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ABSTRACT

MEN2B is a very rare autosomal dominant hereditary tumor syndrome associated with medullary thyroid carcinoma (MTC) in 100% cases, pheochromocytoma in 50% cases, and multiple extra-endocrine features, many of which can be quite disabling. Only few data are available in the literature. The aim of this review is to try to give further insights into the natural history of the disease and to point out the missing evidence that would help clinicians optimize the management of such patients. MEN2B is mainly characterized by the early occurrence of MTC, which led the American Thyroid Association to recommend preventive thyroidectomy before the age of 1. However, as the majority of mutations are *de novo*, improved knowledge of the non-endocrine signs would help to lower the age of diagnosis and improve long-term outcomes. Future large scale studies will be aimed at characterizing more in detail the main characteristics and outcomes of MEN2B.

1. INTRODUCTION

While the first clinical association between pheochromocytoma and medullary thyroid carcinoma (MTC) was described in 1961 by Sipple *et al.* (Sipple 1961), it was only 5 years later that the first description of oral mucosal neuromas in 2 patients with MTC and pheochromocytoma was reported in the English literature (Williams and Pollock 1966). The term multiple endocrine neoplasia type 2 was proposed by Steiner *et al.* in 1968 (Steiner, et al. 1968) and the rare association of hereditary MTC with a mucosal neuroma phenotype was eventually named multiple endocrine neoplasia type 2B (MEN2B) by Chong *et al.* in 1975 (Chong, et al. 1975). The phenotypic specificities of MEN2B compared with MEN2A include prominent extra-endocrine features, a more aggressive presentation of MTC, and the lack of primary hyperparathyroidism. Because of its very low point prevalence (0.9-1.65 per million) and incidence (1.4-2.6 per million live births per year) (Machens, et al. 2013; Mathiesen, et al. 2017b; Znaczko, et al. 2014), MEN2B (OMIM #162300) remains poorly described in the literature. As a consequence, the information provided for MEN2B is derived from large studies that primarily address MEN2A or from large, poorly described genetic studies with little clinical detail. Three large genetic registries of patients with *RET* mutations have identified MEN2B in 21/141 (15%, Germany), 20/246 (8.1%, Italy), and 4/145 (2.8%, EUROMEN) patients (Frank-Raue, et al. 1996; Machens, et al. 2003; Romei, et al. 2010), emphasizing the difficulty in collecting robust data about the outcome of patients with MEN2B. Up to now, the largest published descriptive study of MEN2B was based on only 44 patients (Brauckhoff, et al. 2014). The aim of this review is to give detailed insights

on the genetics, natural history and management of MEN2B, in addition to specific points that should be detailed in future large-scale studies.

2. GENETICS OF MEN2B

MEN2B, similar to MEN2A, is due to autosomal dominant, activating germline mutations of the *RET* proto-oncogene.(Carlson, et al. 1994b; Hofstra, et al. 1994). The *RET* proto-oncogene has 21 exons and encodes a tyrosine-kinase receptor expressed in thyroid parafollicular C cells. Hyperactivation of the receptor leads to the induction of downstream signals responsible for oncogenesis (Mise, et al. 2006). While it has long been held that the parafollicular C cells are of neural crest origin, a recent lineage tracing study suggested that anterior endoderm, and not the neural crest, is the only source of differentiated C cells in mice (Johansson, et al. 2015).

MEN2B is primarily due to a methionine to threonine substitution at codon 918 in the tyrosine-kinase domain of RET (Carlson et al. 1994b; Hofstra et al. 1994). Over 90% of patients present with *de novo* mutations (Brauckhoff et al. 2014), which might be due to a reduced fertility of the patients carrying the MEN2B phenotype. *De novo* mutations have also been found to be of paternal origin and associated with advanced paternal age, suggesting a differential susceptibility of *RET* to mutation in paternally and maternally derived DNA (Carlson, et al. 1994a; Choi, et al. 2012). Other mutations involving codon 883 (*A883F*) or rare double heterozygotes involving mutations of codon 804 in combination with other *RET* mutations have also been reported in patients with a MEN2B phenotype. The increased aggressiveness of *M918T* compared with other *RET* mutations may in part be due to the location of amino-acid 918 within the catalytic core of the RET kinase, where it leads

to increases in ATP binding and the enzymatic activity of the kinase and the ability to be activated without receptor dimerization (Gujral, et al. 2006; Plaza-Menacho, et al. 2014). *In vitro* studies indeed suggest that the M918T-mutated receptor can be activated and autophosphorylated in the absence of any ligand. Finally, the M918T-mutated receptor can be further activated by its endogenous ligand, even if it is already hyperactivated spontaneously, and this might explain the more aggressive clinical presentation, or at least the very early age of MTC diagnosis, in patients with the *M918T* mutation (Gujral et al. 2006).

