

Review**Medullary thyroid carcinoma**

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Summary

Medullary thyroid carcinoma (MTC) arises from parafollicular or C cells that produce calcitonin (CT), and accounts for 5–10% of all thyroid cancers. MTC is hereditary in about 25% of cases. The discovery of a MTC in a patient has several implications: disease extent should be evaluated, pheochromocytoma and hyperparathyroidism should be screened for and whether the MTC is sporadic or hereditary should be determined by a direct analysis of the RET proto-oncogene. In this review, pathological characteristics, tumour markers and genetic abnormalities in MTC are discussed. The diagnostic and therapeutic modalities applied to patients with clinical MTC and those identified with preclinical disease through familial screening are also described. Progresses concerning genetics, initial treatment, follow-up, screening and treatment of pheochromocytoma have permitted an improvement in the long-term outcome. However, there is no effective treatment for distant metastases, and new therapeutic modalities are urgently needed.

Medullary thyroid carcinoma (MTC) arises from parafollicular or C cells that produce calcitonin (CT), and accounts for 5–10% of all thyroid cancers. MTC is hereditary in about 25% of cases (Ball *et al.*, 2000; Gagel & Marx, 2002).

The discovery of a MTC in a patient has several implications: disease extent should be evaluated, pheochromocytoma and hyperparathyroidism should be screened for and whether the MTC is sporadic or hereditary should be determined by a direct analysis of the RET proto-oncogene.

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and those identified with preclinical disease through familial screening are also described.

The normal C cell

C cells migrate during embryonic life probably from the neural crest to the thyroid gland, along with the ultimobranchial body. The parafollicular or C cells are located inside the follicles between the basal layer and the follicular cells. C cells account for about 0.1% of thyroid cells and are most numerous at the junction of the upper third and the lower two-thirds of the thyroid lobes.

Pathology of MTC

MTC is typically located at the junction of the upper third and the lower two-thirds of the thyroid lobes. It is usually firm in consistency and either whitish or red in colour. On histological examination, MTC consists in sheets of spindle-shaped, round or polygonal cells separated by fibrous stroma. The nuclei are usually uniform in shape with rare mitotic figures. The cytoplasm is eosinophilic with a finely granular appearance. Amyloid deposits are seen between tumour cells in about 75% of tumours. In all MTCs, there is positive immunohistochemical staining for calcitonin (CT) and carcinoembryonic antigen (CEA). Mixed MTC are uncommon and combine C cell and follicular features (Hedinger *et al.*, 1988; Rosai *et al.*, 1992).

Atypical MTC should be differentiated from thyroid metastases from a foregut-derived endocrine tumour that produces CT in 15% of cases, mainly when metastases are present; diagnosis relies on clinical presentation, histological features and serum marker levels (Leboulleux *et al.*, 1999; Guignat *et al.*, 2001).

Metastatic dissemination to both central and latero-cervical lymph nodes occurs at similar high frequencies; lymph node metastases are found in 20–30% of patients with an MTC of less than 1 cm in diameter, in 50% of patients with a tumour > 1–4 cm in diameter and in up to 90% of patients with a tumour > 4 cm in diameter (Moley & De Benedetti, 1999; Scollo *et al.*, 2003). Metastases outside the neck may arise in the liver, lungs, bones and less frequently in the brain and skin. Distant metastases are usually diffuse and multiple in involved organs, and generally affect multiple organs.

Histological findings in hereditary MTC

The first histological abnormality observed in hereditary disease is C cell hyperplasia, that is usually detected exclusively through CT immunostaining (Guyétant *et al.*, 1997; LiVolsi, 1997).

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In the general population, C cell hyperplasia is present in 20–30% of thyroid glands, either with a normal histological appearance or with an autoimmune thyroiditis or a follicular tumour. It is more frequent in men than in women, and in adults its prevalence does not change with age (Guyétant *et al.*, 1997).

C cell hyperplasia is present in virtually all patients with hereditary MTC. Hereditary MTCs derived from C cell hyperplasia are bilateral, multicentric neoplasms. In contrast, sporadic MTCs are usually unifocal, but C cell hyperplasia may be present as observed in the general population (Hedinger *et al.*, 1988; Rosai *et al.*, 1992).

Secretory products

Calcitonin

CT is a small monocatenar peptide (32 amino acids). Several molecular forms of CT are present in the blood. Two-site immunoradiometric assay (IRMA) methods, using monoclonal antipeptide antibodies are specific for mature CT (Demers & Spencer, 2003), with normal circulating CT concentration below 7 ng/l. High CT levels are normally found in neonates, followed by an age-related decline from birth to about 1 year of age. Elevated basal CT concentrations are found in subjects with MTC, C cell hyperplasia and in rare subjects without any C cell abnormalities. Also, elevated CT concentrations are found in patients with severe renal insufficiency, during lactation and in 15% of patients with foregut-derived endocrine tumours, mainly when metastatic. In contrast, in patients with C cell disease, increased basal CT level occurs early during the course of the disease and the highest concentrations are observed in patients with the greatest tumour burden. There are, however, exceptions, and there is no strict relationship between basal CT level and tumour burden. Finally, in some patients with minimal C cell disease, basal CT level may be normal but will be abnormally elevated with a provocative test.

