



Distinct Impacts of Clinicopathological and Mutational Profiles on Long-Term Survival and Recurrence in Medullary Thyroid Carcinoma

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Background: Medullary thyroid carcinoma (MTC) has a poorer prognosis than differentiated thyroid cancers; however, comprehensive data on the long-term outcomes of MTC remain scarce. This study investigated the extended clinical outcomes of MTC and aimed to identify prognostic factors.

Methods: Patients diagnosed with MTC between 1980 and 2020 were retrospectively reviewed. Their clinical characteristics, long-term clinical outcomes, and prognostic factors for recurrence and mortality were analyzed.

Results: The study included 226 patients (144 women, 82 men). The disease-specific survival (DSS) rates for all MTC patients at 5-, 10-, 20-, and 30-year intervals were 92.7%, 89.4%, 74.3%, and 68.1%, respectively. The recurrence-free survival (RFS) rates were 71.1%, 56.1%, 40.2%, and 32.1% at these intervals. DSS was comparable between the groups from 1980–2009 and 2010–2020 ($P=0.995$); however, the 1980–2009 group had significantly lower RFS rates ($P=0.031$). The 2010–2020 group exhibited greater extents of surgical and lymph node dissection ($P=0.003$) and smaller tumors ($P=0.003$). Multivariate analysis identified extrathyroidal extension as the strongest prognostic factor for both RFS and DSS. Age > 55 years and tumor size of ≥ 2 cm were also significant prognostic factors for DSS, while hereditary disease and lymph node metastasis were significant for RFS. Survival analysis after propensity-score matching of rearranged during transfection (*RET*)-negative and non-screened *RET*-positive groups showed comparable DSS but longer RFS in the *RET*-negative group.

Conclusion: Extrathyroidal extension was identified as the strongest prognostic factor for RFS and DSS. Older age and larger tumor size were associated with decreased DSS, while *RET* mutation and lymph node metastasis significantly impacted RFS.

Keywords: Long-term outcomes; Medullary thyroid carcinoma; RET mutation; Survival analysis; Thyroid neoplasms

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INTRODUCTION

Medullary thyroid carcinoma (MTC), which accounts for 2% to 4% of thyroid cancers, arises from calcitonin-secreting C cells and is responsible for up to 13.4% of deaths related to thyroid cancer [1]. Studies have shown that the 5- and 10-year overall survival (OS) rates for MTC patients range from 70%–94% and 57%–90%, respectively [2–8], while the 5- and 10-year recurrence-free survival (RFS) rates vary between 83%–86% and 65%–66% [4,5]. Distant metastasis, which was initially detected in 7%–23% of MTC patients, is a well-established strong predictor of disease-specific mortality [4,9]. Previous studies have identified several risk factors for recurrence and mortality in MTC patients, including age at diagnosis, specific types of the rearranged during transfection (*RET*) mutation, lymph node status, tumor stage, and postoperative biochemical remission (BCR) status [4,10].

However, advances in preoperative diagnostics, incorporating genetic, biochemical, radiological, and cytological examinations, have facilitated the early-stage detection of MTC, leading to diagnoses at earlier tumor stages [11,12]. This progress, along with evolving guidelines and clinical practices such as increased surgical extents, has changed the clinicopathological characteristics and prognosis of MTC over time [13–16]. Given these temporal changes, there is a critical need for updated information on the long-term outcomes of MTC and the identification of prognostic factors to guide tailored management. Although retrospective analyses of MTC patients from our center have been conducted, covering the period from 1982 to 2012 [4,17], a decade has since passed, necessitating a comprehensive reassessment. This reassessment includes data from newly diagnosed patients and extend the follow-up data for existing patients over subsequent years. Therefore, the objective of this study was to evaluate the long-term outcomes of surgically treated MTC patients over more than three decades, analyzing their characteristics and identifying prognostic factors related to recurrence and survival.

METHODS

Patient selection and data collection

A retrospective review was conducted on 226 MTC patients diagnosed and followed from 1980 to 2020 at Seoul National University Hospital. The study excluded cases with incomplete medical records ($n=40$), resulting in a total of 226 patients included for analysis. Data were collected on patient and disease

characteristics, including *RET* mutation testing, surgical extents, histopathology, and clinical outcomes such as recurrences and deaths. Additionally, information on the survival status of patients, duration of survival, and cause of death up to December 31, 2019, was obtained from the registered data of Korea Statistics [18]. The causes of death were categorized as either death from all causes, defined as patient death for any reason, or MTC-specific death, defined as patient death related to MTC. To assess changes in clinical characteristics and long-term outcomes over time, patients were grouped by their year of diagnosis. The year 2009/2010 was selected as the reference point for dividing the patient cohorts into different time periods, coinciding with the publication of the American Thyroid Association (ATA) guidelines in 2009 and 2015 [15,16], which may have significantly influenced clinical practice for managing MTC. Patients were then grouped into 10-year intervals based on their year of diagnosis (Supplemental Table S1). Our observations indicated that the clinical characteristics were similar between the periods 1980–2009 and 2010–2020, supporting this grouping approach. The study was conducted in accordance with the Declaration of Helsinki and received approval from the Institutional Review Board of Seoul National University Hospital (No. 2012-081-1181), with a waiver for individual consent for the retrospective analysis.

Definitions and outcomes of interest

Initial distant metastasis was defined as the detection of structural metastasis to distant organs beyond the neck, based on imaging findings at the time of the initial diagnosis of primary MTC. Recurrence was identified through the detection of structural recurrence, pathologically confirmed via reoperation, or through evidence of metastasis to distant organs outside the neck on imaging. Persistent disease was indicated by evidence of metastasis at the time of surgery that persisted postoperatively, or by evidence of ongoing structural disease within 6 months following surgery. For the postoperative status, patients were classified based on their serum calcitonin levels and the presence of structural disease on imaging studies conducted within 6 months of surgery. BCR was characterized by a serum calcitonin level of <10 pg/mL with no evidence of structural disease, while biochemical disease (BCD) was indicated by a serum calcitonin level of ≥ 10 pg/mL, also with no evidence of structural disease. At the last follow-up, patients were categorized into one of the following statuses: BCR, BCD, stable disease (SD), progressive disease (PD), or death due to MTC. SD was defined as a recurrence of the disease with a serum calcitonin level of ≥ 10

pg/mL and structural disease, but without significant changes in the disease status during the last follow-up. PD was defined as a recurrence of the disease with an observed increase in size or extent during the last follow-up. Death due to MTC was defined as a death resulting from the progression of MTC.

Long-term oncologic outcomes, including disease-specific survival (DSS) and RFS, were analyzed. DSS was defined as

the time from surgery to death from MTC, and RFS was defined as the time from surgery to clinically evident recurrence. Prognostic factors for DSS and RFS of MTC patients, as well as OS, defined as the time from surgery to death from any cause, and distant metastasis-free survival (DMFS), defined as the time from surgery to clinically evident distant metastasis, were analyzed.