Notably, the precise mechanisms leading to MTC remain imperfectly understood. While it is obvious that activating *RET* mutations lead to C-cell hyperplasia, the precise mechanisms leading to MTC tumorigenesis remain to be determined. As *RET* is a proto-oncogene, a single germline mutation should be enough to lead to MTC. However, the phenotypic variability of MEN2B patients could be explained by the occurrence of associated somatic mutations that might accelerate the oncogenic process and modify the aggressiveness of MTC (Eng 1996; Mulligan 2014) or differentially regulate targeted genes (Maliszewska, et al. 2013). Recently, a whole gene-expression profile of MEN2A and MEN2B MTC samples identified 3 genes differently deregulated, which might explain the different phenotypes: *NNAT* (tumor suppressor gene), *CD14B* (cell cycle control) and *NTRK3* (tyrosine receptor kinase) (Oczko-Wojciechowska, et al. 2017).

Finally, the underlying *RET* mutation also determines the extra-thyroidal features of MEN2, likely imparting a different susceptibility of adrenal or parathyroid cells to a given mutant receptor; this might explain the lack of primary hyperparathyroidism in MEN2B or, for example, the difference in the prevalence of pheochromocytoma in *M918T* patients compared with patients who harbor exon 10 germline *RET* mutations (Frank-Raue, et al.

2011; Gujral and Mulligan 2006). Of note, the *M918T RET* mutation has also been identified somatically in a high percentage of sporadic MTC (Eng and Mulligan 1997; Marsh, et al. 1996). The fact that MEN2A mutations are uncommon in sporadic MTC might suggest that they have their effects primarily in specific developmental windows, while the *M918T* mutation would be a stronger oncogenic driver with similar effects whether occurring in the germline or somatically.

3. PHENOTYPE OF PATIENTS WITH A GERMLINE *M918T RET* MUTATION

The *M918T RET* mutation is the most frequent etiology (>95%) of MEN2B (Hansford and Mulligan 2000). Patients with MEN2B usually present with very early-onset MTC, a 50% lifetime risk of pheochromocytoma and universal extra-endocrine features, mainly bowel problems due to diffuse intestinal ganglioneuromatosis (constipation, feeding difficulties in infancy, megacolon) and alacrima, both of which can be the earliest presenting features, in addition to mucosal neuromas and a marfanoid body habitus (note that both may not be become clinically apparent until several years of age) (Brauckhoff, et al. 2008; Cohen, et al. 2002; Eng, et al. 1996; O'Riordain, et al. 1994) (Table 1).

a. Medullary Thyroid Carcinoma in M918T-mutated MEN2B

<u>Epidemiology, diagnosis and prognosis</u>

MTC is a malignant neuroendocrine tumor arising from the parafollicular C-cells of the thyroid gland. It develops within the first years of life in 100% of MEN2B patients and remains the leading cause of death in MEN2B. It is usually the first disease to be diagnosed, and the average age of diagnosis of MTC is in the second decade of life, about 10 years

earlier than that seen in individuals with MEN2A (Brandi, et al. 2001; O'Riordain et al. 1994). The size of the tumor at diagnosis is not significantly different between MEN2A and MEN2B (Thosani, et al. 2013). Compared with sporadic MTC, MEN2B-associated MTC is usually multifocal, bilateral, and accompanied by C-cell hyperplasia, the initial step of tumorigenesis in a well-defined oncologic cascade similar to MEN2A (Figure 1) (Mete and Asa 2013; Waguespack, et al. 2011). MTC has been reported as early as the first few months of life and lymph node metastases have been identified within the first year (Zenaty, et al. 2009).

