Increased Mortality in Patients With Differentiated Thyroid Cancer Associated With Graves' Disease

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Context: We previously reported that differentiated thyroid cancer (DTC) has higher aggressiveness and poorer prognosis in patients with Graves' disease (GD) than DTC in euthyroid control patients. Subsequent studies on this issue reached controversial conclusions. Genetic and environmental factors, as well as the lack of appropriate control subjects and/or inadequate patient follow-up, may account for these discrepancies.

Objective: The aim of this study was to investigate the long-term disease-specific mortality of nonoccult DTCs occurring in patients with GD compared with DTCs in matched euthyroid control patients.

Patients and Design: The previously described cohorts of nonoccult DTCs occurring in either patients with GD (DTC-GD, n = 21) or matched euthyroid DTC control patients (n = 70) were compared again after a longer follow-up (50–363.6 months; median, 165.6 months) to compare the major clinical endpoints of persistent/recurrent disease and overall survival. Both cohorts were recruited in 1982–1994 at a single institution. All patients had undergone total thyroidectomy and were followed up according to a standardized protocol.

Results: Persistent/recurrent disease was more frequent in DTC-GD patients than in control patients ($P = .0119$). Disease-specific mortality was also significantly higher in DTC-GD patients (6 of 21, 28.6%) than in euthyroid control patients (2 of 70, 2.9%) ($P = .0001$). At the last visit, the percentage of disease-free patients was 57.1% (12 of 21) in the DTC-GD group vs 87.1% (61 of 70) in the control group ($P = .0025$).

Conclusions: Nonoccult DTCs occurring in patients with GD cause increased disease-specific mortality compared with DTCs in matched euthyroid control patients. These findings emphasize the need for early diagnosis and aggressive treatment of nonoccult DTCs in patients with GD. (***J Clin Endocrinol Metab* 98: 1014–1021, 2013**)

Palpable thyroid nodules occur in approximately 15% of patients with Graves' disease (GD) (1–7) compared with the usual rate of 5% observed in the general population (8). When more sensitive echographic screening is used, thyroid nodules are still found more frequently in patients with GD than in the general population (9–13). Moreover, in patients with GD, the malignancy rate of thyroid nodules ranges from 10% to 46%, somewhat higher than that observed in euthyroid patients (2–7). These and other observations strongly suggest that patients with GD have an increased risk of thyroid cancer (14).

We reported previously that differentiated thyroid cancer (DTC) occurring in patients with GD tends to be larger

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Abbreviations: CT, computed tomography; DTC, differentiated thyroid cancer; DTC-GD, patient with DTC and concomitant Graves' disease; FNAB, fine-needle aspiration biopsy; IL, interleukin; GD, Graves' disease; Tg, thyroglobulin; TSHR-Ab, TSH receptor-stimulating antibody.

that DTC occurring in patients with hyperfunctioning thyroid nodules or in euthyroid subjects (15). Moreover, DTCs occurring in patients with GD may occasionally originate distant metastases that respond to TSH receptor–stimulating antibodies (TSHR-Abs) and induce thyrotoxicosis even after total thyroidectomy (16). Moreover, when GD occurs in patients operated on for DTC, it may be associated with rapid growth of local DTC relapse several years after thyroidectomy (17). In addition to the effect of stimulating TSHR-Abs, interleukins (ILs) 4 and 10, locally produced in thyroid glands affected by GD, also have a strong antiapoptotic effect on malignant thyrocytes, despite negatively affecting mitogenesis in other malignant cells (18, 19). Antiapoptotic ILs, therefore, may contribute to thyroid cancer progression and unfavorable prognosis in patients with GD.

At present, neck ultrasound scans and fine-needle aspiration biopsy (FNAB) are established diagnostic procedures for thyroid nodules and may lead to early cancer diagnosis and treatment (20). In a previous study, carried out in patients with DTCs diagnosed in the years 1982–1994, we found that occult DTCs incidentally found at surgery in patients with GD had a good prognosis (15). In contrast, nonoccult DTCs showed a significantly higher frequency of persisting/relapsing disease than DTCs occurring in matched euthyroid control patients (15). Herein, we report that, after prolonged follow-up, patients with GD with nonoccult DTCs have a significantly higher cancer-related mortality than matched control patients.