Table 1. Clinical Characteristics of Medullary Thyroid Carcinoma Patients by Year of Diagnosis

Characteristic	Total (n=226)	1980–2009 (n=120)	2010–2020 (n=106)	P value
Female sex	144 (64)	78 (65)	66 (62)	0.669
Age, yr	48.1±15.5	44.9±14.8	51.7±15.5	<0.001
Hereditary	53 (24)	27 (23)	26 (25)	0.719
RET mutation, positive/tested	46 (20)/132 (58)	22 (18)/47 (39)	24 (23)/85 (80)	0.524/<0.001
Operation				
Surgical extent, less than TT/TT	17 (8)/209 (92)	15 (13)/105 (88)	2 (2)/104 (98)	0.003
Lymph node extent ^a , CND/LND/none	88 (42)/83 (40)/39 (19)	26 (25)/45 (42)/35 (33)	62 (58)/38 (36)/4 (4)	<0.001
Pathology				
Tumor size in greatest length, cm	2.1±1.8	2.3±1.5	2.0±2.0	0.003
Tumor size, <2 cm/≥2 cm	111 (49)/85 (38)	43 (36)/52 (43)	68 (64)/33 (31)	0.003
ETE, none/microscopic/gross	129 (68)/41 (22)/19 (10)	62 (69)/19 (21)/9 (10)	67 (68)/22 (22)/10 (10)	0.981
Lymph node metastasis ^b , none/CN/LN	64 (28)/42 (19)/56 (25)	20 (17)/16 (13)/29 (24)	44 (42)/26 (25)/27 (25)	0.072
Resection margin positive status	12 (6)	7 (8)	5 (5)	0.805
Combined pathology, PTC/FTC	28 (14)/2 (1)	8 (8)/1 (1)	20 (19)/1 (1)	0.006
Initial distant metastasis	8 (4)	2 (2)	6 (6)	0.151
Postoperative neck radiotherapy	41 (18)	22 (18)	19 (18)	0.994
Recurrence outcomes				
Follow-up period, mo	70.5 (36.0–134.5)	74.5 (37.75–158.3)	64.5 (34.5–119.3)	<0.001
Recurrence/persistence	92 (41)/8 (4)	65 (54)/2 (2)	27 (25)/6 (6)	<0.001
Cumulative recurrence rate at 5/10/20/30 years, %	28.9/43.9/59.8/67.9	32.6/50.9/65.4/72.3	23.8/33.2/-/-	0.031
Sites of recurrence ^c				
LN/mediastinum/distant organs	83 (89)/25 (27)/69 (74)	60 (91)/18 (27)/44 (67)	23 (85)/7 (26)/25 (93)	0.265
Single/multiple	44 (44)/56 (56)	33 (49)/35 (51)	11 (34)/21 (66)	0.186
Final disease status, BCR/BCD/CD	113 (50)/40 (18)/41 (18)	50 (42)/26 (22)/22 (18)	63 (59)/14 (13)/19 (18)	0.139
Survival outcomes				
Follow-up period, mo	116.0 (57.8–170.0)	157.5 (72.8–227.3)	89.5 (48.3–132.8)	<0.001
Mortality, MTC-specific/all causes	32 (14)/43 (19)	22 (18)/31 (26)	10 (9)/12 (11)	0.016/0.005
Cumulative disease specific death rate at 5/10/20/30 years, %	7.3/10.6/25.7/31.9	7.8/10.1/25.5/31.7	7.3/11.5/-/-	0.995

Values are expressed as number (%), mean±standard deviation, or median (interquartile range).

RET, rearranged during transfection; TT, total thyroidectomy; CND, central node dissection; LND, lateral node dissection; ETE, extrathyroidal extension; CN, central node; LN, lateral node; PTC, papillary thyroid carcinoma; FTC, follicular thyroid carcinoma; BCR, biochemical remission; BCD, biochemical disease; CD, clinical disease; MTC, medullary thyroid carcinoma.

^aAvailable information for 210 patients (106 patients in the 1980–2009 period and 104 patients in the 2010–2020 period); ^bAvailable information for 166 patients (67 patients in the 1980–2009 period and 99 patients in the 2010–2020 period); ^cPercentages are relative to only 93 recurrent patients (66 patients in the 1980–2009 period and 27 patients in the 2010–2020 period). Data includes cases with recurrence at multiple sites, with each site counted separately, which may have resulted in duplicate counts. Specific recurrences to distant organs are summarized in Supplemental Table S2.

RET mutation and screening

Medical and family histories, along with *RET* proto-oncogene mutation analysis, were reviewed to determine whether the disease was sporadic or hereditary. Genetic testing for *RET* proto-oncogene mutations (exons 8, 10, 11, 13, 14, 15, 16) was performed only in selected patients. Patients who tested positive for *RET* mutations or those who exhibited clinical suspicion for multiple endocrine neoplasia type 2 (MEN2) syndromes—either through a family history of MTC or the presence of MEN2 phenotypes—were classified as having hereditary MTC. Conversely, those who tested negative were classified as having sporadic MTC. Among the patients tested for *RET* mutations, those with detected mutations were labeled *RET*-positive, while those without detected mutations were labeled *RET*-negative. *RET*-positive patients were further categorized as either screened or non-screened, depending on whether MTC was identified during screening examinations prompted by positive *RET* results. *RET* mutations were classified as moderate, high, or highest risk, according to the guidelines set forth by the ATA [16].

Statistical analysis

Data were presented as mean $\pm$ standard deviation or as percentages (*n*%) for descriptive statistics. For continuous variables, either the Student *t* test or the Mann-Whitney *U* test was employed. Categorical variables were analyzed using the Pearson chi-square test or logistic regression analysis. Kaplan–Meier survival analysis was utilized to estimate the DSS and RFS rates, with the log-rank test comparing these rates between groups. To compare the long-term oncologic outcomes between

RET-negative patients and those not screened but *RET*-positive, 1:1 propensity-score matching was conducted to adjust for significant variables such as age and sex. This matching was executed using the MatchIt package (R Foundation for Statistical Computing, Vienna, Austria). The Cox proportional hazard regression model was then applied to identify prognostic factors for recurrence and disease-specific death. In all analyses, a *P* value of less than 0.05 was considered to indicate statistical significance. All data were analyzed using R software version 4.3.1 (R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

Clinical characteristics and long-term oncologic outcomes over time

The clinical characteristics of all MTC patients, categorized by the year of diagnosis, are summarized in Table 1. Between 1980 and 2020, a total of 226 patients were diagnosed with MTC. The cohort included 62 (27%) men and 164 (73%) women, with a mean age of 48.1 ± 15.5 years. When comparing the two groups by diagnosis period (1980–2009 vs. 2010–2020) patients diagnosed in the latter period were older, although the sex distribution was similar ($P=0.669$). Furthermore, 24% of cases were hereditary, although no significant differences were observed between the groups ($P=0.719$). *RET* mutation testing was conducted more frequently in the later period (39% vs. 80%, $P<0.001$), but the prevalence of *RET* mutations did not differ significantly between the groups (18% vs. 23%, $P=0.524$). Surgical interventions were more extensive in the 2010–2020 group,