Pentagastrin testing is the most widely used stimulation test for CT secretion (Demers & Spencer, 2003). After overnight fasting, basal serum CT is measured. The patient is then given a slow intravenous injection of 0.5 µg pentagastrin per kg body weight, diluted in 5 ml normal saline over 3 min while lying supine. Serum CT is measured again 3 and 5 min after the beginning of the injection. Some authors advocate a bolus injection of pentagastrin, a calcium infusion or a combined calcium–pentagastrin stimulation test. Other calcitonin stimulation tests with TRH or omeprazole have a much lower sensitivity and cannot be recommended as a routine (Vitale *et al.*, 2002).

The pentagastrin stimulation test is contraindicated during pregnancy, and in patients with asthma, coronary disease, severe hypertension or a duodenal ulcer. Side-effects include dizziness, tachycardia or bradycardia, nausea and substernal tightness. Pentagastrin-stimulated CT remains below 10 ng/l in 80% of

normal adult subjects, and in nearly all subjects before the age of 20 years. The peak CT value is observed 3 min later and remains below 30 ng/l in 95% of the normal population. This low peak may be due to C cell hyperplasia but may also occur in the absence of any detectable C cell abnormality. Mean basal as well as stimulated CT values are higher in men than in women. In MTC patients with an elevated basal CT level, the peak value is usually 5–10 fold higher, but in some patients with high basal CT level, the increase may be less important. In patients with an endocrine tumour of another origin, an increase in CT following pentagastrin stimulation is either limited or absent, the peak/basal ratio being < 2. The main clinical interest of pentagastrin stimulation testing is in subjects belonging to an MTC family in whom it may allow to schedule appropriate surgery, and to ascertain cure in MTC patients in whom basal serum CT concentration is undetectable postoperatively.

Measurement of procalcitonin, C-terminal peptide (katecalcin), N-terminal peptide and CGRP does not provide any further information in patients with C cell disease (Ball *et al.*, 2000; Demers & Spencer, 2003).

Carcinoembryonic antigen

Carcinoembryonic antigen (CEA) is produced by neoplastic C cells. Measurement of serum CEA concentration is useful during follow-up because high concentrations or rapidly increasing concentrations indicate disease progression (Mendelsohn *et al.*, 1984).

Other peptides

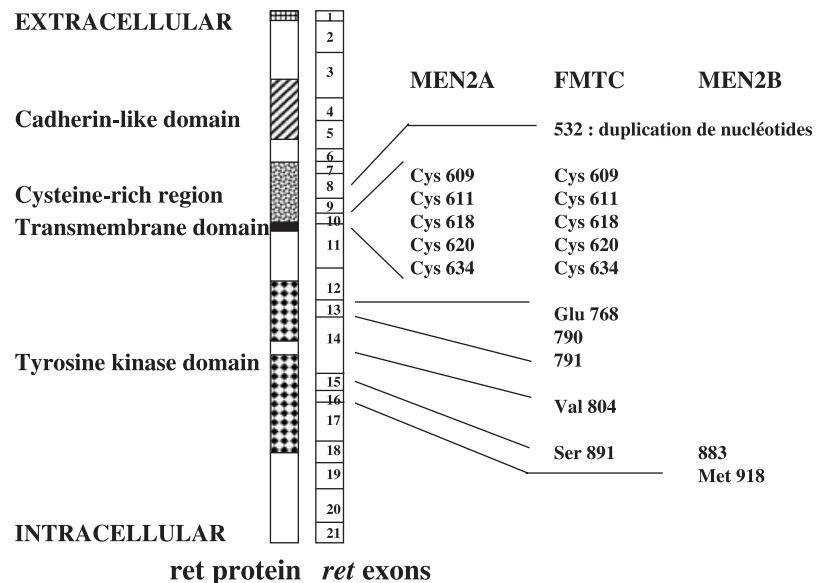
MTC may express a number of genes that are not normally expressed, or expressed at low levels in the normal C cell. The protein products of these genes include somatostatin, proopiomelanocortin, vasoactive intestinal peptide, gastrin-releasing peptide, neurotensin, prostaglandins, kinins, serotonin and histaminase. They can produce clinical syndromes, including Cushing's syndrome, flushing and diarrhoea. Circulating levels of the alpha subunit of the pituitary glycopeptidic hormones are normal in most MTC patients. In MTC patients, chromogranin A (CgA) level may be elevated in the presence of large metastases (Guignat *et al.*, 2001). An elevated CgA level in a patient with a moderately elevated CT level suggests the presence of a pheochromocytoma or of an endocrine tumour of another origin. None of these peptides are comparable to CT in terms of sensitivity.

Genetic abnormalities in MTC

There are four types of clinical MTCs. MTC may be sporadic or hereditary. Hereditary MTC is transmitted as an autosomal dominant trait and may be either transmitted alone (FMTC) or as part of a multiple endocrine neoplasia (MEN) type 2A or 2B syndrome (Table 1).

Table 1 Medullary thyroid carcinoma: clinical findings (from Schlumberger & Pacini, 2003)

Ret mutation	FMTC germinal	MEN-2A germinal	MEN-2B germinal	Sporadic MTC somatic
Exons	10,11,13,14,15	10,11	16 (15)	13,15,16
MTC	100%	100%	100%	100%
Age at clinical occurrence (years)	< 20 → 50	< 20	< 1	< 40
Multicentricity	100%	100%	100%	~30%
Bilaterality	100%	100%	100%	~30%
C cell hyperplasia	100%	100%	100%	rare
Phaeochromocytoma	0%	10–60%	50%	0%
Hyperparathyroidism	0%	5–20%	0%	0%
Cutaneous lichen amyloidosis	0%	< 5%	0%	0%
Ganglioneuromatosis	0%	0%	100%	0%
Dysmorpby	0%	0%	100%	0%

**Fig. 1** The RET gene and protein.

Germline mutations

Germline mutations of the RET proto-oncogene were identified in MEN-2A, FMTC and MEN-2B (Hofstra *et al.*, 1994; Mulligan *et al.*, 1994; Santoro *et al.*, 1995).