Patients and Methods

Patients

In the period 1982–1994, we selected a group of patients with DTCs and concomitant GD (DTC-GD) and also a group of euthyroid patients with DTCs matched with the DTC-GD patients on the basis of host and tumor characteristics (15). In brief, the diagnosis of GD was based on history and signs of hyperthyroidism with increased ^{131}I thyroid uptake and the absence of "hot" nodules at scintiscan. The presence of ophthalmopathy and autoimmune involvement of the thyroid (circulating TSHR-Abs, anti-microsomal antibodies and/or anti-thyroglobulin antibodies) confirmed the diagnosis. Patients with a thyroid nodule were subjected to FNAB and referred to surgery unless the nodule contained abundant colloid and a scarce number of follicular cells with pyknotic nuclei. In addition, all patients with GD younger than 40 years with hyperthyroidism relapsing after 10 to 16 months of treatment with methimazole were referred for surgery unless surgical treatment was contraindicated.

Among the 550 patients with GD operated on in the period 1982–1994, 35 DTCs were diagnosed. Among them, 14 cases were occult carcinomas incidentally found at pathology examination after surgery. They were considered separately because they were previously found to have a favorable evolution and no remarkable clinical consequences, and only the 21 nonoccult DTCs were included in the present study. An occult cancer has been defined as follows: a single focus of papillary carcinoma measuring $\leq$1 cm in diameter contained within the thyroid gland of an adult patient found incidentally at thyroidectomy done for other reasons. This definition excludes cancers with multiple foci, lymph node metastases, invasion of the thyroid capsule or blood vessels or tall cell features, and cancer detected preoperatively because of investigation (eg, FNAB) for suspected cancer or metastases (21).

In the DTC-GD group 5 cancers were subcentimetric (range, 4–8 mm; median, 6 mm). Two of them were diagnosed preoperatively because of lymph node metastases. Three were diagnosed preoperatively as suspicious nodules on the basis of ultrasound and cytological findings; all of them were multifocal and 1 had capsule invasion. In the control group, 15 cancers were subcentimetric (range, 2–9 mm; median, 8 mm). Four of them were diagnosed preoperatively because of lymph node metastases; a 2-mm cancer belonged to this group. Eleven were diagnosed preoperatively as suspicious nodules on the basis of ultrasound and cytological findings; 2 of them had lymph node invasion, 2 were multifocal, and 3 had capsule invasion. One was both multifocal and with capsule invasion.

Circulating TSHR-Abs were measured as described previously (4, 16) and were present in 20 of 21 patients with GD with cancer. In the single patient with undetectable TSHR-Abs, signs and symptoms were typical of GD.

Seventy euthyroid control patients operated on for thyroid cancer in the same period ($\pm$2 years) were also selected to match the 21 DTC-GD patients on the basis of sex, age ($\pm$2 years), class of tumor size ($\leq$1, 1.1–4.0, or $>$4 cm), and tumor histotype. This control series included patients with cancer arising as a solitary cold nodule or cancer occurring in a euthyroid multinodular goiter.

The study was approved by the local ethics committee (Ospedale Garibaldi, Catania, Italy) and all participating patients gave written informed consent.

Surgery and postoperative follow-up

All patients with DTCs included in this study underwent total or near-total thyroidectomy plus central compartment lymph node dissection. Later, cervical lymph nodes were dissected only when macroscopically involved or in the presence of extensive invasion of central nodes.

For each patient all histological slides were reviewed, and histopathological diagnoses were graded according to the seventh edition of the World Health Organization thyroid malignancy classification (22). Tumor stage was assessed according to pTNM system (23), where T (size of the primary tumor) and N (regional lymph node metastases) were determined on the basis of pathological data and M (evidence of distant metastases) was based on chest and bone X-rays, computed tomography (CT) scan, magnetic resonance imaging, bone scan, and/or total body scan with ^{131}I.