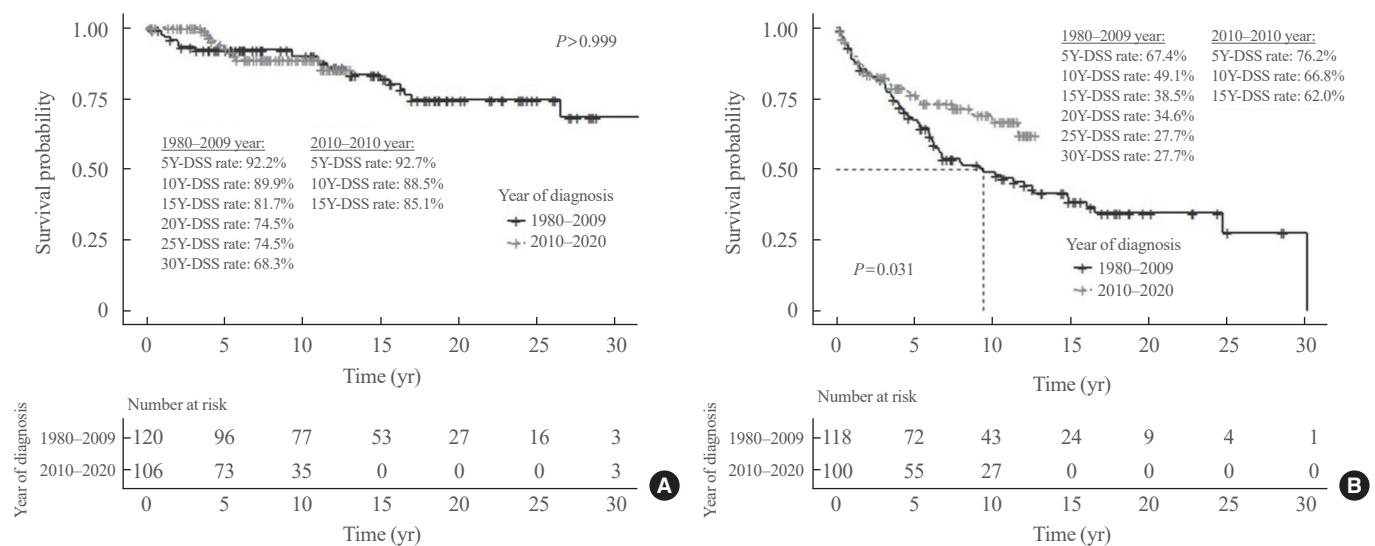


Fig. 1. Long-term survival analysis by year of diagnosis. (A) Disease-specific survival (DSS). (B) Recurrence-free survival (RFS).

including greater surgical extent ($P=0.003$) and more extensive lymph node dissection ($P<0.001$). Pathological findings revealed that the mean tumor size was larger in the earlier group (2.3 cm vs. 2.0 cm, $P=0.003$), and lymph node metastasis status was worse in 1980–2009 than in 2010–2020, but with marginal statistical significance ($P=0.072$). There were also more combined carcinomas, especially papillary thyroid carcinomas, in the 2010–2020 group ($P<0.006$). There were no significant differences in extrathyroidal extension and resection margin status between the two groups ($P>0.050$). Eight cases (4%) exhibited

initial distant metastasis, with no significant difference between the two groups ($P=0.151$) (Table 1).

Survival analysis of all MTC patients revealed 5-, 10-, 20-, and 30-year RFS rates of 71.1%, 56.1%, 40.2%, and 32.1%, respectively, along with 5-, 10-, 20-, and 30-year DSS rates of 92.7%, 89.4%, 74.3%, and 68.1%, respectively (Supplemental Fig. S1). The period from 1980–2009 saw more recurrences than the 2010–2020 period (54% vs. 25%, $P<0.001$), with 5- and 10-year RFS rates of 66.2% and 48.2% for the earlier period, and 75.5% and 61.2% for the latter period (Table 1). The sites of re-

Table 2. Multivariate Survival Analysis^a Using a Cox Proportional Hazards Model to Identify Prognostic Factors in Medullary Thyroid Carcinoma

Variable	OS			DSS			RFS			DMFS		
	HR	95% CI	P value	HR	95% CI	P value	HR	95% CI	P value	HR	95% CI	P value
Age, yr												
<55	Ref			Ref								
≥55	2.826	1.069–7.468	0.036	6.447	1.848–22.483	0.003						
Sex												
Male	Ref			Ref			Ref			Ref		
Female	1.003	0.381–2.641	0.995	1.564	0.485–5.046	0.454	0.683	0.404–1.155	0.155	0.679	0.349–1.321	0.255
Hereditary												
Sporadic	Ref			Ref			Ref					
Hereditary	0.348	0.070–1.734	0.198	0.358	0.041–3.147	0.354	1.966	1.058–3.651	0.032			
Tumor size, cm												
<2	Ref			Ref	Ref		Ref			Ref		
≥2	2.227	0.671–7.390	0.191	5.166	1.053–25.343	0.043	1.801	0.987–3.288	0.055	1.919	0.870–4.235	0.106
Extrathyroidal extension												
None	Ref			Ref			Ref			Ref		
Microscopic	4.722	1.283–17.374	0.020	10.500	1.977–55.774	0.006	4.960	2.495–9.857	<0.001	5.000	2.087–11.981	<0.001
Gross	3.624	0.771–17.034	0.103	7.942	1.217–51.809	0.030	3.448	1.491–7.973	0.004	3.034	1.049–8.771	0.041
Resection margin												
Negative										Ref		
Positive	2.395	0.628–9.131	0.201	1.771	0.413–7.607	0.442				1.958	0.709–5.405	0.194
Lymph node metastasis												
None	Ref			Ref			Ref			Ref		
Central nodes	2.318	0.432–12.368	0.325	1.718	0.295–9.995	0.547	2.691	1.088–6.657	0.032	3.402	0.918–12.613	0.067
Lateral and mediastinal nodes	2.400	0.417–13.315	0.317	0.799	0.121–5.263	0.815	4.020	1.635–9.885	0.002	4.833	1.314–17.914	0.018
Initial distant metastasis												
None	Ref			Ref								
Metastasis	1.662	0.374–7.380	0.504	2.885	0.6051–13.845	0.186						

OS, overall survival; DSS, disease-specific survival; RFS, recurrence-free survival; DMFS, distant metastasis-free survival; HR, hazard ratio; CI, confidence interval.

^aAdjusted for age, sex, hereditary/sporadic, tumor size, resection margin, lymph node metastasis, and initial distant metastasis (summarized data of multivariate analysis of model 2 in Supplemental Tables S4–S7).

currence, including distant sites, did not differ significantly (Supplemental Table S2). Additionally, there were more deaths attributed to MTC in the 1980–2009 period than in the 2010–2020 period (18% vs. 9%, $P=0.016$). The RFS rate showed a significant difference between the two periods ($P=0.031$), but the DSS rate did not ($P=0.995$) (Fig. 1).

Clinical characteristics according to last follow-up status

When grouped by last follow-up status, 113 patients (50.0%) were in the BCR group, 64 patients (28.3%) were in the BCD or SD group, and 49 patients (21.7%) had PD or died due to MTC. The period from 2010 to 2020 saw a higher proportion of MTC patients with a better last follow-up status (BCR > BCD or SD > PD or death due to MTC) compared to the period from 1980 to 2009 ($P<0.001$). The male sex ratio and age were higher in patients with PD and those who died due to MTC than in the BCR and BCD or SD groups. Although the extent of surgery did not significantly differ between the groups, the extent of lymph node dissection increased with the severity of the disease status. The prevalence of lymph node metastasis and distant metastasis at diagnosis was higher as the disease status worsened. Poorer pathology findings, including larger tumor size, extrathyroidal extension, and resection margins, were also more commonly associated with advanced disease status. Recurrence and death were more frequent among patients with advanced disease status (Supplemental Table S3).

Prognostic factors for recurrence and survival

The multivariate analyses using the Cox proportional hazards model to identify prognostic factors for OS, DSS, RFS, and DMFS are summarized in Table 2. The univariate and multivariate analyses for each OS, DSS, RFS, and DMFS are summarized in Supplemental Tables S4–S7, respectively.

Multivariate analysis identified extrathyroidal extension as the strongest independent prognostic factor for both DSS and RFS. For DSS, the hazard ratios (HRs) were 10.500 (95% confidence interval [CI], 1.977 to 55.774; $P=0.006$) for microscopic extension and 7.942 (95% CI, 1.217 to 51.809; $P=0.030$) for gross extension. For RFS, the HRs were 4.960 (95% CI, 2.495 to 9.857; $P<0.001$) for microscopic extension and 3.448 (95% CI, 1.491 to 7.973; $P=0.004$) for gross extension. Additionally, extrathyroidal extension was associated with increased tumor size, lymph node metastasis, and initial distant metastasis ($P<0.001$) (Supplemental Table S8).

