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Original Article

Diagnostic value of combined detection of serological biomarkers in thyroid carcinoma

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Abstract: Objective: To evaluate the diagnostic value of combined detection of serum carcinoembryonic antigen (CEA), thyroglobulin (Tg), calcitonin (CT), and thyroid-stimulating hormone (TSH) using a chemiluminescence assay in thyroid carcinoma (TC). Methods: A total of 320 inpatients with TC - including 261 with papillary TC, 37 with follicular TC, 19 with medullary TC, and 3 with undifferentiated TC - were enrolled as the TC group. Meanwhile, 120 healthy individuals undergoing routine examinations and 120 patients with benign thyroid diseases were included as the control group. Serum levels of CEA, Tg, CT, and TSH were compared between groups and among different pathological types of TC. ROC curves were constructed to assess the diagnostic performance of each biomarker alone and in combination. Results: The combined detection of the four biomarkers yielded a sensitivity of 75.63%, accuracy of 75.54%, and negative predictive value of 69.88%, all higher than those of any single biomarker. ROC analysis showed that the AUC for the combined test of four markers and for the combination of CEA and Tg were 0.840 and 0.768, respectively, both exceeding those of individual tests. The four-marker combination demonstrated the highest diagnostic value. Conclusion: Combined measurement of serum CEA, Tg, CT, and TSH significantly enhances the diagnostic efficacy for TC, reducing both misdiagnosis and missed diagnosis rates, and provides a reliable basis for early clinical detection and intervention.

Keywords: Thyroid carcinoma, carcinoembryonic antigen, thyroglobulin, calcitonin, serum thyroid-stimulating hormone, diagnostic value

Introduction

Thyroid carcinoma (TC) is the most common malignant tumor of the endocrine system. With population aging, lifestyle changes, and advances in medical technology, it has become a major public health concern [1-3]. Addressing this challenge requires not only a deeper understanding of TC pathogenesis but also the development of more accurate and efficient diagnostic strategies. Substantial variations exist among medical institutions in China regarding diagnostic practices, treatment protocols, and disease management, particularly in the use of serum biomarkers for TC diagnosis.

The early symptoms of TC are often nonspecific and are usually detected incidentally during routine physical examinations [4]. Ultrasonography, the most commonly used imaging modality, facilitates early detection, accurate staging, and timely treatment of TC [5, 6]. However, it remains limited in distinguishing benign from malignant nodules. Although histopathological examination provides a definitive diagnosis, it is invasive and requires tissue sampling.

Among available diagnostic approaches, serological biomarker testing is notable for its rapidity, accuracy, non-invasiveness, and reproducibility [7]. Thyroglobulin (Tg), thyroglobulin antibody (Tg-Ab), and calcitonin (CT) are associated with thyroid function and often show alterations in thyroid diseases [8, 9]. Carcinoembryonic antigen (CEA) and thymidine kinase 1 (TK1) are related to thyroid tumor proliferation, while thyroid-stimulating hormone (TSH) assists in distinguishing between benign and malignant nodules [10, 11]. However, no specific biomarker has yet been established for thyroid carcinoma, and the role of Tg in differential diagnosis remains controversial [12, 13].