Postoperative evaluation was performed as described previously (15). Persistent/recurrent disease was defined by 1 or more than 1 of the following criteria: (*a*) serum thyroglobulin (Tg) under either suppressive L-thyroxine therapy or after TSH stimulation at detectable levels and/or higher than the value defined on the basis of the Tg assay sensitivity used at the time of measurement; (*b*) evidence of suspicious lymph nodes during the

neck ultrasound scan, confirmed by fine-needle biopsy with Tg measured in the washout of the aspirate; (*c*) positive posttherapeutic ^{131}I total body scan. In patients with persistent/recurrent disease, other imaging procedures (CT scan, magnetic resonance imaging, bone scan, and positron emission tomography) were performed when appropriate.

Nonsurgically removable distant metastases were treated with ^{131}I (3700–5550 MBq) every 8 to 12 months. Disease progression was defined as enlargement of metastases or tumor masses in the neck (as evaluated by total body scan and/or CT imaging plus serum Tg increase) and/or appearance of new metastatic foci and/or cancer-related death.

Statistical analysis

The distribution of pathological stage and other variables in GD and control groups was compared with the use of contingency tables and χ^2 test or Fisher exact test. Recurrence-free survival and disease-free survival in the 2 patient groups were calculated from the date of surgery and compared using Kaplan-Meier plots. The log-rank test was used to evaluate the differences between curves. Univariate and multivariate analysis of prognostic variables was performed according to the Cox proportional hazards model. The following variables were analyzed for correlation with the occurrence of persisting/relapsing disease or death: patient age (<45 vs ≥45 years), sex, multifocal or bilateral disease, extrathyroidal invasion, pTNM stage, and GD. Only variables identified to be potentially significant at univariate analysis were included in the multivariate model to evaluate their independent effect. Hazard ratios and their 95% confidence intervals were calculated with the Cox proportional hazards regression model. Data analysis was performed using the SPSS version 11 statistical package for Macintosh.

Results

Clinical and histopathological characteristics of the 21 nonoccult DTCs associated with GD and of the 70 DTCs occurring in matched euthyroid control patients are shown in Table 1 and indicate that, at cancer presentation, no significant difference between the 2 groups was present. Follow-up time was also similar. The papillary histotype was highly prevalent in both groups (90.4% in DTC-GD and 92.8% in euthyroid DTC). Tumor size was similar in the 2 groups with a median size of 1.5 cm in both groups (Table 1). Tumor pTNM stage distribution was also similar between the 2 patient groups (Table 2). Although there was a trend toward a higher frequency of high-stage tumors in the DTC-GD group, the difference did not reach statistical significance (Table 2).

Table 1. Clinical and Histopathological Characteristics of DTCs in Patients With GD and in Matched Euthyroid Control Patients

	Patients With GD (n = 21)	Control Euthyroid Patients (n = 70)	*P* Value
Age at diagnosis, y			
Range	20–69	19–67	.358
Median	43	38	
Sex (female/male)	17/4	62/8	.365
Follow-up, mo			
Range	50–343	76–364	.808
Median	165.6	173.3	
Papillary histotype, % (n)	85.71 (18/21)	92.8 (65/70)	.311
Tumor size, cm			
Range	0.4–8.0	0.2–10.0	.336
Median	1.5	1.5	

During postoperative follow-up, the DCT-GD group showed more frequent relapse/persistent disease (11 of 21 vs 19 of 70 adverse events in the DTC-GD and euthyroid DTC group, respectively) and a shorter disease-free survival (78 ± 17 vs 148 ± 10 months) ($P = .0119$, log-rank test) (Figure 1A). Six patients (28.6%) in the DCT-GD group and 2 (2.9%) in the control group died because of the disease (odds ratio = 9.91; 95% confidence interval = 1.99–49.34, Cox regression). Mean survival time was 108 ± 17 months in the DCT-GD group and 195 ± 5 months in the control group. This difference between the 2 groups is highly significant ($P = .0001$, log-rank test) (Table 3 and Figure 1B). Among survivors, 3 patients in the DTC-GD group (3 of 15 [20%]) and 7 of 68 (10.3%) in the DTC control group had recurrent/persistent disease at the last follow-up visit. Overall, 12 of 21 (57.1%) in the DTC-GD group vs 61 of 70 (87.1%) in the euthyroid group ($P = .0025$, χ^2 test) remained disease-free after a mean follow-up period of 214 ± 18 and 187 ± 12 months, respectively (Table 3). Circulating TSHR-Abs were present in all patients who developed a recurrence with the exception of 1 patient. In those patients, circulating TSHR-Abs persisted as long as signs of disease were evident. The only patient with negative TSHR-Abs at recurrence also had negative results at diagnosis, although the diagnosis of GD was obvious.