Age >55 years (HR, 6.447; 95% CI, 1.848 to 22.483; $P=0.003$) and tumor size ≥ 2 cm (HR, 5.166; 95% CI, 1.053 to 25.343;

$P=0.043$) were also significant prognostic factors for DSS. Hereditary disease (HR, 1.966; 95% CI, 1.058 to 3.651; $P=0.030$) and lymph node metastasis (HR, 2.691; 95% CI, 1.088 to 6.657; $P=0.032$ for central nodes; HR, 4.020; 95% CI, 1.635 to 9.885; $P=0.002$ for lateral and mediastinal nodes) were also significant prognostic factors for RFS (Table 2).

Survival graphs for each of the prognostic factors for DSS and RFS are shown in Figs. 2, 3, while survival graphs for each of the prognostic factors for OS and DMFS are shown in Supplemental Figs. S2, S3.

Genetic characteristics of MTC patients and clinical characteristics and survival of RET mutation-tested patients

Of the one MTC patients, 173 (77%) were clinically sporadic, and 53 (24%) were clinically hereditary, with no significant difference between the two time periods ($P=0.719$). *RET* mutation tests were conducted on 132 patients, with 86 (65%) testing *RET*-negative and 46 (35%) testing *RET*-positive. The proportion of *RET*-positive results among those tested was higher in the 1980–2009 group than in the 2010–2020 group (35% vs. 25%, $P=0.032$). Among the *RET*-positive patients, 27 (59%) were classified as moderate risk, 15 (33%) as high risk, and three (9%) as highest risk according to the ATA risk classification, with no significant difference in the distribution of ATA risks between the year groups ($P=0.318$) (Supplemental Table S9).

RET-positive patients were categorized based on whether MTC was identified through screening due to a family history of MEN2 or not. There were 32 non-screened *RET*-positive patients and 14 screened *RET*-positive patients. The *RET*-negative patients were significantly older than both the non-screened *RET*-positive patients (52.3 vs. 39.3, $P<0.001$) and the screened *RET*-positive patients (52.3 vs. 25.2, $P<0.001$). Additionally, the non-screened *RET*-positive patients were significantly older than their screened counterparts (39.3 vs. 25.2, $P=0.004$). The screened *RET*-positive patients had, on average, significantly smaller tumors than both the *RET*-negative patients (0.8 cm vs. 1.8 cm, $P<0.001$) and non-screened *RET*-positive patients (0.8 cm vs. 2.5 cm, $P<0.001$). The recurrence rate was significantly lower in the *RET*-negative patients than in non-screened *RET*-positive patients (31% vs. 81%, $P<0.001$), but similar to that of the screened *RET*-positive patients (31% vs. 7%, $P=0.126$) (Table 3). There were no significant differences in recurrence sites, including distant recurrence sites (Supplemental Table S10). The survival graphs indicated no significant difference in DSS rates between the *RET*-negative and non-screened *RET*-positive

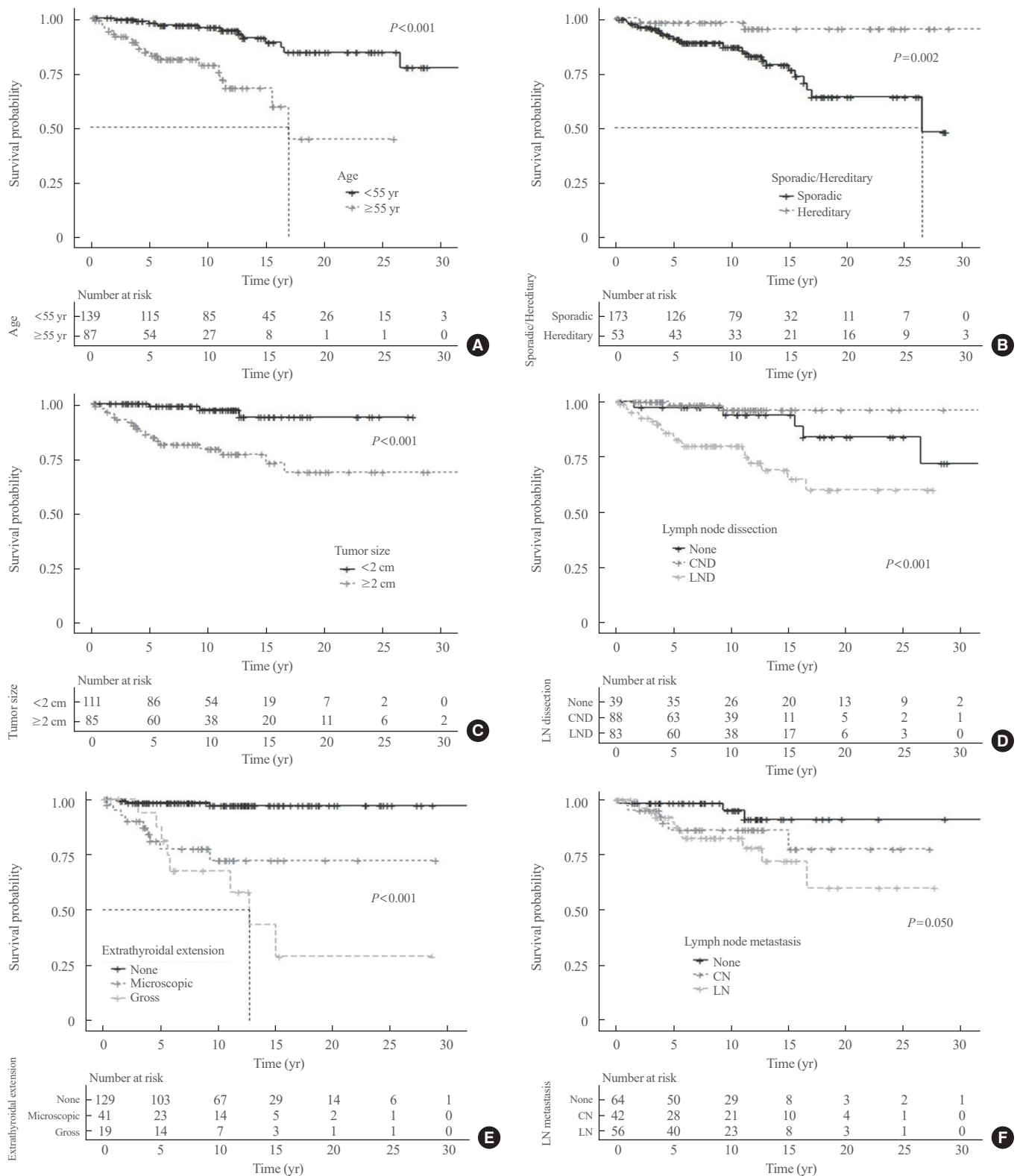


Fig. 2. Survival graphs according to risk factors for disease-specific survival in patients with medullary thyroid carcinoma. (A) Disease-specific survival by age. (B) Disease-specific survival by sporadic versus hereditary disease. (C) Disease-specific survival by tumor size. (D) Disease-specific survival by lymph node (LN) dissection extent. (E) Disease-specific survival by the presence of extrathyroidal extension. (F) Disease-specific survival by the presence of LN metastasis. CND, central node dissection; LND, lateral node dissection; CN, central node.