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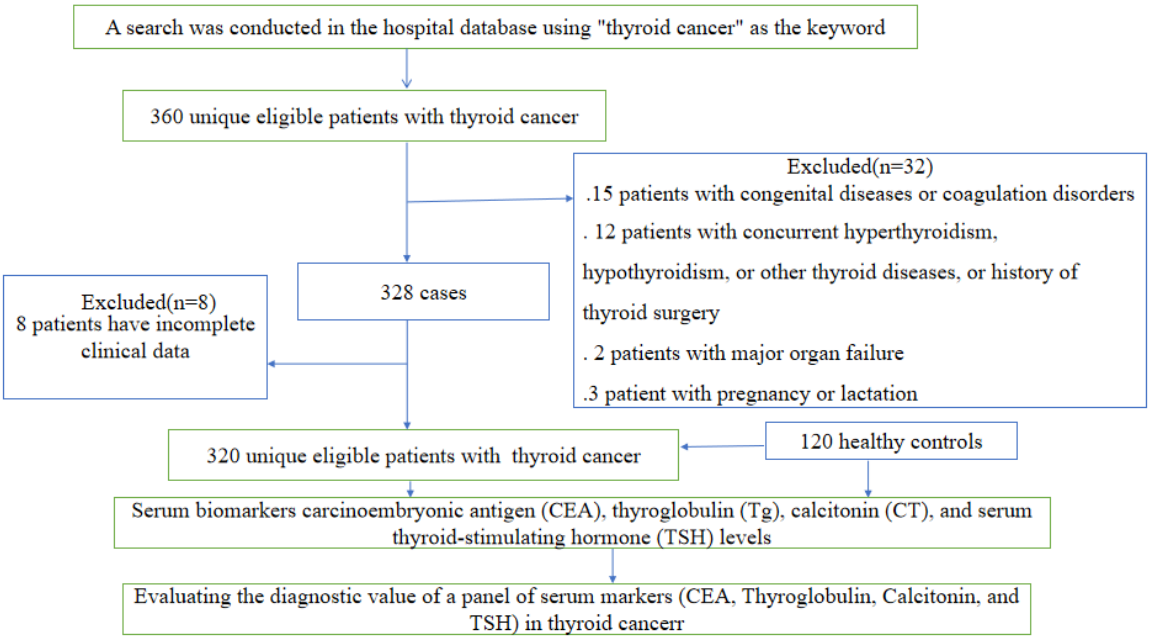

Figure 1. Illustration of the present study.

Recently, various diagnostic models for TC have been proposed, integrating clinical, imaging, and serological parameters [14]. However, most rely on single biomarkers or simple combinations, leading to limited diagnostic accuracy and clinical applicability. Current serological studies remain focused primarily on individual tumor markers, whereas comprehensive evaluations of multiple serological indicators are still scarce [15]. This highlights the need for integrated diagnostic approaches with enhanced efficacy. Therefore, in this study, we systematically assessed the diagnostic performance of CEA, Tg, CT, and TSH individually and in combination, aiming to establish a combined serological model for improving the diagnostic accuracy of TC.

Information and methods

Data collection

TC Group: A total of 320 inpatients with pathologically confirmed TC were recruited from Shenzhen Hospital, Cancer Hospital, Chinese Academy of Medical Sciences, between March 2021 and March 2024.

Inclusion Criteria: ① Age ≥18 years; ② Presence of single or multiple thyroid nodules confirmed by examination; ③ First-time diagnosis with complete clinical data, including preoperative ultrasonography and serum marker tests (Tg, CEA, CT, and TSH); ④ Provision of written informed consent.

Exclusion Criteria: ① Congenital disorders or coagulation abnormalities; ② Coexisting hyperthyroidism, hypothyroidism, or other thyroid diseases, or a history of thyroid surgery; ③ Severe dysfunction of major organs; ④ Pregnancy or lactation.

Control Group: During the same period, 120 healthy controls (HC) and 120 patients with benign thyroid diseases (BTD) were included as controls (**Figure 1**).

Patients with benign thyroid nodules: Inclusion Criteria: ① Age ≥18 years; ② Presence of one or more benign thyroid nodules confirmed by ultrasonography and/or fine-needle aspiration cytology (FNAC); ③ First-time participation with complete clinical data. Exclusion Criteria: ① Congenital disorders or coagulation abnormalities; ② History of thyroid dysfunction (e.g., hyperthyroidism, hypothyroidism, thyroiditis) or prior thyroid surgery/intervention; ③ Confirmed malignant thyroid nodules; ④ Severe dysfunction of major organs (heart, liver, kidneys, etc.); ⑤ Pregnancy or lactation; ⑥ Inability to comply with study procedures.