The relevant clinical and histopathological characteristics of patients who developed distant metastases or died of the disease or showed persisting disease at the last visit are shown in Tables 4 and 5 for the DTC-GD and the euthyroid control groups, respectively.

To evaluate whether the effect of GD on cancer outcome was independent of cancer stage at diagnosis, we analyzed the effect of GD in patients subdivided in 2 categories including those with stage I–II and stage III–IV cancer. In patients with stage III–IV but not in those with stage I–II cancer, cancer relapses were significantly more frequent ($P = .0062$, log-rank test) in patients with GD than in euthyroid control patients. Similarly, cancer-specific deaths were higher in patients with GD and stage I–II cancer although not significantly ($P = .069$) and reached borderline statistical significance ($P = .0586$ and $P = .0285$, log-rank

Table 2. Tumor Stage, as Evaluated According Both to the Fifth pTNM Edition (in use at the Time of Previous Report) and to the Currently Used Seventh pTNM Edition[a]

Tumor Stage	5th pTNM Edition		7th pTNM Edition	
	Patients With GD (n = 21)	Euthyroid Control (n = 70)	Patients With GD (n = 21)	Euthyroid Control (n = 70)
I	12 (57.1)	49 (70.0)	13 (61.9)	53 (75.7)
II	1 (4.8)	8 (11.4)	0	2 (2.9)
III	5 (23.8)	12 (17.2)	1 (4.8)	8 (11.4)
IV	3 (14.3)	1 (1.4)	7 (33.3)	7 (10.0)

[a] Data are n (%). $P = .0554$ and .0584; χ^2 test applied to the tumor stage distribution in patients with GD vs euthyroid patients, respectively, for the fifth and the seventh pTNM edition.

and Breslow test, respectively) in patients with GD and stage III–IV. By applying multivariate Cox analysis, we found that only 2 variables, stage and GD, were significantly and independently associated with total relapses and with cancer-specific deaths. (Please see Supplemental Table 1 published on The Endocrine Society's Journals Online web site at http://jcem.endojournals.org.) Only 1 of 21 (4.8%) patients in the GD group (female, 56 years, T2N1M0) was lost to follow-up after 109 months. She did not show persisting/relapsing disease at any time and was disease-free at the last visit.

In the control group, 12 of 70 (17.1%) patients were lost to follow-up (11 female and 1 male, mean age, 41 ± 9.4 years). Mean follow-up was 84.4 ± 9.2 months. Ten patients were classified as T1N1M0, 1 as T3N1M0, and 1 as T4N0M0. Only this last patient (64 years, female) (Table 5) had developed persisting disease. However, after repeated radioiodine therapy, the patient became disease-free and remained disease-free up to the last visit (101 months of follow-up). The patient died of unrelated (heart) problems.

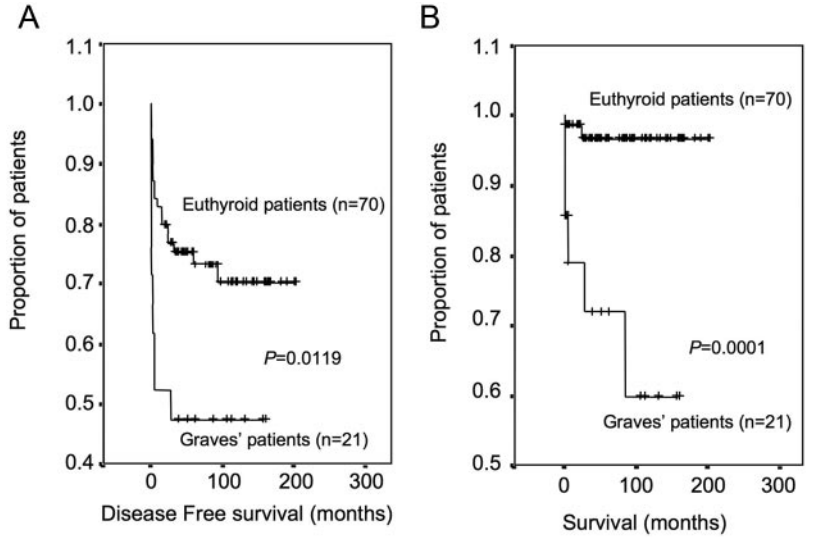

Figure 1. Disease-free time (A) and survival time (B) (Kaplan-Meier plots) in DTC-GD patients and in matched euthyroid control patients. *P* values are calculated according to log-rank test.