Among MEN2 patients, those with MEN2B have a worse prognosis, with a 10-year survival of 75.5% compared with 97.4% in MEN2A (Modigliani, et al. 1998). Some analyses have suggested that the more aggressive MEN2B phenotype is due to the stage at diagnosis (more advanced disease due to its earlier onset), rather than an intrinsically more aggressive tumor due to the *M918T* mutation. This hypothesis may also hold true for MEN2A, given a recent study showing that high-risk *RET* exon 11 and moderate-risk exon 10 mutations had similar overall survival and time to development of distant metastatic disease after the diagnosis of MTC (Voss, et al. 2017). In the series by Leboulleux *et al.*, based on 18 patients with MEN2B, the overall outcome of MTC did not appear more aggressive than for sporadic or other heritable MTC (Leboulleux, et al. 2002). The majority of patients presented with stage III MTC at diagnosis, and only three had an undetectable plasma calcitonin level after surgery, including one who had a further increase of calcitonin 8 years after surgery. The probability of surgical cure is undoubtedly lower in MEN2B than in MEN2A, but it is likely due to an earlier age of MTC appearance and a delayed clinical diagnosis (Wray, et al. 2008). Of note, 5- and 10-year overall survival rates were 85 and 75%, respectively (Leboulleux et al. 2002). The largest series ever reported on MEN2B was based on 44 patients (including three with inherited mutations, and 41 with *de novo* mutations). The three patients with an inherited

M918T mutation were operated on before 1 year of age and were biochemically cured. In the remaining 41 cases, patients were cured only when the diagnosis was made before 4 years of age. To avoid the risk of incurable MTC, the current American Thyroid Association (ATA) guidelines recommend early thyroidectomy before the age of 1 in children carrying the *M918T* mutation (Wells, et al. 2015). However, except for familial cases, this is difficult to achieve in current practice, as the vast majority of patients with MEN2B carry *de novo* mutations, leading to delayed diagnosis and making early intervention difficult. Improving the outcome of MEN2B is thus mainly based on appropriate education of pediatricians and other health care professionals to recognize the early, nonendocrine manifestations of disease (Brauckhoff et al. 2008; O'Riordain, et al. 1995).

Management

Surgery is the first line treatment of MTC in the majority of patients with MEN2, as it can modify the clinical course and long-term prognosis of the tumor (Brandi et al. 2001). The surgical treatment of clinical disease in hereditary MTC is the same as for sporadic MTC (Wells et al. 2015). However, one of the main issues with MEN2B-related MTC is the very early age of appearance of the MTC precursor lesion, C Cell hyperplasia (Sanso, et al. 2002). Indeed, Zenaty *et al.* reported that bilateral millimetric MTC associated with unilateral lymph node micrometastases were already present in some of the patients with MEN2B operated before the age of 1 (Zenaty et al. 2009).

Prophylactic thyroidectomy (i.e. thyroidectomy in presymptomatic carriers) before the appearance of MTC was first advised in patients with MEN2 more than 20 years ago (Wells, et al. 1994) and then confirmed to be a viable approach 10 years later (Skinner, et al. 2005). Less data is available regarding MEN2B patients in this setting, although it appears that long-term cure can also be achieved with prophylactic thyroidectomy in this group (Waguespack,

et al. 2011). In any case, the oncologic goal of early surgery is not so much to prevent malignancy from occurring in the first place but to remove the thyroid before MTC metastasis occurs, and recent data would suggest that this goal may be achieved in a majority of MEN2B patients treated before age 4 years (Brauckhoff et al. 2014). The reality remains, however, that very few children with MEN2B will actually have a true prophylactic thyroidectomy.

As previously mentioned, in MEN2B, the ATA recommends thyroidectomy before 1 year of age (Wells et al. 2015). At this age, surgical complications can be high (Kluijfhout, et al. 2015), underscoring the need for these surgeries to be performed only by experienced thyroid surgeons in tertiary care centers where multidisciplinary expertise exists. The extent of surgery should be based on the clinical data available, including the presence of thyroid nodules/abnormal lymph nodes on ultrasonographic examination and the basal calcitonin level, recognizing that calcitonin levels are higher in children less than age three (Basuyau, et al. 2004). In the prophylactic setting and in the absence of suspicious lymph nodes, the performance of a central neck dissection should be determined by the treating surgeon (Wells et al. 2015). This decision depends on whether the parathyroid glands can be identified and left *in situ* or autotransplanted, recognizing that the parathyroid glands are very difficult to localize in infants (Moley, et al. 2015). A central neck dissection is recommended for overt clinical disease, and lateral cervical lymph node dissection can also be considered in children with MEN2B and clinically apparent disease or significantly elevated calcitonin levels (Jin and Moley 2016; Waguespack et al. 2011; Wells et al. 2015).

