N=59 family members (degree not stated) from 7 MEN2A families:  20 symptomatic; 39 ‘at risk’ | RET mutation testing by direct DNA sequencing of RET exons 10 and 11, and restriction site polymorphism analysis of exons 10 and exon 11 |
| ([Pacini et al. 1995](#_ENREF_152))  Italy | IV diagnostic evidence | Diagnostic yield | N=58 family members (degree not stated) from 9 MEN2 families:  16 affected; 42 at risk of disease | RET mutation testing by restriction site polymorphism analysis of exons 10, 11 or 16 |
| ([Pasini et al. 2002](#_ENREF_154))  Italy | IV diagnostic evidence | Diagnostic yield | N=26 family members (degree not stated) of a patient with Hirschsprung’s disease and MEN2 (RET C618R mutation). | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10, 11, 13, 14 and 16, and restriction site polymorphism analysis of exon 15, with confirmatory direct DNA sequencing of exon 10 in the proband  Restriction site polymorphism analysis and direct DNA sequencing of exon 10 was used to identify RET mutation in family members |
| ([Patocs et al. 2004](#_ENREF_156))  Hungary | IV diagnostic evidence | Diagnostic yield | N=41 patients with PCCs | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11 and 14 (also checked for mutations in VHL) |
| ([Pinna et al. 2007](#_ENREF_159))  Italy | IV diagnostic evidence | Diagnostic yield | N=22 patients with MTC  N=43 first-degree relatives of 7 index cases with RET | RET mutation testing by direct DNA sequencing of exons 8–16 in index case and appropriate exon in family members |
| ([Prazeres et al. 2006](#_ENREF_160))  Portugal | IV diagnostic evidence | Diagnostic yield | N=53 patients with apparently sporadic MTC | RET mutation testing by direct DNA sequencing of exons 8, 10, 11 and 13–16, plus restriction site polymorphism analysis when possible |
| ([Punales et al. 2003](#_ENREF_161))  Brazil | IV diagnostic evidence | Diagnostic yield | N=160 individuals  133 family members plus 17 index cases from 17 MEN2 families  113 family members were from families with MEN2A  54 (37 + 17 index cases) had clinical signs of disease  10 patients with apparently sporadic MTCs | RET mutation testing by single-strand conformation polymorphism analysis, restriction enzyme analysis and direct DNA sequencing of exons 10, 11, 13, 14 or 15 |
| ([Radien et al. 1997](#_ENREF_164))  France | IV diagnostic evidence | Diagnostic yield | N=120 patients with apparently sporadic PCC | RET mutation testing by denaturing gradient gel electrophoresis analysis of exons 10, 11, 13 and 16 |
| ([Romei et al. 2011](#_ENREF_173))  Italy | IV diagnostic evidence | Diagnostic yield | N=729 patients with apparently sporadic MTC (no familial history of MTC or other endocrine disease)  N=146 relatives (degree not stated) of 47 RET M+ index cases who had MTCs without family history of endocrine disorders | RET mutation testing method changed over 15 years  Initially used DNA sequencing of exons 10, 11 and 16; later added exons 13–15; and recently added exons 5 and 8  RET mutation testing of relatives by direct DNA sequencing of exon affected in index case |
| ([Sanso et al. 2002](#_ENREF_178))  Argentina | IV diagnostic evidence | Diagnostic yield | N=98 relatives from 17 MEN2A index patients  N=13 relatives from 5 MEN2B index patients | RET mutation testing by direct DNA sequencing of exons 10, 11 and 16, and confirmed by restriction site polymorphism analysis |
| ([Shan et al. 1998](#_ENREF_183))  Japan – China | IV diagnostic evidence | Diagnostic yield | N=40 patients with apparently sporadic MTCs | RET mutation testing by restriction site polymorphism analysis of codon 918 mutations |
| ([Sharma & Saranath 2011](#_ENREF_184))  India | IV diagnostic evidence | Diagnostic yield | N=51 MTC patients  N=25 first-degree relatives from 7 index cases with family history and a RET mutation | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 for index cases, and specific exons for family members |
| ([Shifrin et al. 2009](#_ENREF_185))  USA | IV diagnostic evidence | Diagnostic yield | N=107 family members from family with V804M mutation | RET mutation testing by direct DNA sequencing of exons 10, 11 and 13–16 (not clear if relatives tested for specific mutation or all) |
| ([Shimotake et al. 1996](#_ENREF_186))  Japan | IV diagnostic evidence | Diagnostic yield | N=37 first-degree relatives in a MEN2 family with a RET C634R mutation  6 without clinical signs | RET mutation testing by restriction site polymorphism analysis and direct DNA sequencing of exon 11 |
| ([Shirahama et al. 1998](#_ENREF_187))  Japan | IV diagnostic evidence | Diagnostic yield | N=71 patients with MTC:  44 from MEN2 or FMTC families; 22 apparently sporadic MTCs; 5 patients without familial information | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10 and 11; if no mutations found, then restriction site polymorphism analysis of codons 768 and 918 |
| ([Siggelkow et al. 2001](#_ENREF_189))  Germany | IV diagnostic evidence | Diagnostic yield | N=34 first- and second-degree relatives of an index case with FMTC and a RET C611F mutation:  8/14 at risk members of the 3rd generation  17/17 at risk members of the 4th generation  9/15 at risk members of the 5th generation | RET mutation testing by direct DNA sequencing of exon 10 to identify mutation in proband, and restriction site polymorphism analysis of exon 10 in family members |
| ([Tsai et al. 1994](#_ENREF_202))  USA | IV diagnostic evidence | Diagnostic yield | N=109 members (degree not stated) of 13 kindreds:  9 MEN2A; 2 MEN2B; 2 FMTC  47 clinically affected, 62 non-affected | RET mutation testing by direct DNA sequencing of exons 10 and 11 |
| ([Uchino et al. 1999](#_ENREF_204))  Japan | IV diagnostic evidence | Diagnostic yield | N=27 members (degree not stated) from 5 MEN2A families whose clinical status was unknown | RET mutation testing by single-strand conformation polymorphism analysis and direct DNA sequencing of exons 10, 11, 13, 14 or 16 |
| ([Uchino et al. 1998](#_ENREF_203))  Japan | IV diagnostic evidence | Diagnostic yield | N=40 patients of apparently sporadic MTCs who had surgery between 1965 and 1996 | RET mutation testing by direct DNA sequencing of exons 10, 11, 13, 14 and 16, and confirmed with restriction site polymorphism analysis |
| ([Vestergaard et al. 2007](#_ENREF_206))  Denmark | IV diagnostic evidence | Diagnostic yield | N=27 first-degree family members (children of RET M+ patients) from a large kindred with a RET Y791F mutation | RET mutation testing by direct DNA sequencing of exon 13 |
| ([Wells Jr & Skinner 1998](#_ENREF_208))  USA | IV diagnostic evidence | Diagnostic yield | N=58 first-degree family members from 7 kindreds with MEN2A, showing no clinical signs/symptoms | RET mutation testing by restriction site polymorphism analysis or direct DNA sequencing of exons 10 and 11 |
| ([Woodward et al. 1997](#_ENREF_213))  UK | IV diagnostic evidence | Diagnostic yield | N=16 kindreds with familial PCC | RET mutation testing of exons 10 and 11 (method not stated) (also checked for mutations in GDNF and VHL) |
| ([Wu et al. 1998](#_ENREF_214))  Taiwan | IV diagnostic evidence | Diagnostic yield | N=26 first- and second-degree relatives of 2 probands from 2 unrelated MEN2A families | RET mutation testing by direct DNA sequencing of exons 10 and 11 |

CCH = C-cell hyperplasia; HPT = hyperparathyroidism; MTC = medullary thyroid cancer; PCC = phaeochromocytoma; RET M+ = RET-mutation-positive

Table 100 Study profiles of studies showing change in management

| Study and location | Level of evidence and quality assessment | Study design | Study population | Intervention | Comparator | Inclusion/exclusion criteria | Outcomes assessed | Duration of follow-up |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ([Spinelli et al. 2010](#_ENREF_193))  Italy | III-2 interventional evidence  High risk of bias (16/26) | Cohort study | N=13 patients (8–17 years of age) with MEN2 who underwent surgery for MTC:  7 (54%) MEN2A  4 (31%) FMTC  2 (15%) MEN2B | Prophylactic thyroidectomy performed on clinically asymptomatic patients, basal calcitonin <100 pg/mL with pentagastrin-stimulated calcitonin test lower than 250 pg/mL | Total thyroidectomy based on clinical signs (thyroid nodulation, altered basal calcitonin >100 pg/mL, pentagastrin-stimulated calcitonin test >250 pg/mL, characteristic phenotype (in MEN2B)) | Inclusion  Patients with MEN2 who underwent surgery for MTC, aged ≤17 years | Incidence and severity of MTC  Safety of thyroidectomy | Mean = 8.6 years  (range 1.5–15 years) |

MTC = medullary thyroid cancer

# Appendix Excluded studies

#### Data could not be extracted:

Eisenhofer, G, Lenders, JWM et al. 1999, 'Plasma normetanephrine and metanephrine for detecting pheochromocytoma in von Hippel-Lindau disease and multiple endocrine neoplasia type 2', *New England Journal of Medicine,*vol. 340, no. 24, pp. 1872–1879.

Heshmati, HM, Gharib, H et al. 1997, 'Genetic testing in medullary thyroid carcinoma syndromes: mutation types and clinical significance', *Mayo Clinic Proceedings,*vol. 72,no. 5, pp. 430–436.

International *RET* mutation consortium 2006, 'The relationship between specific RET proto-oncogene mutations and disease phenotype in multiple endocrine neoplasia type 2', *JAMA: Journal of the American Medical Association,*vol. 276,no. 19, pp. 1575–1579.

Kaldrymides, P, Mytakidis, N et al. 2006, 'A rare RET gene exon 8 mutation is found in two Greek kindreds with familial medullary thyroid carcinoma: implications for screening', *Clinical Endocrinology,*vol. 64,no. 5, pp. 561–566.

Lips, CJ, Landsvater, RM et al. 1995, 'From medical history and biochemical tests to presymptomatic treatment in a large MEN2A family', *Journal of Internal Medicine,*vol. 238,no. 4, pp. 347–356.

Machens, A & Dralle, H 2008, 'Familial prevalence and age of RET germline mutations: implications for screening', *Clinical Endocrinology,*vol. 69,no. 1, pp. 81–87.

Moore, SW & Zaahl, MG 2010, 'Chasing the ubiquitous RET proto-oncogene in South African MEN2 families: implications for the surgeon', *South African Journal of Surgery,*vol. 48,no. 4, pp. 127–131.

Rodriguez, JM, Balsalobre, M et al. 2008, 'Pheochromocytoma in MEN2A syndrome: study of 54 patients', *World Journal of Surgery,* vol.32,no. 11, pp. 2520–2526.

Santos, MAC, Quedas, E et al. 2007, 'Screening of RET gene mutations in multiple endocrine neoplasia type-2 using conformation sensitive gel electrophoresis (CSGE)', *Arquivos Brasileiros de Endocrinologia e Metabologia,*vol. 51,no. 9, pp. 1468–1476.

#### Unable to be located for analysis:

Correia, MJ, Lopes, LO et al. 2000, 'Multiple endocrine neoplasia type 2A: study of a family', *Revista Portuguesa de Cardiologia,*vol. 19,no. 1, pp. 11–31.

Gagel, RF & Cote, GJ 1996, ‘Ret proto-oncogene mutations in multiple endocrine neoplasia type 2’*,* in: JP Bilezikian, LG Raisz & GA Rodan (eds), *Principles of bone biology*. *‘*San Diego, Academic Press, pp. 799–807.

Gonzalez-Yebra, B, Medrano, ME et al. 2003, 'Penetrance of inherited medullary thyroid carcinoma and genotype-phenotype correlation in a large multiple endocrine neoplasia type 2A family with C634Y RET mutation', *Endocrine Pathology,*vol. 14,no. 1, pp. 71–80.

Hatzl-Griesenhofer, M, Pichler, R et al. 2002, 'Results of calcitonin screening in a central upper Austrian region', *Journal of Endocrine Genetics,*vol. 3,no. 2, pp. 75–85.

Palasi, R, Orellana, C et al. 1998, 'Analysis of the ret proto-oncogene in families with multiple endocrine neoplasia type 2A', *British Journal of Surgery,*vol. 85, pp. 60-60.

Robledo, M, Cebrian, A et al. 1998, 'Genetic analysis of the proto-oncogene RET in 41 cases with', *European Journal of Human Genetics,*vol. 6, pp. 131–131.

Vestergaard, P, Kroustrup, JP et al. 1999, 'Neuromas in multiple endocrine neoplasia type 2A with a RET codon 611 mutation', *Journal of Endocrine Genetics,*vol. 1,no. 1, pp. 33–37.

Zehnbauer, BA, Baty, BJ et al. 1998, 'A new mutation in the RET proto-oncogene associated with multiple endocrine neoplasia type 2A', *American Journal of Pathology,*vol. 153,no. 5, pp. 1661–1661.

#### Duplicated articles or data

Aubert-Petit, G, Baudin, E et al. 1999, 'Neuroendocrine tumors and von Hipple-Lindau disease: three cases', *Presse Medicale,*vol. 28,no. 23, pp. 1231–1234.

Baudin, E, Travagli, JP & Schlumberger, M 2006, 'How effective is prophylactic thyroidectomy in asymptomatic multiple endocrine neoplasia type 2A?', *Nature Clinical Practice Endocrinology and Metabolism,*vol. 2,no. 5, pp. 256–257.

Erdogan, MF & Gursoy, A 2006, 'Multiple endocrine neoplasia type 2 and sporadic medullary thyroid carcinoma: Turkish experience', *Pediatric Endocrinology Reviews,*vol. 3,(suppl. 3), pp. 503–507.

Fabien, N, Paulin, C et al. 1994, 'The RET proto-oncogene is expressed in normal human parafollicular thyroid cells', *International Journal of Oncology,* vol.4,no. 3, pp. 623–626.

Hofstra, RMW, Landsvater, RM et al. 1994, 'A mutation in the RET proto-oncogene associated with multiple endocrine neoplasia type 2B and sporadic medullary thyroid carcinoma', *Nature,*vol. 367,no. 6461, pp. 375–376.

Hoppner, W, Frank-Raue, K & Raue, F 1996, *Mutations in the RET proto-oncogene in German families with multiple endocrine neoplasia type 2,* Hereditary Cancer.

Komminoth, P, Kunz, EK et al. 1995, 'Analysis of RET proto-oncogene point mutations distinguishes heritable from nonheritable medullary thyroid carcinomas', *Cancer,*vol. 76,no. 3, pp. 479–489.

Neocleous, V, Passalaris, T et al. 2004, 'Description of the first two seemingly unrelated Greek Cypriot families with a common C618R RET proto-oncogene mutation', *Genetic Testing,*vol. 8,no. 2, pp. 163–168.

Wells Jr, SA, Chi, DD et al. 1994, 'Predictive DNA testing and prophylactic thyroidectomy in patients at risk for multiple endocrine neoplasia type 2A', *Annals of Surgery,*vol. 220,no. 3, pp. 237–250.

Yulin, Z, Yongju, Z et al. 2007, 'RET proto-oncogene mutations are restricted to codons 634 and 918 in mainland Chinese families with MEN2A and MEN2B', *Clinical Endocrinology,*vol. 67,no. 4, pp. 570–576.

Yun, JC, Kim, HH et al. 2004, 'RET proto-oncogene mutations are restricted to codon 634 and 618 in Korean families with multiple endocrine neoplasia 2A', *Thyroid,*vol. 14,no. 10, pp. 813–818.

#### Total sample size for diagnostic yield of less than 20, or less than 10 for health outcomes

Bartsch, DK, Hasse, C et al. 2000, 'A RET double mutation in the germline of a kindred with FMTC', *Experimental and Clinical Endocrinology and Diabetes,*vol. 108,no. 2, pp. 128–132.

Boedeker, CC, Erlic, Z, et al. 2009, 'Head and neck paragangliomas in von Hippel-Lindau disease and multiple endocrine neoplasia type 2', Journal of Clinical Endocrinology and Metabolism, vol. 94, no. 6, pp. 1938-1944.

D'Aloiso, L, Carlomagno, F et al. 2006, 'In vivo and in vitro characterization of a novel germline RET mutation associated with low-penetrant nonaggressive familial medullary thyroid carcinoma', *Journal of Clinical Endocrinology & Metabolism,*vol. 91,no. 3, pp. 754–759.

Gul, K, Ozdemir, D et al. 2010, 'Coexistent familial nonmultiple endocrine neoplasia medullary thyroid carcinoma and papillary thyroid carcinoma associated with RET polymorphism', *American Journal of the Medical Sciences,* vol.340,no. 1, pp. 60–63.

Haecker, FM, Oertli, D et al. 2003, 'Multiple endocrine neoplasias type 2A and thyroid medullary carcinoma: an interdisciplinary challenge', *Pediatric Surgery International,*vol. 19,no. 1–2, pp. 62–64.

Hammond, PJ, Murphy, D et al. 2010, 'Childhood phaeochromocytoma and paraganglioma: 100% incidence of genetic mutations and 100% survival', *Journal of Pediatric Surgery,*vol. 45,no. 2, pp. 383–386.

Hansen, HS, Torring, H et al. 2000, 'Is thyroidectomy necessary in RET mutations carriers of the familial medullary thyroid carcinoma syndrome?', *Cancer,*vol. 89,no. 4, pp. 863–867.

Huang, CN, Wu, SL et al. 1998, 'RET proto-oncogene mutations in patients with apparently sporadic medullary thyroid carcinoma', *Journal of the Formosan Medical Association,*vol. 97,no. 8, pp. 541–546.

Jackson, MB, Guttenberg, M et al. 2005, 'Multiple endocrine neoplasia type 2A in a kindred with C634Y mutation', *Pediatrics,*vol. 116,no. 3, pp. e468–e471.

Jacobs, JM & Hawes, MJ 2001, 'From eyelid bumps to thyroid lumps: report of a MEN type IIb family and review of the literature', *Ophthalmic Plastic and Reconstructive Surgery,*vol. 17, no. 3, pp. 195–201.

Jasim, S, Ying, AK et al. 2011, 'Multiple endocrine neoplasia type 2B with a RET proto-oncogene A883F mutation displays a more indolent form of medullary thyroid carcinoma compared with a RET M918T mutation', *Thyroid,*vol. 21, no. 2, pp. 189–192.

Kawasaki, T, Tomita, Y et al. 1997, 'Absence of RET proto-oncogene mutations in a father and son with pheochromocytoma and pancreatic islet cell tumor', *International Journal of Urology,*vol. 4,no. 2, pp. 169–171.

Krueger, JE, Maitra, A & Albores-Saavedra, J 2000, 'Inherited medullary microcarcinoma of the thyroid: a study of 11 cases', *American Journal of Surgical Pathology,*vol. 24,no. 6, pp. 853–858.

Landsvater, RM, Rombouts, AGM et al. 1993, 'The clinical implications of a positive calcitonin test for C-cell hyperplasia in genetically unaffected members of an men2a kindred', *American Journal of Human Genetics,*vol. 52,no. 2, pp. 335–342.

Lee, MS, Hwang, DY et al. 1998, 'Mutations of ret proto-oncogene in 3 Korean families with MEN2A: clinical use of new restriction sites for genetic diagnosis', *Endocrine Journal,*vol. 45,no. 4, pp. 555–561.

Magalhaes, PKR, De Castro, M et al. 2004, 'Polymorphisms in the RET proto-oncogene and the phenotypic presentation of familial medullary thyroid carcinoma', *Thyroid,*vol. 14,no. 10, pp. 848–852.

Nakao, A, Naomoto, Y et al. 2001, 'A family of multiple endocrine neoplasia type 2A with the RET proto-oncogene mutation in codon 618 (Cys->Arg)', *Japanese Journal of Clinical Oncology,*vol. 31,no. 4, pp. 157–161.

Nunes, AB, Ezabella, MCL et al. 2002, 'A novel Val648Ile substitution in RET proto-oncogene observed in a Cys634Arg multiple endocrine neoplasia type 2A kindred presenting with an adrenocorticotropin-producing pheochromocytoma', *Journal of Clinical Endocrinology & Metabolism,* vol.87,no. 12, pp. 5658–5661.

Oishi, S, Sato, T et al. 1995, 'Mutations of the RET proto-oncogene in multiple endocrine neoplasia type 2A (Sipple's syndrome)', *Endocrine Journal,*vol. 42,no. 4, pp. 527–536.

Okada, Y, Suchi, M et al. 1999, 'Noncardiogenic pulmonary edema as the chief manifestation of a pheochromocytoma: a case report of MEN2A with pedigree analysis of the RET proto-oncogene', *The Tohoku Journal of Experimental Medicine,*vol. 188,no. 2, pp. 177–187.

O'Keeffe, DA, Hill, ADK et al. 1998, 'RET M-proto-oncogene analysis in medullary thyroid carcinoma', *Irish Journal of Medical Science,*vol. 167,no. 4, pp. 226–230.

Oriordain, DS, Obrien, T et al. 1995, 'Multiple endocrine neoplasia type 2B: more than an endocrine disorder', *Surgery,*vol. 118,no. 6, pp. 936–942.