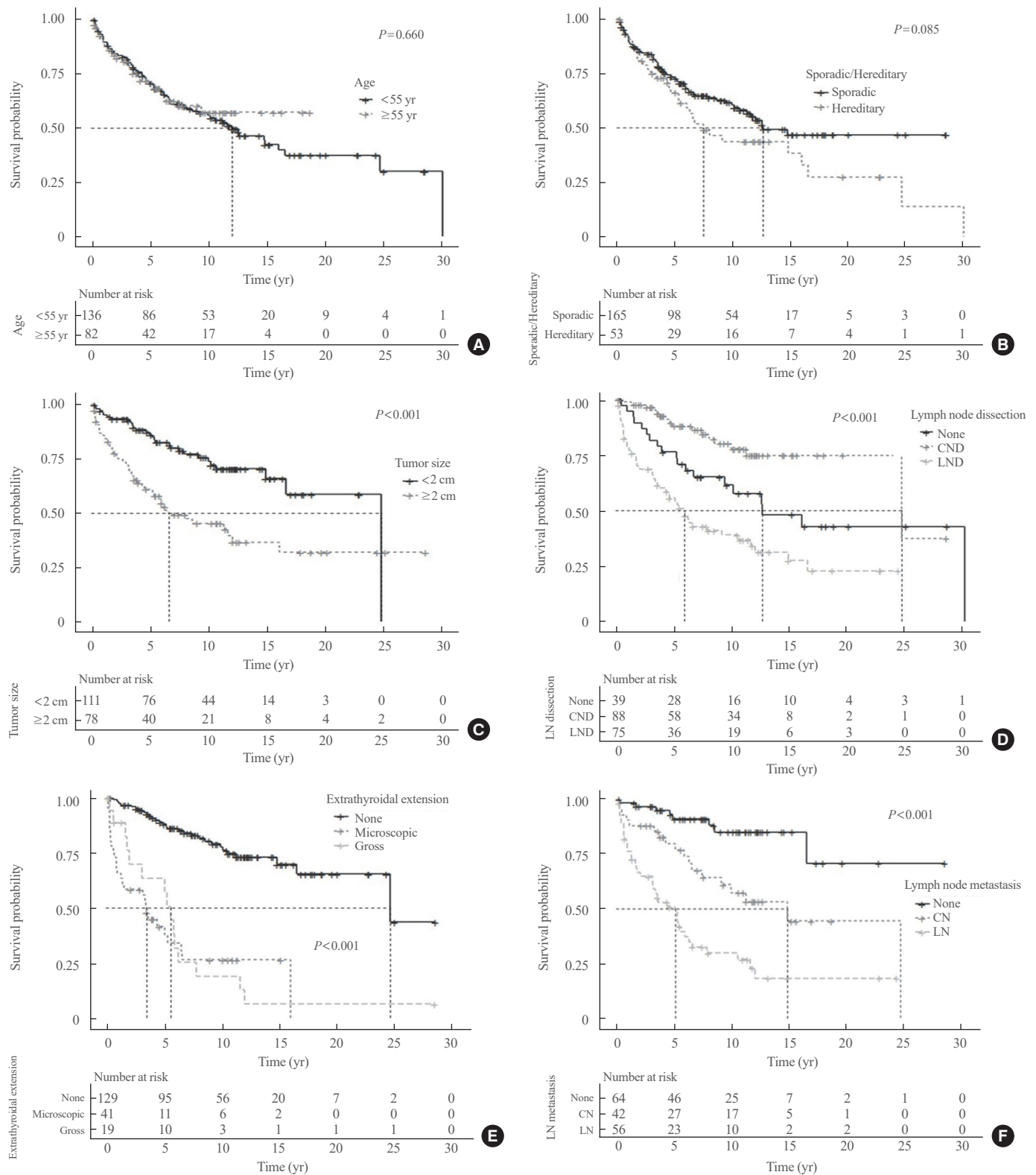


Fig. 3. Survival graphs according to risk factors for recurrence-free survival in patients with medullary thyroid carcinoma. (A) Recurrence-free survival by age. (B) Recurrence-free survival by sporadic versus hereditary disease. (C) Recurrence-free survival by tumor size. (D) Recurrence-free survival by lymph node (LN) dissection extent. (E) Recurrence-free survival by the presence of extrathyroidal extension. (F) Recurrence-free survival by the presence of LN metastasis. CND, central node dissection; LND, lateral node dissection; CN, central node.

Table 3. Clinical Characteristics between *RET*-Negative and Non-Screened *RET*-Positive Patients before and after Propensity Score Matching

Characteristic	Before propensity-score matching				After propensity-score matching			
	<i>RET</i> -negative (n = 86)	<i>RET</i> -positive (non- screened) (n = 32)	P value ^a	<i>RET</i> -positive (screened) (n = 14)	P value ^b	<i>RET</i> -negative (n = 32)	<i>RET</i> -positive (non-screened) (n = 32)	P value
Sex								
Male	28 (33)	12 (38)	0.614	7 (50)	0.204	12 (38)	12 (38)	>0.999
Female	58 (67)	20 (63)		7 (50)		20 (63)	20 (63)	
Age, yr	52.3±11.8	39.3±13.2	<0.001	25.2±16.9	<0.001	42.4±10.2	39.3±13.2	0.216
Follow-up duration, yr	103 (61–149)	149 (104–307)	<0.001	139 (59–188)	0.201	128 (87–156)	149 (104–307)	0.055
Medullary thyroid cancer								
Basal calcitonin, pg/mL ^d	1,369±2,950	1,469±1,214	0.036	3,742±12,860	0.504	2,385±4,396	1,469±1,214	0.569
Tumor size, cm ^e	1.8±1.5	2.5±1.4	0.351	0.8±0.6	<0.001	2.4±2.0	2.5±1.4	0.340
Tumor size > 2 cm	31/84 (37)	15/26 (58)	0.099	3/14 (21)	0.411	15/30 (50)	15/26 (58)	0.759
Extrathyroidal extension	27/82 (63)	8/25 (32)	0.931	1/13 (8)	0.064	11/30 (37)	8/25 (32)	0.717
Lymph node metastasis	43/77 (56)	17/22 (77)	0.070	6/14 (55)	0.935	19/28 (68)	17/22 (77)	0.462
Distant metastasis at diagnosis	3/86 (4)	0/32 (0)	0.562	0/14 (0)	0.478	2/32 (6)	0/32 (0)	0.492
Recur	26/83 (31)	26/32 (81)	<0.001	1/14 (7)	0.062	14/32 (44)	26/32 (81)	0.005
Cumulative recurrence rate at 5/10/20/30 years, %	21.9/31.4/44.1/44.1	48.9/78.0/92.7/92.7	<0.001	0.0/90.0/90.0/90.0	0.063	38.0/52.5/52.5/52.5	48.9/78.0/92.7/92.7	0.040
Cumulative disease-specific death rate at 5/10/20/30 years, %	4.1/9.5/19.6/19.6	0.0/0.0/4.8/4.8	0.159	0.0/0.0/0.0/0.0	0.250	3.4/11.9/11.9/11.9	0.0/0.0/4.8/4.8	0.250

Values are expressed as number (%), mean ± standard deviation, or median (interquartile range). *RET*, rearranged during transfection.

^aP value for comparison between *RET*-negative and *RET*-positive (non-screened) patients; ^bP value for comparison between *RET*-negative and *RET*-positive (screened) patients; ^cP value for comparison between *RET*-positive (non-screened) and *RET*-positive (screened) patients; ^dAvailable for 71 *RET*-negative, 22 *RET*-positive (non-screened) patients, and 17 *RET*-positive (screened) patients before matching, and available for 26 *RET*-negative and 22 *RET*-positive patients after matching; ^eAvailable for 84 *RET*-negative, 26 *RET*-positive (non-screened) patients, and 17 *RET*-positive (screened) patients before matching, and available for 30 *RET*-negative and 26 *RET*-positive patients after matching.