Occult carcinomas

Fourteen occult carcinomas occurred in patients with GD (2 male and 12 female). Age at diagnosis ranged from 20 to 71 years (46.1 ± 12.7 years, mean ± SD) and tumor size ranged from 7 to 10 mm (8.9 ± 1.2 mm, mean ± SD). These patients were matched to 16 euthyroid control patients with similar characteristics. Patients were followed for a mean period of 127.1 ± 72.5 months (in the GD group) and for 181.7 ± 85.5 months (in the control group). No patient had evidence of relapse at any time and all of them were disease-free at the last control visit. Individual data are shown in Supplemental Table 2.

Discussion

In the present study, we observe that, despite similar characteristics at presentation and similar treatment, after an average follow-up period of 14 years, patients with non-occult DTC associated with GD have a much higher cancer-related mortality than euthyroid patients with DTC matched for age, sex, and tumor size, all factors known to affect cancer prognosis. In the same series, we previously reported that the cumulative risk for recurrent/progressive distant metastases was approximately 3-fold higher in patients with GD than in euthyroid patients. After a follow-up period ranging from 50 to 363 months, nearly 30% of patients in the GD group had died of cancer, a very high proportion when compared with 3% of matched control subjects. We did not match patients for pTNM stage because such a matching would prevent observing a possible effect of GD on tumor aggressiveness at diagnosis. Our approach has the advantage of allow-

Table 3. Outcome of DTCs in Patients With GD and in Matched Euthyroid Control Patients

Outcome	Patients With GD (n = 21)	Euthyroid Control (n = 70)	*P* Value
Dead, n (%)	6 (28.6)	2 (2.9)	.002[a]
Patients with persistent disease, (%)	3 (14.3)	7 (10)	NS
Detectable Tg	3 (14.3)	2 (2.9)	
Lymph nodes metastases		4 (5.7)	
Distant metastases		1 (1.4)	
Disease-free	12 (57.1)	61 (87.1)	.0025[b]

Abbreviation: NS, not significant.

[a] Fisher's exact test.

[b] χ^2 test.

ing observation of possible differences in metastatic and invasive behavior associated with GD hyperthyroidism between tumors with similar size and with the same histotype. A similar approach has been followed by others (24). However, cancer stage was not significantly different between patients with GD and control patients. Moreover, we observed that GD and stage were the only 2 variables independently related to cancer mortality.

Whether DTCs occurring in patients with GD are generally more aggressive than DTCs occurring in euthyroid patients is debated because controversial data have been reported (13). However, very few studies have addressed this issue in controlled retrospective studies (13). Discordant results can be explained by changing criteria in the preoperative selection of patients, in the thyroid pathologic examination, and in the surgical and postsurgical management of the patient. To our knowledge, only 2 other studies have used a control group of euthyroid patients (24, 25). Both these studies did not show a worse outcome of DTCs associated with GD.

In the first study, from Australia (25), control patients were only matched for the time of diagnosis. Tumor size was 0.9 ± 0.4 cm in the GD group and 2.3 ± 1.7 cm in the control group. More importantly, the GD group contained 15 papillary cancers and 1 follicular cancer, whereas the control group contained 82 papillary, 23 follicular, and 5 anaplastic cancers. However, despite the unfavorable baseline situation in the control group, patients with GD reached a similar outcome. When only papillary cancers were compared, tumor size was 0.64 ± 0.4 cm in the GD group but 1.98 ± 1.3 cm in the control group. Again, no significant difference was found between the 2 groups, suggesting that very small tumors, occurring in patients with GD, behave like larger tumors in euthyroid patients. In the second study, carried out in Japan (24), the thyroid cancers studied were small, with a median diameter of 0.9 and 1.0 cm, in the GD and in the control group, respectively. Median follow-up was approximately 50 months, a short period to observe cancer-related mortality for small thyroid cancers. Mortality, in fact, was very low in both groups (1 of 154 and 2 of 176, respectively, in the GD and the euthyroid group). Moreover, both these studies may have included a large proportion of occult cancers incidentally found at the postsurgical pathology examination. In this regard, we also found that occult DTCs incidentally found in patients with GD at surgery all had a favorable prognosis and none of them relapsed at any time. It is noteworthy that occult thyroid cancers occur very frequently in the population (up to 35.6% in some autopsy studies), and, although sharing the same architectural and cytological features of papillary carcinomas, for the great majority they have no