Pazaitou-Panayiotou, K, Giatzakis, C et al. 2010, 'Identification of two novel mutations in the RET proto-oncogene in the same family', *Thyroid,*vol. 20,no. 4, pp. 401–406.

Peacock, ML, Borst, MJ et al. 1996, 'Detection of RET mutations in multiple endocrine neoplasia type 2A and familial medullary thyroid carcinoma by denaturing gradient gel electrophoresis', *Human Mutation,*vol. 7,no. 2, pp. 100–104.

Pegoraro, RJ, Hacking, DJ et al. 1998, 'Molecular diagnosis multiple endocrine neoplasia type 2A', *South African Medical Journal,*vol. 88,no. 1, pp. 39–42.

Peppa, M, Boutati, E et al. 2008, 'Multiple endocrine neoplasia type 2A in two families with the familial medullary thyroid carcinoma associated G533C mutation of the RET proto-oncogene', *European Journal of Endocrinology,*vol. 159,no. 6, pp. 767–771.

Pigny, P, Bauters, C et al. 1999, 'A novel 9-base pair duplication in RET exon 8 in familial medullary thyroid carcinoma', *Journal of Clinical Endocrinology & Metabolism,*vol. 84,no. 5, pp. 1700–1704.

Piolat, C, Dyon, JF et al. 2006, 'Very early prophylactic thyroid surgery for infants with a mutation of the RET proto-oncogene at codon 634: evaluation of the implementation of international guidelines for MEN type 2 in a single centre', *Clinical Endocrinology,*vol. 65,no. 1, pp. 118–124.

Radian, S, Badiu, C et al. 2007, 'Molecular diagnosis of multiple endocrine neoplasia (MEN) type 2A: implementation of mutation detection in RET oncogene and challenges in the management of affected individuals', *Acta Endocrinologica-Bucharest,*vol. 3,no. 1, pp. 13–22.

Ritter, MM, Frilling, A et al. 1996, 'Isolated familial pheochromocytoma as a variant of von Hippel-Lindau disease', *Journal of Clinical Endocrinology & Metabolism,*vol. 81,no. 3, pp. 1035–1037.

Schulten, HJ, Al-Maghrabi, J et al. 2011, 'Mutational screening of RET, HRAS, KRAS, NRAS, BRAF, AKT1, and CTNNB1 in medullary thyroid carcinoma', *Anticancer Research,*vol. 31,no. 12, pp. 4179–4183.

Shaha, AR, Cohen, T et al. 2006, 'Late-onset medullary carcinoma of the thyroid: need for genetic testing and prophylactic thyroidectomy in adult family members', *Laryngoscope,*vol. 116, no. 9, pp. 1704–1707.

Shannon, KE, Gimm, O et al. 1999, 'Germline V804M mutation in the RET proto-oncogene in two apparently sporadic cases of MTC presenting in the seventh decade of life', *Journal of Endocrine Genetics,*vol. 1,no. 1, pp. 39–45.

Simon, S, Pavel, M et al. 2002, 'Multiple endocrine neoplasia 2A syndrome: surgical management', *Journal of Pediatric Surgery,*vol. 37,no. 6, pp. 897–900.

Smith, VV, Eng, E & Milla, PJ 1999, 'Intestinal ganglioneuromatosis and multiple endocrine neoplasia type 2B: implications for treatment', *Gut,*vol. 45,no. 1, pp. 143–146.

Takaya, K, Yoshimasa, T et al. 1996, 'The RET proto-oncogene in sporadic pheochromocytomas', *Internal Medicine (Tokyo, Japan),*vol. 35,no. 6, pp. 449–452.

Toogood, AA, Eng, C et al. 1995, 'No mutation at codon 918 of the RET gene in a family with multiple endocrine neoplasia type 2B', *Clinical Endocrinology,*vol. 43,no. 6, pp. 759–762.

Van Heurn, LWE, Schaap, C et al. 1999, 'Predictive DNA testing for multiple endocrine neoplasia 2: a therapeutic challenge of prophylactic thyroidectomy in very young children', *Journal of Pediatric Surgery,*vol. 34, no. 4, pp. 568–571.

Vandenbosch, K, Renard, M et al. 2005, 'Medullary thyroid carcinoma in a child with a new RET mutation and a RET polymorphism', *Genetic Counseling,*vol. 16, no.1, pp. 95–100.

Vieira, AEF, Mello, MP et al. 2002, 'Molecular and biochemical screening for the diagnosis and management of medullary thyroid carcinoma in multiple endocrine neoplasia type 2A', *Hormone and Metabolic Research,* vol.34,no. 4, pp. 202–206.

Wundrack, I, Reichert, J et al. 1996, 'Molecular screening for RET proto-oncogene mutations in a German MEN2A pedigree', *Endocrine Pathology,*vol. 7,no. 1, pp. 71–76.

Yao, B, Liu, X et al. 2009, 'A novel mutation (D631 del) of the RET gene was associated with MEN2A in a Chinese pedigree', *Endocrine Journal,*vol. 56,no. 1, pp. 99–104.

Yoshimoto, K, Kimura, T et al. 1996, 'Absence of mutations at codon 768 of the RET proto-oncogene in sporadic and hereditary pheochromocytomas', *Endocrine Journal,*vol. 43,no. 1, pp. 109–114.

Zenaty, D, Aigrain, Y et al. 2009, 'Medullary thyroid carcinoma identified within the first year of life in children with hereditary multiple endocrine neoplasia type 2A (codon 634) and 2B', *European Journal of Endocrinology,*vol. 160,no. 5, pp. 807–813.

Zirie, M, Mohammed, I et al. 2001, 'Multiple endocrine neoplasia type IIA: report of a family with a study of three generations in Qatar', *Endocrine Practice,*vol. 7,no. 1, pp. 19–27.

#### Foreign language articles:

Alvandi, E, Pedram, M et al. 2007, 'Detection of RET proto-oncogene Cys634Arg mutation, the cause of medullary thyroid carcinoma, in an Iranian child', *Iranian Journal of Pediatrics,*vol. 17, pp. 301–305.

Arias Miranda, IM, Casal Alvarez, F & Castano, L 2005, 'Multiple endocrine neoplasia 2A type: a genetic study of one family', *Medicina Clinica,*vol. 124,no. 3, p. 116.

Bahlo, M, Schott, M et al. 2008, 'Multiple endocrine neoplasia 2A: newly discovered mutation as a late manifestation', *Deutsche Medizinische Wochenschrift,*vol. 133,no. 10, pp. 464–466.

Belli, S, Storani, ME et al. 2003, 'Analysis of the RET proto-oncogene in multiple endocrine neoplasia 2A and in familial medullary thyroid carcinoma: clinical-pathological findings in asymptomatic carriers', *Medicina,*vol. 63,no. 1, pp. 41–45.

Benazzouz, B, Chraibi, A et al. 2006, 'Characterization of RET proto-oncogene C634Y mutation in a Moroccan family with multiple endocrine neoplasia type 2A', *Annales d'Endocrinologie,*vol. 67,no. 1, pp. 21–26.

Benazzouz, B, Hafidi, A et al. 2008, 'C634R mutation of the proto-oncogene RET and molecular diagnosis in multiple endocrine neoplasia type 2 in a large Moroccan family', *Bulletin du Cancer,*vol. 95,no. 4, pp. 457–463.

Biarnes, J, Miranda, M et al. 1996, 'The molecular pathology of RET proto-oncogene in families with multiple endocrine neoplasm type 2A', *Medicina Clinica,*vol. 107,no. 9, pp. 321–325.

Calbo, L, Spinelli, C et al. 1998, 'Surgical management of hereditary medullary carcinoma of the thyroid in patients with RET proto-oncogene mutation', *Chirurgia Italiana,*vol. 50,no. 5–6, pp. 47–51.

Canizo, A, Fanjul, M et al. 2008, 'Is immediate prophylactic thyroidectomy indispensable in familiar medullary thyroid carcinoma?', *Cirugía Pediátrica: organo oficial de la Sociedad Española de Cirugía Pediátrica,*vol. 21,no. 2, pp. 100–103.

Carreno Villareal, M, Girbes, J et al. 2001, 'Utility of the ret proto-oncogene in the diagnosis of the hereditary medullary thyroid carcinoma: correlation with the surgical results', *Acta Otorrinolaringologica Espanola,*vol. 52,no. 1, pp. 57–63.

Czetwertynska, M, Kozlowicz-Gudzinska, I et al. 2006, 'Clinical and genetic profile of patients with medullary thyroid cancer treated in the Cancer Centre – Institute of Oncology in Warsaw', *Endokrynologia Polska,*vol. 57,no. 4, pp. 415–419.

Dos Santos, MACG, Nunes, AB et al. 2006, 'Genetic screening of multiple endocrine neoplasia type 2: experience of the USP endocrine genetics unit', *Arquivos Brasileiros de Endocrinologia e Metabologia,*vol. 50,no. 1, pp. 7–16.

Dralle, H 2012, 'Subtotal adrenalectomy due to MEN2 pheochromocytoma', *Chirurg,*vol. 83,no. 6, pp. 572–572.

Elenkova, A, Jeunemaitre, X et al. 2010, 'Genotype-phenotype correlations in familial pheochromocytomas', *Endokrinologya,*vol. 15,no. 3, pp. 132–141.

Engelbach, M, Kunt, T et al. 2000, 'Predictive molecular genetics in the diagnosis and treatment of families with multiple endocrine neoplasia type 2', *Deutsche Medizinische Wochenschrift,*vol. 125,no. 3, pp. 37–44.

Fitze, G, Saeger, HD et al. 2004, 'Management of multiple endocrine neoplasia syndrome type 2 families in association with rare germline mutations of the RET proto-oncogene', *Klinische Padiatrie,*vol. 216,no. 5, pp. 270–276.

Frank-Raue, K, Heimbach, C et al. 2003, 'Genotype-phenotype correlation in hereditary medullary thyroid carcinoma', *Deutsche Medizinische Wochenschrift,*vol. 128,no. 39, pp. 1998–2002.

Frank-Raue, K, Hoppner, W et al. 1997, 'Mutation of reT proto-oncogene in medullary thyroid carcinoma', *Deutsche Medizinische Wochenschrift,*vol. 122,no. 6, pp. 143–149.

Frayssinet, C, Vezzosi, D et al. 2008, 'Retroperitoneal laparoscopic adrenalectomy in a pregnant woman presenting MEN2a with a pheochromocytoma: case report and review of the literature', *Annales d'Endocrinologie,*vol. 69,no. 1, pp. 53–57.

Gagel, RF 1997, 'Unresolved issues in the genesis and management of multiple endocrine neoplasia type 2', *Hormone and Metabolic Research,*vol. 29, no. 3, pp. 135–137.

Gomez, FJP, Espinosa, MJB et al. 1999, 'Genetic analysis of RET mutations in MEN2 families from the community of Murcia, Spain', *Medicina Clinica,*vol. 112,no. 17, pp. 646–650.

Hashizume, K, Suzuki, S et al. 2006, 'Multiple endocrine neoplasia type IIa (MEN2A)', *Nippon Rinsho – Japanese Journal of Clinical Medicine,*suppl. 3, pp. 337–342.

Hoie, J, Heimdal, K et al. 2000, 'Prophylactic thyroidectomy in carriers of RET oncogene mutation carriers', *Tidsskrift for den Norske Laegeforening,*vol. 120,no. 27, pp. 3249–3252.

Ishizu, K, Shiraishi, K et al. 1999, 'A case of multiple endocrine neoplasia type 2A (MEN2A) with a mutation in the RET gene', *Hinyokika Kiyo – Acta Urologica Japonica,*vol. 45,no. 6, pp. 407–410.

Ito, T, Shirahama, S et al. 2004, 'The RET gene in multiple endocrine neoplasia type 2 (MEN2)', *Nippon Rinsho – Japanese Journal of Clinical Medicine,*vol. 62,no. 5, pp. 883–888.

Jiang, C-x, Zeng, Z et al. 2011, 'Pheochromocytomas in adrenal medulla or extra-adrenal and multiple endocrine neoplasms: a clinicopathologic analysis of 181 cases', *Zhonghua Bing Li Xue Za Zhi – Chinese Journal of Pathology,*vol. 40,no. 11, pp. 762–766.

Komminoth, P, Muletta-Feurer, S et al. 1996, 'Detection of RET proto-oncogene mutation in the diagnosis of multiple endocrine neoplasia type 2 (MEN2)', *Schweizerische Medizinische Wochenschrift,*vol. 126,no. 31–32, pp. 1329–1338.

Kuznetsov, NS, Bel'tsevich, DG et al. 2002, 'Diagnosis and treatment of syndrome of multiple endocrine neoplasia type 2', *Khirurgiia,* vol. 2, pp. 4–9.

Mazura, I, Vcelak, J et al. 1996, 'Ret proto-oncogene mutation found in the Czech population and its predictive value for offspring of patients with medullary carcinoma of the thyroid gland', *Vnitrni Lekarstvi,*vol. 42,no. 11, pp. 751–756.

Menon, MM & Simha, MR 2005, 'RET mutation status in medullary thyroid cancer (MTC) patients and the significance of genetic screening for mutations in their immediate relatives: a preliminary report', *Indian Journal of Pathology and Microbiology,*vol. 48,no. 2, pp. 161–165.

Miranda, IMA, Alvarez, FC & Castano, L 2005, 'Multiple endocrine neoplasia 2A type: a genetic study of one family', *Medicina Clinica,*vol. 124,no. 3, pp. 116–116.

Morimoto, I 1995, 'Multiple endocrine neoplasia', *Nippon Rinsho – Japanese Journal of Clinical Medicine,*vol. 53,no. 4, pp. 899–903.

Ning, Z-w, Wang, O et al. 2006, 'RET gene Cys634Trp mutation in a multiple endocrine neoplasia type 2A kindred', *Zhongguo Yi Xue Ke Xue Yuan Xue Bao – Acta Academiae Medicinae Sinicae,*vol. 28,no. 6, pp. 799–802.

Obara, T, Yamashita, T et al. 1995, 'Multiple endocrine neoplasia type 2A, type 2B and familial medullary thyroid carcinoma syndrome', *Nippon Rinsho – Japanese Journal of Clinical Medicine,*vol. 53, no. 11, pp. 2708–2715.

Peczkowska, M, Januszewicz, A et al. 2002, 'RET proto-oncogene germline mutation in pheochromocytoma patients: incidence and clinical consequences', *Nadcisnienie Tetnicze,*vol. 6,no. 4, pp. 279–284.

Pomares Gomez, FJ, Bernabe Espinosa, MJ et al. 1999, 'Genetic analysis of RET mutations in families with multiple endocrine neoplasia type II in the community of Murcia', *Medicina Clinica,*vol. 112,no. 17, pp. 646–650.

Qi, Y & Wang, WQ 2010, 'Genetic screening in pheochromocytoma and paraganglioma', *Journal of Shanghai Jiaotong University (Medical Science),*vol. 30no. 5, pp. 503–507.

Rafecas, A, Ribas, Y et al. 1998, 'Usefulness of the genetic screening in the diagnosis of medullary thyroid carcinoma', *Medicina Clinica,*vol. 111,no. 16, pp. 619–622.

Raffel, A, Cupisti, K et al. 2005, 'Decision making in postoperative incidentally found small C-cell-carcinoma', *Zentralblatt fur Chirurgie,*vol. 130,no. 5, pp. 434–439.

Ramos SC, Ojeda SD et al. 2005, 'Prophylactic thyroidectomy in children and young people with hereditary medullary thyroid carcinoma: a Chilean experience', *Revista Medica de Chile,*vol. 133,no. 9, pp. 1029–1036.

Raue, F, Geiger, S et al. 1993, 'Prognostic-significance of calcitonin screening in familial medullary thyroid carcinoma', *Deutsche Medizinische Wochenschrift,*vol. 118,no. 3, pp. 49–52.

Real, S, Gomez, L et al. 2005, 'Detection of a non-standard mutation in the ret proto-oncogene by site directed mutagenesis', *Medicina,*vol. 65,no. 1, pp. 41–46.

Roeher, HD, Simon, D et al. 1995, 'Prophylactic radical operation of C-cell carcinoma in MEN-II syndrome on the basis of genetic screening', vol. *Chirurg,* vol.66,no. 12, pp. 1196–1202.

Rumyantseva, UV, Rumyantsev, PO et al. 2007, 'Diagnosis and treatment policy in familial thyroid cancer', *Problemy Endokrinologii,*vol. 53, no. 4, pp. 32–37.

Sanso, G, Domene, HM et al. 1998, 'Early diagnosis of multiple endocrine neoplasia type 2 (MEN2) by detection of mutated RET proto-oncogene carriers', *Medicina,*vol. 58,no. 2, pp. 179–184.

Santos, MAC, Nunes, AB et al. 2006, 'Genetic screening of multiple endocrine neoplasia type 2: experience of the USP Endocrine Genetics Unit', *Arquivos Brasileiros de Endocrinologia e Metabologia,*vol. 50,no. 1, pp. 7–16.

Schafer, K, Senninger, N et al. 1996, 'Operative strategy in children and adolescents with medullary carcinoma of the thyroid (MTC) including multiple endocrine neoplasia (MEN) type II', *Langenbeck’s Archiv fur Chirurgie*, pp. 202–204.

Schuffenecker, I, Chambe, B & Lenoir, G 1996, 'Analysis of the RET gene and medullary cancer of the thyroid: contribution to the diagnosis and treatment', *Annales d'Endocrinologie,*vol. 57,no. 1, pp. 9–14.

Shibukawa, S, Noshiro, T et al. 1997, 'A family of multiple endocrine neoplasia type 2A (Sipple's syndrome)', *Folia Endocrinologica Japonica,*vol. 73,no. 4, pp. 583–590.

Skinner, MA & Wells Jr, SA 1997, 'Medullary carcinoma of the thyroid gland and the MEN2 syndromes', *Seminars in Pediatric Surgery,*vol. 6,no. 3, pp. 134–140.

Spinelli, C, Puccini, M et al. 2002, 'Prophylactic total thyroidectomy in children and adolescents with genetic mutations in the RET M-proto-oncogene', *La Pediatria Medica e Chirurgica – Medical and Surgical Pediatrics,*vol. 24,no. 1, pp. 53–57.

Stuhrmann, M 1995, 'The direct gene test in familial medullary thyroid gland carcinoma and in MEN syndromes: detection of mutations in the ret proto-oncogene saves screening studies', *Fortschritte der Medizin,*vol. 113,no. 35–36, pp. 503–506.

Todorov, G, Petkov, R et al. 2006, 'Medullary thyroid carcinoma: current surgical aspects. Overview and retrospective analysis of 22 cases', *Khirurgiia,*vol. 4–5, pp. 5–8.

Torre, GC, Varaldo, E & Bottaro, P 2004, 'State-of-the-art in the diagnosis and therapy of the MEN1 and MEN2 syndromes', *Il Giornale di Chirurgia,*vol. 25,no. 4, pp. 109–115.

Tratzmuller, R, Irle, U et al. 1999, 'Multiple endocrine neoplasia type 2 (MEN2) in children and adolescents', *Monatsschrift fur Kinderheilkunde,*vol. 147,no. 8, pp. 733–743.

Valsamaki, P, Gerali, S et al. 2004, 'A patient with MEN-IIA syndrome due to de novo mutation and papillary thyroid carcinoma: the role of 99mTc-depreotide in diagnosing metastases and brief review of the literature', *Hellenic Journal of Nuclear Medicine,*vol. 7,no. 3, pp. 168–173.

Wallin, G, Bondesson, AG et al. 2001, 'Hereditary thyroid cancer can be cured by prophylactic surgery', *Läkartidningen,*vol. 98,no. 25, pp. 3024–3028.

Wiesli, P, Rojas, J et al. 1999, 'Identification and screening of a family with type 2A multiple endocrine neoplasia with DNA analysis', *Schweizerische Rundschau für Medizin Praxis – Revue Suisse de Médecine Praxis,* vol.88,no. 41, pp. 1667–1673.

Wohllk, N, Becker, P et al. 2000, 'Multiple endocrine neoplasia: a clinical model for testing molecular genetic techniques', *Revista Medica de Chile,*vol. 128,no. 7, pp. 791–800.

Wohllk, N, Becker, P et al. 2001, 'Germline mutations of the ret proto-oncogene in Chilean patients with hereditary and sporadic medullary thyroid carcinoma', *Revista Medica de Chile,*vol. 129, no. 7, pp. 713–718.

Wygoda, Z, Oczko-Wojciechowska, M et al. 2006, 'Medullary thyroid carcinoma: the comparison of the hereditary and sporadic types of cancer', *Endokrynologia Polska,*vol. 57,no. 4, pp. 407–414.

Wygoda, Z, Wloch, J et al. 2001, 'Results of treating medullary thyroid carcinoma: the differences between sporadic and inherited forms', *Wiadomości Lekarskie (Warsaw, Poland: 1960),*vol. 54, suppl. 1, pp. 422–431.

Yao, B, Liu, X et al. 2007, 'Mutation of the RET proto-oncogene in type 2A multiple endocrine neoplasia Chinese families and the application of pentagastrin stimulation test in diagnosis and follow-up', *Zhonghua Nei Ke Za Zhi – Chinese Journal of Internal Medicine,*vol. 46, no. 11, pp. 914–918.