Specific complications of thyroidectomy in very young MEN2 patients remain difficult to determine as few studies have evaluated this specific issue, and previous studies generally included older patients (primarily MEN2A patients in whom thyroidectomy should be

performed around 5 years of age). We can thus only extrapolate the potential consequences of surgery for MEN2B at a very young age. In 50 patients with MEN2A aged less than 19 at the time of surgery, 3 patients presented with permanent hypocalcemia after surgery (Skinner et al. 2005). In another series of 44 children aged 17 years or younger, including three with MEN2B, 4 of 11 patients younger than 3 (44%) suffered from transient hypocalcemia, while 2 had permanent hypocalcemia. This rate was significantly higher than that observed for older patients (Kluijfhout et al. 2015). Finally, in a recent series dealing with a large number of preventive thyroidectomies, permanent hypoparathyroidism occurred in only 1 of 102 children (<1%) operated on by an experienced surgeon with the intent to preserve the parathyroid glands *in situ* with an intact vascular pedicle (Moley et al. 2015).

Post-surgical follow-up should incorporate calcitonin and carcinoembryonic antigen (CEA) levels, cervical ultrasound (if calcitonin positive), and further imaging dictated by the level and trend of calcitonin and CEA (Taieb, et al. 2014; Wells et al. 2015). In the majority of cases, residual disease will primarily be in cervical lymph nodes, but other sites of metastatic disease include intrathoracic lymph nodes, lungs, liver, bones, and rarely the brain. While the specific treatment of advanced, metastatic MTC in the context of MEN2B is beyond the scope of this review, it is however necessary to discuss the two commercially available, molecular targeted small molecule kinase inhibitors approved for the treatment of MTC: vandetanib and cabozantinib. In adults, Wells *et al.* reported in 2012 (Wells, et al. 2012) the results of a randomized, double blind placebo-controlled phase III study on Vandetanib in 331 patients with metastatic MTC. The median progression free survival was significantly different in the vandetanib vs placebo groups (30.5 [predicted] vs 19.3 months, respectively; $p<0.001$). Specifically, in the subgroup of patients with nonhereditary MTC and a somatic

M918T mutation (n=101), an objective response rate of 54.5% was observed. Fox and coworkers also reported in 2013 the first phase I/II clinical trial of vandetanib in 16 children and adolescents with metastatic MTC, 15 of whom had MEN2B and the *M918T* mutation. All 15 patients had a decrease in tumor size, and 7 had a confirmed partial response, including two who had ultimate progression of disease after 44 and 48 cycles of vandetanib. Clinical response was also documented by a mean decline of 59% (35-84) in calcitonin levels (Fox, et al. 2013). Cabozantinib was studied in a randomized, double blind placebo-controlled phase III study (Elisei, et al. 2013) of 330 patients, which showed a significant improvement of median progression-free survival (11.2 vs 4.0 months in the cabozantinib and placebo groups, respectively). Sherman *et al.* subsequently reported, in a subgroup analysis of the cabozantinib phase III trial, that patients with a germline or somatic *M918T* mutation had the greatest progression free survival benefit vs placebo, in comparison with patients with no identified *RET* mutation (Sherman, et al. 2016).

In summary, MTC is the major component of MEN2B. Presumably more aggressive, it appears much earlier than in any other form of hereditary MTC. ATA guidelines suggest thyroidectomy before age 1, a surgical procedure that should be performed only by highly-experienced thyroid surgeons. As the majority of the patients present with *de novo* mutations and hence have a delayed clinical diagnosis, thyroid surgery is rarely curative. For patients presenting with symptomatic or progressive MTC metastases, targeted therapy using the commercially available tyrosine kinase inhibitors has provided hope for better long-term outcomes.