The RET gene has 21 exons. It encodes a membrane tyrosine kinase receptor. This membrane-associated receptor is composed of an extracellular domain with a distal cadherin-like region and a cysteine-rich region just outside the membrane, a single transmembrane region and an intracellular tyrosine kinase domain (Fig. 1). The ret receptor couples with GFR α -1 [the glial cell line-derived neurotrophic factor (GDNF) receptor] to form a receptor for GDNF (and other ligands). The addition of GDNF results in dimerization of the receptor system, and this activation

induces autophosphorylation of the tyrosine kinase and activation of downstream signalling pathways (Manie *et al.*, 2001).

Mutations causing MEN-2A affect the cysteine-rich extracellular domain. Each mutation converts a cysteine into another amino acid and activates the tyrosine kinase receptor by ligand-independent dimerization and cross-phosphorylation. These mutations are located in codon 634 (exon 11) or in codons 609, 611, 618 and 620 (exon 10). They account for 98% of all mutations associated with MEN-2A. The most common mutation, accounting for over 80% of all mutations associated with MEN-2A, affects codon 634. A single mutation, a codon 634 cysteine substituted for an arginine (TGC to CGC), accounts for 50% of all MEN-2A mutations.

In about half of FMTC kindreds, mutations affect codons in exon 10 (mainly codons 618 and 620). In a limited number of

families, mutations affect exon 11 (codons 630, 631 or 634). In an increasing proportion of FMTC kindreds, mutations affect exon 13 (codons 768, 790 and 791), exon 14 (codons 804 and 844) and exon 15 (codon 891) in the intracellular domain of the gene. These mutations may interfere with intracellular ATP binding of the tyrosine kinase domain.

Over 95% of kindreds with MEN-2A and FMTC have a mutation of one of these codons. Finally, rare mutations have recently been reported, such as a 9-base pair duplication in exon 8 (codon 532) in a FMTC family. In the rare families in which no mutation of the RET proto-oncogene has been identified, other genetic abnormalities may exist in the RET gene or in other genes (Lima *et al.*, 2003).

In approximately 95% of patients with MEN-2B, a single mutation converting a methionine into a threonine at codon 918 (exon 16) has been identified (Hofstra *et al.*, 1994). This mutation causes alterations in the substrate recognition pocket of the catalytic probe. Other rare intracellular mutations associated with MEN-2B involve codon 883 (exon 15), or a double RET germline mutation at codons 804 and 904 (Menko *et al.*, 2002).

De novo mutations (i.e. a germline mutation that does not exist in parents) have been found in 4–10% of index cases with MEN-2A and FMTC, and in most cases of MEN-2B. It is frequently a *de novo* mutation that is located on the allele inherited from the patient's father (Schuffenecker *et al.*, 1997).

There is a close relationship between the genotype and phenotype (Eng *et al.*, 1996; Yip *et al.*, 2003). The risk of pheochromocytoma is about 50% in subjects with a codon 634 or 918 mutation and 8% in subjects with a mutation in exon 10, and is low in patients with mutations in codons 790, 791, 804 and 891. Most families with parathyroid neoplasia and all families with cutaneous lichen amyloidosis syndrome have a codon 634 mutation. Families with MEN-2A syndrome and Hirschsprung's disease have a mutation in codons 609, 618 or 620.

In subjects with a codon 634 mutation, the cumulative risk of MTC rises linearly with age, between fewer than 2 and 20 years of age. The mean age at diagnosis was 10 years among patients with MTC, and once malignant transformation has taken place, nodal metastases occur an average of 6.6 years later (Machens *et al.*, 2003). The identification of a single child with MTC and lymph node metastases at the age of 6 years guided consensus recommendations for thyroidectomy at the age of 5 years. In subjects with an exon 10 mutation, MTC may occur later.

In subjects with mutations in exons 13, 14 or 15, the mean age at diagnosis was significantly older (16.6 years) and lymph node metastases may occur even later, and C cell disease has a less aggressive course (Frohnauer & Decker, 2000; Lombardo *et al.*, 2002; Machens *et al.*, 2003). There are, however, exceptions, and an MTC was diagnosed at the age of 6 years in a child with an exon 14 mutation. Clinical MTC may occur earlier in the MEN-2B syndrome, usually during the first 6 months of life.

Somatic mutations

Somatic mutations (exclusively in the tumour) in codon 918 of the RET proto-oncogene have been identified in 25–33% of sporadic MTC (Romei *et al.*, 1996). Codon 618, 634, 768, 804 and 883 mutations of the RET gene have been identified in a few tumours. Loss of the normal RET allele or duplication and amplification of the mutant allele contributes to transformation in a high percentage of tumours (Huang *et al.*, 2000).

Modifiers of the transformation process, such as polymorphisms of RET at codons 691 and 836 are poorly known (Robledo *et al.*, 2003).

Clinical syndromes

Sporadic MTC

Sporadic MTC can arise clinically at any age but its incidence peaks during the fourth and sixth decades of life.

Patients with sporadic MTC usually present with a palpable thyroid nodule. Clinical neck lymph node metastases are detected in half of patients and may reveal the disease. Metastases outside the neck, in the liver, lungs or bones are present initially in 20% of cases.