Zakrzewska, A, Makowska, AM & Bar-Andziak, E 2004, 'Late onset of medullary thyroid carcinoma with bilateral adrenal pheochromocytomas in the case of patient with MEN2', *Polski Merkuriusz Lekarski,*vol. 17,no. 102, pp. 633–637.

Zhou, Y-L, Zhu, S-X et al. 2007, 'The clinical patterns and RET proto-oncogene in fifteen multiple endocrine neoplasia type 2A pedigrees', *Zhonghua Nei Ke Za Zhi – Chinese Journal of Internal Medicine,*vol. 46,no. 6, pp. 466–470.

#### Outdated systematic reviews:

Machens, A & Dralle, H 2007, 'Genotype-phenotype based surgical concept of hereditary medullary thyroid carcinoma', *World Journal of Surgery,*vol. 31,no. 5, pp. 957–968.

Szinnai, G, Meier, C et al. 2003, 'Review of multiple endocrine neoplasia type 2A in children: therapeutic results of early thyroidectomy and prognostic value of codon analysis', *Pediatrics,*vol. 111,no. 2, pp. E132–139.

#### Wrong index test:

Bockhorn, M, Frilling, A et al. 1999, 'No correlation between RET immunostaining and the codon 918 mutation in sporadic medullary thyroid carcinoma', *Langenbeck's Archives of Surgery,*vol. 384,no. 1, pp. 60–64.

Dolan, SJ & Russell, CFJ 2000, 'Medullary thyroid carcinoma in Northern Ireland, 1967-1997', *Annals of the Royal College of Surgeons of England,*vol. 82,no. 3, pp. 156–161.

Eng, C, Foster, KA et al. 1996, 'Mutation analysis of the c-mos proto-oncogene and the endothelin-B receptor gene in medullary thyroid carcinoma and phaeochromocytoma', *British Journal of Cancer,*vol. 74, no. 3, pp. 339–341.

Honda, M, Tsukada, T et al. 2000, 'A novel mutation of the MEN1 gene in a Japanese kindred with familial isolated primary hyperparathyroidism', *European Journal of Endocrinology,*vol. 142, no. 2, pp. 138–143.

Kraimps, JL, Denizot, A et al. 1996, 'Primary hyperparathyroidism in multiple endocrine neoplasia type IIa: retrospective French multicentric study', *World Journal of Surgery,*vol. 20,no. 7, pp. 808–813.

Marsh, DJ, Zheng, ZM et al. 1997, 'Mutation analysis of glial cell line-derived neurotrophic factor, a ligand for an RET/coreceptor complex, in multiple endocrine neoplasia type 2 and sporadic neuroendocrine tumors', *Journal of Clinical Endocrinology & Metabolism,*vol. 82,no. 9, pp. 3025–3028.

Robledo, M, Gil, L et al. 2003, 'Polymorphisms G691S/S904S of RET as genetic modifiers of MEN2A', *Cancer Research,*vol. 63,no. 8, pp. 1814–1817.

Stefanaki, A, Maris, T et al. 1994, 'Characterization of pheochromocytomas using quantitative analysis of the parameter T2 of the mass (T2-QMRI)', *Magma,*vol. 2,no. 1.

Takano, T, Miyauchi, A et al. 2001, 'Large-scale analysis of mutations in RET exon 16 in sporadic medullary thyroid carcinomas in Japan', *Japanese Journal of Cancer Research,*vol. 92,no. 6, pp. 645–648.

Zedenius, J, Wallin, G et al. 1994, 'Somatic and MEN2A de novo mutations identified in the RET proto-oncogene by screening of sporadic MTCs', *Human Molecular Genetics,*vol. 3,no. 8, pp. 1259–1262.

#### Wrong intervention:

Barbieri, RB, Bufalo, NE et al. 2011, 'The inheritance of CYP1A1m1 and CYP1A2 polymorphisms may increase the susceptibility to medullary thyroid carcinoma', *Endocrine Reviews,*vol. 32,no. 3.

Margraf, RL, Crockett, DK et al. 2009, 'Multiple endocrine neoplasia type 2 RET proto-oncogene database: repository of MEN2-associated RET sequence variation and reference for genotype/phenotype correlations', *Human Mutation,*vol. 30,no. 4, pp. 548–556.

#### Wrong outcomes:

Baumgartner-Parzer, SM, Lang, R, et al. 2005, 'Polymorphisms in exon 13 and intron 14 of the RET proto-oncogene: genetic modifiers of medullary thyroid carcinoma?', *Journal of Clinical Endocrinology & Metabolism*, vol. 90, no. 11, pp. 6232–6236.

Benej, M, Bendlova, B et al. 2012, 'Establishing high resolution melting analysis: method validation and evaluation for c-RET proto-oncogene mutation screening', *Clinical Chemistry and Laboratory Medicine,*vol. 50,no. 1, pp. 51–60.

Bugalho, MJ, Domingues, R & Sobrinho, L 2003, 'MEN2A families: from hot spots to hot regions', *International Journal of Molecular Medicine,*vol. 11,no. 1, pp. 71–74.

Cebrian, A, Lesueur, F, Martin, S, Leyland, J, Ahmed, S, Luccarini, C, Smith, PL, Luben, R, Whittaker, J, Pharoah, PD, Dunning, AM & Ponder, BAJ 2005, 'Polymorphisms in the initiators of RET (Rearranged during transfection) signaling pathway and susceptibility to sporadic medullary thyroid carcinoma', Journal of Clinical Endocrinology and Metabolism, vol. 90, no. 11, pp. 6268-6274.

Chi, DK, Dou, S et al. 1994, 'A rapid presymptomatic genetic test for the detection of mutations in the RET proto-oncogene associated with men2A and FMTC', *Surgical Forum,*vol. 45, pp. 510–513.

Cohen, MS, Phay, JE et al. 2002, 'Gastrointestinal manifestations of multiple endocrine neoplasia type 2', *Annals of Surgery,* vol.235,no. 5, pp. 648–654.

Dang, GT, Cote, GJ et al. 1999, 'A codon 891 exon 15 RET proto-oncogene mutation in familial medullary thyroid carcinoma: a detection strategy', *Molecular and Cellular Probes,*vol. 13,no. 1, pp. 77–79.

Decker, RA 1993, 'Expression of papillary thyroid carcinoma in multiple endocrine neoplasia type 2A', *Surgery,*vol. 114,no. 6, pp. 1059–1063.

Decker, RA, Peacock, ML & Watson, P 1998, 'Hirschsprung disease in MEN2A: increased spectrum of RET exon 10 genotypes and strong genotype-phenotype correlation', *Human Molecular Genetics,*vol. 7,no. 1, pp. 129–134.

Durick, K, Yao, VJ et al. 1995, 'Tyrosines outside the kinase core and dimerization are required for the mitogenic activity of RET/ptc2', *Journal of Biological Chemistry,*vol. 270,no. 42, pp. 24642–24645.

Eng, C, Clayton, D et al. 1996, 'The relationship between specific ret proto-oncogene mutations and disease phenotype in multiple endocrine neoplasia type 2: International RET Mutation Consortium analysis', *Journal of the American Medical Association,*vol. 276,no. 19, pp. 1575–1579.

Fabien, N, Paulin, C et al. 1994, 'The ret proto-oncogene is expressed in normal human parafollicular thyroid-cells', *International Journal of Oncology,*vol. 4, no. 3, pp. 623–626.

Fernandez, RM, Antinolo, G et al. 2003, 'The RET C620S mutation causes multiple endocrine neoplasia type 2A (MEN2A) but not Hirschsprung’s disease (HSCR) in a family cosegregating both phenotypes', *Human Mutation,* vol.22,no. 5, pp. 412–415.

Frank-Raue, K, Rondot, S et al. 2007, 'Change in the spectrum of RET mutations diagnosed between 1994 and 2006', *Clinical Laboratory,* vol.53,no. 5–6, pp. 273–282.

Gardner, E, Mulligan, LM et al. 1994, 'Haplotype analysis of MEN2 mutations', *Human Molecular Genetics,* vol.3,no. 10, pp. 1771–1774.

Gardner, E, Papi, L et al. 1993, 'Genetic-linkage studies map the multiple endocrine neoplasia type-2 loci to a small interval on chromosome 10q11.2', *Human Molecular Genetics,*vol. 2,no. 3, pp. 241–246.

Hofstra, RM, Landsvater, RM et al. 1994, 'A mutation in the RET proto-oncogene associated with multiple endocrine neoplasia type 2B and sporadic medullary thyroid carcinoma', *Nature,*vol. 367,no. 6461, pp. 375–376.

Hoppner, W & Ritter, MM 1997, 'A duplication of 12 bp in the critical cysteine rich domain of the RET proto-oncogene results in a distinct phenotype of multiple endocrine neoplasia type 2A', *Human Molecular Genetics,*vol. 6,no. 4, pp. 587–590.

Iihara, M, Yamashita, T et al. 1997, 'A nationwide clinical survey of patients with multiple endocrine neoplasia type 2 and familial medullary thyroid carcinoma in Japan', *Japanese Journal of Clinical Oncology,* vol.27,no. 3, pp. 128–134.

Inoue, K, Shimotake, T et al. 1999, 'Mutational analysis of the RET proto-oncogene in a kindred with multiple endocrine neoplasia type 2A and Hirschsprung's disease', *Journal of Pediatric Surgery,* vol.34,no. 10, pp. 1552–1554.

Kambouris, M, Jackson, CE & Feldman, GL 1996, 'Diagnosis of multiple endocrine neoplasia [MEN] 2A, 2B and familial medullary thyroid cancer [FMTC] by multiplex PCR and heteroduplex analyses of RET proto-oncogene mutations', *Human Mutation,* vol.8,no. 1, pp. 64–70.

Kameyama, K, Okinaga, H & Takami, H 2004, 'RET oncogene mutations in 75 cases of familial medullary thyroid carcinoma in Japan', *Biomedicine and Pharmacotherapy,* vol.58,no. 6–7, pp. 345–347.

Kebebew, E, Ituarte, PHG et al. 2000, 'Medullary thyroid carcinoma: clinical characteristics, treatment, prognostic factors, and a comparison of staging systems', *Cancer,*vol. 88,no. 5, pp. 1139–1148.

Kim, IJ, Chung Kang, H et al. 2002, 'RET oligonucleotide microarray for the detection of RET mutations in multiple endocrine neoplasia type 2 syndromes', *Clinical Cancer Research,*vol. 8, no. 2, pp. 457–463.

Kruckeberg, KE & Thibodeau, SN 2004, 'Pyrosequencing technology as a method for the diagnosis of multiple endocrine neoplasia type 2', *Clinical Chemistry,*vol. 50,no. 3, pp. 522–529.

Margraf, RL, Mao, R et al. 2007, 'RET proto-oncogene genotyping using unlabeled probes, the masking technique, and amplicon high-resolution melting analysis', *Journal of Molecular Diagnostics,* vol.9, no. 2, 184–196.

Margraf, RL, Mao, R & Wittwer, CT 2006, 'Masking selected sequence variation by incorporating mismatches into melting analysis probes', *Human Mutation,* vol. 27no. 3, 269–278.

Margraf, RL, Mao, R & Wittwer, CT 2008, 'Rapid diagnosis of MEN213 using unlabeled probe melting analysis and the LightCycler 480 instrument', *Journal of Molecular Diagnostics,*vol. 10,no. 2, pp. 123–128.

Marsh, DJ, Theodosopoulos, G et al. 2001, 'Rapid mutation scanning of genes associated with familial cancer syndromes using denaturing high-performance liquid chromatography', *Neoplasia,*vol. 3,no. 3, pp. 236–244.

Moers, AM, Landsvater, RM et al. 1996, 'Familial medullary thyroid carcinoma: not a distinct entity? Genotype-phenotype correlation in a large family', *American Journal of Medicine,*vol. 101,no. 6, pp. 635–641.

Mole, SE, Mulligan, LM et al. 1993, 'Localisation of the gene for multiple endocrine neoplasia type 2A to a 480 kb region in chromosome band 10q11.2', *Human Molecular Genetics,*vol. 2,no. 3, pp. 247–252.

Moore, SW, Appfelstaedt, J & Zaahl, MG 2007, 'Familial medullary carcinoma prevention, risk evaluation, and RET in children of families with MEN2', *Journal of Pediatric Surgery,*vol. 42,no. 2, pp. 326–332.

Mulligan, LM, Eng, C et al. 1994, 'Diverse phenotypes associated with exon 10 mutations of the RET proto-oncogene', *Human Molecular Genetics,*vol. 3,no. 12, pp. 2163–2167.

Park, JG 1999, ‘Hereditary tumor registry in Korea’*,* in: J Utsunomiya, JJ Mulvihill, W Weber and Y Yuasa (eds), ‘*Familial cancer and prevention – molecular epidemiology: a new strategy toward cancer control*’. Wiley-Liss, New York.

Pazaitou-Panayiotou, K, Kaprara, A et al. 2005, 'Efficient testing of the RET gene by DHPLC analysis for MEN2 syndrome in a cohort of patients', *Anticancer Research,* vol.25,no. 3B, pp. 2091–2095.

Rios, A, Rodriguez, JM et al. 2011, 'Histological and immunohistochemical profile of sporadic and familial medullary thyroid carcinoma', *Endocrinologia y Nutricion: organo de la Sociedad Espanola de Endocrinologia y Nutricion,*vol. 58,no. 10, pp. 521–528.

Torrente, I, Arturi, F et al. 2004, 'Evaluation of a DHPLC-based assay for rapid detection of RET germline mutations in Italian patients with medullary thyroid carcinoma', *Journal of Endocrinological Investigation,* vol.27,no. 2, pp. 111–116.

#### Wrong population (e.g. MEN1 or patients not suspected of MEN2, but already diagnosed):

Abu-Amero, KK, Alzahrani, AS et al. 2006, 'Association of mitochondrial DNA transversion mutations with familial medullary thyroid carcinoma/multiple endocrine neoplasia type 2 syndrome', *Oncogene,*vol. 25,no. 5, pp. 677–684.

Agarwal, S, Agarwal, A et al. 2012, 'MEN2A family-prophylactic thyroidectomy for asymptomatic siblings with positive 634 codon mutation', *Journal of the Association of Physicians of India,*vol. 60, pp. 127–129.

Ainahi, A, Kebbou, M et al. 2006, 'Study of the RET gene and his implication in thyroid cancer: Morocco case family', *Indian Journal of Cancer,*vol. 43,no. 3, pp. 122–126.

Alemi, M, Lucas, SD et al. 1997, 'A complex nine base pair deletion in RET exon 11 common in sporadic medullary thyroid carcinoma', *Oncogene,*vol. 14, no. 17, pp. 2041–2045.

Arlt, DH, Baur, B et al. 2000, 'A novel type of mutation in the cysteine rich domain of the RET receptor causes ligand independent activation', *Oncogene,*vol. 19,no. 30, pp. 3445–3448.

Arum, SM, Dahia, PLM et al. 2005, 'A RET mutation with decreased penetrance in the family of a patient with a "sporadic" pheochromocytoma', *Endocrine,*vol. 28,no. 2, pp. 193–198.

Bolino, A, Schuffenecker, I et al. 1995, 'RET mutations in exons 13 and 14 of FMTC patients', *Oncogene,*vol. 10,no. 12, pp. 2415–2419.

Borrego, S, Wright, FA et al. 2003, 'A founding locus within the RET proto-oncogene may account for a large proportion of apparently sporadic Hirschsprung’s disease and a subset of cases of sporadic medullary thyroid carcinoma', *American Journal of Human Genetics,*vol. 72,no. 1, pp. 88–100.

Calender, A, Giraud, S et al. 1995, 'Multiple endocrine neoplasia type-1 in France: clinical and genetic studies', *Journal of Internal Medicine,* vol.238,no. 3, pp. 263–268.

Cascon, A, Ruiz-Llorente, S et al. 2004, 'Genetic and epigenetic profile of sporadic pheochromocytomas', *Journal of Medical Genetics,*vol. 41,no. 3.

Cho, NH, Lee, HW et al. 2005, 'Genetic aberrance of sporadic MEN2A component tumours: analysis of RET', *Pathology,*vol. 37,no. 1, pp. 10–13.

Dvoraka, S, Vaclavikova, E et al. 2006, 'New multiple somatic mutations in the RET proto-oncogene associated with a sporadic medullary thyroid carcinoma', *Thyroid,*vol. 16,no. 3, pp. 311–316.

Edstrom, E, Grondal, S et al. 1999, 'Long term experience after subtotal adrenalectomy for multiple endocrine neoplasia type IIa', *European Journal of Surgery,*vol. 165,no. 5, pp. 431–435.

Elisei, R, Cosci, B et al. 2004, 'RET exon 11 (G691S) polymorphism is significantly more frequent in sporadic medullary thyroid carcinoma than in the general population', *Journal of Clinical Endocrinology & Metabolism,* vol.89,no. 7, pp. 3579–3584.

Eng, C, Mulligan, LM et al. 1995, 'Mutation of the RET proto-oncogene in sporadic medullary thyroid carcinoma', *Genes Chromosomes Cancer,*vol. 12,no. 3, pp. 209–212.

Eng, C, Mulligan, LM et al. 1996, 'Heterogeneous mutation of the RET proto-oncogene in subpopulations of medullary thyroid carcinoma', *Cancer Research,*vol. 56,no. 9, pp. 2167–2170.

Eng, C, Smith, DP et al. 1994, 'Point mutation within the tyrosine kinase domain of the RET proto-oncogene in multiple endocrine neoplasia type 2B and related sporadic tumours', *Human Molecular Genetics,*vol. 3,no. 2, pp. 237–241.

Eng, C, Smith, DP et al. 1994b, 'Erratum: Point mutation within the tyrosine kinase domain of the RET proto-oncogene in multiple endocrine neoplasia type 2B and related sporadic tumours, *Human Molecular Genetics*, vol. 3 no. 4, p. 686.

Eng, C, Thomas, GA et al. 1998, 'Mutation of the RET proto-oncogene is correlated with ret immunostaining in subpopulations of cells in sporadic medullary thyroid carcinoma', *Journal of Clinical Endocrinology & Metabolism,*vol. 83,no. 12, pp. 4310–4313.

Ferraris, AM, Mangerini, R et al. 1997, 'Polyclonal origin of medullary carcinoma of the thyroid in multiple endocrine neoplasia type 2', *Human Genetics,*vol. 99,no. 2, pp. 202–205.

Frank-Raue, K, Kratt, T et al. 1996, 'Diagnosis and management of pheochromocytomas in patients with multiple endocrine neoplasia type 2-relevance of specific mutations in the RET proto-oncogene', *European Journal of Endocrinology,*vol. 135,no. 2, pp. 222–225.

Frilling, A, Hoeppner, W et al. 1994, 'Screening for mutations of the RET proto-oncogene in men2A families', *Surgical Forum,*vol. 45,pp. 480–481.

Godballe, C. Jorgensen, G et al. 2010, 'Medullary thyroid cancer: RET testing of an archival material', *European Archives of Oto-Rhino-Laryngology,*vol. 267,no. 4, pp. 613–617.

Ishida, O, Zeki, K et al. 1995, 'Germ line mutation in the RET proto-oncogene associated with familial multiple endocrine neoplasia type 2B: a case report', *Japanese Journal of Clinical Oncology,*vol. 25,no. 3, pp. 104–108.

Komminoth, P 1995, 'Rudolf-Virchow-Preis 1995. The role of RET proto-oncogene mutation analysis in the diagnosis of multiple endocrine neoplasia type 2 (MEN2) gene carriers and in the discrimination of sporadic and familial medullary thyroid carcinomas and pheochromocytomas', *Verhandlungen der Deutschen Gesellschaft für Pathologie,*vol. 79, pp. L–LV.

Kroustrup, JP, Laurberg, P & Madsen, P H 1999, 'Rapid MEN2A gene carrier identification using primer-specific PCR amplification', *Scandinavian Journal of Clinical and Laboratory Investigation,*vol. 59,no. 8, pp. 643–647.

Larsson, C, Weber, G et al. 1994, 'Genetics of multiple endocrine neoplasia type-1', in: B Wiedenmann et al. (eds), *'Molecular and cell biological aspects of gastroenteropancreatic neuroendocrine tumor disease',* *Annals of the New York Academy of Sciences,* vol. 733, pp. 453–463.

Libroia, A, Verga, U et al. 1998, 'Seventeen-year-long follow-up of a family affected by type 2A multiple endocrine neoplasia (MEN2A)', *Journal of Endocrinological Investigation,*vol. 21,no. 2, pp. 87–92.

Margraf, RL, Durtschi, JD et al. 2012, 'Determination of RET sequence variation in an MEN2 unaffected cohort using multiple-sample pooling and next-generation sequencing', *Journal of Thyroid Research,* vol.2012, pp. 318232.

Margraf, RL, Mao, R et al. 2006, 'Mutation scanning of the RET proto-oncogene using high-resolution melting analysis', *Clinical Chemistry,* vol.52,no. 1, pp. 138–141.

Marsh, DJ, Robinson, BG et al. 1994, 'A rapid screening method for the detection of mutations in the RET proto-oncogene in multiple endocrine neoplasia type 2A and familial medullary thyroid carcinoma families', *Genomics,*vol. 23,no. 2, pp. 477–479.