b. Pheochromocytoma in M918T-mutated MEN2B

Limited data are available on the outcomes of pheochromocytoma in MEN2B. This explains why some physicians may fear the possibility of a more aggressive pheochromocytoma clinical outcome in MEN2B compared with MEN2A, as if the outcome was comparable to that of MTC. In MEN2B, the youngest age reported for pheochromocytoma was 12 years old (Nguyen, et al. 2001), and the current ATA guidelines suggest starting routine pheochromocytoma screening (plasma or 24-hour urine fractionated metanephrines) at age 11 years (Wells et al. 2015). There have also been rare cases of adrenal ganglioneuroma identified in MEN2B, a tumor that can be mistaken for pheochromocytoma (Lora, et al. 2005). The largest dedicated study on MEN2B pheochromocytoma was based on 15 patients. The median age at pheochromocytoma diagnosis was 25 years (18-40) compared to 34 years (17-60) in the 70 patients with MEN2A ($p<0.05$) (Thosani et al. 2013). At diagnosis, the median size of pheochromocytoma in the MEN2B patients was smaller than for patients with MEN2A (25 vs 38 mm) ($p<0.01$), but this could be due to stricter surveillance in patients with MEN2B, thus leading to an earlier diagnosis in asymptomatic patients. After a mean follow-up of 57 months, none of the patients presented with delayed metastasis.

c. Extra-endocrine features of M918T-mutated MEN2B

The penetrance of the extra-endocrine features in MEN2B may be incomplete for a given patient, but all MEN2B patients will have one or more of these nonendocrine manifestations (Brauckhoff et al. 2014; O'Riordain et al. 1995). The association of several of these clinical signs in a patient should prompt the astute clinician to measure a calcitonin level and pursue genetic testing for MEN2B. Early recognition of these signs should decrease the age at which thyroidectomy will be performed and theoretically lead to better long-term outcomes.

Marfanoid habitus and other skeletal features

The skeletal phenotype (Figure 2) usually includes a variable expression of a marfanoid body habitus characterized by taller stature, long limbs, a thin elongated face and arachnodactyly of the fingers and toes. Other skeletal abnormalities such as lordosis, kyphosis, scoliosis, joint hypermobility, pes cavus, pectus excavatum (linked to overgrowth of the ribs), high-arched palate, and slipped capital femoral epiphysis (SCFE) can also be associated.

Mucosal neuromas

Mucosal neuromas (Figure 2) may be evident in many MEN2B cases at birth. Although their appearance may be delayed until an older age (Brauckhoff et al. 2008), they occur in the majority of cases within the first decade (Gorlin, et al. 1968). Mucosal neuromas are described as multiple, small soft papules in or around the oral cavity, including the tip of the tongue and lips, the nasal and laryngeal mucosae, and the conjunctivae. The lesions appear as multiple 2–7 mm yellow to white sessile painless nodules. When in enough numbers, labial lesions give a “blubbery” appearance.

Ophthalmological signs

In the series reported by Brauckhoff et al. (Brauckhoff et al. 2014), ocular manifestations (Figure 2) were present in all patients with detailed clinical information, the most frequent sign being alacrima (“tearless crying”). Mild ptosis, eversion of upper eyelids, conjunctival neuromas and prominent corneal nerves are also a component of the MEN2B phenotype (Parker, et al. 2004).

Gastrointestinal signs

Over 40 years ago, Carney *et al.* first focused on the gastrointestinal signs associated with MEN2B (Carney, et al. 1976). Out of 16 patients with likely MEN2B, symptomatic gastrointestinal signs were present in 10 cases at birth or shortly thereafter, while a total of