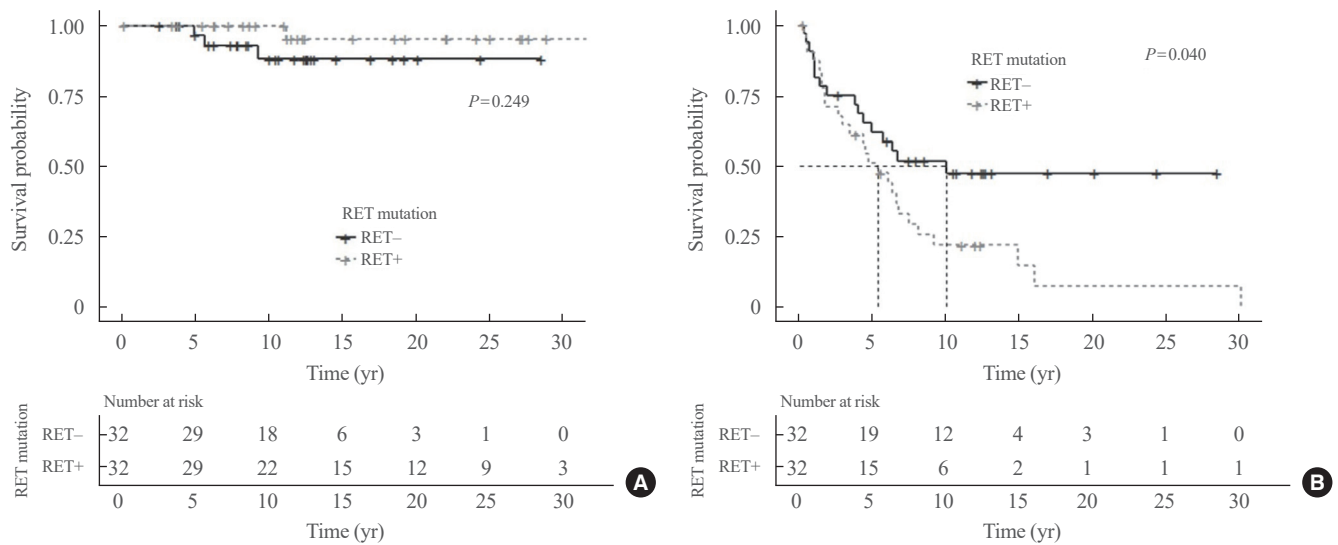


Fig. 4. Long-term survival analysis of rearranged during transfection (*RET*)-negative and non-screened *RET*-positive patients after propensity-score matching. (A) Disease-specific survival. (B) Recurrence-free survival.

patients ($P=0.159$). However, there was a significant difference in RFS rates between these two groups ($P<0.001$) (Supplemental Fig. S4).

After 1:1 propensity-score matching of the *RET*-negative and non-screened *RET*-positive patients, the clinical characteristics were similar in terms of sex, age, basal calcitonin level, tumor size, extrathyroidal extension, lymph node involvement, distant metastasis at diagnosis, recurrence sites, and disease-specific death ($P>0.050$). However, there was a significant difference in the recurrence rate (44% vs. 81%, $P=0.005$) (Table 3). Survival graphs indicated no significant difference in the DSS rate ($P=0.249$), but a significant difference in the RFS rate ($P=0.040$) was noted, even after propensity-score matching of *RET*-negative and non-screened *RET*-positive patients (Fig. 4).

DISCUSSION

This long-term follow-up study showed that the patients treated in 2010–2020 had significantly improved RFS but no meaningful difference in DSS compared to those treated in 1980–2009. Despite decreases, the recurrence rate remained remarkably high, at 23.8% at 5 years and 33.2% at 10 years. Notably, MTC-related mortality remained unchanged over 40 years, at 7.3%–7.8% at 5 years and 10.1%–11.5% at 10 years. The risk factors for recurrence or mortality varied somewhat, with older age being associated only with mortality, and lymph node metastases or *RET* proto-oncogene mutations being associated only with recurrence. Extrathyroidal extension and larger tumor size in-

creased both risks, but the association between tumor size and recurrence was not statistically significant.

The reasons for the observed decrease in recurrence rates without a corresponding decrease in mortality remain unclear. This trend was significant across different time periods and between hereditary and non-hereditary cases. The risk factors for DSS included age, extrathyroidal extension, and tumor size greater than 2 cm. In contrast, RFS was associated with extrathyroidal extension and lymph node metastasis. From 1980 to 2009, tumors were generally larger, whereas from 2010 to 2020, the average age of patients was higher, with similar rates of extrathyroidal extension in both periods. The contrasting trends in age and tumor size may account for the minimal changes in DSS observed between these periods. The increased frequency of lymph node dissection during 2010–2020 likely improved RFS by reducing lymph node recurrence, but did not significantly affect DSS, as lymph node metastasis has a weak association with DSS. These findings underscore the importance of early detection at a younger age or before significant tumor growth and extrathyroidal extension to reduce mortality. Additionally, lymph node dissection plays a crucial role in lowering the recurrence rate.

The presence of metastatic lymph nodes during initial surgery is widely recognized as a negative prognostic indicator for achieving complete remission [6,19]. Our study also found that lymph node metastasis at diagnosis was a strong prognostic factor for both recurrence and distant metastasis. However, lymph node dissection itself was not an independent prognostic factor for re-

currence and death. Nonetheless, the observed decrease in lymph node metastasis rates after 2010, alongside an increase in lymph node dissection, suggests the importance of this procedure in reducing lymph node recurrence at initial surgery, despite no change in the rates of distant metastases compared to previous years. The increase in lymph node dissection during the 2010–2020 period, likely in response to ATA guideline recommendations [15,16], may have contributed to improved prognosis by removing small or occult lymph node metastases that could otherwise lead to recurrence if left untreated. Survival curves, according to the extent of lymph node dissection, highlighted a superior prognosis among patients undergoing central node dissection (CND). CND may facilitate the removal of clinically undetected micro-metastases during surgery, consequently reducing lymph node recurrence rates. Hence, guidelines advocate for prophylactic CND even in clinically node-negative cases due to the heightened prevalence of occult disease [16,20]. In advanced stages of disease, the benefits of extensive nodal dissection remain debatable since it does not guarantee cure or prolonged survival and may lead to increased morbidity and complications [21]. Thus, the extent of lymph node dissection must be tailored according to the presurgical evaluation [22,23]. Additionally, for those with lymph node metastasis, close follow-up is crucial for detecting any recurrence, enabling timely and appropriate management of recurrent disease.

Extrathyroidal extension has emerged as the most potent prognostic factor for both RFS and DSS, with no significant differences observed between the two time periods studied. It is associated with increasing age, larger tumor size, the extent of lymph node metastasis, and the presence of initial distant metastasis. Given that extrathyroidal extension correlates with advanced disease characteristics, our findings underscore the need for earlier detection of MTC to improve prognosis. However, since extrathyroidal extension independently increases the risk of both recurrence and death, more rigorous follow-up and management are crucial for MTC patients with either microscopic or gross extrathyroidal extension, even if they do not exhibit other advanced disease characteristics. While some previous studies have identified extrathyroidal extension as a risk factor for recurrence and death [4,24,25], others have not [26]. Due to these conflicting results, further research is necessary to confirm our findings regarding the prognostic significance of extrathyroidal extension for recurrence and death.