In the presence of a thyroid nodule, several clinical features may prompt the clinician to suspect an MTC: its location in the upper third of a lobe, pain on palpation, a diarrhoeal syndrome and flushes that are more frequently present in patients with a large tumour burden. Ultrasonography typically reveals a hypoechogenic solid nodule with frequent microcalcification, and there may be lymph node abnormalities. A familial history of thyroid tumours, pheochromocytoma, diarrhoea or sudden death should be sought, whenever a MTC is suspected. Fine-needle biopsy has made it possible to diagnose MTC prior to surgery, but cytology may only diagnose malignancy. Virtually all patients with clinically detectable MTC have elevated basal circulating CT, and several prospective studies have suggested that routine measurement of circulating calcitonin should be performed in all patients with thyroid nodules (Pacini *et al.*, 1994; Rieu *et al.*, 1995; Niccoli *et al.*, 1997; Vierhapper *et al.*, 1997). It detected MTC in 0.4% of approximately 10 000 patients with thyroid nodules. In this population, the cytologist made the correct diagnosis of MTC in only 50% of the cases. Compared to a historical group of MTC patients not submitted to CT screening, patients detected through measurement of CT had a significantly lower tumour stage at diagnosis, CT normalized more frequently after surgery and the 10-years outcome was better. However, 30–60% of subjects with elevated basal or pentagastrin-stimulated CT levels had either C cell hyperplasia only or even a normal C cell population. As a consequence, a number of patients with elevated CT level were operated upon with no MTC found at surgery. Thus, further

studies are still needed to evaluate precisely the appropriate strategy in subjects with thyroid nodule, and possibly to measure CT in selected patients only, and also to determine which patients with detectable CT should undergo surgery.

MEN-2A

MEN-2A is a syndrome associating MTC, pheochromocytoma and hyperparathyroidism.

C cell disease

In patients with MEN-2A, there is an age-specific penetrance of MTC and of nodal metastasis. At present, genetic testing is recommended before the age of 5 in all subjects at risk. It identifies gene carriers and also eliminates the likelihood of the risk in 50% of first-degree relatives.

Pheochromocytoma

Pheochromocytoma occurs in approximately 10–50% of MEN-2A gene carriers depending on the mutation.

In prospectively screened families, the diagnosis of clinically significant adrenal medullary disease invariably follows the diagnosis of C cell disease. Thus, in patients with apparently sporadic pheochromocytoma, CT level should be measured and when found elevated a RET mutation should be sought. There is histological progression from adrenal medullary hyperplasia to pheochromocytoma, which is almost always benign and is located in an adrenal gland. Pheochromocytoma is bilateral in 60–80% of cases, but often after an interval of several years. Extra-adrenal paragangliomas are rare.

Clinical symptoms (intermittent jitteriness, headache, palpitation, increased sweating, sense of anxiety) fluctuate. Hypertension is rarely present at an early stage. Pheochromocytoma should therefore be screened routinely by imaging and by measuring plasma catecholamines or 24-h urinary excretion of metanephrine and normetanephrine (Lenders *et al.*, 2002). CgA is frequently elevated in subjects with pheochromocytoma. If biochemical tests are abnormal, ¹²³I-meta-iodobenzylguanidine (MIBG) scintigraphy may demonstrate abnormal uptake in one or both adrenal glands.

Hyperparathyroidism

Hyperparathyroidism occurs in 10–25% of known MEN-2A gene carriers mainly with a codon 634 mutation, usually after the third decade of life. It often consists of parathyroid hyperplasia, with one or more adenomas in older patients. Hyperparathyroidism develops slowly and is usually mild and should be screened by yearly measurements of serum ionized calcium and

eventually PTH 1–84 once a year throughout life in subjects with a codon 634 mutation (Ball *et al.*, 2000; Gagel & Marx, 2002).

Other abnormalities

A pruritic and pigmented papular lesion of the skin on the upper portion of the back has been observed in some MEN-2A families and is called notalgia (Gagel *et al.*, 1989).

Hirschsprung's disease has been observed in rare families with MEN-2A.

Enlarged corneal nerves have been observed in MEN-2A families (Yip *et al.*, 2003).

Familial medullary thyroid carcinoma

A number of FMTC families have been described in which hereditary MTC was the only manifestation observed. Diagnosis of MTC at a later age and a more favourable outcome compared to MTC patients with the codon 634 mutation have been reported in families with mutations in exons 13, 14 or 15.

MEN-2B

MEN-2B is a syndrome associating MTC, pheochromocytoma, ganglioneuromatosis, marfanoid features and muscular and skeletal abnormalities. Hyperparathyroidism is not observed in this syndrome (Vasen *et al.*, 1992; Leboulleux *et al.*, 2002).

MTC was found in MEN-2B children operated during the 6 first months of life, and is frequently associated with extension beyond the thyroid capsule and with lymph node and distant metastases.

Pheochromocytomas are identified in about half of the MEN-2B individuals, and should be managed in a similar manner to that described for MEN-2A.

Marfanoid features, muscular and skeletal abnormalities with a typical facies are frequent. Ganglioneuromatosis includes mucosal neuromas occurring on the distal portion of the tongue, in thickened lips, throughout the intestinal tract and eventually in the urinary tract. Gastrointestinal disorders include colonic cramping, obstructive symptoms and diarrhoea. Hypertrophy of corneal nerves is frequent and is evaluated by slit lamp examination.