Healthy controls: Inclusion Criteria: ① Age ≥18 years; ② No history of thyroid disease or thy-

Table 1. Baseline data the thyroid cancer group, healthy control group, and benign disease control group

Variable	Group A (n = 320)	Group B (n = 120)	Group C (n = 120)	*P*-value
Sex (n, %)				0.458
Male	168 (52.5%)	58 (48.3%)	66 (55.0%)	
Female	152 (47.5%)	62 (51.7%)	54 (45.0%)	
Age (years)	56.8 ± 9.5	58.2 ± 8.7	57.3 ± 10.1	0.327
BMI (kg/m^2)	24.8 ± 3.2	25.1 ± 2.9	24.5 ± 3.4	0.215
TI-RADS Grading (n, %)				<0.001
Grade I	22 (6.9%)	5 (4.2%)	-	
Grade II/III	88 (49.4%)	68 (56.7%)	-	
Grade IV	210 (43.7%)	47 (39.1%)	-	
Family genetic history				0.778
Yes	110 (34.3)	50 (41.7)	-	
No	210 (63.6)	70 (58.3)	-	
Radiation exposure				0.942
Yes	5 (0.3)	3 (2.5)	-	
No	315 (97.7)	2 (97.5)	-	
Metabolic syndrome				0.884
Yes	12 (3.75)	8 (6.67)	-	
No	308 (96.25)	112 (93.34)	-	
Hypertension (n, %)				0.642
Yes	134 (41.9%)	53 (44.2%)	47 (39.2%)	
No	186 (58.1%)	67 (55.8%)	73 (60.8%)	
Diabetes (n, %)				0.751
Yes	67 (20.9%)	28 (23.3%)	23 (19.2%)	
No	253 (79.1%)	92 (76.7%)	97 (80.8%)	

BMI: Body mass index; TI-RADS: Thyroid Imaging, Reporting and Data System.

roid surgery; ③ Normal thyroid function and ultrasonography findings. Exclusion Criteria: ① Severe chronic systemic disease; ② History of neck irradiation; ③ Recent use of medications affecting thyroid function; ④ Pregnancy or lactation; ⑤ Inability to comply with study procedures.

Due to the retrospective nature of the study, the requirement for informed consent was waived by the Ethics Committee of the National Cancer Center/National Clinical Research Center (Approval No.: KYKT2024-40-1).

Baseline demographic and clinical characteristics of the three groups are summarized in **Table 1**.

Experimental methods

Sample Collection and Processing: Fasting venous blood (5 mL) were collected from each participant. After centrifugation for 10 minutes, 600 μL of serum were retained following routine clinical testing and stored at -80°C for further analysis.

Biomarker Detection: All biomarkers were measured using chemiluminescence immunoassays. The instruments used were the cobas e602 analyzer (for CEA) and the CL-6000i analyzer (for Tg, CT, and TSH). All reagents and consumables were manufacturer-matched original kits. Standardized operating procedures were strictly followed to ensure test reliability.

The validated reference ranges were: CEA = 0-5 ng/mL; Tg = 1.28-50 ng/mL; CT = 0-9.2 pg/mL; TSH = 0.35-5.1 μIU/mL.

Judgment Criteria: Values exceeding the upper limit of the reference range were defined as positive. In the combined test, positivity was determined if any of the four biomarkers was positive; results were considered negative only

Table 2. Comparison of biomarker levels in the thyroid cancer group, healthy control group, and benign disease control group

Groups	CEA (ng/ml)	Tg (ng/ml)	CT (pg/ml)	TSH (uIU/ml)
Thyroid cancer Group	14.40 ± 2.31	86.53 ± 24.76	19.56 ± 3.18	6.94 ± 1.76
Benign Disease Controls	2.43 ± 1.18	40.33 ± 12.28	2.98 ± 1.65	3.45 ± 1.27
Healthy Controls	1.47 ± 1.11	14.72 ± 6.23	1.51 ± 1.20	2.06 ± 1.06
t-value				
Thyroid cancer vs. Benign	7.993	2.895	8.008	2.790
Thyroid cancer vs. Healthy	8.738	4.872	9.172	4.102
Benign vs. Healthy	1.026	3.221	1.231	1.525
P-value				
Thyroid cancer vs. Benign	<0.001	0.045	<0.001	0.049
Thyroid cancer vs. Healthy	<0.001	<0.001	<0.001	0.015
Benign vs. Healthy	0.363	0.032	0.286	0.202

Table 3. Comparison of biomarker positivity rates among three groups: thyroid cancer group, healthy control group, and benign disease control group