Similar to trends observed in differentiated thyroid cancer [27–29], the tumor size in greatest length decreased over time when comparing the 1980–2009 and 2010–2020 groups. This

trend may be attributed to the early detection of MTC due to advances in imaging techniques, increased screening, and enhanced follow-up in recent years, aligning with previous reports [30–32]. Throughout the study period, clinical diagnostic tools for MTC, including serum calcitonin measurements, neck ultrasonography, and *RET* genetic testing, continued to evolve [33]. These changes in standard diagnostic procedures could have influenced the detection rates of initial MTCs or recurrent diseases. It is noteworthy that the mean age in the 2010–2020 group was older, despite expectations of more common early detection over time. Although this trend has been observed in other recent studies, the reasons for the increased incidence of older patients remain unclear [34,35]. Possible explanations may include the demographic shift towards an aging population and increased life expectancy due to better healthcare. Additionally, the gradual advancement of MTC, which may result in delayed symptom onset and diagnosis among older patients, along with alterations in thyroid tissue induced by environmental factors or the natural aging process, could also contribute to the development of MTC in older individuals. Nevertheless, further research is needed to fully understand these factors.

The extent of surgery and lymph node dissection was greater in the 2010–2020 group than in the 1980–2009 group. This increase can likely be attributed to changes in surgical practices and the influence of new guidelines published over the years, particularly after the 2009 MTC management guidelines were released by the ATA [15]. Both the early diagnosis and a comprehensive surgical approach, which includes at least a total thyroidectomy and central neck lymph node dissection, are known to significantly improve outcomes for MTC patients [30,36]. These practices may have contributed to the observed decrease in recurrence rates in patients diagnosed with MTC in more recent years. However, our multivariate analysis indicated that the extent of surgery was not an independent predictor of recurrence or death, suggesting that other factors may have played a more significant role in influencing our results.

The genetic landscape of MTC, particularly the role of *RET* mutations, continues to be a significant focus of both research and clinical interest. Our analysis revealed differential outcomes between *RET*-negative and *RET*-positive patients, with *RET*-negative patients exhibiting higher recurrence rates. This highlights the critical role of genetic profiling in guiding risk assessment and treatment decisions in MTC. The integration of *RET* mutation testing for family members of MTC patients allows clinicians to identify individuals at risk, providing the opportunity for prophylactic surgeries before the onset of overt clinical

disease [37]. Furthermore, the genetic signature within a specific lineage offers prognostic insights into the potential aggressiveness of the disease [16]. Testing for the *RET* proto-oncogene has increased over time, as documented in our results. This increase was particularly notable following the 2009 ATA guidelines, which recommended *RET* mutation genetic testing for patients with MTC [15]. However, the proportion of those undergoing *RET* gene testing still does not meet the 100% recommendation. Possible reasons for this include patients' reluctance to undergo genetic testing, the high costs and lack of insurance coverage, and issues with the accessibility, convenience, and efficiency of the test.

The implementation of *RET* gene mutation screening has led to the earlier identification of patients with hereditary MTC, resulting in a higher frequency of early-stage hereditary MTC [38]. Consequently, we observed potential differences in outcomes between screened and non-screened *RET*-positive patients, necessitating a separate analysis. To accurately assess whether the *RET* proto-oncogene mutation is associated with recurrence or disease-specific death, we excluded screened patients from the comparative analysis between *RET*-positive and *RET*-negative patients. Additionally, we employed propensity-score matching to adjust for age and sex, which are recognized as significant variables. After matching, no significant differences were observed between *RET*-negative and non-screened *RET*-positive patients in terms of disease-specific outcomes. However, *RET*-positive status in non-screened patients was associated with a higher recurrence rate than in *RET*-negative patients. This finding contrasts with previous studies suggesting the *RET* mutation does not impact long-term outcomes [26,39]. It is noteworthy, however, that those previous studies did not exclude screened patients from their analyses. Therefore, more research specifically focusing on non-screened *RET*-positive MTC patients is essential to validate our findings.

This study has several limitations worth noting. As a retrospective analysis, it is subject to the inherent variability in patient management practices over three decades. This variability includes changes in clinical protocols and the evolving quality of data, which occasionally resulted in missing information. Additionally, there was a lack of standardized systematic follow-up evaluations for all patients. Furthermore, the single-center study design limits the robustness and generalizability of the findings. Despite these limitations, our study provides valuable insights into the evolving landscape of MTC management and the factors influencing long-term outcomes. By identifying key prognostic factors and highlighting the impact of surgical interven-

tions and genetic characteristics, our findings contribute to the ongoing efforts to optimize personalized approaches for MTC patients. Although the study included a large cohort, considering the rarity of MTC, further multi-center studies with larger sample sizes are essential to validate and extend these outcomes.

In conclusion, extrathyroidal extension emerged as a strong prognostic factor for both RFS and DSS in MTC. Additionally, hereditary disease and lymph node metastasis significantly impacted RFS, while older age and larger tumor size were associated with decreased DSS. Non-screened *RET*-positive patients demonstrated higher recurrence rates than *RET*-negative patients. These findings underscore the importance of early cancer detection, as well as rigorous follow-up and management for MTC patients with adverse prognostic factors. Moreover, despite the stable mortality rate, the observed decline in recurrence over the past decade highlights the need to identify individuals at high risk of MTC-related death to ensure timely and appropriate diagnosis and treatment.

CONFLICTS OF INTEREST

Young Joo Park is an editor-in-chief and Sun Wook Cho is a deputy editor of the journal. But they were not involved in the peer reviewer selection, evaluation, or decision process of this article. No other potential conflicts of interest relevant to this article were reported.

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AUTHOR CONTRIBUTIONS

Conception or design: M.Y.O., Y.J.P., H.K.Y. Acquisition, analysis, or interpretation of data: M.Y.O., Y.J.P., H.K.Y. Drafting the work or revising: M.Y.O., K.Y.J., H.C., Y.J.C., S.W.C., S.K., K.E.L., E.J.C., D.J.P., Y.J.P., H.K.Y. Final approval of the manuscript: M.Y.O., K.Y.J., H.C., Y.J.C., S.W.C., S.K., K.E.L., E.J.C., D.J.P., Y.J.P., H.K.Y.

