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Research Article

Familial Medullary Thyroid Cancer: Five-year Review of the Most Frequent Mutations in the RET Gene: An Update

Blanca Estela Díaz Castañeda[1], Mariela Eulogio Vences[1], Brian Emmanuel Díaz Gordillo[1], Miriam Deyanira Rodríguez Piña[1], and Hugo Mendieta Zerón[1,2]*

[1]Faculty of Medicine, Autonomous University of the State of Mexico, Mexico
[2]Ciprés Grupo Médico CGM SC

ORCID:
Blanca Estela Díaz Castañeda: https://orcid.org/0000-0001-7330-1427
Mariela Eulogio Vences: https://orcid.org/0000-0003-2788-6174
Brian Emmanuel Díaz Gordillo: https://orcid.org/0000-0002-0367-6343
Miriam Deyanira Rodríguez Piña: https://orcid.org/0000-0003-1882-2814
Hugo Mendieta Zerón: https://orcid.org/0000-0003-3492-8950

Abstract

Background: Familial Medullary Thyroid Cancer (FMTC) is hereditary in 25% of cases. Patients with an inherited form of FMTC usually have a germline mutation in the *RET* proto-oncogene (10q11.2); these mutations generally occur in exons 10 (codons 618 and 620) and 11 (codons 630, 631, and 634).

Methods: A narrative review of articles focused on the pathology of familial medullary thyroid cancer was carried out using the next databases PubMed, ScienceDirect, BMC, Springer, Frontiers, PMC, Wiley Online Library, Cold Spring Harbor and ELSEVIER. This search was carried out between August and September 2021.

Results: 19 studies were selected in which the following mutations were found: five studies (26.31%) reported mutation in exon 10; three studies (15.78%) in exon 11; three studies in exon 13 (one of them associated with a rare mutation in exon 7) (10.52% plus 5.26%); three studies (15.78%) in exon 14; two studies (10.52%) in exon 15; two (10.52%) in exon 16; and one (5.26%) rare FMTC NO *RET*. The two most frequent mutations were in codons 620 of exon 10 and 804 of exon 14.

Conclusion: The findings of this review are consistent with the medical literature, finding the most common *RET* mutations in exon 10 and codon 620. It is essential that in patients with a presumptive diagnosis, genetic studies (identification of germline mutations in the *RET* proto-oncogene, located on chromosome 10q11.2) be performed.

Keywords: familial medullary thyroid cancer, *RET* proto-oncogene, thyroidectomy

Two points why this article is of interest:

1. It is an update on the most frequent *RET* mutations, associated with medullary thyroid cancer.
2. From a clinical point of view, knowing the RET two main mutations are in codons 620 and 804 of exons 10 and 14, respectively, would allow to request a specific molecular biology study to confirm the mutations when there is a case with criteria for familial medullary thyroid cancer.

Corresponding Author: Hugo Mendieta Zerón; email: drmendietaz@yahoo.com

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1. Introduction

Medullary thyroid cancer (MTC) is a neuroendocrine tumor of parafollicular or C cells of the thyroid gland, responsible for calcitonin production [1]. It accounts for 5% of all thyroid cancers and frequently occurs sporadically (75% of MTCs are sporadic and unilateral) or hereditarily (bilateral multifocal). Since the latter is associated with genetic conditions that depend on clinical data or other extra-thyroid manifestations, they have been classified into multiple endocrine neoplasia type 2 (MEN2) or familial medullary thyroid cancer (FMTC), with 25% of understudied patients corresponding to the familial type or MEN2 [2, 3].

Approximately 25% of MTC cases associated with pathogenic variants at the germline level are based on *RET* proto-oncogenes (10q11.2 locus), representing etiologic factors for FMTC or MEN2 development, including genetic conditions associated with autosomal dominant inheritance patterns [3].