Maruyama, S, Iwashita, T et al. 1994, 'Germ line mutations of the ret proto-oncogene in Japanese patients with multiple endocrine neoplasia type 2A and type 2B', *Japanese Journal of Cancer Research,*vol. 85,no. 9, pp. 879–882.

Matias-Guiu, X, Colomer, A et al. 1995, 'Expression of the ret proto-oncogene in phaeochromocytoma: an in situ hybridization and northern blot study', *Journal of Pathology,*vol. 176,no. 1, pp. 63–68.

Migita, M, Hiromatsu, Y et al. 1997, 'Mutation of RET proto-oncogene in Japanese patients with multiple endocrine neoplasia type 2b and sporadic medullary thyroid carcinoma', *Endocrine Journal,* vol.44,no. 4, pp. 559–565.

Mulligan, LM, Eng, C et al. 1994, 'Specific mutations of the RET proto-oncogene are related to disease phenotype in MEN2A and FMTC', *Nature Genetics,*vol. 6,no. 1, pp. 70–74.

Mulligan, LM, Kwok, JBJ et al. 1993, 'Germ-line mutations of the RET proto-oncogene in multiple endocrine neoplasia type 2A', *Nature,*vol. 363,no. 6428, pp. 458–460.

Musholt, PB, Musholt, TJ et al. 1997, '"Cold" single-strand conformational variants for mutation analysis of the RET proto-oncogene', *Surgery,*vol. 122,no. 2, pp. 363–370; discussion pp. 370–361.

Padberg, BC, Schroder, S et al. 1995, 'Absence of RET proto-oncogene point mutations in sporadic hyperplastic and neoplastic lesions of the parathyroid gland', *American Journal of Pathology,*vol. 147,no. 6, pp. 1600–1607.

Paun, DL, Mohora, M et al. 2008, 'Genetic testing for multiple endocrine neoplasia type 2', *Revue Roumaine de Médecine Interne – Romanian Journal of Internal Medicine,*vol. 46,no. 2, pp. 159–163.

Quadro, L, Panariello, L et al. 1994, 'Frequent RET proto-oncogene mutations in multiple endocrine neoplasia type 2A', *Journal of Clinical Endocrinology & Metabolism,*vol. 79,no. 2, pp. 590–594.

Romei, C, Mariotti, S et al. 2010, 'Multiple endocrine neoplasia type 2 syndromes (MEN2): results from the ItaMEN network analysis on the prevalence of different genotypes and phenotypes', *European Journal of Endocrinology,*vol. 163,no. 2, pp. 301–308.

Rossel, M, Pasini, A et al. 1997, 'Distinct biological properties of two RET isoforms activated by MEN2A and MEN2B mutations', *Oncogene,*vol. 14,no. 3, p. 265.

Rossel, M, Schuffenecker, M et al. 1995, 'Detection of a germline mutation at codon 918 of the RET proto-oncogene in French MEN2B families', *Human Genetics,*vol. 95,no. 4, pp. 403–406.

Sanchez, B, Robledo, M et al. 1999, 'High prevalence of the C634Y mutation in the RET proto-oncogene in MEN2A families in Spain', *Journal of Medical Genetics,*vol. 36,no. 1, pp. 68–70.

Takiguchi-Shirahama, S, Koyama, K et al. 1995, 'Germline mutations of the RET proto-oncogene in eight Japanese patients with multiple endocrine neoplasia type 2A (MEN2A)', *Human Genetics,*vol. 95,no. 2, pp. 187–190.

van Naderveen, FH, Korpershoek, E et al. 2009, 'Array-comparative genomic hybridization in sporadic benign pheochromocytomas', *Endocrine-Related Cancer,*vol. 16,no. 2, pp. 505–513.

Willeke, F, Hauer, MP et al. 1998, 'Multiple endocrine neoplasia type 2-associated ret proto-oncogene mutations do not contribute to the pathogenesis of sporadic parathyroid tumors', *Surgery,*vol. 124,no. 3, pp. 484–490.

Williams, GH, Rooney, S et al. 1996, 'Analysis of the RET proto-oncogene in sporadic parathyroid adenomas', *Journal of Pathology,*vol. 180,no. 2, pp. 138–141.

Xue, F, Yu, H et al. 1994, 'Germline RET mutations in MEN2A and FMTC and their detection by simple DNA diagnostic tests', *Human Molecular Genetics,*vol. 3,no. 4, pp. 635–638.

Yip, L, Cote, GJ et al. 2003, 'Multiple endocrine neoplasia type 2: evaluation of the genotype-phenotype relationship', *Archives of Surgery,*vol. 138,no. 4, pp. 409–416; discussion p. 416.

Yoshimoto, K, Endo, H et al. 1998, 'Familial isolated primary hyperparathyroidism with parathyroid carcinomas: clinical and molecular features', *Clinical Endocrinology,*vol. 48,no. 1, pp. 67–72.

Yoshimoto, K, Iwahana, H & Itakura, M 1993, 'Relatively good prognosis of multiple endocrin neoplasia type-2b in japanese: review of cases in Japan and analysis of genetic changes in tumors', *Endocrine Journal,*vol. 40,no. 6, pp. 649–657.

Zhou, G-w, Wei, Y et al. 2009, 'Diagnosis and surgical treatment of multiple endocrine neoplasia', *Chinese Medical Journal,*vol. 122,no. 13, pp. 1495–1500.

Zhou, YL, Zhao, YJ et al. 2007, 'RET proto-oncogene mutations are restricted to codons 634 and 918 in mainland Chinese families with MEN2A and MEN2B', *Clinical Endocrinology,* vol.67,no. 4, pp. 570–576.

#### Articles with the wrong study type:

Ahmed, M & Al-Hindi, H 2010, 'Early diagnosis of medullary thyroid cancer: a case for early prophylactic thyroidectomy', *Endocrine Reviews,*vol. 31,no. 3.

Arnold, A 1996, 'RET mutation screening in sporadic pheochromocytoma', *Journal of Clinical Endocrinology & Metabolism,*vol. 81,no. 1, p. 430.

Beck, O, Fassbender, WJ et al. 2004, 'Pheochromocytoma in childhood: implication for further diagnostic procedures', *Acta Paediatrica,*vol. 93,no. 12, pp. 1630–1634.

Brauckhoff, M & Gimm, O 2009, 'Extrathyroidal manifestations of multiple endocrine neoplasia type 2', *Thyroid: official journal of the American Thyroid Association,*vol. 19,no. 6, pp. 555–557.

Castellani, MR, Seregni, E et al. 2008, 'MIBG for diagnosis and therapy of medullary thyroid carcinoma: is there still a role?', *Quarterly Journal of Nuclear Medicine and Molecular Imaging,*vol. 52,no. 4, pp. 430-–440.

Chen, H, Luo, JW & Zhang, ZJ 2007, 'Detection of the mutation of ret proto-oncogene in a pedigree with multiple endocrine neoplasia 2A', *Journal of Hypertension,*vol. 25, pp. S129–S130.

Chen, H-D, Chao, K et al. 1993, 'Familial medullary thyroid carcinoma: report of three cases in a family', *Journal of the Formosan Medical Association,*vol. 92, no. 1, pp. 68–71.

Chen, H-M & Fang, J-Y 2009, 'Genetics of the hamartomatous polyposis syndromes: a molecular review', *International Journal of Colorectal Disease,*vol. 24, no. 8, pp. 865–874.

Chew, SL 2009, 'Use of mutation analysis in endocrine neoplasia syndromes', *Clinical Medicine – journal of the Royal College of Physicians of London,*vol. 9,no. 4, pp. 362–363.

Chikouche, A, Ait Abdallah, M et al. 2012, 'RET proto-oncogene mutations analysis in Algerian patients with a medullary thyroid carcinoma', *Clinical Chemistry and Laboratory Medicine,*vol. 50,no. 4, pp. A101–A102.

Clowes, VE, Shaw-Smith, C et al. 2008, 'MEN2 screening dilemmas in a family with a novel RET mutation in the MEN2 susceptibility region of the gene, a family history of Hirschsprung’s disease, and no family history of MEN2-related tumours', *Clinical Endocrinology,*vol. 68,no. 4, pp. 666–667.

Colombo-Benkmann, M, Bramswig, J et al. 2002, 'Surgical strategy in a kindred with a rare RET proto-oncogene mutation of variable penetrance with regard to multiple endocrine neoplasia', *World Journal of Surgery,*vol. 26,no. 10, pp. 1286–1290.

Cordella, D, Muzza, M et al. 2006, 'An in-frame complex germline mutation in the juxtamembrane intracellular domain causing RET activation in familial medullary thyroid carcinoma', *Endocrine-Related Cancer,*vol. 13,no. 3, pp. 945–953.

Cordella, D, Muzza, M et al. 2010, 'Functional analyses of four novel ret germline mutations: juxtamembrane mutations display the highest level of autophosphorylation', *Endocrine Abstracts,*vol. 20, HTC4.

Day, PF, Gianotti, TF & Picasso, MFR 2004, 'Familial medullary thyroid carcinoma (FMTC): first family described with FMTC associated to Cys611Trp mutation of proto-oncogene RET', *Journal of Internal Medicine,*vol. 255,no. 6, pp. 711–712.

Dotto, J & Nose, V 2008, 'Familial thyroid carcinoma: a diagnostic algorithm', *Advances in Anatomic Pathology,*vol. 15,no. 6, pp. 332–349.

Dralle, H 2010, 'Medullary thyroid carcinoma: the primary, the pacemaker in recurrences, and the gene carrier', *Endocrine Journal,*vol. 57, pp. S248–S248.

Dvorakova, S, Dvorakova, K et al. 2005, 'A novel Czech kindred with familial medullary thyroid carcinoma and Hirschsprung's disease', *Journal of Pediatric Surgery,*vol. 40,no. 6, pp. E1–E6.

Edery, P, Attie, T et al. 1994, 'A novel polymorphism in the coding sequence of the human RET proto-oncogene', *Human Genetics,*vol. 94,no. 5, pp. 579–580.

Egawa, S, Futami, H et al. 1997, 'Point mutations of the RET proto-oncogene in patients with multiple endocrine neoplasia 2 in Japan', *Proceedings of the American Association for Cancer Research Annual Meeting,*vol. 38, pp. 266–267.

Elise, R, Bottici, V et al. 2011, 'Medullary thyroid cancer: from genetic to therapy', *Tumor Biology,*vol. 32, p. S20.

Ellard, S 2004, 'Multiple endocrine neoplasia types 1 and 2', *Methods in Molecular Medicine,*vol. 92, pp. 267–283.

Eng, C 1997, 'Genetic screening in hereditary medullary thyroid carcinoma', *Acta Chirurgica Austriaca,*vol. 29,no. 1, pp. 5–8.

Eng, C, Smith, DP et al. 1995, 'A novel point mutation in the tyrosine kinase domain of the RET proto-oncogene in sporadic medullary thyroid carcinoma and in a family with FMTC', *Oncogene,*vol. 10,no. 3, pp. 509–513.

Eng, C, Toogood, AA et al. 1995, 'A family with multiple endocrine neoplasia type 2B which does not have a mutation at codon 928 of exon 16 of the RET proto-oncogene', *Journal of Endocrinology,*vol. 144,suppl., p. P44.

Engiz, O, Ocal, G et al. 2009, 'Early prophylactic thyroidectomy for RET mutation-positive MEN2B', *Pediatrics International,*vol. 51,no. 4, pp. 590–593.

Erbilgin, Y, Tutuncu, Y et al. 2010, 'Ret oncogene genotypes in multiple endocrine neoplasia type 2: studies in four Turkish families', *Clinical Genetics,*vol. 78, p. 61.

Fialkowski, EA, DeBenedetti, MK et al. 2008, 'RET proto-oncogene testing in infants presenting with Hirschsprung’s disease identifies 2 new multiple endocrine neoplasia 2A kindreds', *Journal of Pediatric Surgery,*vol. 43,no. 1, pp. 188–190.

Fink, A, Lapidot, M & Spierer, A 1998, 'Ocular manifestations in multiple endocrine neoplasia type 2B', *American Journal of Ophthalmology,*vol. 126,no. 2, pp. 305–307.

Forster-Gibson, CJ & Mulligan, LM 1994, 'Multiple endocrine neoplasia type 2', *European Journal of Cancer,*vol. 30A,no. 13, pp. 1969–1974.

Frank-Raue, K 1999, 'Clinical implications of molecular analysis of the RET proto-oncogene in medullary thyroid carcinoma, multiple endocrine neoplasia type 2, and Hirschsprung's disease', *Clinical Laboratory,*vol. 45,no. 3–4, pp. 117–122.

Franz, C, Chi, D et al. 1995, 'Spectrum of RET proto-oncogene mutations in 52 MEN2A and FMTC families', *American Journal of Human Genetics,*vol. 57,4 suppl., p. A64.

Gagel, RF 1994, ‘Multiple endocrine neoplasia type II’, in:JP Bilezikian, R Marcus & MA Levine (eds), ‘*The parathyroids: basic and clinical concepts*’. New York, Raven Press.

Gallegos-Martinez, J, Herrera, MF et al. 1998, 'A false-positive diagnosis of C-cell hyperplasia in a member of a family with multiple endocrine neoplasia type 2A and familial colonic polyposis', *Surgery,*vol. 123,no. 5, pp. 587–588.

Giacche, M, Mori, L et al. 2011, 'Clinical follow-up for pheochromocytoma/paraganglioma in a population of susceptible subjects identified by genetic analysis', *High Blood Pressure and Cardiovascular Prevention,*vol. 18,no. 3, p. 131.

Giacche, M, Panarotto, A et al. 2010, 'Pheochromocytoma may occur in kindreds with ret mutation associated with familial medullary thyroid carcinoma (Ser891Ala)', *Journal of Hypertension,*vol. 28, p. e347.

Gill, JR, Reyes-Mugica, M et al. 1996, 'Early presentation of metastatic medullary carcinoma in multiple endocrine neoplasia, type IIA: implications for therapy', *Journal of Pediatrics,*vol. 129,no. 3, pp. 459–464.

Gimenez-Roqueplo, AP, Lehnert, H et al. 2006, 'Phaeochromocytoma, new genes and screening strategies', *Clinical Endocrinology,*vol. 65,no. 6, pp. 699–705.

Giraud, S, Pigny, P et al. 1999, 'Mutational analysis of the RET proto-oncogene in 200 French MEN2 families: a genotype-phenotype correlation', *American Journal of Human Genetics,*vol. 65,no. 4, pp. A63–A63.

Goretzki, PE, Hoppner, W et al. 1998, 'Genetic and biochemical screening for endocrine disease', *World Journal of Surgery,*vol. 22,no. 12, pp. 1202–1207.

Grobmyer, SR, Guillem, JG et al. 1999, 'Colonic manifestations of multiple endocrine neoplasia type 2B: report of four cases', *Diseases of the Colon & Rectum,* vol.42, no. 9, pp. 1216–1219.

Hasse-Lazar, K, Krawczyk, A et al. 2010, 'Pheochromocytomas in the RET proto-oncogene mutations carriers', *Endocrine Abstracts,*vol. 22, p. P449.

Hoeppner, W, Dralle, H & Brabant, G 1998, 'Duplication of 9 base pairs in the critical cysteine-rich domain of the RET proto-oncogene causes multiple endocrine neoplasia type 2A', *Human Mutation,*vol. 1,suppl. 1, pp. S128–S130.

Hoeppner, W, Frank-Raue, K & Raue, F 1996a, 'Molecular genetics, diagnostic and prophylactic therapy in multiple endocrine neoplasia type 2A', *European Journal of Clinical Investigation,*vol. 26,suppl. 1, p. A52.

Hoeppner, W 2007, 'Mutations in the RET proto-oncogene in medullary thyroid carcinoma', *Clinical Laboratory,*vol. 53,no. 5–6, pp. 283–284.

Igaz, P, Patocs, A et al. 2002, 'Occurrence of pheochromocytoma in a MEN2A family with codon 609 mutation of the RET proto-oncogene', *Journal of Clinical Endocrinology & Metabolism,* vol.87,no. 6, p. 2994.

Iyengar, S, Tallini, G et al. 1994, 'Mutation analysis of the RET gene in individuals with sporadic and familial pheochromocytoma', *American Journal of Human Genetics,*vol. 55,no. 3, suppl., p. A60.

Jafri, M & Maher, ER 2012, 'The genetics of phaeochromocytoma: using clinical features to guide genetic testing', *European Journal of Endocrinology,*vol. 166,no. 2, pp. 151–158.

Jansson, SKG, Lindskog, S et al. 2000, 'Medullary thyroid carcinoma caused by a RET M-mutation at codon 618 in a large MEN-2A family mapped over 9 generations', *Endocrine Journal,*vol. 47,suppl. August, p. 215.

Kaczmarek-Rys, M, Hoppe-Golebiewska, J et al. 2010, 'RET proto-oncogene mutations in Polish patient with medullary thyroid cancer', *Medizinische Genetik,*vol. 22,no. 1, 96.

Kasprzak, L, Nolet, S et al. 1999, 'Familial medullary thyroid carcinoma and prominent corneal nerves associated with a codon 804 germline mutation of the RET gene', *American Journal of Human Genetics,* vol.65,no. 4, pp. A132–A132.

Lairmore, TC & Wells Jr, SA 1993, 'Genetic testing for multiple endocrine neoplasia', *British Journal of Surgery,*vol. 80,no. 9, pp. 1092–1093.

Learoyd, DL, Delbridge, LW & Robinson, BG 2000, 'Multiple endocrine neoplasia', *Australian and New Zealand Journal of Medicine,*vol. 30,no. 6, pp. 675–682.

Learoyd, DL & Robinson, BG 2005, 'Do all patients with RET mutations associated with multiple endocrine neoplasia type 2 require surgery?', *Nature Clinical Practice: Endocrinology & Metabolism,*vol. 1,no. 2, pp. 60–61.

Lee, MJ, Chung, KH et al. 2010, 'Multiple endocrine neoplasia type 2B: early diagnosis by multiple mucosal neuroma and its DNA analysis', *Annals of Dermatology,*vol. 22,no. 4, pp. 452–455.

Lee, YJ, Li, HC et al. 2001, 'Picture of the month: Multiple endocrine neoplasia 2B syndrome', *Archives of Pediatrics & Adolescent Medicine,*vol. 155,no. 7, pp. 845–846.

Lemos, M, Regateiro, FJ et al. 2000, 'Molecular characterisation of a family with MEN2A associated with cutaneous lichen amyloidosis', *European Journal of Human Genetics,*vol. 8,suppl. 1, p. 109.

Li, JJ, Zhou, YL & Yang, YP 2007, 'Genetic and clinical research of familial medullary thyroid carcinoma', *Chinese Journal of Cancer Prevention and Treatment,*vol. 14,no. 24, pp. 1851–1853+1856.

Lips, CJM 1998, 'Clinical management of the multiple endocrine neoplasia syndromes: results of a computerized opinion poll at the sixth international workshop on multiple endocrine neoplasia and von Hippel-Lindau disease', *Journal of Internal Medicine,*vol. 243,no. 6, pp. 589–594.

Lo, CY, Wat, NMS et al. 2003, 'Multiple endocrine neoplasia type 2A in Chinese families', *Clinical Endocrinology,*vol. 58,no. 4, p. 528.

Macdonald, F 2000, 'The impact of presymptomatic molecular testing in hereditary cancers', *Medical Principles and Practice,*vol. 9,no. 4, pp. 221–248.

Machens, A & Dralle, H 2008, 'Pheochromocytoma penetrance varies by RET mutation in MEN2A', *Surgery,*vol. 143,no. 5, p. 697.

Machens, A & Dralle, H 2010, 'Simultaneous medullary and papillary thyroid carcinomas in carriers of the V804M RET germline mutation-a spurious association?', *Surgery,*vol. 147,no. 6, pp. 895–896.

Martinez, JJ, Bailey, A et al. 1996, 'Gene sequencing as a diagnostic tool in multiple endocrine neoplasia type 2 and familial medullary thyroid carcinoma', *Clinical Chemistry,*vol. 42,no. 11, pp. 6–6.

Martucciello, G, Lerone, M et al. 2012, 'Multiple endocrine neoplasias type 2B and RET proto-oncogene', *Italian Journal of Pediatrics,*vol. 38, pp. 9.

Massoll, N & Mazzaferri, EL 2004, 'Diagnosis and management of medullary thyroid carcinoma', *Clinics in Laboratory Medicine,*vol. 24,no. 1, pp. 49–83.

Matias-Guiu, X 1998, 'RET proto-oncogene analysis in the diagnosis of medullary thyroid carcinoma and multiple endocrine neoplasia type II', *Advances in Anatomic Pathology,*vol. 5,no. 3, pp. 196–201.

Matias-Guiu, X & Garrastazu, MT 1998, 'Composite phaeochromocytoma-ganglioneuroblastoma in a patient with multiple endocrine neoplasia type IIA', *Histopathology,*vol. 32,no. 3, pp. 281–282.

Matias-Guiu, X, Lagarda, E et al. 1997, 'Identification of a novel somatic mutation in the RET proto oncogene in a patient with sporadic medullary thyroid carcinoma', *Human Mutation,* vol.9,no. 5, pp. 476–476.

Mavraki, E, McConachie, M & Baty, D 2010, 'Genetic testing in phaeochromocytoma/ paraganglioma referrals: a seven year audit', *Journal of Medical Genetics,*vol. 47, pp. S73–S73.