297 12 patients complained of constipation and/or diarrhea during follow-up. Gastrointestinal
298 signs, especially constipation, usually represent the first nonspecific manifestation of
299 MEN2B; feeding intolerance can be observed in infancy. The gastrointestinal issues are due
300 to diffuse intestinal ganglioneuromatosis and impaired colonic motility that leads to an
301 intestinal pseudo-obstruction and the ultimate development of megacolon. Indeed, in
302 contrast with Hirschsprung disease, in which a part of the bowel (typically the rectosigmoid
303 colon) is aganglionic, megacolon is due to enteric and extrinsic nerve hyperplasia and
304 ganglioneuromas of the submucosal and myenteric plexuses that lead to distension of the
305 colon by loss of normal bowel tone (Gibbons, et al. 2016). Rectal biopsy can lead to the
306 diagnosis of intestinal ganglioneuromatosis (Gfroerer, et al. 2016). Of note, increased
307 secretion of catecholamines (such as would be seen in MEN2B-associated
308 pheochromocytoma) can worsen constipation and rarely lead to toxic megacolon (Thosani,
309 et al. 2015). MEN2B patients can also present with upper GI symptomatology and
310 esophageal manifestations (Cohen et al. 2002; Gibbons et al. 2016). In a previously reported
311 study of 28 MEN2B patients, 39% had difficulty swallowing and 14% had vomiting,
312 suggesting a diagnosis of esophageal abnormalities. Medical management (dietary
313 adjustments, laxatives, fiber supplements) of MEN2B patients can be difficult and some
314 patients (about a third) will ultimately require hospital admission and/or surgery (Cohen et
315 al. 2002; Gibbons et al. 2016). Finally, intestinal manifestations may also be predictive of
316 MTC aggressiveness, as suggested by some authors showing that a worse MTC prognosis
317 was associated with more severe gastrointestinal signs (Brauckhoff, et al. 2004).
318 Other signs: Other associations have also been reported in MEN2B, including coarse facies,
319 tooth malposition, abnormal feet with a long first toe, and an increased space between the
320 1st and the 2nd toe (Martucciello, et al. 2012). Underweight, chronic pain and asthenia, and

delayed puberty can also be seen.

4. THE SPECIFIC FEATURES OF PATIENTS CARRYING THE *M918V RET* MUTATION

Recently, the phenotype of 50 Brazilian patients from eight kindreds carrying the rare *M918V RET* mutation was reported (Martins-Costa, et al. 2016). None of the patients presented with extra-endocrine features characteristics of MEN2B. Age at diagnosis of MTC varied from 24 to 59 years, with incomplete penetrance identified. While only two patients presented with distant metastases at last follow-up, the majority of operated patients (12/20) had lymph node metastases at the time of surgery. None of the patients presented with pheochromocytoma or hyperparathyroidism. Together, these data suggest that this variant is not responsible for a true MEN2B phenotype. Thus, the authors recommended classifying this variant as a moderate risk in the current ATA classification.

5. THE SPECIFIC FEATURES OF PATIENTS CARRYING THE *A883F RET* MUTATION

The codon 883 alanine to phenylalanine (A883F) *RET* mutation is responsible for less than 5% cases of MEN2B. International guidelines on MTC management have recently reclassified the *A883F RET* mutation as a high risk variant, recommending early thyroidectomy by 5 years of age. Until recently, however, only isolated case reports had been published with patients carrying this rare mutation. Mathiesen and coworkers recently reported the outcome of 13 unique *A883F* carriers from 8 different families (Mathiesen, et al. 2017a). Three patients with C-cell hyperplasia and one with normal thyroid pathology who were operated on at a median age of 7.5 years were considered cured at last follow-up. Only 4/11 evaluable patients (two patients without original pathology available) had lymph node metastases at the time of initial surgery (Median age, 20.5 years; range 10-39 years). The

earliest age at distant metastasis was 20 years. Ten-year survival of the nine patients with MTC (median follow-up, 12 years) was 88%. At last follow-up, 38% of the patients had presented with pheochromocytoma (median age at diagnosis, 34 years). Other extra-endocrine features of MEN2B were inconsistently observed. Mucosal neuromas were present in 11 patients and marfanoid body habitus in 6. These data thus suggest that the clinical phenotype and MTC aggressiveness of the *RET A883F* mutation is not as severe as the classical *M918T* mutation. Rather, the onset and disease course of MTC can be highly variable, and very early thyroidectomy (< 1 year old) is not necessary.

6. THE SPECIFIC FEATURES OF PATIENTS CARRYING TANDEM *RET* MUTATIONS

Four reports have described MEN2B phenotypes in patients without a mutation in *RET* codon 883 or 918. Instead, the patients presented with compound *RET* mutations including a common mutation in codon 804 in combination with a second substitution mutation in codon 781, 806, 904 or 905 (Cranston, et al. 2006; Menko, et al. 2002; Miyauchi, et al. 1999; Nakao, et al. 2013). In all these cases, the patient presented with MTC, extra-endocrine features suggesting MEN2B, but no pheochromocytoma. These cases are rare and clearly much less severe than classical MEN2B, but the possibility of a double *RET* mutation should be kept in mind in patients with a MEN2B phenotype but without a classical *RET* 883 or 918 mutation.