Usually, ligand binding activates the *RET* receptor, promoting the signal transduction pathway that results in cell proliferation. However, the *RET* protein could become a nonphosphorylated tyrosine kinase receptor when no ligand is bound. Contrastively, mutations in the *RET* proto-oncogene of cancer cells could lead to autophosphorylation of tyrosine residues, causing the *RET* receptor to be activated. Hence, there is a relationship between the site of *RET* mutations and the phenotype. In addition, these mutations present diverse transformations, which could depend on their location in the *RET* molecule [4].

The *RET* protein comprises two regions: (i) an extracellular region with a ligand-binding domain that consists of a cadherin-like domain and a highly conserved cysteine-rich domain and (ii) an intracellular region comprising two tyrosine kinase domains. Ligand binding causes the cysteine-rich domain to facilitate receptor dimerization and autophosphorylation, activating the tyrosine kinase and signaling pathways, including JAK/STAT, PI3K/AKT, and RAS/RAF/MAPK. Subsequently, *RET* produces neoplasms in two main ways: *RET* mutations or *RET* fusions [5]. Furthermore, typical FMTC mutations occur in exons 10 (codons 618 and 620) and 11 (codons 630, 631, and 634) of the *RET* gene, they mainly affect cysteine-rich extracellular domains and are less associated with the tyrosine kinase domain. Finally, MTC accounts for 13% of all thyroid cancer-related deaths, with an overall survival rate and prognosis that is intermediate to those of patients with differentiated thyroid and anaplastic thyroid cancers [3].

2. Materials and Methods

A narrative review of articles that focused on FMTC pathology was conducted using the following databases: PubMed, ScienceDirect, BMC, Springer, Frontiers, PMC, Wiley Online Library, Cold Spring Harbor, and ELSEVIER. This search was performed between August 2, 2021 and September 10, 2021.

Three independent authors conducted the search and selection of articles. The final choices of the included articles were based first on the title and abstract and then on the specified inclusion and exclusion criteria. Finally, once all necessary information had been collected, full texts were read and the main ideas extracted.

The inclusion criteria were studies that focused on FMTC, which was related to genetic mutations and in association with the *RET* proto-oncogene, its presenting pathology, and/or related family lines. Furthermore, studies considered included case reports, case series, retrospective studies, letters to the editor, cohort studies, and peer reviews. However, the exclusion criteria were as follows: studies with publication dates older than five years, those related to sporadic MTC and familial non-MTCs, clinical trials, systematic reviews, narrative reviews, meta-analyses, and multicenter/cross-sectional studies.

3. Results

Following the systematic search, 118 related articles were obtained, 19 of which were of interest and importance to this study. Of the 19 included studies, 5 reported mutations in exon 10 (26.31%); 3 studies each reported mutations in exons 11 (15.78%), 13 (one of them associated with a rare mutation in exon 7) (10.52% plus 5.26%), and 14 (15.78%); 2 studies each reported mutations in exons 15 (10.52%) and 16 (10.52%); and then 1 rare FMTC NO *RET* (5.26%) was reported. The two most frequent mutations were in codons 620 and 804 of exons 10 and 14, respectively.

Interestingly, of the 17 clinical cases reported, 12 (70.58%) were female and 5 (29.42%) were male. The mean age within the clinical cases was 38 years and the median was 44 years (age range, 2–70 years). Table 1 shows the most critical findings of the MTC investigation identified in this review.

Table 1: MTC clinical cases.