Mayr, B, Brabant, G & Von Zur Muhlen, A 1999, 'Incidental detection of familial medullary thyroid carcinoma by calcitonin screening for nodular thyroid disease', *European Journal of Endocrinology,*vol. 141,no. 3, pp. 286–289.

Mazura, I, Vcelak, J et al. 1996, 'Direct DNA analysis of medullary thyroid carcinoma in Czech and Slovak families', *Chemicke Listy,*vol. 90,no. 9, pp. 672–673.

McMahon, R, Dow, DJ et al. 1996, 'An evaluation of direct mutation screening in MEN2A, MEN2B, FMTC and 'sporadic' MTC families', *European Journal of Human Genetics,*vol. 4,suppl. 1, p. 46.

Miltenburg, DM, Conklin, L & Sastri, S 2000, 'The role of genetic screening and prophylactic surgery in surgical oncology', *Journal of the American College of Surgeons,*vol. 190,no. 5, pp. 619–628.

Miyauchi, A 2010, 'Treatment of medullary thyroid carcinoma based on germline RET mutation analysis', *Endocrine Journal,*vol. 57, pp. S206–S206.

Mori, L, Giacche, M et al. 2010, 'Asymptomatic bilateral pheochromocytoma in VHL mutation (Val84Leu) carrier identified by family genetic screening', *Journal of Hypertension,*vol. 28, p. e347.

Morkane, C, Raptis, D et al. 2010, 'Prophylactic thyroidectomy in children with multiple endocrine neoplasia type 2', *Regulatory Peptides,*vol. 164,no. 1, p. 9.

Mulligan, LM & Ponder, BA 1995, 'Genetic basis of endocrine disease: multiple endocrine neoplasia type 2', *Journal of Clinical Endocrinology & Metabolism,*vol. 80,no. 7, pp. 1989–1995.

Nakata, S, Okugi, H et al. 2001, 'Multiple endocrine neoplasia type 2B', *International Journal of Urology,* vol.8,no. 7, pp. 398–400.

Nasser, T, Qari, F et al. 2010, 'RET codon 618 mutations is the most frequent phenotype in Saudi families with multiple endocrine neoplasia type 2A', *Endocrine Abstracts,*vol. 22, pp. P380.

Nelkin, BD & Baylin, SB 1993, 'RET oncogene responsible for men2A', *Current Biology,*vol. 3,no. 7, pp. 477–480.

Nilsson, O, Ahlman, H et al. 2000, 'Adrenal and extra-adrenal pheochromocytomas in a family with germline RET V804L mutation, previously associated only with familial medullary thyroid carcinoma', *Laboratory Investigation,* vol.80,no. 1, p. 73A.

Nilsson, O, Tisell, LE et al. 1999, 'Adrenal and extra-adrenal pheochromocytomas in a family with germline RET V804L mutation', *Journal of the American Medical Association,*vol. 281,no. 17, pp. 1587–1588.

Nishikawa, M, Murakumo, Y et al. 2003, 'Cys611Ser mutation in RET proto-oncogene in a kindred with medullary thyroid carcinoma and Hirschsprung's disease', *European Journal of Human Genetics,*vol. 11,no. 5, pp. 364–368.

Oliva-Rodriguez, R, Guerrero-Vazquez, R et al. 2011, 'Descriptive study of the clinical behavior of multiple endocrine neoplasia 2A', *Endocrine Reviews,*vol. 32,no. 3.

Paun, DL, Radian, S et al. 2010, 'RET mutation screening in multiple endocrine neoplasia type 2 (MEN2) and medullary thyroid carcinoma patients (MTC) in Romania', *Endocrine Abstracts,*vol. 22, p. P437.

Petri, BJ, van Eijck, CHJ et al. 2009, 'Phaeochromocytomas and sympathetic paragangliomas', *British Journal of Surgery,*vol. 96,no. 12, pp. 1381–1392.

Przybylik-Mazurek, E, Hubalewska-Dydejczyk, A et al. 2010, 'Clinical feature and genetic testing in patients with multiple endocrine neoplasia syndrome type 2', *Endocrine Abstracts,*vol. 20, p. P211.

Rabl, W, Strom, TM et al. 1996, 'C-cell hyperplasia in multiple endocrine neoplasia 2A (MEN2A): molecular genetic diagnosis versus calcitonin levels and immunohistochemistry', *Hormone Research (Basel),*vol. 46,suppl. 2, p. 74.

Raue, F & Frank-Raue, K 1997, 'Biochemical parameters in diagnosis and follow-up of patients with multiple endocrine neoplasia type 2', *Acta Chirurgica Austriaca,*vol. 29,no. 1, pp. 9–11.

Raue, F, Rondot, S et al. 2011, 'Molecular genetic work up for thyroid and parathyroid diseases', *Clinical Chemistry and Laboratory Medicine,*vol. 49, p. S64.

Riccialdelli, L, Biondi, E et al. 2000, 'Follow-up of medullary thyroid carcinoma: our data', *Journal of Endocrinological Investigation,*vol. 23,6 suppl., p. 36.

Roman, S, Mehta, P & Sosa, JA 2009, 'Medullary thyroid cancer: early detection and novel treatments', *Current Opinion in Oncology,* vol.21,no. 1, pp. 5–10.

Shiba, E, Watanabe, T et al. 1996, *Surgical treatment of MEN2A in the era of DNA diagnosis,* World Congress of the International College of Surgeons, Kyoto, Japan, vols 1–2.

Siderova, M, Boyadzhieva, M et al. 2010, 'Six members of a family with multiple endocrine neoplasia type 2A', *Endocrine Abstracts,*vol. 22, p. P399.

Sobrino, PS, Fernandez, CP et al. 2010, 'Clinical features and outcome of sporadic medullary thyroid carcinoma', *Endocrine Abstracts,*vol. 22, p. P425.

Speel, EJM, Aarts, M et al. 2005, 'Conventional and 1p-specific microarray-based CGH analysis of sporadic and syndrome-related pheochromocytomas', *Virchows Archiv – official journal of the European Society of Pathology,*vol. 447,no. 2, pp. 189–190.

Spyer, G, Ellard, S et al. 2006, 'Phenotypic multiple endocrine neoplasia type 213, without endocrinopathy or RET gene mutation: implications for management', *Thyroid,*vol. 16,no. 6, pp. 605–608.

Stewart, S, Watkinson, J et al. 2009, 'Genetics in mainstream medicine: best practice review', *Endocrine Abstracts,*vol. 19, p. P189.

Stipancic, G, La Grasta Sabolic, L et al. 2011, 'Medullary thyroid carcinoma in two children from a family with multiple endocrine neoplasia 2a syndrome: a case report', *Hormone Research in Paediatrics,*vol. 76, p. 277.

Takami, H, Takiguchi-Shirahama, S et al. 1996, 'Germline mutation of the RET proto-oncogene in patients with hereditary medullary thyroid carcinoma', *Proceedings of the American Association for Cancer Research Annual Meeting,*vol. 37,pp. 208–209.

Tang, NL, Yeung, VT et al. 1998, 'Determination of the risk for familial disease in RET mutation-negative patients with medullary thyroid cancer', *Journal of Internal Medicine,*vol. 244, no. 2, pp. 185–187.

Thibodeau, SN, Lindor, NM et al. 1994, 'Mutations in the RET proto-oncogene in sporadic pheochromocytomas', *American Journal of Human Genetics,*vol. 55,3 suppl., p. A71.

Traugott, AL & Moley, JF 2010, 'Multiple endocrine neoplasia type 2: clinical manifestations and management', *Cancer Treatment and Research,*vol. 153, pp. 321–337.

Uchino, S, Noguchi, S et al. 2000, 'Absence of somatic RET gene mutation in sporadic parathyroid tumors and hyperplasia secondary to uremia, and absence of somatic MEN1 gene mutation in MEN2A-associated hyperplasia', *Biomedicine and Pharmacotherapy,*vol. 54,suppl. 1, pp. 100s–103s.

Vanhorne, JB, Harrison, KJ et al. 2001, 'Structure and localization of the GFRA4 locus and investigation of the gene in human thyroid cancer', *American Journal of Human Genetics,* vol.69,no. 4, pp. 357–357.

Vasil'ev, EV, Polyakova, EY et al. 2002, 'Molecular analysis of the RET proto-oncogene on MEN II patients', *European Journal of Human Genetics,*vol. 10, pp. 247–248.

Walshe, P, Seaberg, RM et al. 2008, 'Management of medullary carcinoma of the thyroid during pregnancy in a patient with an intron substitution', *Journal of Otolaryngology-Head & Neck Surgery,*vol. 37,no. 2, pp. E39–E41.

Welander, J, Soderkvist, P & Gimm, O 2011, 'Genetics and clinical characteristics of hereditary pheochromocytomas and paragangliomas', *Endocrine-Related Cancer,*vol. 18,no. 6, pp. R253–R276.

Wheeler, MH 2007, 'Impact of genetic screening on the diagnosis and management of medullary thyroid carcinoma', *Expert Review of Endocrinology and Metabolism,*vol. 2,no. 2, pp. 117–119.

Wieringa, GE, Rafferty, JA et al. 2000, 'The use of real-time PCR technology for the investigation of multiple endocrine neoplasia type 2A', *Journal of Endocrinology,*vol. 167,suppl., OC3.

Wiggins, J 2004, 'Predictive genetic testing for MEN2 in children: a case report', *Journal of Medical Genetics,*vol. 41, pp. S20–S20.

Wohllk, N, Cote, GJ et al. 1996, 'Application of genetic screening information to the management of medullary thyroid carcinoma and multiple endocrine neoplasia type 2', *Endocrinology and Metabolism Clinics of North America,*vol. 25,no. 1, pp. 1–25.

Wolf, A, Willenberg, HS et al. 2005, 'Adrenal pheochromocytoma with contralateral cortisol-producing adrenal adenoma: diagnostic and therapeutic management', *Hormone and Metabolic Research,*vol. 37,no. 6, pp. 391–395.

Yamashita, T, Lihara, M et al. 1997, 'Treatment of minute medullary thyroid carcinoma in multiple endocrine neoplasia 2A families first diagnosed by DNA analysis of RET proto-oncogene mutations: a case report', *Japanese Journal of Clinical Oncology,*vol. 27,no. 1, pp. 42–45.

Yin, M, King, SK et al. 2006, 'Multiple endocrine neoplasia type 2B diagnosed on suction rectal biopsy in infancy: a report of 2 cases', *Pediatric and Developmental Pathology,*vol. 9,no. 1, pp. 56–60.

Yonekawa, H, Sugitani, I et al. 2007, 'A family of multiple endocrine neoplasia type 2A (MEN2A) with Cys630Tyr RET germline mutation: report of a case', *Endocrine Journal,*vol. 54,no. 4, pp. 531–535.

Zbuk, KM & Eng, C 2007, 'Cancer phenomics: RET and PTEN as illustrative models', *Nature Reviews Cancer,*vol. 7,no. 1, pp. 35–45.

# References

Abdelhakim, A, Anne, B, Mohamed, K, Nadia, B, Mohammed, T, Taoufiq, F, Catherine, R & Said, E 2009, 'RET genetic screening in patients with medullary thyroid cancer: The Moroccan experience', *Journal of Cancer Research and Therapeutics*, vol. 5, no. 3, Jul-Sep, pp. 198-202.

ABS 2011, *Population Clock*, Australian Bureau of Statistics, Canberra, viewed 8th July 2011, <<http://www.abs.gov.au/ausstats/abs%40.nsf/94713ad445ff1425ca25682000192af2/1647509ef7e25faaca2568a900154b63?OpenDocument>>.

ABS 2012, *Table 1.9 Life Tables, States, Territories and Australia, 2009-2011*, Australian Bureau of Statistics, Canberra, viewed 10 April 2013,

<<http://www.abs.gov.au/AUSSTATS/abs@.nsf/DetailsPage/3302.0.55.0012009-2011?OpenDocument>>.

AIHW 2010, 'Australian Cancer Incidence and Mortality workbooks', *Thyroid cancer for Australia (ICD 10 C73)*, Australian Institute of Health and Welfare.

Alderazi, Y, Yeh, MW, Robinson, BG, Benn, DE, Sywak, MS, Learoyd, DL, Delbridge, LW & Sidhu, SB 2005, 'Phaeochromocytoma: current concepts', *Med J Aust*, vol. 183, no. 4, Aug 15, pp. 201-204.

Algun, E, Abaci, N, Kosem, M, Kotan, C, Koseoglu, B, Boztepe, H, Sekeroglu, R, Aslan, H, Topal, C, Ayakta, H, Uygan, I, Alagol, F, Erginel-Unaltuna, N & Aksoy, H 2002, 'Clinical characteristics and genetic screening of an extended family with MEN2A', *Journal of Endocrinological Investigation*, vol. 25, no. 7, pp. 603-608.

ALRC 2003, *Essentially Yours: The Protection of Human Genetic Information in Australia (ALRC Report 96)*, Australian Law Reform Commission, Canberra, viewed 11 August 2011, <<http://www.alrc.gov.au/publications/report-96>>.

Alvandi, E, Akrami, SM, Chiani, M, Hedayati, M, Nayer, BN, Tehrani, MR, Nakhjavani, M & Pedram, M 2011, 'Molecular analysis of the RET proto-oncogene key exons in patients with medullary thyroid carcinoma: a comprehensive study of the Iranian population', *Thyroid*, vol. 21, no. 4, Apr, pp. 373-382.

Alvares Da Silva, AM, Maciel, RMB, Dias Da Silva, MR, Toledo, SRC, De Carvalho, MB & Cerutti, JM 2003, 'A Novel Germ-Line Point Mutation in RET Exon 8 (Gly533Cys) in a Large Kindred with Familial Medullary Thyroid Carcinoma', *Journal of Clinical Endocrinology and Metabolism*, vol. 88, no. 11, pp. 5438-5443.

Amar, L, Bertherat, J, Baudin, E, Ajzenberg, C, Bressac-de Paillerets, B, Chabre, O, Chamontin, B, Delemer, B, Giraud, S, Murat, A, Niccoli-Sire, P, Richard, S, Rohmer, V, Sadoul, J-L, Strompf, L, Schlumberger, M, Bertagna, X, Plouin, P-F, Jeunemaitre, X & Gimenez-Roqueplo, A-P 2005, 'Genetic Testing in Pheochromocytoma or Functional Paraganglioma', *Journal of Clinical Oncology*, vol. 23, no. 34, December 1, 2005, pp. 8812-8818.

Ameur, N, Lacroix, L, Roucan, S, Roux, V, Broutin, S, Talbot, M, Dupuy, C, Caillou, B, Schlumberger, M & Bidart, J-M 2009, 'Aggressive inherited and sporadic medullary thyroid carcinomas display similar oncogenic pathways', *Endocrine-Related Cancer*, vol. 16, no. 4, Dec, pp. 1261-1272.

Bandolier 1999, 'Diagnostic testing emerging from the gloom', *Bandolier 70*, vol. 6, no. 12, p. 2.

Bar, M, Friedman, E, Jakobovitz, O, Leibowitz, G, Lerer, I, Abeliovich, D & Gross, DJ 1997, 'Sporadic phaeochromocytomas are rarely associated with germline mutations in the von Hippel-Lindau and RET genes', *Clinical Endocrinology*, vol. 47, no. 6, pp. 707-712.

Bayer, R 1991, 'Public health policy and the AIDS epidemic. An end to HIV exceptionalism?', *N Engl J Med*, vol. 324, no. 21, May 23, pp. 1500-1504.

Beauchamp, TL & Childress, JF 2001, *Principles of Biomedical Ethics*, 5th edn, Oxford University Press, New York.

Beldjord, C, Desclaux-Arramond, F, Raffin-Sanson, M, Corvol, JC, De Keyzer, Y, Luton, JP, Plouin, PF & Bertagna, X 1995, 'The RET protooncogene in sporadic pheochromocytomas: Frequent MEN 2-like mutations and new molecular defects', *Journal of Clinical Endocrinology and Metabolism*, vol. 80, no. 7, pp. 2063-2068.

Bergant, D, Hocevar, M, Besic, N, Glavac, D, Korosec, B & Caserman, S 2006, 'Hereditary medullary thyroid cancer in Slovenia - Genotype-phenotype correlations', *Wiener Klinische Wochenschrift*, vol. 118, no. 13-14, pp. 411-416.

Bihan, H, Baudin, E, Meas, T, Leboulleux, S, Al Ghuzlan, A, Hannoteaux, V, Travagli, JP, Valleur, P, Guillausseau, PJ & Cohen, R 2012, 'Role of prophylactic thyroidectomy in RET 790 familial medullary thyroid carcinoma', *Head and Neck*, vol. 34, no. 4, pp. 493-498.

Blaugrund, JE, Johns, MM, Jr., Eby, YJ, Ball, DW, Baylin, SB, Hruban, RH & Sidransky, D 1994, 'RET proto-oncogene mutations in inherited and sporadic medullary thyroid cancer', *Hum Mol Genet*, vol. 3, no. 10, Oct, pp. 1895-1897.

Boer, A, Szakall Jr, S, Klein, I, Kasler, M, Vincze, B, Tron, L, Godeny, M, Herzog, H, Peter, I & Esik, O 2003, 'FDG PET imaging in hereditary thyroid cancer', *European Journal of Surgical Oncology*, vol. 29, no. 10, pp. 922-928.

Borry, P, Goffin, T, Nys, H & Dierickx, K 2008, 'Attitudes regarding predictive genetic testing in minors: A survey of European clinical geneticists', *American Journal of Medical Genetics Part C: Seminars in Medical Genetics*, vol. 148C, no. 1, pp. 78-83.

Brandi, ML, Gagel, RF, Angeli, A, Bilezikian, JP, Beck-Peccoz, P, Bordi, C, Conte-Devolx, B, Falchetti, A, Gheri, RG, Libroia, A, Lips, CJ, Lombardi, G, Mannelli, M, Pacini, F, Ponder, BA, Raue, F, Skogseid, B, Tamburrano, G, Thakker, RV, Thompson, NW, Tomassetti, P, Tonelli, F, Wells, SA, Jr. & Marx, SJ 2001, 'Guidelines for diagnosis and therapy of MEN type 1 and type 2', *J Clin Endocrinol Metab*, vol. 86, no. 12, Dec, pp. 5658-5671.

Bugalho, MJ, Domingues, R, Santos, JR, Catarino, AL & Sobrinho, L 2007, 'Mutation analysis of the RET proto-oncogene and early thyroidectomy: results of a Portuguese cancer centre', *Surgery*, vol. 141, no. 1, pp. 90-95.

Bugalho, MJ, Frade, JP, Santos, JR, Limbert, E & Sobrinho, L 1997, 'Molecular analysis of the RET proto-oncogene in patients with sporadic medullary thyroid carcinoma: A novel point mutation in the extracellular cysteine-rich domain', *European Journal of Endocrinology*, vol. 136, no. 4, pp. 423-426.

Burke, W, Pinsky, LE & Press, NA 2001, 'Categorizing genetic tests to identify their ethical, legal, and social implications', *Am J Med Genet*, vol. 106, no. 3, Fall, pp. 233-240.

Burke, W & Press, NA 2006, 'Genetics as a tool to improve cancer outcomes: ethics and policy', *Nat Rev Cancer*, vol. 6, no. 6, pp. 476-482.

Burzynski, GM, Nolte, IM, Bronda, A, Bos, KK, Osinga, J, Plaza Menacho, I, Twigt, B, Maas, S, Brooks, AS, Verheij, JB, Buys, CH & Hofstra, RM 2005, 'Identifying candidate Hirschsprung disease-associated RET variants', *Am J Hum Genet*, vol. 76, no. 5, May, pp. 850-858.

Calva, D, O'Dorisio, TM, O'Dorisio, MS, Lal, G, Sugg, S, Weigel, RJ & Howe, JR 2009, 'When Is Prophylactic Thyroidectomy Indicated for Patients with the RET Codon 609 Mutation?', *Annals of Surgical Oncology*, vol. 16, no. 8, Aug, pp. 2237-2244.

Caron, P, Attie, T, David, D, Amiel, J, Brousset, F, Roger, P, Munnich, A & Lyonnet, S 1996, 'C618R mutation in exon 10 of the RET proto-oncogene in a kindred with multiple endocrine neoplasia type 2A and Hirschsprung's disease', *Journal of Clinical Endocrinology and Metabolism*, vol. 81, no. 7, pp. 2731-2733.

Cascon, A, Pita, G, Burnichon, N, Landa, I, Lopez-Jimenez, E, Montero-Conde, C, Leskela, S, Leandro-Garcia, LJ, Leton, R, Rodriguez-Antona, C, Diaz, JA, Lopez-Vidriero, E, Gonzalez-Neira, A, Velasco, A, Matias-Guiu, X, Gimenez-Roqueplo, AP & Robledo, M 2009, 'Genetics of pheochromocytoma and paraganglioma in Spanish patients', *Journal of Clinical Endocrinology and Metabolism*, vol. 94, no. 5, pp. 1701-1705.

Caulfield, T & McGuire, AL 2012, 'Direct-to-Consumer Genetic Testing: Perceptions, Problems, and Policy Responses', in CT Caskey, CP Austin & JA Hoxie (eds), *Annual Review of Medicine, Vol 63*, vol. 63, pp. 23-33.