Screening for FMTC

The first step in the management of a kindred with MEN-2A or FMTC is to perform a RET proto-oncogene analysis on the index-case (Ball *et al.*, 2000; Brandi *et al.*, 2001; Gagel & Marx, 2002; Fig. 2). As all known mutations are located in seven exons (exons 8, 10, 11, 13–16), polymerase chain reaction amplification and direct DNA sequencing are straightforward and practical. When a mutation is found, it is screened in all first-degree

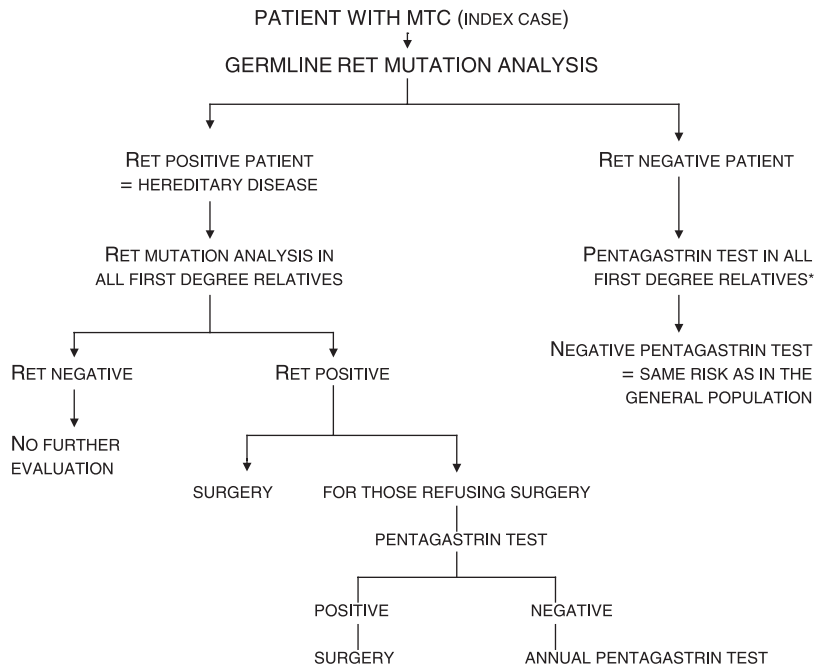


Fig. 2 Genetic testing in the diagnosis and management of multiple endocrine neoplasia. *, if features suggestive of hereditary tumor (from Schlumberger & Pacini, 2003).

relatives to identify gene carriers. This analysis is performed after informed consent has been obtained and should be repeated twice on separate blood samples to exclude the possibility of a sample mix-up or laboratory errors.

In a family with an identified mutation, 50% of first-degree relatives do not carry the mutated gene and the risk of developing the disease in these individuals is similar to that of the general population. These subjects are reassured and require no further evaluation.

In MEN-2A families, a positive genetic test signifies that the affected child has a 90% or greater probability of developing MTC at some point during life. A consensus has been reached to offer a total thyroidectomy to all patients with a germline RET mutation based exclusively on the genetic test result. In experienced hands, the morbidity caused by total thyroidectomy is not worse in children than in adults and treatment with thyroxine is simple and devoid of long-term side-effects, and, in particular, does not stunt growth. Furthermore, family members usually have a positive attitude towards genetic testing, and early surgery in an affected child is usually well accepted by parents (Freyer *et al.*, 2001). At this point in time, however, it is not clear whether long-term studies of children who are thyroidectomized at an earlier age, based exclusively on genetic testing will result in a higher cure rate than the 90% found in children in whom the decision was based on pentagastrin testing. The consensus guidelines have divided hereditary MTC into three different risk categories (Brandi *et al.*, 2001; Cote & Gagel, 2003):

Category 1 – Highest risk. This category includes patients with MEN-2B. When faced with typical phenotypic abnormalities associated with gastrointestinal disorders in a child, the physician should promptly consider the diagnosis of MEN-2B. The diagnosis should then be confirmed by a RET proto-oncogene analysis. Surgery may be decided during the first 6 months of life, and preferably during the first month.

Category 2 – High risk. This category includes cases with a RET mutation in codon 634, who should be submitted to surgery at the age of 5 years. In subjects with an exon 10 mutation, the age at which thyroidectomy should be performed, depends on the mutation, at 5 years for mutations 611, 618 and 620, and possibly later in life for mutation 609.

Category 3 – Intermediate risk. In families with mutations in exons 13, 14 or 15, clinical C cell disease may emerge later in life, usually after the second decade of life. MTC then usually has an indolent course. There are exceptions and there is no consensus regarding the appropriate age for thyroidectomy. Some researchers recommend surgery at the age of 5, some by the age of 10 and for others, pentagastrin testing should be performed every other year, starting around the age of 5, and surgery should be performed only when the basal and/or pentagastrin stimulated CT level becomes abnormal or during the third or even fourth decade of life (Frohnauer & Decker, 2000; Lombardo *et al.*, 2002).

Other screening studies

Screening for pheochromocytoma should be performed in all gene carriers before thyroidectomy because the occurrence of a pheochromocytoma has been associated with practically all types of mutations.

Patients with apparently sporadic MTC

Approximately 5% of patients with apparently sporadic MTC have a germline mutation of the RET proto-oncogene, and consequently hereditary MTC. Cases discovered include several *de novo* germline mutations and members of previously unidentified kindreds, but this is becoming unusual with the routine search for familial disease.