Table 4. Clinicopathological Characteristics and Outcome of DTCs in Patients With GD, Who Had Developed Distant Metastases or Had Shown Evidence of Persisting/Relapsing Disease at the Last Visit

	Baseline				Outcome				
Age, y	Sex	Tumor Diameter, cm	Histotype and pTNM Stage	Ophthalmopathy/ TSHR-Ab	Follow-Up Duration, mo	Distant Metastases	^{131}I, MBq	Other Therapies	Outcome
59	M	6.0	Foll pT4N0M1	+/2 + +	251	Lung, parotid, brain	52 170	Parotid surgery, RT, Chem	Dead
69	F	6.0	Foll pT4N1M1	+/2 + +	50	Lung	18 500		Dead
46	M	0.7	Pap pT4N1M0	−/+	67	Lung	7400		Tg
66	F	8.0	Pap pT4N1M1	+/+	62	Lung, bone, MED-lymph	22 200		Dead
29	F	2.0	Pap pT4N1M0	+/+	186	Lung, MED-lymph	3700		Dead
60	M	8.0	Pap pT3N0M0		168	Lung	7400		Disease-free
29	F	1.5	Pap pT4mN1M0	−/2+	343	Lung, bone	35 520	Lung surgery	Tg
53	M	8.0	Foll pT4N1M0	−/+	189	Lung, bone	31 450	Lung surgery, Chem	Dead
63	F	2.0	Pap pT4N1M0	−/+	126	Lung	7400	LR-lymph surgery	Dead
20	F	0.4	Pap pT1mN1M0	+/+	189		14 800		Tg

Abbreviations: Chem, chemotherapy; F, female; Foll, follicular cancer; LR-lymph, locoregional lymph nodes; m, multifocal; M, male; MED-lymph, mediastinum lymph nodes; Pap, papillary cancer; RT, external radiotherapy; Tg, detectable serum Tg.

Table 5. Clinicopathological Characteristics and Outcome of DTCs in Euthyroid Control Patients, Who Had Developed Distant Metastases or Had Shown Evidence of Persisting/Relapsing Disease at the Last Visit

Baseline					Outcome				
Age, y	Sex	Tumor Diameter, cm	Histotype and pTNM Stage	Ophthalmopathy/ TSHR-Ab	Follow-Up Duration, mo	Distant Metastases	I-131, MBq	Other Therapies	Outcome
30	F	1.5	Pap pT4N1M0	—	202	Lung	7585		Disease-free
29	F	3.0	Pap pT2N1M0	—	302	Lung	555	MED-lymph surgery	Disease-free
37	F	1.5	Pap pT2N1M0	—	180	Lung	1480		Disease-free
65	F	4.5	Foll pT4mN0M1	—	153	Lung, bone	35 150	Fourth rib resection	Dead
29	F	2.0	Pap pT2N1M0	—	202	Lung	3940		Disease-free
64	F	10	Foll pT4N0M0	—	101	Jugular vein thrombus	7400	Surgery	Disease-free; died of unrelated problems
23	F	3.0	Pap pT2N1M0	—	228		3700		LR-lymph
30	F	2.0	Pap pT2N1M0	—	245	Lung	11 840		Tg
35	F	4.0	Pap pT2N1M0	—	329		20 350	LR-lymph surgery	LR-lymph
54	F	0.5	Pap pT3mN0Mx	—	200	MED-lymph	3700	MED-lymph surgery	LR-lymph
56	F	1.3	Pap pT1mN0M0	—	344		3700		LR-lymph