Centre for Genetics Education 2013, *Family Cancer Services*, NSW Government, viewed 28 February 2013,

<<http://www.genetics.edu.au/Genetics-Services/family-cancer-services#SAFCC>>.

Chang, CF, Yang, WS, Su, YN, Wu, IL & Chang, TC 2009, 'Mutational spectrum of multiple endocrine neoplasia type 2 and sporadic medullary thyroid carcinoma in Taiwan', *Journal of the Formosan Medical Association*, vol. 108, no. 5, pp. 402-408.

Chi, DD, Toshima, K, Donis-Keller, H, Wells Jr, SA, Buenaventura, P, Lillemoe, KD & Thompson, JC 1994, 'Predictive testing for multiple endocrine neoplasia type 2A (MEN 2A) based on the detection of mutations in the RET protooncogene', *Surgery*, vol. 116, no. 2, pp. 124-133.

Chiefari, E, Chiarella, R, Crocetti, U, Tardio, B, Arturi, F, Russo, D, Trischitta, V, Filetti, S & Zingrillo, M 2001, 'A large family with hereditary MTC: Role of RET genetic analysis in differential diagnosis between MEN 2A and FMTC', *Hormone and Metabolic Research*, vol. 33, no. 1, pp. 52-56.

Chiefari, E, Russo, D, Giuffrida, D, Zampa, GA, Meringolo, D, Arturi, R, Chiodini, I, Bianchi, D, Attard, M, Trischitta, V, Bruno, R, Giannasio, P, Pontecorvi, A & Filetti, S 1998, 'Analysis of RET proto-oncogene abnormalities in patients with MEN 2A, MEN 2B, familial or sporadic medullary thyroid carcinoma', *Journal of Endocrinological Investigation*, vol. 21, no. 6, pp. 358-364.

Chung, YJ, Kim, HH, Kim, HJ, Min, YK, Lee, MS, Lee, MK, Kim, KW, Ki, CS, Kim, JW & Chung, JH 2004, 'RET proto-oncogene mutations are restricted to codon 634 and 618 in Korean families with multiple endocrine neoplasia 2A', *Thyroid*, vol. 14, no. 10, Oct, pp. 813-818.

Costante, F, Durante, C, Francis, Z, Schlumberger, M, Filetti, S 2009, 'Determination of calcitonin levels in C-cell disease: clinical interest and potential pitfalls' *Nat Clin Pract Endocrinol Metab*, vol. 5, no. 1, pp. 35-44.

de Groot, JW, Plukker, JT, Wolffenbuttel, BH, Wiggers, T, Sluiter, WJ & Links, TP 2006, 'Determinants of life expectancy in medullary thyroid cancer: age does not matter', *Clin Endocrinol*, vol. 65, no. 6, pp. 729-736.

De Krijger, RR, Van Nederveen, FH, Korpershoek, E, De Herder, WW, Keizer-Schrama, SMPFDM & Dinjens, WNM 2006, 'Frequent genetic changes in childhood pheochromocytomas', in KEG Pacak (ed.), *Pheochromocytoma*, vol. 1073, pp. 166-176.

Decker, RA, Geiger, JD, Cox, CE, Mackovjak, M, Sarkar, M & Peacock, ML 1996, 'Prophylactic surgery for multiple endocrine neoplasia type IIa after genetic diagnosis: Is parathyroid transplantation indicated?', *World Journal of Surgery*, vol. 20, no. 7, pp. 814-821.

Decker, RA, Peacock, ML, Borst, MJ, Sweet, JD & Thompson, NW 1995, 'Progress in genetic screening of multiple endocrine neoplasia type 2A: is calcitonin testing obsolete?', *Surgery*, vol. 118, no. 2, Aug, pp. 257-263; discussion 263-254.

Delatycki, MB 2008, 'Population screening for reproductive risk for single gene disorders in Australia: now and the future', *Twin Res Hum Genet*, vol. 11, no. 4, Aug, pp. 422-430.

Delbridge, L & Robinson, B 1998, 'Genetic and biochemical screening for endocrine disease: III. Costs and logistics', *World Journal of Surgery*, vol. 22, no. 12, pp. 1212-1217.

Diaz, RE & Wohllk, N 2012, 'Multiple endocrine neoplasia: the Chilean experience', *Clinics*, vol. 67, 2012, pp. 7-11.

Diergaarde, B, Bowen, DJ, Ludman, EJ, Culver, JO, Press, N & Burke, W 2007, 'Genetic information: Special or not? Responses from focus groups with members of a health maintenance organization', *American Journal of Medical Genetics Part A*, vol. 143A, no. 6, Mar, pp. 564-569.

Donis-Keller, H 1995, 'The RET proto-oncogene and cancer', *Journal of Internal Medicine*, vol. 238, no. 4, pp. 319-325.

Dos Santos, MACG, De Quedas, EPS, Toledo, RDA, Lourenco Jr, DM & De Toledo, SPA 2007, 'Screening of RET gene mutations in multiple endocrine neoplasia type-2 using conformation sensitive gel electrophoresis (CSGE)', *Arquivos Brasileiros de Endocrinologia e Metabologia*, vol. 51, no. 9, pp. 1468-1476.

Dourisboure, RJ, Belli, S, Domenichini, E, Podesta, EJ, Eng, C & Solano, AR 2005, 'Penetrance and clinical manifestations of non-hotspot germline RET mutation, C630R, in a family with medullary thyroid carcinoma', *Thyroid*, vol. 15, no. 7, pp. 668-671.

Downs, SH & Black, N 1998, 'The feasibility of creating a checklist for the assessment of the methodological quality both of randomised and non-randomised studies of health care interventions', *Journal of Epidemiology and Community Health*, vol. 52, no. 6, June 1, 1998, pp. 377-384.

Dralle, H, Gimm, O, Simon, D, Frank-Raue, K, Gortz, G, Niederle, B, Wahl, RA, Koch, B, Walgenbach, S, Hampel, R, Ritter, MM, Spelsberg, F, Heiss, A, Hinze, R & Hoppner, W 1998, 'Prophylactic thyroidectomy in 75 children and adolescents with hereditary medullary thyroid carcinoma: German and Austrian experience', *World Journal of Surgery*, vol. 22, no. 7, pp. 744-751.

Duthie, K & Bond, K 2011, 'Improving ethics analysis in health technology assessment', *Int J Technol Assess Health Care*, vol. 27, no. 1, Jan, pp. 64-70.

Eisenhofer, G, Lenders, JWM, Timmers, H, Mannelli, M, Grebe, SK, Hofbauer, LC, Bornstein, SR, Tiebel, O, Adams, K, Bratslavsky, G, Linehan, WM & Pacak, K 2011, 'Measurements of Plasma Methoxytyramine, Normetanephrine, and Metanephrine as Discriminators of Different Hereditary Forms of Pheochromocytoma', *Clinical Chemistry*, vol. 57, no. 3, Mar, pp. 411-420.

Elisei, R, Alevizaki, M, Conte-Devolx, B, Frank-Raue, K, Leite, V & Williams, GR 2012, '2012 European Thyroid Association Guidelines for Genetic Testing and Its Clinical Consequences in Medullary Thyroid Cancer', *European Thyroid Journal*, vol. 1, no. 4, pp. 216-231.

Elisei, R, Romei, C, Cosci, B, Agate, L, Bottici, V, Molinaro, E, Sculli, M, Miccoli, P, Basolo, F, Grasso, L, Pacini, F & Pinchera, A 2007, 'RET genetic screening in patients with medullary thyroid cancer and their relatives: experience with 807 individuals at one center', *The Journal of clinical endocrinology and metabolism*, vol. 92, no. 12, pp. 4725-4729.

Eng, C 1999, 'RET proto-oncogene in the development of human cancer', *J Clin Oncol*, vol. 17, no. 1, Jan, pp. 380-393.

Eng, C, Crossey, PA, Mulligan, LM, Healey, CS, Houghton, C, Prowse, A, Chew, SL, Dahia, PL, O'Riordan, JL, Toledo, SP & et al. 1995, 'Mutations in the RET proto-oncogene and the von Hippel-Lindau disease tumour suppressor gene in sporadic and syndromic phaeochromocytomas', *J Med Genet*, vol. 32, no. 12, Dec, pp. 934-937.

Eng, C, Mulligan, LM, Smith, DP, Healey, CS, Frilling, A, Raue, F, Neumann, HP, Ponder, MA & Ponder, BA 1995, 'Low frequency of germline mutations in the RET proto-oncogene in patients with apparently sporadic medullary thyroid carcinoma', *Clin Endocrinol (Oxf)*, vol. 43, no. 1, Jul, pp. 123-127.

Erdogan, MF, Gursoy, A, Gullu, S, Aydintug, S, Kucuk, B, Baskal, N, Kamel, N, Hoppner, W & Erdogan, G 2007, 'Clinical and genetic experience in Turkish multiple endocrine neoplasia type 2 families', *Endocrinologist*, vol. 17, no. 5, pp. 273-277.

Erdogan, MF, Gursoy, A, Ozgen, G, Cakir, M, Bayram, F, Ersoy, R, Algun, E, Cetinarslan, B, Comlekci, A, Kadioglu, P, Balci, MK, Yetkin, I, Kabalak, T & Erdogan, G 2005, 'Ret proto-oncogene mutations in apparently sporadic Turkish medullary thyroid carcinoma patients: Turkmen study', *J Endocrinol Invest*, vol. 28, no. 9, Oct, pp. 806-809.

Erlic, Z, Hoffmann, MM, Sullivan, M, Franke, G, Peczkowska, M, Harsch, I, Schott, M, Gabbert, HE, Valimaki, M, Preuss, SF, Hasse-Lazar, K, Waligorski, D, Robledo, M, Januszewicz, A, Eng, C & Neumann, HPH 2010, 'Pathogenicity of dna variants and double mutations in multiple endocrine neoplasia type 2 and Von Hippel-Lindau syndrome', *Journal of Clinical Endocrinology and Metabolism*, vol. 95, no. 1, pp. 308-313.

Erlic, Z, Rybicki, L, Peczkowska, M, Golcher, H, Kann, PH, Brauckhoff, M, Mussig, K, Muresan, M, Schaffler, A, Reisch, N, Schott, M, Fassnacht, M, Opocher, G, Klose, S, Fottner, C, Forrer, F, Plockinger, U, Petersenn, S, Zabolotny, D, Kollukch, O, Yaremchuk, S, Januszewicz, A, Walz, MK, Eng, C & Neumann, HP 2009, 'Clinical predictors and algorithm for the genetic diagnosis of pheochromocytoma patients', *Clin Cancer Res*, vol. 15, no. 20, pp. 6378-6385.

Etit, D, Faquin, WC, Gaz, R, Randolph, G, DeLellis, RA & Pilch, BZ 2008, 'Histopathologic and clinical features of medullary microcarcinoma and C-cell hyperplasia in prophylactic thyroidectomies for medullary carcinoma: A study of 42 cases', *Archives of Pathology and Laboratory Medicine*, vol. 132, no. 11, pp. 1767-1773.

Feldman, GL, Edmonds, MW, Ainsworth, PJ, Schuffenecker, I, Lenoir, GM, Saxe, AW, Talpos, GB, Roberson, J, Petrucelli, N & Jackson, CE 2000, 'Variable expressivity of familial medullary thyroid carcinoma (FMTC) due to a RET V804M (GTG->ATG) mutation', *Surgery*, vol. 128, no. 1, pp. 93-98.

Fernandez, RM, Navarro, E, Antinolo, G, Ruiz-Ferrer, M & Borrego, S 2006, 'Evaluation of the role of RET polymorphisms/haplotypes as modifier loci for MEN 2, and analysis of the correlation with the type of RET mutation in a series of Spanish patients', *International journal of molecular medicine*, vol. 17, no. 4, pp. 575-581.

Fialkowski, EA & Moley, JF 2006, 'Current approaches to medullary thyroid carcinoma, sporadic and familial', *J Surg Oncol*, vol. 94, no. 8, pp. 737-747.

Fink, M, Weinhausel, A, Niederle, B & Haas, OA 1996, 'Distinction between sporadic and hereditary medullary thyroid carcinoma (MTC) by mutation analysis of the RET proto-oncogene', *International Journal of Cancer*, vol. 69, no. 4, pp. 312-316.

Fitze, G, Schierz, M, Bredow, J, Saeger, HD, Roesner, D & Schackert, HK 2002, 'Various penetrance of familial medullary thyroid carcinoma in patients with RET protooncogene codon 790/791 germline mutations', *Annals of Surgery*, vol. 236, no. 5, pp. 570-575.

Fleming, B 2011, *Multiple Endocrine Neoplasia syndrome - Genetics*, Melbourne, viewed 3rd June 2011, <<http://www.endocrinesurgery.net.au/men-syndrome-genetics/>>.

Frank-Raue, K, Hoppner, W, Buhr, H, Herfarth, C & Raue, F 1997, 'Results and follow-up in eleven MEN 2A gene carriers after prophylactic thyroidectomy', *Experimental and Clinical Endocrinology and Diabetes*, vol. 105, no. SUPPL. 4, pp. 76-78.

Frank-Raue, K, Hoppner, W, Buhr, H, Herfarth, C, Ziegler, R & Raue, F 1996, 'Application of genetic screening in families with hereditary medullary thyroid carcinoma', *Exp Clin Endocrinol Diabetes*, vol. 104 Suppl 4, pp. 108-110.

Frank-Raue, K, Rybicki, LA, Erlic, Z, Schweizer, H, Winter, A, Milos, I, Toledo, SP, Toledo, RA, Tavares, MR, Alevizaki, M, Mian, C, Siggelkow, H, Hufner, M, Wohllk, N, Opocher, G, Dvorakova, S, Bendlova, B, Czetwertynska, M, Skasko, E, Barontini, M, Sanso, G, Vorlander, C, Maia, AL, Patocs, A, Links, TP, De Groot, JW, Kerstens, MN, Valk, GD, Miehle, K, Musholt, TJ, Biarnes, J, Damjanovic, S, Muresan, M, Wuster, C, Fassnacht, M, Peczkowska, M, Fauth, C, Golcher, H, Walter, MA, Pichl, J, Raue, F, Eng, C & Neumann, HP 2011, 'Risk profiles and penetrance estimations in multiple endocrine neoplasia type 2A caused by germline RET mutations located in exon 10', *Human Mutation*, vol. 32, no. 1, pp. 51-58.

Franz, C & Wells Jr, SA 1997, 'Prophylactic thyroidectomy in MEN 2 gene carriers', *Acta Chirurgica Austriaca*, vol. 29, no. 1, pp. 12-14.

Frilling, A, Dralle, H, Eng, C, Raue, F, Broelsch, CE, Moley, J, Evans, D, Clark, O & Norton, J 1995, 'Presymptomatic DNA screening in families with multiple endocrine neoplasia type 2 and familial medullary thyroid carcinoma', *Surgery*, vol. 118, no. 6, pp. 1099-1104.

Frohnauer, MK, Decker, RA, Weber, C, Dralle, H, Moley & Russell, C 2000, 'Update on the MEN 2A c804 RET mutation: Is prophylactic thyroidectomy indicated?', *Surgery*, vol. 128, no. 6, pp. 1052-1058.

Fugazzola, L, Cerutti, N, Mannavola, D, Ghilardi, G, Alberti, L, Romoli, R & Beck-Peccoz, P 2002, 'Multigenerational familial medullary thyroid cancer (FMTC): Evidence for FMTC phenocopies and association with papillary thyroid cancer', *Clinical Endocrinology*, vol. 56, no. 1, pp. 53-63.

Gagel, RF, Cote, GJ, Martins Bugalho, MJ, Boyd, AE, 3rd, Cummings, T, Goepfert, H, Evans, DB, Cangir, A, Khorana, S & Schultz, PN 1995, 'Clinical use of molecular information in the management of multiple endocrine neoplasia type 2A', *J Intern Med*, vol. 238, no. 4, Oct, pp. 333-341.

Genetics Subcommittee of PSTC 2010, *Application for genetic testing for hereditary mutations in the RET gene*.

Giarelli, E 2001, 'Ethical issues in genetic testing. The experiences of one family diagnosed with an inherited cancer syndrome', *Journal of infusion nursing : the official publication of the Infusion Nurses Society*, vol. 24, no. 5, pp. 301-310.

Gil, L, Azanedo, M, Pollan, M, Cristobal, E, Arribas, B, Garcia-Albert, L, Garcia-Saiz, A, Maestro, ML, Torres, A, Menarguez, J & Rojas, JM 2002, 'Genetic analysis of RET, GFr(alpha)1 and GDNF genes in Spanish families with multiple endocrine neoplasia type 2A', *International Journal of Cancer*, vol. 99, no. 2, pp. 299-304.

Gilchrist, DM, Morrish, DW, Bridge, PJ & Brown, JL 2004, 'Cost analysis of DNA-based testing in a large Canadian family with multiple endocrine neoplasia type 2', *Clinical Genetics*, vol. 66, no. 4, pp. 349-352.

Gimm, O, Niederle, BE, Weber, T, Bockhorn, M, Ukkat, J, Brauckhoff, M, Thanh, PN, Frilling, A, Klar, E, Niederle, B & Dralle, H 2002, 'RET proto-oncogene mutations affecting codon 790/791: A mild form of multiple endocrine neoplasia type 2A syndrome?', *Surgery*, vol. 132, no. 6, pp. 952-959.

Gonzalez, B, Salcedo, M, Medrano, ME, Mantilla, A, Quinonez, G, Benitez-Bribiesca, L, Rodriguez-Cuevas, S, Cabrera, L, De Leon, B, Altamirano, N, Tapia, J & Dawson, B 2003, 'RET oncogene mutations in medullary thyroid carcinoma in Mexican families', *Archives of Medical Research*, vol. 34, no. 1, pp. 41-49.

Gosnell, JE, Sywak, MS, Sidhu, SB, Gough, IR, Learoyd, DL, Robinson, BG & Delbridge, LW 2006, 'New era: Prophylactic surgery for patients with multiple endocrine neoplasia-2A', *ANZ Journal of Surgery*, vol. 76, no. 7, pp. 586-590.

Gostin, LO & Hodge, JG 1999, 'Genetic privacy and the law: an end to genetics exceptionalism', *Jurimetrics*, pp. 21-58.

Green, MJ & Botkin, JR 2003, '"Genetic exceptionalism" in medicine: Clarifying the differences between genetic and nongenetic tests', *Annals of Internal Medicine*, vol. 138, no. 7, Apr, pp. 571-575.

Guerrero, IM, Pessoa, CH, Olmedo, DB, Pontes, ER, Matos, LC, Tilli, TM, Barcinski, MA & Gimba, ER 2006, 'Analysis of inherited genetic variants in ret proto-oncogene of Brazilian patients with apparently sporadic medullary thyroid carcinoma', *Thyroid*, vol. 16, no. 1, Jan, pp. 9-15.

Guyetant, S, Josselin, N, Savagner, F, Rohmer, V, Michalak, S & Saint-Andre, JP 2003, 'C-cell hyperplasia and medullary thyroid carcinoma: Clinicopathological and genetic correlations in 66 consecutive patients', *Modern Pathology*, vol. 16, no. 8, pp. 756-763.

Halling, KC, Bufill, JA, Cotter, M, Artz, SA, Carpenter, AB, Schaid, D, Hartman-Adams, H, Chang, HH, Boustany, MM, Fithian, L, Jhiang, SM & Thibodeau, SN 1997, 'Age-Related Disease Penetrance in a Large Medullary Thyroid Cancer Family With a Codon 609 RET Gene Mutation', *Mol Diagn*, vol. 2, no. 4, Dec, pp. 277-286.

Hedayati, M, Nabipour, I, Rezaei-Ghaleh, N & Azizi, F 2006, 'Germline RET mutations in exons 10 and 11: An Iranian survey of 57 medullary thyroid carcinoma cases', *Medical Journal of Malaysia*, vol. 61, no. 5, pp. 564-569.

Heizmann, O, Haecker, FM, Zumsteg, U, Muller, B, Oberholzer, M & Oertli, D 2006, 'Presymptomatic thyroidectomy in multiple endocrine neoplasia 2a', *European Journal of Surgical Oncology*, vol. 32, no. 1, pp. 98-102.

Hernandez, C, Simo, R, Oriola, J & Mesa, J 1997, 'False-positive results of basal and pentagastrin-stimulated calcitonin in non-gene carriers of multiple endocrine neoplasia type 2A', *Thyroid*, vol. 7, no. 1, pp. 51-54.

HGSA 2008, *Pre-symptomatic and Predictive Testing in Children and Young People*, Human Genetics Society of Australasia, viewed 11 August 2011,

<https://[www.hgsa.org.au/website/wp-content/uploads/2009/12/2008-PS02.pdf](http://www.hgsa.org.au/website/wp-content/uploads/2009/12/2008-PS02.pdf)>.

Hofmann, B 2005, 'Toward a procedure for integrating moral issues in health technology assessment', *Int J Technol Assess Health Care*, vol. 21, no. 3, Summer, pp. 312-318.

Iacobone, M, Schiavi, F, Bottussi, M, Taschin, E, Bobisse, S, Fassina, A, Opocher, G & Favia, G 2011, 'Is genetic screening indicated in apparently sporadic pheochromocytomas and paragangliomas?', *Surgery*, vol. 150, no. 6, Dec, pp. 1194-1201.