A blood RET proto-oncogene analysis is advocated for all patients with apparently sporadic MTC. If the test is negative, the probability that this individual may be a member of a kindred with hereditary MTC without RET proto-oncogene mutation is less than 1%. In case of a negative RET proto-oncogene analysis, pentagastrin screening in first-degree relatives (preferably adults) is usually performed in the presence of features suggestive of a hereditary tumour (young age at diagnosis, multifocal and bilateral tumours, C cell hyperplasia). If pentagastrin test results are normal in three first-degree relatives, the probability of hereditary MTC drops to an insignificant level (Ball *et al.*, 2000; Gagel & Marx, 2002). In contrast, finding a positive CT response to pentagastrin stimulation in a first-degree relative does not necessarily mean that the MTC is hereditary, because such a response may be related to benign C cell hyperplasia, especially in men (Guyétant *et al.*, 1997).

Initial treatment of MTC

Surgery

The primary treatment of MTC is surgical removal of all neoplastic tissue present in the neck, and should be performed after careful exclusion of a pheochromocytoma. Several studies have shown that survival in patients with MTC is dependent upon the adequacy of the initial surgical procedure. This is accomplished by total thyroidectomy and bilateral lymph node dissection.

Total thyroidectomy is indicated for both the hereditary and sporadic forms of MTC for several reasons. C cell distribution is diffuse and bilateral. Bilateral MTC is observed in 30% of sporadic cases, and C cell disease is bilateral in hereditary MTC. Finally, approximately 5% of apparently sporadic MTC prove to be index cases for hereditary forms signifying that they will inexorably lead to bilateral disease.

Lymph node dissection is indicated during primary surgery because of the notoriously high incidence of regional lymphatic

involvement in MTC (Moley & De Benedetti, 1999; Scollo *et al.*, 2003). In patients with lymph node involvement, central and ipsilateral neck compartments are involved at a similar frequency (75%); involvement of the upper third of the jugulocarotid chain is observed in 20% of patients without involvement of the lower two-thirds. Among patients with unilateral thyroid tumours, 20% had contralateral jugulocarotid involvement and even those with tumours of less than 1 cm in diameter. However, contralateral jugulocarotid involvement was found only in patients with involvement of the central and ipsilateral neck compartments. Findings were similar in patients with either sporadic or hereditary MTC. Thus, dissection of both central neck and of bilateral neck compartments should be performed. Contralateral lymph node dissection may be omitted only in patients with a unilateral thyroid tumour and with no ipsilateral and central lymph node involvement. Dissection of the antero-superior mediastinum should be performed if the central compartment is involved. 'En bloc' dissection is the only procedure recommended.

In gene carriers detected through prospective family screening who have no overt thyroid lesion, total thyroidectomy and dissection of the central compartment and bilateral lower two-thirds of the lateral neck compartments may be curative. Whether lymph node dissection should be routinely performed in young gene carriers with no other evidence of disease, including normal basal or pentagastrin-stimulated CT level is still controversial.

The four parathyroid glands should be identified during surgery; this is easier with methylene blue staining. In patients with a codon 634 mutation, if they appear to be normal, they can be left in place or be implanted in a muscle. In a patient with evidence of hyperparathyroidism, if a gland appears to be enlarged or if an adenoma is clearly present, it should be removed; if hyperplasia is present in all glands, subtotal parathyroidectomy should be performed.

In patients with proven adrenal medullary disease, the abnormal adrenal medullary tissue should be resected prior to thyroidectomy, if possible by laparoscopy. The risk of corticosteroid deficiency has become more life-threatening in MEN-2 patients than a pheochromocytoma and the strategy is to remove only the affected adrenal glands. Approximately 60–80% of the patients who undergo a unilateral adrenalectomy eventually develop a contralateral pheochromocytoma over a period of 10 years. Cortical sparing adrenalectomy is aimed to maintain adrenocortical function, but exposes the patient to an increased risk of local recurrence.

Postoperative management

After total thyroidectomy, thyroxine treatment is given to maintain serum TSH concentration within the normal range.

When measured 5 days after surgery, basal CT concentrations may still be elevated in patients with high preoperative CT

concentrations (Fugazzola *et al.*, 1994) and may normalize later. Similarly, CEA has a long half-life in blood. Consequently, serum CT and CEA are measured 2–3 months after surgery: if basal CT is then undetectable, a pentagastrin stimulation test is performed. Patients with no detectable calcitonin concentrations with pentagastrin stimulation testing are likely to be free of disease. However, serum CT again became detectable during follow-up in 5% of these patients (Modigliani *et al.*, 1998; Kebebew *et al.*, 2000). This underlines the need for long-term clinical and biological follow-up.

In patients with clinical disease, biochemical cure is obtained in 75–90% of patients without lymph node involvement, but even with aggressive surgery, in only 20–30% of patients with lymph node metastases, and rarely (4%) in patients with more than 10 lymph node metastases (Scollo *et al.*, 2003). This clearly demonstrates how treatment at an early stage is a highly significant prognostic factor in MTC patients.

In patients with persistently elevated CT levels, the challenge is finding the site of residual disease (Fig. 3). Tumour localization techniques include ultrasonography of the neck and liver, CT or MRI of the neck, chest and liver, and bone scintigraphy. Isotopic scanning with thallium 201, dimercaptosuccinic acid labelled with ^{99m}Tc , ^{123}I MIBG, somatostatin analogues labelled with Indium 111 (Baudin *et al.*, 1996) or monoclonal antibodies directed against CT or CEA, and even positron emission tomography (PET) with FDG are poorly sensitive. The use of fluorodopamine PET may be more sensitive (Gourgiotis *et al.*, 2003). Liver metastases may be difficult to visualize by these imaging modalities and may be seen only at laparoscopy.