Abbreviations: Chem, chemotherapy; F, female; Foll, follicular cancer; LR-lymph, locoregional lymph nodes; m, multifocal; M, male; MED-lymph, mediastinum lymph nodes; Pap, papillary cancer; Tg, detectable serum Tg.

clinical significance. Actually, it has been proposed that these lesions be indicated as "papillary microtumors" to underline the uncertain malignant potential of these lesions (21). In addition to these remarkable methodological differences between the Australian and the Japanese studies and our present study, it is possible that genetic and environmental factors also may have influenced the different results. It is worth mentioning that our study has been performed in eastern Sicily, a region including a volcanic area with a high incidence of thyroid cancer (26). Environmental factors, possibly related to the volcanic activity of Mount Etna, may interact with the risk factors operating in patients with GD and specifically contribute to the high mortality observed in our study. Moreover, we excluded occult cancers, which may include tumors with uncertain malignant potential and have an excellent prognosis (21).

The mechanisms leading to the increased frequency and aggressiveness of DTCs in patients with GD are not entirely understood. Several lines of evidence strongly suggest that TSHR-Abs may play a major role in thyroid cancer initiation and progression. TSHR-Abs and TSH activate the same intracellular pathways including both the adenylate cyclase and the phospholipase C cascades (27). These 2 cascades ultimately lead to the activation of the extracellular signal−regulated kinase 1/2, Akt, and nuclear factor-κB pathways (27). This result explains the mitogenic and antiapoptotic effects elicited by both TSH and TSHR-Abs in thyroid follicular cells, especially in the presence of insulin or IGF-I (28–30). Moreover, TSH and TSHR-Abs stimulate angiogenesis in the thyroid, by up-regulating vascular endothelial growth factor, placenta growth factor, and their cognate receptors (flt-1 and Flk-1/KDR) (31, 32). In accordance with these in vitro findings, we and others have observed that concomitant with GD and the appearance of circulating TSHR-Abs, thyroid cancer may relapse years after thyroidectomy and DTC distant metastases may cause thyrotoxicosis (16, 33). Interestingly, cultured thyroid cancer cells from patients with GD respond to the patient's own TSHR-Abs by cAMP accumulation (16). It worth mentioning that in our study circulating TSHR-Abs were present in all patients with GD, who developed a recurrence with the exception of 1 patient. In those patients, circulating TSHR-Abs persisted as long as signs of disease were evident. In accordance with our findings, it is well know that antithyroid antibodies persist for years after thyroidectomy and complete ablation with ^{131}I administration (34, 35) and have a close relationship with persisting/metastatic cancer (36, 37).

We believe that, although our findings are suggestive of a causal relationship between circulating TSHR-Abs and cancer prognosis and mortality, further work is necessary to formally prove it. TSHR-Abs, however, are heterogeneous with regard to serum levels, binding characteristics to the TSH receptor, and biological effects on thyroid cells (38). Therefore, they may affect thyroid cancer in different patients variously.

In addition to TSHR-Abs, IL-4 produced by Th2 lymphocytes in thyroid glands affected by GD may play a role in protecting DTC cells from apoptosis. IL-4 has strong antiapoptotic effects in DTC cells by up-regulating the antiapoptotic molecule Bcl-2 and down-regulation of the proapoptotic molecule Bax (18), and we found previously that thyroid cancer specimens express the IL-4 receptor

(18). Other authors have obtained similar results (19). Finally, we cannot exclude the possibility that the autoimmune process of GD may affect the clinical evolution of thyroid cancer by altering the host immune response to the tumor (39, 40).

In conclusion, nonoccult DTCs associated with GD may develop distant metastases more frequently than DTCs occurring in matched euthyroid patients and cause higher cancer-specific mortality. There is evidence that TSHR-Abs, antiapoptotic ILs and possibly other factors linked to autoimmune inflammation may play a role in DTC progression (39, 40). Further studies are needed to fully clarify how these GD−related factors interact with other genetic and environmental factors. From the clinical point of view, our observations suggest addition of GD to the list of risk factors that should be considered in the screening for early diagnosis of thyroid cancer in thyroid nodules (20).

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