7. PERSPECTIVES AND CONCLUSIONS

In contrast with MEN2A, the overall outcome of MEN2B remains relatively poor because MTC, the disease that primarily determines overall prognosis, is not readily modified by early thyroidectomy, given the typical delay in clinical diagnosis (due to the high rate of *de novo*

disease) and the early development of metastatic disease in MEN2B-associated MTC. Nevertheless, total thyroidectomy and clinically-appropriate compartment-oriented neck dissection should still be considered for most patients with MEN2B, whatever the age at diagnosis. The one exception might be the patient with an extensive distant metastatic burden (in whom there is no immediate concern about symptomatic progression in the neck) who might benefit from upfront systemic therapy. The difficulty in properly evaluating the clinical features and outcomes of MEN2B relates to the low number of patients reported. This explains why, despite its first description over 50 years ago, the natural history of MEN2B remains imprecise. In particular, the outcome of patients undergoing surgery at different ages seems highly variable.

Our understanding of the clinical spectrum, the underlying genetic causes and the optimal management of patients MEN2B has progressed, but there is still much that needs to be clarified. Future retrospective studies will require a large-scale, international network of specialized centers, given the rarity of MEN2B, and should be aimed at:

- Educating pediatricians and other primary care providers: improving their knowledge of the extra-endocrine features of MEN2B to allow earlier patient identification and management. As such, better description of the non-endocrine features, specifically their prevalence and clinical presentation, will be of major help. Though MEN2B is a rare disease, each provider should be aware of the red flags (marfanoid habitus and other skeletal features, mucosal neuromas, constipation, alacrima) that will lead to the clinical and genetic diagnosis of MEN2B.
- Improving understanding of the outcomes of MTC in MEN2B, defining the predictors of a more aggressive clinical course, and determining how patient outcomes differ depending on the age at initial surgery. While MEN2B has always been considered a

fatal disease, better knowledge of the natural history of MTC may inform the development of new treatments and provide better understanding as to the timing of therapy. Interestingly, some patients with MEN2B, despite diagnosis at a later age, can present with stable disease for many years whereas others who were identified and operated on at a younger age will develop progressive metastatic disease on long-term follow-up. The major advances introduced by tyrosine kinase inhibitors will be more fully exploited when the outcome of operated MEN2B MTC is better known.

- Improving understanding of the outcome of MEN2B pheochromocytoma. The hypothesis that the aggressiveness of MTC could be extrapolated to the aggressiveness of pheochromocytoma has been raised. The natural history of pheochromocytoma in MEN2B and the optimal surgical approach should be major goals of future studies.

Finally, future prospective studies should be aimed at understanding the reasons for the wide phenotypic variability observed in patients with MEN2, especially MEN2B. Transcriptomic studies have begun to suggest reasons why the phenotype might be different depending on the mutation. MEN2 is also characterized by an intra-familial variability that may be explained by modifying genes or polymorphisms. A complete dataset of these modifiers will help tailor the treatment and the follow-up of such patients. In conclusion, despite the rather old age of MEN2B, there are still lots of things to explore before we achieve a more complete understanding of the pathophysiology of this rare disease.

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642

TABLE 1: Multiple Endocrine Neoplasia Type 2A and type 2B phenotypic characteristics and lifetime risk of development.

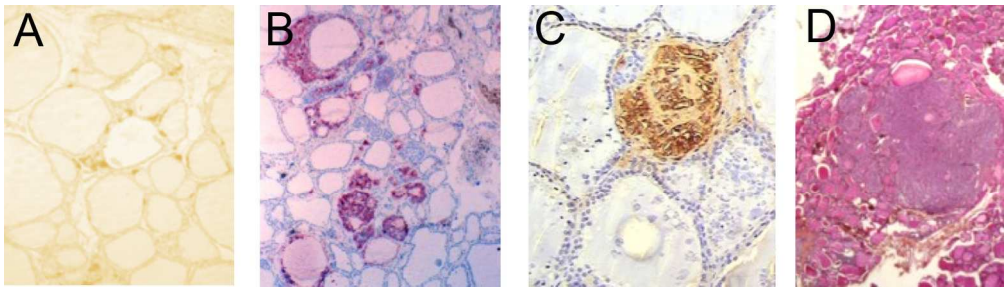
	Risk of Medullary thyroid carcinoma	**Pheochromocytoma**	**Primary Hyperpara-thyroidism**	**Other extra-endocrine signs**
MEN2A	90-100%	0-50% (risk depends on genotype)	0-20% (risk depends on genotype)	<5% (Hirschprung disease, Cutaneous lichen amyloidosis)
MEN2B (M918T, A883F, Tandem mutations)	100%	50% (risk depends on genotype)	0%	100% Gastrointestinal Ophthalmological Skeletal Manifestations Mucosal neuromas

For further details on the clinical manifestations of MEN2B, please refer to the text.