Selective venous sampling catheterization with CT measurements appears to be the most sensitive and specific technique for localizing occult metastatic disease (Wells *et al.*, 1982; Frank-Raue *et al.*, 1992; Abdelmoumene *et al.*, 1994). At the Institut Gustave-Roussy, Villejuif, it is performed in MTC patients with detectable postoperative CT levels below 500 ng/l and no evidence of distant metastases at imaging. Patients with CT levels higher than 500 ng/l are likely to have distant metastases. A gradient in the suprahepatic veins suggests metastatic disease in the liver and may avoid re-operation. In the absence of evidence of distant spread, gradients in the neck or in the mediastinum pinpoint neoplastic foci that need to be removed. Re-operation allowed the removal of neoplastic tissue and a subsequent decrease in CT levels in most patients. It is likely that postoperative CT level will not decrease significantly in patients with thyroid tumour extension beyond the thyroid capsule (pT4) or with more than 10 lymph node metastases or with large mediastinal involvement in the primary operative specimen (Tisell *et al.*, 1986; Abdelmoumene *et al.*, 1994).

This strategy is not unanimously advocated, because the 5-year relapse-free survival was only 60% (Pellegriti *et al.*, 2003) and was similar to that reported with a more conservative approach

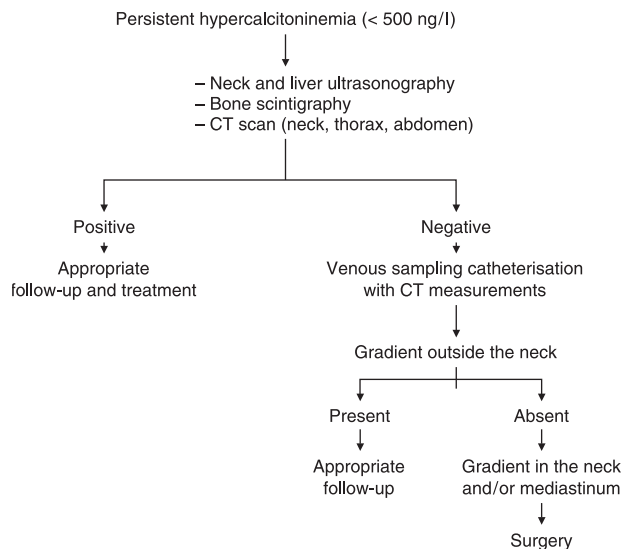


Fig. 3 Follow-up of MTC patients with postoperative hypercalcaemia (from Schlumberger & Pacini, 2003).

(Van Heerden *et al.*, 1990). Therefore, in some centres, only patients with palpable or easily visualized lymph node metastases are operated.

External radiation therapy

External radiation therapy (50 Gy in 25 sessions and 5 weeks) to the neck and mediastinum appeared to be effective in two selected groups of patients: in some patients with inoperable tumours it induced long-term stabilization; it reduced, by a factor of 2–4 the risk of neck recurrences in patients with persistently elevated CT after what appeared to be complete surgery (Schlumberger *et al.*, 1991; Brierley *et al.*, 1996; Fersht *et al.*, 2001).

Long-term follow-up

Follow-up protocol

After initial treatment, follow-up is performed every 6 months for 2 years and annually thereafter. It includes serum CT and CEA measurements (Ball *et al.*, 2000; Gagel & Marx, 2002). The probability of finding a recurrence with imaging techniques increases with higher serum CT or CEA concentrations. CT concentration may fluctuate by 20–30% at short-term intervals. As previously discussed, when serum CT and CEA concentrations are elevated, a complete work-up is performed. In the absence of detectable lesions, another work-up is performed after an interval of 6 months to 1 year.

A detectable CT level is compatible with long-term survival, during which CT may remain stable with time or slowly increases. Among patients with detectable CT, a high serum CEA or a rapid increase in CEA concentration signals disease progression.

Screening for pheochromocytoma and hyperparathyroidism is performed in all patients with hereditary MTC, at an interval that is related to the risk.

Local and regional recurrences

Surgery is the main treatment for local and regional recurrences. The extent of surgery depends on the type of previous surgical procedures and on the nature of the recurrence: if initial surgery was complete, recurrent disease is resected; if the extent of initial surgery was incomplete, the primary surgery protocol is completed.

In the absence of known distant metastases, external radiation therapy to the neck and mediastinum may be indicated after surgery when tumour markers remain elevated, which is generally the case.

Distant metastases

Clinical presentation

Distant metastases are the main cause of MTC-related death. Half are present initially. They are often multiple in involved organs and simultaneously affect multiple organs, such as the liver, lungs and bones (Schlumberger *et al.*, 1991).

Lung metastases are macro- or micro-nodular and are generally diffuse throughout both lungs. Bone metastases are osteolytic or osteoblastic on radiographs and bone scintigraphy shows increased uptake. Liver metastases are hyperechoic at ultrasonography and when they are small, they present the same features as hepatic haemangiomas; CT scan, or preferably MRI with injection of contrast medium, may be helpful to confirm the presence of liver metastases (Dromain *et al.*, 2003).

Survival after the discovery of distant metastases is about 20% at 10 years. A few patients with metastatic disease have been long-term survivors, even without any systemic treatment.

Treatment of metastatic disease

Management of metastatic disease is first orientated towards the relief of symptoms. Loperamide is used to treat diarrhoea, and usual treatment modalities are used for pain.

Bone surgery is indicated in patients with orthopaedic or neurological complications or a high risk of such complications. Surgery may also be indicated in case of a single or a few metastases located in the brain, lungs or liver. However, metastases are often diffuse and multiple.