Groups	Number of cases	CEA Number of cases (positivity rate %)	Tg Number of cases (positivity rate %)	CT Number of cases (positivity rate %)	TSH Number of cases (positivity rate %)	Combined tests Number of cases (positivity rate %)
Thyroid cancer Group	320	139 (43.44)	67 (20.94)	87 (27.19)	48 (15.00)	242 (75.63)
Benign Disease Controls	120	8 (6.67)	6 (21.67)	32 (26.67)	17 (12.5)	49 (40.83)
Healthy Controls	120	1 (0.83)	5 (4.17)	1 (0.83)	3 (2.50)	10 (8.33)
χ^2 value		112.146	18.838	38.490	13.368	169.219
P value		<0.001	<0.001	<0.001	<0.001	<0.001

CEA: Carcinoembryonic Antigen; Tg: thyroglobulin; CT: calcitonin; TSH: thyroid-stimulating hormone.

when all markers were within the reference ranges.

Primary Observation: To determine whether combined biomarker detection provides superior diagnostic value compared with individual markers.

Secondary Observation: To compare differences in serum biomarker levels (CEA, CT, TSH) among the groups.

Statistical analysis

All analyses were performed using SPSS 19.0. Continuous variables were expressed as mean ± standard deviation (x±sd). Data normality and homogeneity of variance were evaluated using the Shapiro-Wilk and Levene tests, respectively. Normally distributed data with equal variances were analyzed using the independent-samples t-test; data violating variance homogeneity were analyzed using Welch's t-test.

Categorical variables were expressed as counts and percentages (%) and compared using the chi-square test. Multivariate logistic regression was performed to develop a predictive model for TC, using backward stepwise selection with inclusion and exclusion significance levels of $\alpha\_in = 0.05$ and $\alpha out = 0.10$, respectively.

The DeLong test was used to compare the AUCs of different diagnostic models. Multiple comparisons were adjusted using the Bonferroni correction, setting the significance threshold at $\alpha = 0.05/6 \approx 0.008$.

Because only three cases of undifferentiated TC were included, these patients were analyzed within the overall TC group.

Results

Comparison of CEA, Tg, CT, and TSH levels among groups

The levels of all four biomarkers (CEA, Tg, CT, and TSH) were generally higher in the TC group compared with both control groups (all P<0.05). Specifically, Tg levels showed statistically sig-

Table 4. Comparison of biomarker levels among three pathological subtypes of thyroid cancer

Pathological types	CEA (ng/ml)	Tg (ng/ml)	CT (pg/ml)	TSH (uIU/ml)
Papillary thyroid cancer	12.91 ± 3.52	92.32 ± 20.28	18.21 ± 1.08	3.52 ± 1.51
Follicular thyroid carcinoma	28.61 ± 5.34	70.76 ± 10.23	19.00 ± 2.81	4.27 ± 1.86
Medullary thyroid carcinoma	7.39 ± 2.05	78.21 ± 13.05	26.55 ± 3.50	2.45 ± 1.15
t1 value	4.252	1.269	0.455	0.327
p1 value	0.013*	0.273	0.673	0.759
t2 value	2.347	1.279	3.944	0.976
p2 value	0.079	0.270	0.017*	0.384
t3 value	6.426	0.262	2.913	1.442
p3 value	<0.001*	0.870	0.044*	0.223

CEA: Carcinoembryonic Antigen; Tg; thyroglobulin; CT: calcitonin; TSH: thyroid-stimulating hormone. *: $P < 0.05$ is statistically significant.

nificant differences between the TC group and each control group (benign thyroid disease and healthy controls, $P<0.05$). Similarly, CEA, CT, and TSH levels were significantly higher in the TC group than in either control group (all $P<0.05$). However, when comparing the benign thyroid disease group with the healthy control group, only Tg demonstrated a significant difference. Detailed results are shown in **Table 2**.

Comparison of biomarker positivity rates among groups

In the TC group, the positivity rate of CEA was the highest among single tests (43.44%). When the four biomarkers were tested in combination, the positivity rate increased markedly to 75.63%. Across all three groups, the differences between the single and combined tests were statistically significant ($P<0.05$). The detailed results are presented in **Table 3**.