LEGEND TO FIGURES

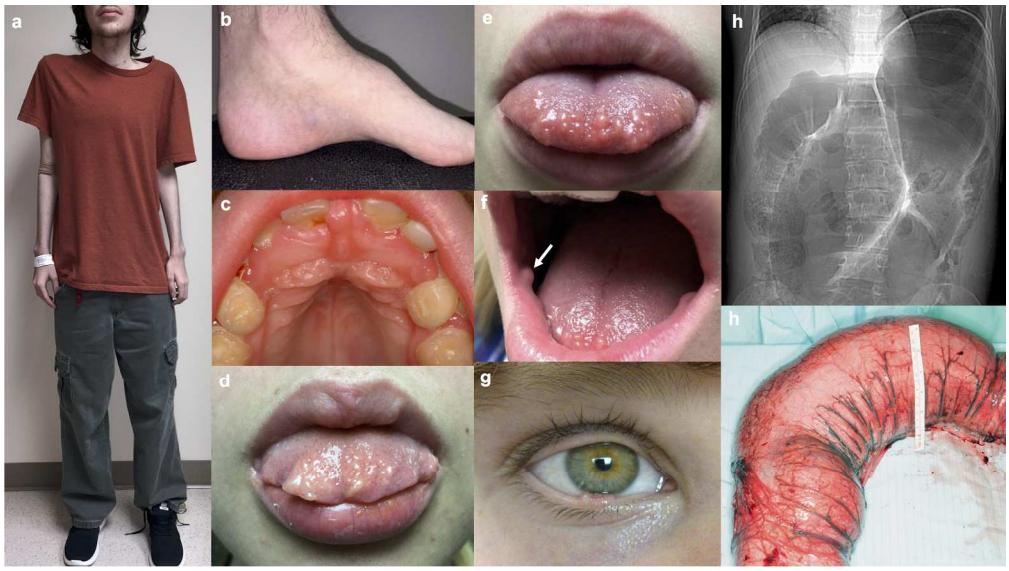
Figure 1: The development of Medullary thyroid carcinoma (MTC). From normal C-cells (A), the tumor follows an asymptomatic hyperplasic state (B) before the developement of microMTC (C, size ≤ 1 cm) and then macroMTC (D, size > 1 cm).

Figure 2: The pathognomonic clinical phenotype of MEN2B: 1) a marfanoid body habitus (a) and other skeletal features including scoliosis (a), pes cavus (b), and high-arched palate (c); 2) thickened lips and neuromas affecting the tongue (d and e), the oral mucosa, (f) the conjunctiva (g), and other mucosal surfaces; 3) ophthalmological signs including ptosis and everted upper eyelids (g); and 4) gastrointestinal problems primarily related to impaired colonic motility due to diffuse intestinal ganglioneuromatosis that can lead to megacolon (h). For additional clinical manifestations of MEN2B, please refer to the text.



The development of Medullary thyroid carcinoma (MTC). From normal C-cells (A), the tumor follows an asymptomatic hyperplasic state (B) before the developement of microMTC (C, size ≤ 1 cm) and then macroMTC (D, size > 1 cm).

187x52mm (300 x 300 DPI)



The pathognomonic clinical phenotype of MEN2B: 1) a marfanoid body habitus (a) and other skeletal features including scoliosis (a), pes cavus (b), and high-arched palate (c); 2) thickened lips and neuromas affecting the tongue (d and e), the oral mucosa, (f) the conjunctiva (g), and other mucosal surfaces; 3) ophthalmological signs including ptosis and everted upper eyelids (g); and 4) gastrointestinal problems primarily related to impaired colonic motility due to diffuse intestinal ganglioneuromatosis that can lead to megacolon (h). For additional clinical manifestations of MEN2B, please refer to the text.

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