Country	Patient's age (yr)	Sex	Family history	Exon(s)	Codon(s)/ Variant(s)
Peru [3]	24	Female	Fourth generation with FMTC	11	Cys630ser
Spain [6]	2	Female	The patient is the index case	10	Cys620ser
China [7]	57	Male	Deceased sister	10	C620Y
China [7]	61	Male	Deceased sister	10	C620Y
Israel [8]	45	Male	Consanguinity history	7 y 13	A432A & L769L
India [9]	44	Female	Four sisters and two members of the next generation	15	Ser891Ala
Turquía [10]	44	Female	Big brother	11	634
Lebanon [11]	14	Female	Mother	13	L790F (c.2370G>T)
Denmark [11]	70	Female	Mother and aunt	13	L790F (c.2370G>T)
USA [12]	62	Female	Sister, maternal aunt, and son	14	p.V804M
Italy [13]	66	Male	Older sister	16	p.Met918Thr
Italy [13]	68	Female	Younger brother	16	p.Met918Thr
USA [14]	3	Female	Father and three siblings	14	V804M
Japan [15]	5	Female	Mother	11	634/Cys634Gly
China [16]	48	Female	Patient is the index case	10	p.C611Y
Germany [17]	17	Male	Eight family members	15	p.S891A
United Kingdom [18]	22	Female	Patient is index case	Familial non-*RET* MTC	Familial non-*RET* MTC
Hungary [19]	NA	Six members of the second generation	Several generations	14	V804M and the variant S836S
China [20]	NA	Six *RET* mutation-positive members	Three-generation family	10	Y606C

4. Discussion

Functions of *RET* proto-oncogene germline mutations in the pathogenesis of FMTC cause presenting clinical manifestations to depend on the organs, specific mutation, and age of follow-up. Therefore, diagnosis in patients with FMTC requires a procedure involving a biopsy of the thyroid nodule or thyroidectomy to detect the final pathology.

Likewise, its evaluation requires imaging methods, such as ultrasound with lymph-node mapping, calcitonin and carcinoembryonic antigen-level measurements, and genetic testing (to rule out *RET* proto-oncogene germline mutations). These assessments also evaluate the presence of hyperparathyroidism and rule out the possibility of pheochromocytoma (PHEO) [5].

Although symptomatic thyroid nodule is a frequent reason for consultation, it is estimated that 35% of the population will have a nodule during their lifetime. However, only 10% of these will be associated with some thyroid cancer types. Based on the above hypothesis, there are different ways to determine whether the nodule is neoplastic. For this purpose, ultrasonographic characteristics of the nodule can be estimated with a biopsy, obtained using a fine needle guided by ultrasound. This procedure reduces false negatives and the fact that, more frequently, a nodule of fixed and complex characteristics is mostly associated with a neoplastic etiology, including the existence of ipsilateral cervical lymphadenopathies, which should also be considered a warning sign.

In clinical history, it is also prevalent to find data of altered thyroid functions, such as hypothyroidism or hyperthyroidism, leading to hormone measurements, such as triiodothyronine (T3/T3L), thyroxine (T4L), and thyroid-stimulating hormone (TSH). However, it has been estimated that more than half of the thyroid cancer cases are clinically asymptomatic. Therefore, it is important to inquire about voice alterations (dysphonia), cervical, digestive, and airway compression, dysphagia, and/or foreign body sensations during interrogations. Furthermore, since thyroid hyperfunction is closely related to cancer, serum TSH levels are useful to rule out nodule hyperfunction. Nevertheless, elevated thyroglobulin levels are often considered a warning sign (particularly useful in following up patients undergoing total thyroidectomy and radioactive iodine ablation), including hormone estimation.

Ultrasound is considered important for evaluating nodule characteristics. Therefore, this procedure should be performed in the thyroid gland and lateral necks. If the findings of this last study are a hypogenic nodule, microcalcifications, intranodular vascular flow, disproportionate measures, infiltrating margins, or the presence of cervical nodes with loss of the fatty hilum, then, a fine needle biopsy guided by ultrasound should be conducted. Although a thyroid scan should not be a routine, it is essential when there is evidence of hyperthyroidism or a functioning nodule (decrease in TSH, with or without elevation of thyroid hormones in the blood). In addition, supposing there is a need to assess metastasis or neoplastic extensions in the mediastinum, deep cervical fascia,

or upper airway, computerized tomography or magnetic resonance imaging studies are recommended [21].

During diagnosis, parathyroid hyperplasia (PPH) and/or PHEO should be ruled out during the preoperative stage in any patient with suspected MTC. If the study confirms *RET* mutations, PHEO should be ruled out by measuring 24-hr urinary metanephrines and evaluating the presence of PPH through the parathyroid hormone, calcium, and phosphorus in the blood, ruling out an association with MEN2. However, if performing an *RET* proto-oncogene study before surgery is impossible, the presence of PHEO and PPH should always be ruled out [21].