External radiation therapy is indicated for bone metastases not amenable to surgery, especially when they are painful or located in the spine, the base of the skull, and in pelvic or long bones. External radiation therapy procures rapid relief of bone pain and slower recalcification of lesions. It can also be useful for brain metastases. Embolization of bone metastases and injection of cement can efficiently counteract pain, and can be repeated.

In case of liver metastases from other endocrine tumours, embolization or chemoembolization has proven efficient for both symptoms and tumour masses. It may be beneficial in MTC patients with predominant liver involvement, and is more efficient when metastases are smaller than 3 cm and when liver involvement is less than 30% (Roche *et al.*, 2003).

Treatment with radioactive iodine is pointless, because C cells do not take up radioiodine. Radioactive iodine covalently linked to MIBG offers no significant benefit. Treatment with bi-specific antibodies directed against CEA and DTPA and with DTPA labelled with ¹³¹I appeared poorly effective in patients with large metastases (Kraeber-Bodéré *et al.*, 2003). Treatment with somatostatin analogues labelled with Yttrium 90 or other radionuclides of patients with high metastatic uptake on octreoscan are still under evaluation.

When high-dose doxorubicin (75 mg/m² every 3–4 weeks) is used as a single agent or in association with cisplatin, the response rate is probably less than 20%, and major toxic effects were observed (Schlumberger *et al.*, 1991). Furthermore, all responses were partial and short-lived.

MTC is a well-differentiated endocrine tumour, and MTC patients have been treated with drugs demonstrated to be active in other well-differentiated endocrine tumours. No tumour responses have been obtained with etoposide. Various combinations of 5-fluorouracil (5-FU), dacarbazine, streptozocin, cyclophosphamide, vincristine have produced similar response rates (about 20%) with symptomatic improvement in some patients, but no benefit was found on survival rate (Schlumberger *et al.*, 1995). The addition of doxorubicin did not improve the response rate but resulted in higher toxicity (Nocera *et al.*, 2000). The use of chemotherapy should therefore be limited to the few patients with rapidly progressive metastatic disease, who should then be included in prospective controlled trials. In more typical patients with stable or slowly progressive disease, the regimens currently available appear to offer little benefit, if any.

Somatostatin analogues used either alone or in combination interferon $\alpha 2b$ simply produced an inconstant and transient effect on diarrhoea. They did not produce any tumour regression and their use is not recommended in metastatic MTC patients (Modigliani *et al.*, 1992).

The identification of activating mutations of the RET gene has led to efforts to identify inhibitors of tyrosine kinase or of the message transduction pathway, some of which are currently investigated in clinical trials.

Prognostic factors

Survival rate

The 10-year survival rate in patients with clinical MTC is approximately 65%. Approximately 90% of patients in whom disease is detected at an early stage through familial screening, will remain free of disease.

Prognostic factors

Prognostic factors relevant to outcome in MTC include age at diagnosis, male gender, the initial extent of the disease including lymph node and distant metastases, tumour size, extra-thyroid invasion, vascular invasion, calcitonin immunoreactivity and amyloid staining in tumour tissue (Brierley *et al.*, 1996; Dottorini *et al.*, 1996; Modigliani *et al.*, 1998; Kebebew *et al.*, 2000).

The TNM classification is frequently used and individualizes four stages, but age is not taken into account: stage I, T1N0M0; stage II, T2N0M0; stage III, T3N0M0, T1–T3N1aM0; and stage IV, T1–T3N1bM0, T4M1. Survival rates at 10 years are 100% in stage I, 93% in stage II, 71% in stage III and 20% in stage IV.

In multivariate analysis, only the age of the patient at initial treatment and the disease stage remain significantly independent indicators of survival. This suggests that in routine practice, clinicians attempting to predict outcome in MTC, should take into account not only the disease stage at presentation, as assessed by the pTNM system, but also the age of the patient at diagnosis. A poor outcome is observed in older patients at initial treatment and in cases with an extensive tumour. When the clinical stage of disease is considered, the significant difference in survival between patients with sporadic and hereditary disease disappears.

Among patients with hereditary disease, a multivariate analysis showed that greater age at thyroidectomy and increasing risk group (Brandt *et al.*, 2001) are associated with an increased risk of advanced disease at the time of diagnosis (Yip *et al.*, 2003). This again emphasises the importance of early thyroidectomy.

Calcitonin concentration after initial treatment

Patients with no detectable CT with pentagastrin stimulation testing are likely to be free of disease. In 5% of these patients, serum CT again became detectable during follow-up (Modigliani *et al.*, 1998; Kebebew *et al.*, 2000).

Patients with detectable CT concentrations after initial treatment and who have no other evidence of disease are followed up regularly. With comprehensive follow-up, a recurrence was detected clinically or at imaging in 40% of these patients during the 10 years after initial treatment (Van Heerden *et al.*, 1990; Pellegriti *et al.*, 2003), but a 10-year survival rate of 86% was observed for such patients. Most neck recurrences do in fact benefit from further surgery and most distant metastases found

during follow-up are small at the time of their discovery. They usually progress slowly and are compatible with long survivals, even in the absence of systemic treatment.

Conclusion

Prognosis of MTC has greatly improved with earlier diagnosis of hereditary cases, complete surgical resection of tumour foci and appropriate screening and treatment of pheochromocytoma. There is still a significant proportion of patients with persistent or recurrent disease for whom only palliative treatment modalities are currently available. New agents directed against specific targets will probably be relevant in these patients.

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