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REFERENCES

- Pacini F, Castagna MG, Cipri C, Schlumberger M. Medullary thyroid carcinoma. *Clin Oncol (R Coll Radiol)* 2010; 22:475-85.
- Cupisti K, Wolf A, Raffel A, Schott M, Miersch D, Yang Q, et al. Long-term clinical and biochemical follow-up in medullary thyroid carcinoma: a single institution's experience over 20 years. *Ann Surg* 2007;246:815-21.
- Torresan F, Cavedon E, Mian C, Iacobone M. Long-term outcome after surgery for medullary thyroid carcinoma: a single-center experience. *World J Surg* 2018;42:367-75.
- Jung KY, Kim SM, Yoo WS, Kim BW, Lee YS, Kim KW, et al. Postoperative biochemical remission of serum calcitonin is the best predictive factor for recurrence-free survival of medullary thyroid cancer: a large-scale retrospective analysis over 30 years. *Clin Endocrinol (Oxf)* 2016;84:587-97.
- Lee CR, Lee S, Son H, Ban E, Kang SW, Lee J, et al. Medullary thyroid carcinoma: a 30-year experience at one institution in Korea. *Ann Surg Treat Res* 2016;91:278-87.
- Kebebew E, Ituarte PH, Siperstein AE, Duh QY, Clark OH. Medullary thyroid carcinoma: clinical characteristics, treatment, prognostic factors, and a comparison of staging systems. *Cancer* 2000;88:1139-48.
- Modigliani E, Cohen R, Campos JM, Conte-Devolx B, Maes B, Boneu A, et al. Prognostic factors for survival and for biochemical cure in medullary thyroid carcinoma: results in 899 patients. The GETC Study Group. Groupe d'étude des tumeurs à calcitonine. *Clin Endocrinol (Oxf)* 1998;48: 265-73.
- Brierley J, Tsang R, Simpson WJ, Gospodarowicz M, Sutcliffe S, Panzarella T. Medullary thyroid cancer: analyses of survival and prognostic factors and the role of radiation therapy in local control. *Thyroid* 1996;6:305-10.
- Schlumberger M, Bastholt L, Dralle H, Jarzab B, Pacini F, Smit JW, et al. 2012 European Thyroid Association guidelines for metastatic medullary thyroid cancer. *Eur Thyroid J* 2012;1:5-14.
- Raue F, Bruckner T, Frank-Raue K. Long-term outcomes and aggressiveness of hereditary medullary thyroid carcinoma: 40 years of experience at one center. *J Clin Endocrinol Metab* 2019;104:4264-72.
- Opsahl EM, Akslen LA, Schlichting E, Aas T, Brauckhoff K, Hagen AI, et al. Trends in diagnostics, surgical treatment, and prognostic factors for outcomes in medullary thyroid carcinoma in Norway: a nationwide population-based study. *Eur Thyroid J* 2019;8:31-40.
- Moon S, Lee EK, Choi H, Park SK, Park YJ. Survival comparison of incidentally found versus clinically detected thyroid cancers: an analysis of a nationwide cohort study. *Endocrinol Metab (Seoul)* 2023;38:81-92.
- Filetti S, Durante C, Hartl D, Leboulleux S, Locati LD, Newbold K, et al. Thyroid cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2019; 30:1856-83.
- Kaliszewski K, Ludwig M, Ludwig B, Mikula A, Greniuk M, Rudnicki J. Update on the diagnosis and management of medullary thyroid cancer: what has changed in recent years? *Cancers (Basel)* 2022;14:3643.
- American Thyroid Association Guidelines Task Force; Kloos RT, Eng C, Evans DB, Francis GL, Gagel RF, et al. Medullary thyroid cancer: management guidelines of the American Thyroid Association. *Thyroid* 2009;19:565-612.
- Wells SA Jr, Asa SL, Dralle H, Elisei R, Evans DB, Gagel RF, et al. Revised American Thyroid Association guidelines for the management of medullary thyroid carcinoma. *Thyroid* 2015;25:567-610.
- Jung KY, Kim SM, Kim MJ, Cho SW, Kim BW, Lee YS, et al. Genotypic characteristics and their association with phenotypic characteristics of hereditary medullary thyroid carcinoma in Korea. *Surgery* 2018;164:312-8.
- Ahn JH, Choi H, Kim SJ, Cho SW, Lee KE, Park DJ, et al. The association between vitamin D supplementation and the long-term prognosis of differentiated thyroid cancer patients: a retrospective observational cohort study with propensity score matching. *Front Endocrinol (Lausanne)* 2023;14:1163 671.
- Hyer SL, Vini L, A'Hern R, Harmer C. Medullary thyroid cancer: multivariate analysis of prognostic factors influencing survival. *Eur J Surg Oncol* 2000;26:686-90.
- Asimakopoulos P, Nixon IJ, Shaha AR. Differentiated and medullary thyroid cancer: surgical management of cervical lymph nodes. *Clin Oncol (R Coll Radiol)* 2017;29:283-9.
- Franc S, Niccoli-Sire P, Cohen R, Bardet S, Maes B, Murat A, et al. Complete surgical lymph node resection does not prevent authentic recurrences of medullary thyroid carcinoma. *Clin Endocrinol (Oxf)* 2001;55:403-9.

22. Dralle H. Lymph node dissection and medullary thyroid carcinoma. *Br J Surg* 2002;89:1073-5.
23. Moley JF, DeBenedetti MK. Patterns of nodal metastases in palpable medullary thyroid carcinoma: recommendations for extent of node dissection. *Ann Surg* 1999;229:880-8.
24. de Groot JW, Plukker JT, Wolffenbuttel BH, Wiggers T, Sluiter WJ, Links TP. Determinants of life expectancy in medullary thyroid cancer: age does not matter. *Clin Endocrinol (Oxf)* 2006;65:729-36.
25. Pellegriti G, Leboulleux S, Baudin E, Bellon N, Scollo C, Travagli JP, et al. Long-term outcome of medullary thyroid carcinoma in patients with normal postoperative medical imaging. *Br J Cancer* 2003;88:1537-42.
26. Prabhu M, Shakya S, Ballal S, Ahmed Shamim S, Bal C. RET gene mutation analysis and long-term clinical outcomes of medullary thyroid cancer patients. *Nucl Med Commun* 2020;41:1136-42.
27. Cho BY, Choi HS, Park YJ, Lim JA, Ahn HY, Lee EK, et al. Changes in the clinicopathological characteristics and outcomes of thyroid cancer in Korea over the past four decades. *Thyroid* 2013;23:797-804.
28. Choi H, Lim JA, Ahn HY, Cho SW, Lee KE, Kim KW, et al. Secular trends in the prognostic factors for papillary thyroid cancer. *Eur J Endocrinol* 2014;171:667-75.
29. Elisei R, Molinaro E, Agate L, Bottici V, Masserini L, Ceccarelli C, et al. Are the clinical and pathological features of differentiated thyroid carcinoma really changed over the last 35 years?: study on 4187 patients from a single Italian institution to answer this question. *J Clin Endocrinol Metab* 2010;95:1516-27.
30. Elisei R, Bottici V, Luchetti F, Di Coscio G, Romei C, Grasso L, et al. Impact of routine measurement of serum calcitonin on the diagnosis and outcome of medullary thyroid cancer: experience in 10,864 patients with nodular thyroid disorders. *J Clin Endocrinol Metab* 2004;89:163-8.
31. Karga H, Giagourta I, Papaioannou G, Doumouchtsis K, Polymeris A, Thanou S, et al. Changes in risk factors and tumor node metastasis stage of sporadic medullary thyroid carcinoma over 41 years, before and after the routine measurements of serum calcitonin. *Metabolism* 2011;60:604-8.
32. Alevizaki M, Saltiki K, Rentziou G, Papathoma A, Sarika L, Vasileiou V, et al. Medullary thyroid carcinoma: the influence of policy changing in clinical characteristics and disease progression. *Eur J Endocrinol* 2012;167:799-808.
33. Kim M, Kim BH. Current guidelines for management of medullary thyroid carcinoma. *Endocrinol Metab (Seoul)* 2021;36:514-24.
34. Tao Z, Deng X, Guo B, Ding Z, Fan Y. Subgroup analysis of steadily increased trends in medullary thyroid carcinoma incidence and mortality in the USA, 2000-2020: a population-based retrospective cohort study. *Endocr Relat Cancer* 2024;31:e230319.
35. Sahli ZT, Canner JK, Zeiger MA, Mathur A. Association between age and disease specific mortality in medullary thyroid cancer. *Am J Surg* 2021;221:478-84.
36. Vierhapper H, Niederle B, Bieglmayer C, Kaserer K, Baumgartner-Parzer S. Early diagnosis and curative therapy of medullary thyroid carcinoma by routine measurement of serum calcitonin in patients with thyroid disorders. *Thyroid* 2005;15:1267-72.
37. Machens A, Dralle H. Therapeutic effectiveness of screening for multiple endocrine neoplasia type 2A. *J Clin Endocrinol Metab* 2015;100:2539-45.
38. Elisei R, Romei C. Looking for RET alterations in thyroid cancer: clinical relevance, methodology and timing. *Endocrine* 2023;81:206-15.
39. Xu B, Viswanathan K, Ahadi MS, Ahmadi S, Alzumaili B, Bani MA, et al. Association of the genomic profile of medullary thyroid carcinoma with tumor characteristics and clinical outcomes in an international multicenter study. *Thyroid* 2024;34:167-76.