Diagnosis in patients with MTC can be preoperative during the study of a thyroid nodule that is active in *RET* proto-oncogene mutation carrier relatives. It can even be postoperative. Nevertheless, the high penetrance of *RET* mutation pathogenesis implies the need for early diagnosis.

Since MTC is aggressive and produces calcitonin, therapeutic surgery is essential, with an objective prognosis according to the tumor stage and adequate initial surgical treatment being crucial. Furthermore, based on the dominant condition of inheritance and the close relationship between genotype and phenotype that is associated with the disease aggressiveness, it is important to perform a genetic study on these patient groups.

According to the American Thyroid Association guidelines, three risk groups have been established when identifying an *RET* mutation:

1. ATA-HST→ very high risk
2. ATA-H → high risk
3. ATA-M → moderate risk

The first group, ATA-HST, presents a very high risk of mutations in codon M918T of exon 16 and an invasive MTC phenotype in children under one year. As a result, prophylactic thyroidectomy is recommended before that corresponding age. However, the second group, ATA-H, is characterized by a high risk of mutations in codons C634F/G/R/S/W/Y and A883F of exons 11 and 13. Its phenotype corresponds to MEN2A with a high incidence of OEF at 20 years and MEN2B. Therefore, its management comprises prophylactic thyroidectomy at five years of age. Finally, the third group, corresponding to ATA-M or ATA-MOD, is a moderate risk. Mutations here were distributed in exons 5, 8, 10, 11, 13–15, and 16, and its phenotype was that of later onset of MTC. Hence, less aggressive evolution (NEM2A and MTC) studies for which a prophylactic

thyroidectomy, including PHEO and PPH, is advisable. The above classification guides the time and extent of thyroid surgery. Emphasis has also been placed on exons 10, 11, 13–15, and 16 (90% of the mutations), including exons 5 and 8. Subsequently, *RET* gene studies were conducted through Sanger sequencing of the exons previously amplified by PCR [22].

Surgical treatment is not only the therapeutic alternative for cases of sporadic or hereditary MTC but also the best prophylactic measure to reduce thyroid cancer, including neoplasms associated with MEN2A and 2B. Therefore, for postsurgical management, levothyroxine supplementation (initial dose 1.6–1.8 mcg/kg/day) is recommended. In addition, while TSH should be maintained in the normal range based on age, it should not be suppressed. Besides, radioiodine (radioactive iodine) should not be used in MTC since it has no use [21].

5. Conclusion

Based on the data collected and the understudied literature, FMTC was identified as an essential dominant genetic entity in patients with a presumptive diagnosis. From the genetic studies (identification of germline mutations in the *RET* proto-oncogene, located on chromosome 10q11.2), the characteristic mutations of FMTC notably occurred in exons 10 (codons 618 and 620) and 11 (codons 630, 631, and 634) of the *RET* proto-oncogene, consistent with the findings of this review. However, exon 10 and codon 620 were the sites where mutations occurred most frequently. As for treatments, evidence in favor of radical thyroidectomy was postulated not only as the surgical procedure of choice but also as a prophylactic measure. This procedure was also considered the best alternative to prevent its progression when combined with adequate postsurgical hormone replacement therapy like levothyroxine.

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Ethical Considerations

Not applicable.

Competing Interests

The authors declare no competing interests.

Availability of Data and Material

All data used are available in public repositories and are adequately cited.