International *RET* mutation consortium 2006, 'The relationship between specific RET proto-oncogene mutations and disease phenotype in multiple endocrine neoplasia type 2', *JAMA: the journal of the American Medical Association*, vol. 276, no. 19, pp. 1575-1579.

IVD Australia 2010, *Review of the Funding Arrangements for Pathology Services*, Submission 009, IVD Australia, Parramatta.

Januszewicz, A, Neumann, HP, Lon, I, Szmigielski, C, Symonides, B, Kabat, M, Apel, TW, Wocial, B, Lapinski, M & Januszewicz, W 2000, 'Incidence and clinical relevance of RET proto-oncogene germline mutations in pheochromocytoma patients', *J Hypertens*, vol. 18, no. 8, Aug, pp. 1019-1023.

Jindrichova, S, Vcelak, J, Vlcek, P, Neradilova, M, Nemec, J & Bendlova, B 2004, 'Screening of six risk exons of the RET proto-oncogene in families with medullary thyroid carcinoma in the Czech Republic', *Journal of Endocrinology*, vol. 183, no. 2, pp. 257-265.

Jung, J, Uchino, S, Lee, Y & Park, H 2010, 'A Korean family of familial medullary thyroid cancer with Cys618Ser RET germline mutation', *Journal of Korean medical science*, vol. 25, no. 2, pp. 226-229.

Kakudo, K, Carney, JA & Sizemore, GW 1985, 'Medullary carcinoma of thyroid. Biologic behavior of the sporadic and familial neoplasm', *Cancer*, vol. 55, no. 12, pp. 2818-2821.

Kameyama, K, Okinaga, H & Takami, H 2004, 'Clinical manifestations of familial medullary thyroid carcinoma', *Biomedicine and Pharmacotherapy*, vol. 58, no. 6-7, pp. 348-350.

Kameyama, K & Takami, H 2004, 'Medullary thyroid carcinoma: Nationwide Japanese survey of 634 cases in 1996 and 271 cases in 2002', *Endocrine Journal*, vol. 51, no. 5, pp. 453-456.

Karga, HJ, Karayianni, MK, Linos, DA, Tseleni, SC, Karaiskos, KD & Papapetrou, PD 1998, 'Germ line mutation analysis in families with multiple endocrine neoplasia type 2A or familial medullary thyroid carcinoma', *European Journal of Endocrinology*, vol. 139, no. 4, pp. 410-415.

Keatts, EL & Itano, J 2006, 'Medullary thyroid cancer and the impact of genetic testing', *Clin J Oncol Nurs*, vol. 10, no. 5, Oct, pp. 571-575.

Khan, GTR, Julie Glanville, Amanda J Sowden, Jos Kleijnen 2001, 'Undertaking systematic reviews of research on effectiveness: CRD's guidance for those carrying out or commissioning reviews', *University of York: NHS Centre for reviews and dissemination*, vol. 4, no. 4.

Kimura, T, Yoshimoto, K, Yokogoshi, Y & Saito, S 1995, 'Mutations in the cysteine-rich region of the RET proto-oncogene in patients diagnosed as having sporadic medullary thyroid carcinoma', *Endocrine Journal*, vol. 42, no. 4, pp. 517-525.

Kinder, BK 1998, 'Genetic and biochemical screening for endocrine disease: II. Ethical issues', *World J Surg*, vol. 22, no. 12, Dec, pp. 1208-1211.

Kinlaw, WB, Scott, SM, Maue, RA, Memoli, VA, Harris, RD, Daniels, GH, Porter, DM, Belloni, DR, Spooner, ET, Ernesti, MM & Noll, WW 2005, 'Multiple endocrine neoplasia 2A due to a unique C609S RET mutation presents with pheochromocytoma and reduced penetrance of medullary thyroid carcinoma', *Clinical Endocrinology*, vol. 63, no. 6, pp. 676-682.

Kitamura, Y, Goodfellow, PJ, Shimizu, K, Nagahama, M, Ito, K, Kitagawa, W, Akasu, H, Takami, H, Tanaka, S & Wells Jr, SA 1997, 'Novel germline RET proto-oncogene mutations associated with medullary thyroid carcinoma (MTC): Mutation analysis in Japanese patients with MTC', *Oncogene*, vol. 14, no. 25, pp. 3103-3106.

Klein, I, Esik, O, Homolya, V, Szeri, F & Varadi, A 2001, 'Molecular genetic diagnostic program of multiple endocrine neoplasia type 2A and familial medullary thyroid carcinoma syndromes in Hungary', *Journal of Endocrinology*, vol. 170, no. 3, pp. 661-666.

Komminoth, P, Muletta-Feurer, S, Saremaslani, P, Kunz, EK, Matias-Guiu, X, Hiort, O, Schroder, S, Seelentag, WK, Roth, J & Heitz, PU 1995, 'Molecular Diagnosis of Multiple Endocrine Neoplasia (MEN) in Paraffin-Embedded Specimens', *Endocr Pathol*, vol. 6, no. 4, Winter, pp. 267-278.

Korf, BR 1999, 'Genetic testing for patients with renal disease: Procedures, pitfalls, and ethical considerations', *Seminars in Nephrology*, vol. 19, no. 4, pp. 319-326.

Krawczyk, A, Hasse-Lazar, K, Pawlaczek, A, Szpak-Ulczok, S, Krajewska, J, Paliczka-Cieslak, E, Jurecka-Lubieniecka, B, Roskosz, J, Chmielik, E, Ziaja, J, Cierpka, L, Peczkowska, M, Preibisz, A, Januszewicz, A, Otto, M & Jarzab, B 2010, 'Germinal mutations of RET, SDHB, SDHD, and VHL genes in patients with apparently sporadic pheochromocytomas and paragangliomas', *Endokrynologia Polska*, vol. 61, no. 1, pp. 43-48.

Lallier, M, St-Vil, D, Giroux, M, Huot, C, Gaboury, L, Oligny, L & Desjardins, JG 1998, 'Prophylactic thyroidectomy for medullary thyroid carcinoma in gene carriers of MEN2 syndrome', *Journal of Pediatric Surgery*, vol. 33, no. 6, pp. 846-848.

Lau, GSK, Lang, BHH, Lo, CY, Tso, A, Garcia-Barcelo, MM, Tam, PK & Lam, KSL 2009, 'Prophylactic thyroidectomy in ethnic Chinese patients with multiple endocrine neoplasia type 2A syndrome after the introduction of genetic testing', *Hong Kong Medical Journal*, vol. 15, no. 5, pp. 326-331.

Lazzarini, Z 2001, 'What lessons can we learn from the exceptionalism debate (finally)?', *The Journal of law, medicine & ethics : a journal of the American Society of Law, Medicine & Ethics*, vol. 29, no. 2, pp. 149-151.

Learoyd, DL, Gosnell, J, Elston, MS, Saurine, TJ, Richardson, AL, Delbridge, LW, Aglen, JV & Robinson, BG 2005, 'Experience of prophylactic thyroidectomy in multiple endocrine neoplasia type 2A kindreds with RET codon 804 mutations', *Clin Endocrinol (Oxf)*, vol. 63, no. 6, Dec, pp. 636-641.

Learoyd, DL, Marsh, DJ, Richardson, AL, Twigg, SM, Delbridge, L & Robinson, BG 1997, 'Genetic testing for familial cancer: Consequences of ret proto-oncogene mutation analysis in multiple endocrine neoplasia, type 2', *Archives of Surgery*, vol. 132, no. 9, pp. 1022-1025.

Learoyd, DL & Robinson, B 2005, 'Do all patients with RET mutations associated with multiple endocrine neoplasia type 2 require surgery?', *Nature Clinical Practice: Endocrinology and Metabolism*, vol. 1, no. 2, pp. 60-61.

Lecube, A, Hernandez, C, Oriola, J, Galard, R, Gemar, E, Mesa, J & Simo, R 2002, 'V804M RET mutation and familial medullary thyroid carcinoma: Report of a large family with expression of the disease only in the homozygous gene carriers', *Surgery*, vol. 131, no. 5, May, pp. 509-514.

Lendvai, N, Toth, M, Valkusz, Z, Beko, G, Szuecs, N, Csajbok, E, Igaz, P, Kriszt, B, Kovacs, B, Racz, K & Patocs, A 2012, 'Over-representation of the G12S polymorphism of the SDHD gene in patients with MEN2A syndrome', *Clinics*, vol. 67, 2012, pp. 85-89.

Li, H, Robinson, KA, Anton, B, Saldanha, IJ & Ladenson, PW 2011, 'Cost-Effectiveness of a Novel Molecular Test for Cytologically Indeterminate Thyroid Nodules', *Journal of Clinical Endocrinology & Metabolism*, vol. 96, no. 11, November 1, 2011, pp. E1719-E1726.

Liberati, A, Altman, DG, Tetzlaff, J, Mulrow, C, Gotzsche, PC, Ioannidis, JP, Clarke, M, Devereaux, PJ, Kleijnen, J & Moher, D 2009, 'The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: explanation and elaboration', *PLoS Med*, vol. 6, no. 7, Jul 21, p. e1000100.

Lijmer, JG, Mol, BW, Heisterkamp, S, Bonsel, GJ, Prins, MH, van der Meulen, JH & Bossuyt, PM 1999, 'Empirical evidence of design-related bias in studies of diagnostic tests', *JAMA*, vol. 282, no. 11, Sep 15, pp. 1061-1066.

Lindor, NM, Honchel, R, Khosla, S & Thibodeau, SN 1995, 'Mutations in the RET protooncogene in sporadic pheochromocytomas', *J Clin Endocrinol Metab*, vol. 80, no. 2, Feb, pp. 627-629.

Lindskog, S, Nilsson, O, Jansson, S, Nilsson, B, Illerskog, AC, Ysander, L, Ahlman, H & Tisell, LE 2004, 'Phenotypic expression of a family with multiple endocrine neoplasia type 2A due to a RET mutation at codon 618', *British Journal of Surgery*, vol. 91, no. 6, pp. 713-718.

Lips, CJ, Landsvater, RM, Hoppener, JW, Geerdink, RA, Blijham, G, van Veen, JM, van Gils, AP, de Wit, MJ, Zewald, RA, Berends, MJ & et al. 1994, 'Clinical screening as compared with DNA analysis in families with multiple endocrine neoplasia type 2A', *N Engl J Med*, vol. 331, no. 13, Sep 29, pp. 828-835.

Lombardo, F, Baudin, E, Chiefari, E, Arturi, F, Bardet, S, Caillou, B, Conte, C, Dallapiccola, B, Giuffrida, D, Bidart, JM, Schlumberger, M & Filetti, S 2002, 'Familial medullary thyroid carcinoma: Clinical variability and low aggressiveness associated with RET mutation at codon 804', *Journal of Clinical Endocrinology & Metabolism*, vol. 87, no. 4, Apr, pp. 1674-1680.

Lundgren, CI, Delbridg, L, Learoyd, D & Robinson, B 2007, 'Surgical approach to medullary thyroid cancer', *Arq Bras Endocrinol Metabol*, vol. 51, no. 5, Jul, pp. 818-824.

Machens, A, Brauckhoff, M, Gimm, O & Dralle, H 2006, 'Risk-oriented approach to hereditary adrenal pheochromocytoma', in KEG Pacak (ed.), *Pheochromocytoma*, vol. 1073, pp. 417-428.

Machens, A, Brauckhoff, M, Holzhausen, HJ, Thanh, PN, Lehnert, H & Dralle, H 2005, 'Codon-specific development of pheochromocytoma in multiple endocrine neoplasia type 2', *Journal of Clinical Endocrinology and Metabolism*, vol. 90, no. 7, pp. 3999-4003.

Machens, A, Gimm, O, Hinze, R, Hoppner, W, Boehm, BO & Dralle, H 2001, 'Genotype-phenotype correlations in hereditary medullary thyroid carcinoma: Oncological features and biochemical properties', *Journal of Clinical Endocrinology & Metabolism*, vol. 86, no. 3, Mar, pp. 1104-1109.

Mannelli, M, Castellano, M, Schiavi, F, Filetti, S, Giacche, M, Mori, L, Pignataro, V, Bernini, G, Giache, V, Bacca, A, Biondi, B, Corona, G, Di Trapani, G, Grossrubatscher, E, Reimondo, G, Arnaldi, G, Giacchetti, G, Veglio, F, Loli, P, Colao, A, Ambrosio, MR, Terzolo, M, Letizia, C, Ercolino, T & Opocher, G 2009, 'Clinically guided genetic screening in a large cohort of Italian patients with pheochromocytomas and/or functional or nonfunctional paragangliomas', *Journal of Clinical Endocrinology and Metabolism*, vol. 94, no. 5, pp. 1541-1547.

Margraf, RL, Crockett, DK, Krautscheid, PM, Seamons, R, Calderon, FR, Wittwer, CT & Mao, R 2009, 'Multiple endocrine neoplasia type 2 RET protooncogene database: repository of MEN2-associated RET sequence variation and reference for genotype/phenotype correlations', *Hum Mutat*, vol. 30, no. 4, Apr, pp. 548-556.

Marsh, DJ, McDowall, D, Hyland, VJ, Andrew, SD, Schnitzler, M, Gaskin, EL, Nevell, DF, Diamond, T, Delbridge, L, Clifton-Bligh, P & Robinson, BG 1996, 'The identification of false positive responses to the pentagastrin-stimulation test in RET-mutation-negative members of MEN 2A families', *Clinical Endocrinology*, vol. 44, no. 2, pp. 213-220.

Mastroianno, S, Torlontano, M, Scillitani, A, D'Aloiso, L, Verrienti, A, Bonfitto, N, De Bonis, A, D'Agruma, L, Muscarella, LA, Guarnieri, V, Dicembrino, F, Maranghi, M, Durante, C & Filetti, S 2011, 'Coexistence of multiple endocrine neoplasia type 1 and type 2 in a large Italian family', *Endocrine*, vol. 40, no. 3, pp. 481-485.

McMahon, R, Mulligan, LM, Healey, CS, Payne, SJ, Ponder, M, Ferguson-Smith, MA, Barton, DE & Ponder, BA 1994, 'Direct, non-radioactive detection of mutations in multiple endocrine neoplasia type 2A families', *Hum Mol Genet*, vol. 3, no. 4, Apr, pp. 643-646.

Milos, IN, Frank-Raue, K, Wohllk, N, Maia, AL, Pusiol, E, Patocs, A, Robledo, M, Biarnes, J, Barontini, M, Links, TP, de Groot, JW, Dvorakova, S, Peczkowska, M, Rybicki, LA, Sullivan, M, Raue, F, Zosin, I, Eng, C & Neumann, HP 2008, 'Age-related neoplastic risk profiles and penetrance estimations in multiple endocrine neoplasia type 2A caused by germ line RET Cys634Trp (TGC>TGG) mutation', *Endocr Relat Cancer*, vol. 15, no. 4, Dec, pp. 1035-1041.

Morita, H, Daidoh, H, Nagata, K, Okano, Y, Sudoh, Y, Maruyama, T, Sarui, H, Ishizuka, T, Akagi, K, Nishisho, I & Yasuda, K 1996, 'A family of multiple endocrine neoplasia type 2A: Genetic analysis and clinical features', *Endocrine Journal*, vol. 43, no. 1, pp. 25-30.

MSAC 2005, *Guidelines for the assessment of diagnostic technologies*, Commonwealth of Australia, Canberra, ACT,

<<http://www.msac.gov.au/internet/msac/publishing.nsf/Content/guidelines-1>>.

Munson, R 2000, *Intervention and Reflection: Basic Issues in Medical Ethics*, 6th edn, Wadsworth Thomson Learning, Belmont.

Neocleous, V, Skordis, N, Portides, G, Efstathiou, E, Costi, C, Ioannou, N, Pantzaris, M, Anastasiadou, V, Deltas, C & Phylactou, LA 2011, 'RET proto-oncogene mutations are restricted to codon 618 in Cypriot families with multiple endocrine neoplasia 2', *Journal of Endocrinological Investigation*, vol. 34, no. 10, pp. 764-769.

Neumann, HPH, Bausch, B, McWhinney, SR, Bender, BU, Gimm, O, Franke, G, Schipper, J, Klisch, J, Altehoefer, C, Zerres, K, Januszewicz, A & Eng, C 2002, 'Germ-line mutations in nonsyndromic pheochromocytoma', *New England Journal of Medicine*, vol. 346, no. 19, May 9, pp. 1459-1466.

Neumann, HPH, Eng, C, Mulligan, LM, Glavac, D, Zauner, I, Ponder, BAJ, Crossey, PA, Maher, ER & Brauch, H 1995, 'Consequences of direct genetic testing for germline mutations in the clinical management of families with multiple endocrine neoplasia, type II', *Journal of the American Medical Association*, vol. 274, no. 14, pp. 1149-1151.

Nguyen, L, Niccoli-Sire, P, Caron, P, Bastie, D, Maes, B, Chabrier, G, Chabre, O, Rohmer, V, Lecomte, P, Henry, JF, Conte-Devolx, B & French Calcitonin Tumors Study, G 2001, 'Pheochromocytoma in multiple endocrine neoplasia type 2: a prospective study', *European Journal of Endocrinology*, vol. 144, no. 1, Jan, pp. 37-44.

NHMRC 2000, *How to use the evidence: assessment and application of scientific evidence*, National Health and Medical Research Council, Canberra,

<<http://www.nhmrc.gov.au/guidelines/publications/cp69>>.

NHMRC 2008, *NHMRC additional levels of evidence and grades for recommendations for developers of guidelines. Stage 2 consultation. Early 2008 - end June 2009*, National Health and Medical Research Council, Canberra, ACT, viewed 6/8/08 2008, <<http://www.nhmrc.gov.au/_files_nhmrc/file/guidelines/levels_grades05.pdf>>.

NHMRC 2009a, *NHMRC levels of evidence and grades for recommendations for developers of guidelines.*, National Health and Medical Research Council, Canberra, ACT, viewed 1/10/10 2010, <<http://www.nhmrc.gov.au/guidelines/public-consultations/archived-public-consultations/extension-consultation-period-pilot-pr>>.

NHMRC 2009b, *Use and disclosure of genetic information to a patient’s genetic relatives under section 95AA of the Privacy Act 1988 (Cth). Guidelines for health practitioners in the private sector.*, NHMRC, Canberra, viewed 12 September 2011,

<<http://www.nhmrc.gov.au/guidelines/public-consultations/archived-public-consultations/use-and-disclosure-genetic-information>>.

NPAAC 2007, *Classification of Human Genetic Testing*, Commonwealth of Australia, Canberra, viewed 9 November 2012, Publication No. P3-2319,

<<http://www.health.gov.au/internet/main/publishing.nsf/Content/health-npaac-docs-HumanGenTest.htm>>.

Offit, K, Groeger, E, Turner, S, Wadsworth, EA & Weiser, MA 2004, 'The "duty to warn" a patient's family members about hereditary disease risks', *Jama-Journal of the American Medical Association*, vol. 292, no. 12, Sep, pp. 1469-1473.

Offit, K & Thom, P 2007, 'Ethical and legal aspects of cancer genetic testing', *Seminars in Oncology*, vol. 34, no. 5, Oct, pp. 435-443.

Oriola, J, Hernandez, C, Simo, R, Barcelo, A, Casamitjana, R, Vilardell, E & RiveraFillat, F 1996, 'Genetic analysis of seven Mediterranean families with multiple endocrine neoplasia type 2A', *Clinical Endocrinology*, vol. 44, no. 2, Feb, pp. 207-212.

Pacini, F, Romei, C, Miccoli, P, Elisei, R, Molinaro, E, Mancusi, F, Iacconi, P, Basolo, F, Martino, E & Pinchera, A 1995, 'Early treatment of hereditary medullary thyroid carcinoma after attribution of multiple endocrine neoplasia type 2 gene carrier status by screening for ret gene mutations', *Surgery*, vol. 118, no. 6, Dec, pp. 1031-1035.

PaLMS 2011, *RET Gene Testing Ordering Information*, Pacific Laboratory Medicine Services, Sydney, viewed 6th April 2011,

<<http://www.palms.com.au/php/labinfo/info_index.php?tc=RETMUT&site=RNSH&tn=RET%20Gene%20Testing&s=Blood.whole&sid=4>>.

Pasini, B, Rossi, R, Ambrosio, MR, Zatelli, MC, Gullo, M, Gobbo, M, Collini, P, Aiello, A, Pansini, G, Trasforini, G & Uberti, ECD 2002, 'RET mutation profile and variable clinical manifestations in a family with multiple endocrine neoplasia type 2A and Hirschsprung's disease', *Surgery*, vol. 131, no. 4, pp. 373-381.

Paszko, Z, Sromek, M, Czetwertynska, M, Skasko, E, Czapczak, D, Wisniewska, A, Prokurat, A, Chrupek, M, Jagielska, A & Kozlowicz-Gudzinska, I 2007, 'The occurrence and the type of germline mutations in the RET gene in patients with medullary thyroid carcinoma and their unaffected kindred's from Central Poland', *Cancer Investigation*, vol. 25, no. 8, pp. 742-749.