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References

[1] Kroiss, M., Koehler, V. F., & Spitzweg, C. (2021). Medulläres Schilddrüsenkarzinom. [Medullary thyroid carcinoma: Current clinical progress]. *Deutsche Medizinische Wochenschrift, 146*(23), 1527–1532. https://doi.org/10.1055/a-1495-2991

[2] Cipri, C., Vescini, F., Torresan, F., Pennelli, G., Pelizzo, M. R., Triggiani, V., Guastamacchia, E., & Grimaldi, F. (2019). An unusual case of medullary thyroid carcinoma and a revision of current literature. *Endocrine, Metabolic & Immune Disorders Drug Targets, 19*(2), 226–229. https://doi.org/10.2174/1871530319666181220165350

[3] Carrillo, C. F. B., Rojas, J. A. P., Cornejo, C. B., Zambrano, S. C., Cárdenas, A. P., Santibañez, I. Q., et al. (2018). Cáncer Medular de Tiroides familiar: Reporte de un caso y revisión de la literatura. *Revista de la Facultad de Ciencias Medicas de Cordoba, 75*(4), 303–309. https://doi.org/10.31053/1853.0605.v75.n4.20947

[4] Oczko-Wojciechowska, M., Swierniak, M., Krajewska, J., Kowalska, M., Kowal, M., Stokowy, T., Wojtas, B., Rusinek, D., Pawlaczek, A., Czarniecka, A., Szpak-Ulczok, S., Gawlik, T., Chmielik, E., Tyszkiewicz, T., Nikiel, B., Lange, D., Jarzab, M., Wiench, M., Jarzab, B. (2017). Differences in the transcriptome of medullary thyroid cancer regarding the status and type of RET gene mutations. *Scientific Reports, 7*, 42074.

[5] Román-González, A., Mejía, S., & Zapata, M. (2019). Nuevos tratamientos médicos para el cáncer medular de tiroides. Revista Colomb. *Endocrinology, Diabetes & Metabolism, 6*(4), 306–310.

[6] Morlán Herrador, L., de Arriba, A., Miguel, G., Ferrera, M., & Labarta, J. I. (2016). Estudio de carcinoma medular de tiroides a partir de un caso índice. *Archivos Argentinos de Pediatria, 114*(6), e421–e424.

[7] Huang, Q., Hu, A., & Zhang, M. (2020). Chinese siblings with hereditary medullary thyroid carcinoma caused by RET mutation: Implications for RET oncogene detection. *BMC Endocrine Disorders, 20*(1), 64. https://doi.org/10.1186/s12902-020-0544-3

[8] Azzam, N., Bar-Shalom, R., Saab, A., & Fares, F. (2017). Germline polymorphisms on RET proto-oncogene involved in medullary thyroid carcinoma in a Druze family. *European Journal of Cancer, 82*, 149–152. https://doi.org/10.1016/j.ejca.2017.06.007

[9] Vijayan, R., Nair, V., Menon, U., & Kumar, H. (2021). A rare RET mutation in an Indian pedigree with familial medullary thyroid carcinoma. *Indian Journal of Cancer, 58*(1), 98–100.

[10] Sezer, A., & Çelik, M. (2019). Prophylactic and therapeutic surgery in familial medullary thyroid cancer. In T. Özülker, M. Adaş, S. Günay (Eds.) *Thyroid and parathyroid diseases*. (pp. 371–375). Springer, Cham. https://doi.org/10.1007/978-3-319-78476-2_59

[11] Mathiesen, J. S., Nielsen, S. G., Rasmussen, Å. K., Kiss, K., Wadt, K., Hermann, A. P., Nielsen, M. F., Larsen, S. R., Brusgaard, K., Frederiksen, A. L., Godballe, C., & Rossing, M. (2020). Variability in medullary thyroid carcinoma in RET L790F carriers: A case comparison study of index patients. *Frontiers in Endocrinology, 11*, 251. https://doi.org/10.3389/fendo.2020.00251

[12] Ghazani, A. A., Breen, K. M., Dwan, M., Barletta, J. A., Vatnick, D. R., Stokes, S. M., Block, C., Doherty, G. M., Cohn, A. Y., Marqusee, E., Garber, J. E., & Rana, H. Q. (2020). Unexpected pathogenic RET p.V804M variant leads to the clinical diagnosis and management of medullary thyroid carcinoma. *The American Journal of Case Reports, 21*, e927415–e1. https://doi.org/10.12659/AJCR.927415

[13] Sponziello, M., Benvenuti, S., Gentile, A., Pecce, V., Rosignolo, F., Virzì, A. R., Milan, M., Comoglio, P. M., Londin, E., Fortina, P., Barnabei, A., Appetecchia, M., Marandino, F., Russo, D., Filetti, S., Durante, C., & Verrienti, A. (2018). Whole exome sequencing identifies a germline MET mutation in two siblings with hereditary wild-type RET medullary thyroid cancer. *Human Mutation, 39*(3), 371–377. https://doi.org/10.1002/humu.23378

[14] Pendrick, D. M., Oberg, J. A., Hsiao, S. J., Chung, W. K., Koval, C., Sireci, A., Kuo, J. H., Satwani, P., Glasser, C. L., Sulis, M. L., Mansukhani, M. M., & Glade Bender, J. L. (2019). Identification of a secondary RET mutation in a pediatric patient with relapsed acute myeloid leukemia leads to the diagnosis and treatment of asymptomatic metastatic medullary thyroid cancer in a parent: A case for sequencing the germline. *Cold Spring Harbor Molecular Case Studies, 5(*2), a003889. https://doi.org/10.1101/mcs.a003889

[15] Abe, Y., & Tsukano, S. (2017). Detection of early stage medullary thyroid carcinoma by measuring serum calcitonin using an electro chemiluminescence immuno-assay: A case report of a young Japanese woman with a high-risk RET mutation. *Clinical Pediatric Endocrinology, 26*(4), 243–249. https://doi.org/10.1297/cpe.26.243

[16] Zhang, G., Jiang, Y., Zhang, S., Zhao, L., Fan, J., Zhang, Z., Ma, J., Chen, R., & Xu, Y. (2019). Genetic analysis of a hereditary medullary thyroid carcinoma case with normal preoperative serum calcitonin levels. *Pathology, Research and Practice, 215*(10), 152529. https://doi.org/10.1016/j.prp.2019.152529

[17] Dralle, H., & Machens, A. (2016). Timing and extent of surgery for a pediatric patient with hereditary MTC and positive screening for the S891A RET mutation. In D. Cooper & C. Durante (Eds.) *Thyroid cancer*. Springer. https://doi.org/10.1007/978-3-319-22401-5_45

[18] Smith, J., Read, M. L., Hoffman, J., Brown, R., Bradshaw, B., Campbell, C., Cole, T., Navas, J. D., Eatock, F., Gundara, J. S., Lian, E., Mcmullan, D., Morgan, N. V., Mulligan, L., Morrison, P. J., Robledo, M., Simpson, M. A., Smith, V. E., Stewart, S., … Woodward, E. R. (2016). Germline ESR2 mutation predisposes to medullary thyroid carcinoma and causes up-regulation of RET expression. *Human Molecular Genetics, 25*(9), 1836–1845. https://doi.org/10.1093/hmg/ddw057

[19] Valkusz, Z., Sepp, K., Gardi, J., & Patocs, A. (2016). Screening and follow up of an extended Hungarian family with familial medullary thyroid cancer. *Endocrine Abstracts*, *41*, EP1109. Advance online publication. https://doi.org/10.1530/endoabs.41.EP1109

[20] Wang, J., Yu, Y., Li, D., Wei, S., Yun, X., Jia, Y (2016). The rare RET mutation Y606C in a Chinese family with familial medullary thyroid carcinoma. *Chinese Journal of Clinical Oncology, 43*(21), 947–951.

[21] Letelier, C. (2017). Cáncer hereditario de tiroides, síndromes asociados y estudio genético. *Revista Médica Clínica Las Condes, 28*(4), 610–616. https://doi.org/10.1016/j.rmclc.2017.06.008

[22] Fuenzalida, R. R., Vial, L. I., Rojas, O. V., Pizarro, C. F., Puebla, R. V., & Vial, L. G. (2017). Cirugía profiláctica en cáncer medular de tiroides hereditario. *Revista Chilena de Cirugía, 69*(3), 268–272. https://doi.org/10.1016/j.rchic.2016.11.002