Patocs, A, Karadi, E, Toth, M, Varga, I, Szucs, N, Balogh, K, Majnik, J, Glaz, E & Racz, K 2004, 'Clinical and biochemical features of sporadic and hereditary phaeochromocytomas: An analysis of 41 cases investigated in a single endocrine centre', *European Journal of Cancer Prevention*, vol. 13, no. 5, pp. 403-409.

Patocs, A, Klein, I, Szilvasi, A, Gergics, P, Toth, M, Valkusz, Z, Forizs, E, Igaz, P, Al-Farhat, Y, Tordai, A, Varadi, A, Racz, K & Esik, O 2006, 'Genotype-phenotype correlations in Hungarian patients with hereditary medullary thyroid cancer', *Wiener Klinische Wochenschrift*, vol. 118, no. 13-14, pp. 417-421.

Philips, B, Ball, C, Sackett, D, Badenoch, D, Straus, S, Haynes, B & Dawes, M 2001, *Levels of Evidence* Oxford Centre for Evidence-Based Medicine, vol. May 2001, Oxford Centre for Evidence-Based Medicine, Oxford

Pigny, P, Cardot-Bauters, C, Do Cao, C, Vantyghem, MC, Carnaille, B, Pattou, F, Caron, P, Wemeau, JL & Porchet, N 2009, 'Should genetic testing be performed in each patient with sporadic pheochromocytoma at presentation?', *Eur J Endocrinol*, vol. 160, no. 2, pp. 227-231.

Pinna, G, Orgiana, G, Riola, A, Ghiani, M, Lai, ML, Carcassi, C & Mariotti, S 2007, 'RET proto-oncogene in Sardinia: V804M is the most frequent mutation and may be associated with FMTC/MEN-2A phenotype', *Thyroid*, vol. 17, no. 2, pp. 101-104.

Prazeres, HJ, Rodrigues, F, Figueiredo, P, Naidenov, P, Soares, P, Bugalho, MJ, Lacerda, M, Campos, B & Martins, TC 2006, 'Occurrence of the Cys611Tyr mutation and a novel Arg886Trp substitution in the RET proto-oncogene in multiple endocrine neoplasia type 2 families and sporadic medullary thyroid carcinoma cases originating from the central region of Portugal', *Clinical Endocrinology*, vol. 64, no. 6, pp. 659-666.

Punales, MK, Graf, H, Gross, JL & Maia, AL 2003, 'RET codon 634 mutations in multiple endocrine neoplasia type 2: Variable clinical features and clinical outcome', *Journal of Clinical Endocrinology and Metabolism*, vol. 88, no. 6, pp. 2644-2649.

Quayle, FJ, Benveniste, R, DeBenedetti, MK, Wells, SA & Moley, JF 2004, 'Hereditary medullary thyroid carcinoma in patients greater than 50 years old', *Surgery*, vol. 136, no. 6, pp. 1116-1121.

Quayle, FJ, Fialkowski, EA, Benveniste, R & Moley, JF 2007, 'Pheochromocytoma penetrance varies by RET mutation in MEN 2A', *Surgery*, vol. 142, no. 6, pp. 800-805.

Radien, P, Jeunemaitre, X, Dumont, C, Beldjord, C & Plouin, PF 1997, 'Genetic alterations of the RET proto-oncogene in familial and sporadic pheochromocytomas', *Hormone Research*, vol. 47, no. 4-6, pp. 263-268.

Raue, F & Frank-Raue, K 2009, 'Genotype-phenotype relationship in multiple endocrine neoplasia type 2. Implications for clinical management', *Hormones (Athens)*, vol. 8, no. 1, Jan-Mar, pp. 23-28.

Raue, F & Frank-Raue, K 2010, 'Update multiple endocrine neoplasia type 2', *Fam Cancer*, vol. 9, no. 3, Sep, pp. 449-457.

Raue, F & Frank-Raue, K 2012, 'Genotype-phenotype correlation in multiple endocrine neoplasia type 2', *Clinics (Sao Paulo)*, vol. 67 Suppl 1, pp. 69-75.

Ravine, D & Suthers, G 2012, 'Quality standards and samples in genetic testing', *J Clin Pathol*, vol. 65, no. 5, May, pp. 389-393.

RCPA 2009, *The Catalogue of Genetic Tests and Laboratories*, The Royal College of Pathologists of Australasia, viewed 6th April 2011,

<<http://genetictesting.rcpa.edu.au/component/gene/genetest/labs/AU/255/RET>>.

Rodriguez Gonzalez, JM, Balsalobre, MD, Pomares, F, Torregrosa, NM, Rios, A, Carbonell, P, Glower, G, Sola, J, Tebar, J & Parrilla, P 2002, 'Prophylactic thyroidectomy in MEN 2A syndrome: Experience in a single center', *Journal of the American College of Surgeons*, vol. 195, no. 2, pp. 159-166.

Rogers, WA & Braunack-Mayer, AJ 2004, *Practical Ethics for General Practice*, Oxford University Press, Oxford.

Rohmer, V, Vidal-Trecan, G, Bourdelot, A, Niccoli, P, Murat, A, Wemeau, JL, Borson-Chazot, F, Schvartz, C, Tabarin, A, Chabre, O, Chabrier, G, Caron, P, Rodien, P, Schlumberger, M & Baudin, E 2011, 'Prognostic factors of disease-free survival after thyroidectomy in 170 young patients with a RET germline mutation: a multicenter study of the Groupe Francais d'Etude des Tumeurs Endocrines', *J Clin Endocrinol Metab*, vol. 96, no. 3, Mar, pp. E509-518.

Romei, C, Cosci, B, Renzini, G, Bottici, V, Molinaro, E, Agate, L, Passannanti, P, Viola, D, Biagini, A, Basolo, F, Ugolini, C, Materazzi, G, Pinchera, A, Vitti, P & Elisei, R 2011, 'RET genetic screening of sporadic medullary thyroid cancer (MTC) allows the preclinical diagnosis of unsuspected gene carriers and the identification of a relevant percentage of hidden familial MTC (FMTC)', *Clinical Endocrinology*, vol. 74, no. 2, pp. 241-247.

Rosenthal, M & Diekema, DS 2011, 'Pediatric Ethics Guidelines for Hereditary Medullary Thyroid Cancer', *International Journal of Pediatric Endocrinology*, vol. 2011, no. Feb 6, 2011/02/06/, p. p847603.

Rosenthal, MS & Pierce, HH 2005, 'Inherited medullary thyroid cancer and the duty to warn: Revisiting Pate v. Threlkel in light of HIPAA', *Thyroid*, vol. 15, no. 2, pp. 140-145.

SA Health 2013, *Pathology services*, Government of South Australia, viewed 28 February 2013, <<http://www.sahealth.sa.gov.au/wps/wcm/connect/Public+Content/SA+Health+Internet/Health+services/Pathology+services/>>.

Sanchez Sobrino, P, Paramo Fernandez, C, Gil Gil, P, Mantinan Gil, B, Perez Pedrosa, A, Palmeiro Carballeira, R & Garcia-Mayor, RV 2011, 'Phenotype of the C634Y mutation in the RET Proto-oncogene in MEN2A: Report of a Family', *Endocrinologia y Nutricion*, vol. 58, no. 5, pp. 229-235.

Sanso, GE, Domene, HM, Garcia, R, Pusiol, E, de, M, Roque, M, Ring, A, Perinetti, H, Elsner, B, Iorcansky, S & Barontini, M 2002, 'Very early detection of RET proto-oncogene mutation is crucial for preventive thyroidectomy in multiple endocrine neoplasia type 2 children: presence of C-cell malignant disease in asymptomatic carriers', *Cancer*, vol. 94, no. 2, Jan 15, pp. 323-330.

Schellhaas, E, Koenig, C, Frank-Raue, K, Buhr, H-J & Hotz, HG 2009, 'Long-term outcome of "prophylactic therapy" for familial medullary thyroid cancer', *Surgery*, vol. 146, no. 5, Nov, pp. 906-912.

Schreinemakers, JM, Vriens, MR, Valk, GD, de Groot, JW, Plukker, JT, Bax, KM, Hamming, JF, van der Luijt, RB, Aronson, DC & Borel Rinkes, IH 2010, 'Factors predicting outcome of total thyroidectomy in young patients with multiple endocrine neoplasia type 2: a nationwide long-term follow-up study', *World Journal of Surgery*, vol. 34, no. 4, pp. 852-860.

Schuffenecker, I, Billaud, M, Calender, A, Chambe, B, Ginet, N, Calmettes, C, Modigliani, E & Lenoir, GM 1994, 'RET proto-oncogene mutations in French MEN 2A and FMTC families', *Human Molecular Genetics*, vol. 3, no. 11, pp. 1939-1943.

Schuffenecker, I, Virally-Monod, M, Brohet, R, Goldgar, D, Conte-Devolx, B, Leclerc, L, Chabre, O, Boneu, A, Caron, J, Houdent, C, Modigliani, E, Rohmer, V, Schlumberger, M, Eng, C, Guillausseau, PJ & Lenoir, GM 1998, 'Risk and penetrance of primary hyperparathyroidism in multiple endocrine neoplasia type 2A families with mutations at codon 634 of the RET proto-oncogene. Groupe D'etude des Tumeurs a Calcitonine', *J Clin Endocrinol Metab*, vol. 83, no. 2, Feb, pp. 487-491.

Shan, L, Nakamura, M, Nakamura, Y, Utsunomiya, H, Shou, NH, Jiang, XH, Jing, XF, Yokoi, T & Kakudo, K 1998, 'Somatic mutations in the RET protooncogene in Japanese and Chinese sporadic medullary thyroid carcinomas', *Japanese Journal of Cancer Research*, vol. 89, no. 9, Sep, pp. 883-886.

Sharma, BP & Saranath, D 2011, 'RET gene mutations and polymorphisms in medullary thyroid carcinomas in Indian patients', *Journal of Biosciences*, vol. 36, no. 4, pp. 603-611.

Shifrin, AL, Xenachis, C, Fay, A, Matulewicz, TJ, Kuo, Y-H & Vernick, JJ 2009, 'One hundred and seven family members with the rearranged during transfection V804M proto-oncogene mutation presenting with simultaneous medullary and papillary thyroid carcinomas, rare primary hyperparathyroidism, and no pheochromocytomas: Is this a new syndrome-MEN 2C?', *Surgery*, vol. 146, no. 6, Dec, pp. 998-1005.

Shimotake, T, Iwai, N, Inoue, K, Inazawa, J & Nishisho, I 1996, 'Germline mutations of the RET proto-oncogene in pedigree with MEN type 2A: DNA analysis and its implications for pediatric surgery', *Journal of Pediatric Surgery*, vol. 31, no. 6, pp. 779-781.

Shirahama, S, Ogura, K, Takami, H, Ito, K, Tohsen, T, Miyauchi, A & Nakamura, Y 1998, 'Mutational analysis of the RET proto-oncogene in 71 Japanese patients with medullary thyroid carcinoma', *Journal of human genetics*, vol. 43, no. 2, pp. 101-106.

Shuman, AG, Shaha, AR, Tuttle, RM, Fins, JJ & Morris, LGT 2012, 'Medullary thyroid carcinoma: Ethical issues for the surgeon', *Annals of Surgical Oncology*, vol. 19, no. 7, pp. 2102-2107.

Siggelkow, H, Melzer, A, Nolte, W, Karsten, K, Hoppner, W & Hufner, M 2001, 'Presentation of a kindred with familial medullary thyroid carcinoma and Cys611Phe mutation of the RET proto-oncogene demonstrating low grade malignancy', *European Journal of Endocrinology*, vol. 144, no. 5, pp. 467-473.

Skinner, MA, DeBenedetti, MK, Moley, JF, Norton, JA & Wells Jr, SA 1996, 'Medullary thyroid carcinoma in children with multiple endocrine neoplasia types 2A and 2B', *Journal of Pediatric Surgery*, vol. 31, no. 1, pp. 177-182.

Skinner, MA, Moley, JA, Dilley, WG, Owzar, K, Debenedetti, MK & Wells, SA, Jr. 2005, 'Prophylactic thyroidectomy in multiple endocrine neoplasia type 2A', *N Engl J Med*, vol. 353, no. 11, Sep 15, pp. 1105-1113.

Soini, S 2012, 'Genetic testing legislation in Western Europe - A fluctuating regulatory target', *Journal of Community Genetics*, vol. 3, no. 2, pp. 143-153.

Spinelli, C, Di Giacomo, M, Costanzo, S, Elisei, R & Miccoli, P 2010, 'Role of RET codonic mutations in the surgical management of medullary thyroid carcinoma in pediatric age multiple endocrine neoplasm type 2 syndromes', *Journal of Pediatric Surgery*, vol. 45, no. 8, pp. 1610-1616.

Suthers, GK 2008a, 'Privacy and property issues for a familial cancer service', *Journal of Bioethical Inquiry*, vol. 5, no. 1, pp. 33-37.

Suthers, GK 2008b, *Report of the Australian Genetic Testing Survey 2006*, Royal College of Pathologists of Australasia, Human Genetics Society of Australasia, <[http://www.rcpa.edu.au/static/File/Asset%20library/public%20documents/Media%20Releases/AustralianGeneSurvey2006.pdf>](http://www.rcpa.edu.au/static/File/Asset%20library/public%20documents/Media%20Releases/AustralianGeneSurvey2006.pdf%3e).

Suthers, GK, Armstrong, J, McCormack, J & Trott, D 2006, 'Letting the family know: balancing ethics and effectiveness when notifying relatives about genetic testing for a familial disorder', *J Med Genet*, vol. 43, no. 8, Aug, pp. 665-670.

Suthers, GK, McCusker, EA & Wake, SA 2011, 'Alerting genetic relatives to a risk of serious inherited disease without a patient's consent', *Med J Aust*, vol. 194, no. 8, Apr 18, pp. 385-386.

Szinnai, G, Meier, C, Komminoth, P & Zumsteg, UW 2003, 'Review of Multiple Endocrine Neoplasia Type 2A in Children: Therapeutic Results of Early Thyroidectomy and Prognostic Value of Codon Analysis', *Pediatrics*, vol. 111, no. 2, February 1, 2003, pp. e132-e139.

The President's Council on Bioethics 2008, *The Changing Moral Focus of Newborn Screening*, <<http://bioethics.georgetown.edu/pcbe/reports/newborn_screening/index.html>>.

Therapeutic Goods Administration 2009, *Overview of the new regulatory framework for in vitro diagnostic medical devices (IVDs)*, Commonwealth of Australia, Canberra, <<http://www.tga.gov.au/pdf/ivd-framework-overview.pdf>>.

Toledo, SP, dos Santos, MA, Toledo Rde, A & Lourenco, DM, Jr. 2006, 'Impact of RET proto-oncogene analysis on the clinical management of multiple endocrine neoplasia type 2', *Clinics (Sao Paulo)*, vol. 61, no. 1, Feb, pp. 59-70.

Tsai, MS, Ledger, GA, Khosla, S, Gharib, H & Thibodeau, SN 1994, 'Identification of multiple endocrine neoplasia, type 2 gene carriers using linkage analysis and analysis of the RET proto-oncogene', *Journal of Clinical Endocrinology and Metabolism*, vol. 78, no. 5, pp. 1261-1264.

Uchino, S, Noguchi, S, Adachi, M, Sato, M, Yamashita, H, Watanabe, S, Murakami, T, Toda, M & Murakami, N 1998, 'Novel point mutations and allele loss at the RET locus in sporadic medullary thyroid carcinomas', *Japanese Journal of Cancer Research*, vol. 89, no. 4, pp. 411-418.

Uchino, S, Noguchi, S, Sato, M, Adachi, M, Yamashita, H, Watanabe, S, Murakami, T, Toda, M & Murakami, N 1999, 'Presymptomatic detection and treatment of Japanese carriers of the multiple endocrine neoplasia type 2A gene', *Surgery Today*, vol. 29, no. 9, pp. 862-867.

Vaclavikova, E, Dvorakova, S, Sykorova, V, Bilek, R, Dvorakova, K, Vlcek, P, Skaba, R, Zelinka, T & Bendlova, B 2009, 'RET mutation Tyr791Phe: The genetic cause of different diseases derived from neural crest', *Endocrine*, vol. 36, no. 3, pp. 419-424.

Vestergaard, P, Vestergaard, EM, Brockstedt, H & Christiansen, P 2007, 'Codon Y791F mutations in a large kindred: Is prophylactic thyroidectomy always indicated?', *World Journal of Surgery*, vol. 31, no. 5, May, pp. 996-1001.

Wells Jr, SA & Santoro, M 2009, 'Targeting the RET pathway in thyroid cancer', *Clin Cancer Res*, vol. 15, no. 23, Dec 1, pp. 7119-7123.

Wells Jr, SA & Skinner, MA 1998, 'Prophylactic thyroidectomy, based on direct genetic testing, in patients at risk for the multiple endocrine neoplasia type 2 syndromes', *Experimental and Clinical Endocrinology and Diabetes*, vol. 106, no. 1, pp. 29-34.

Whiting, PF, Rutjes, AW, Westwood, ME, Mallett, S, Deeks, JJ, Reitsma, JB, Leeflang, MM, Sterne, JA & Bossuyt, PM 2011, 'QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies', *Ann Intern Med*, vol. 155, no. 8, Oct 18, pp. 529-536.

Wilcken, B 2011, 'Ethical issues in genetics', *Journal of Paediatrics and Child Health*, vol. 47, no. 9, pp. 668-671.

Wilson, JM & Jungner, YG 1968, '[Principles and practice of mass screening for disease]', *Bol Oficina Sanit Panam*, vol. 65, no. 4, Oct, pp. 281-393.

Winslow, ER, Kodner, IJ & Dietz, DW 2005, 'Ethics and genetic testing', *Seminars in Colon and Rectal Surgery*, vol. 15, no. 3, pp. 186-190.

Woodward, ER, Eng, C, McMahon, R, Voutilainen, R, Affara, NA, Ponder, BAJ & Maher, ER 1997, 'Genetic predisposition to phaeochromocytoma: Analysis of candidate genes GDNF, RET and VHL', *Human Molecular Genetics*, vol. 6, no. 7, pp. 1051-1056.

Wu, SL, Chang, TC, Huang, CN, Chuang, LM & Chang, TJ 1998, 'Germline RET proto-oncogene mutations in two Taiwanese families with multiple endocrine neoplasia type 2A', *Journal of the Formosan Medical Association*, vol. 97, no. 9, pp. 614-618.

Yoshida, S, Imai, T, Kikumori, T, Wada, M, Sawaki, M, Takada, H, Yamada, T, Sato, S, Sassa, M, Uchida, H, Watanabe, R, Kagawa, C, Nakao, A & Kiuchi, T 2009, 'Long Term Parathyroid Function Following Total Parathyroidectomy with Autotransplantation in Adult Patients with MEN2A', *Endocrine Journal*, vol. 56, no. 4, Jul, pp. 545-551.

1. Health Expert Standing Panel (HESP) member and endocrinologist, R Clifton-Bligh, email received on 20 June 2011 [↑](#footnote-ref-1)
2. Horner’s syndrome is a combination of drooping of the eyelid and constriction of the pupil, with redness of the conjunctiva of the eye often present. It indicates a problem with the sympathetic nervous system. [↑](#footnote-ref-2)
3. NSW Government, Centre for Genetics Education (2013). Family Cancer Services. <http://www.genetics.edu.au/Genetics-Services/family-cancer-services#SAFCC> (cited 28/02/13; last updated 07/02/13) [↑](#footnote-ref-3)
4. The initial application proposed the use of genetic testing of the *RET* gene for patients suspected of having MEN2 or in those diagnosed with Hirschsprung’s disease. However, no clinical benefit of using RET testing in Hirschsprung’s disease could be determined through scoping searches of the literature or consultation with clinical experts (HESP members or through public consultation), as there was no ambiguity in the clinical presentation of Hirschsprung’s patients, and so RET testing would be redundant. PASC decided that the assessment of RET testing should not include Hirschsprung’s disease as an indication. [↑](#footnote-ref-4)
5. HESP member and endocrinologist, R Clifton-Bligh, email received on 12 July 2011 [↑](#footnote-ref-5)
6. HESP member and endocrinologist, R Clifton-Bligh, email received on 22 June 2011 [↑](#footnote-ref-6)
7. HESP member and endocrinologist, R Clifton-Bligh, email received on 20 June 2011 [↑](#footnote-ref-7)
8. HESP member and endocrinologist, R Clifton-Bligh, email received on 20 June 2011 [↑](#footnote-ref-8)
9. MESP member and endocrinologist, R Clifton-Bligh, email received on 20 June 2011 [↑](#footnote-ref-9)
10. For further discussion see the Australian Law Reform Commission President’s preliminary account of the 2003 joint inquiry into the protection of human genetic information (Weisbrot 2003). [↑](#footnote-ref-10)
11. Website: [Privacy Legislation Amendment Bill 2006](http://www.comlaw.gov.au/Details/C2006B00109/Explanatory%20Memorandum/Text) (cited 12/03/13) [↑](#footnote-ref-11)
12. Website: [The Royal College of Pathologists of Australasia](http://genetictesting.rcpa.edu.au/component/gene/genetest/labs/AU/255/RET) (cited 12/09/12) [↑](#footnote-ref-12)
13. MESP member and endocrinologist, R Clifton-Bligh, email received on 20 June 2011 [↑](#footnote-ref-13)