Comparison of CEA, Tg, CT, and TSH levels among different pathological types of TC

Tg levels were highest in patients with papillary thyroid carcinoma, although the differences among the three pathological subtypes were not statistically significant. CEA levels were highest in follicular thyroid carcinoma, with statistically significant differences compared to both papillary and medullary TC. CT levels were highest in medullary TC and differed significantly from those in papillary and follicular subtypes. No significant difference in TSH levels was observed among the three pathological types (all $P>0.05$). The detailed results are shown in **Table 4**.

Comparison of biomarker positivity rates in different clinical and pathological subgroups of TC

The combined assay demonstrated higher positivity rates across all subgroups of TC patients compared with single biomarker assays.

By gender: Only Tg positivity showed a significant difference, while other biomarkers and the combined test did not.

By pathological type: Tg and CT positivity rates varied significantly, whereas CEA, TSH, and the combined test showed no significant differences.

By clinical stage, lymph node status, and distant metastasis: Both individual and combined tests showed significant variations.

Detailed subgroup results are provided in **Table 5**.

Comparison of diagnostic efficiency between single and combined biomarker tests

In diagnosing TC, the single-marker tests demonstrated higher specificity than the combined test, whereas the combined test showed superior sensitivity, accuracy, and negative predictive value (NPV). Among individual biomarkers, CEA had the highest positive predictive value (PPV).

As illustrated in **Figure 2**, receiver operating characteristic (ROC) curves were used to evaluate diagnostic performance. The AUCs for CT and TSH were 0.548 and 0.531, respectively, indicating limited diagnostic value. CEA achieved an AUC of 0.758, reflecting better discrim-

Table 5. Comparison of serum biomarker positivity rates in thyroid cancer patients in different characteristic groups

Groups	Number of cases	CEA Number of cases (positivity rate %)	Tg Number of cases (positivity rate %)	CT Number of cases (positivity rate %)	TSH Number of cases (positivity rate %)	Combined tests Number of cases (positivity rate %)
Genders						
Male	120	47 (40.83)	37 (30.83)	38 (31.67)	15 (12.50)	90 (75.00)
Female	200	92 (46.00)	30 (15.00)	49 (24.50)	33 (16.50)	152 (76.00)
χ^2 value		1.425	11.358	1.946	0.941	0.041
P value		0.233	<0.001	0.163	0.332	0.840
Pathological types						
Papillary thyroid cancer	261	114 (43.68)	45 (17.24)	68 (26.05)	42 (16.09)	195 (74.71)
Follicular thyroid carcinoma	37	16 (43.24)	17 (45.95)	9 (24.32)	3 (8.11)	28 (75.51)
Medullary thyroid carcinoma	19	9 (47.37)	5 (26.32)	10 (52.63)	3 (15.79)	16 (83.33)
χ^2 value		0.104	16.344	6.488	1.614	0.863
P value		0.949	<0.001	0.039	0.446	0.650
Clinical Stages						
I+II	212	73 (34.43)	20 (9.43)	27 (12.74)	16 (7.55)	86 (40.57)
III+IV	108	66 (61.11)	47 (43.52)	60 (55.56)	32 (29.63)	92 (85.19)
χ^2 value		20.725	50.215	62.470	27.365	57.709
P value		<0.001	<0.001	<0.001	<0.001	<0.001
lymph node metastasis						
Yes	203	118 (53.20)	51 (25.12)	61 (30.50)	39 (19.21)	163 (80.30)
No	117	31 (26.50)	16 (13,68)	26 (22.22)	9 (7.69)	48 (41.03)
χ^2 value		29.848	5.876	2.297	7.725	50.962
P value		<0.001	0.015	0.030	0.005	<0.001
Distal transfer						
Yes	62	42 (67.74)	23 (37.10)	31 (50.00)	19 (31.15)	52 (83.87)
No	282	97 (34.40)	44 (15.60)	56 (19.86)	29 (10.28)	123 (43.61)
χ^2 value		23.469	14.972	24.439	17.550	32.953
P value		<0.001	<0.001	<0.001	<0.001	<0.001

CEA: Carcinoembryonic Antigen; Tg: thyroglobulin; CT: calcitonin; TSH: thyroid-stimulating hormone.

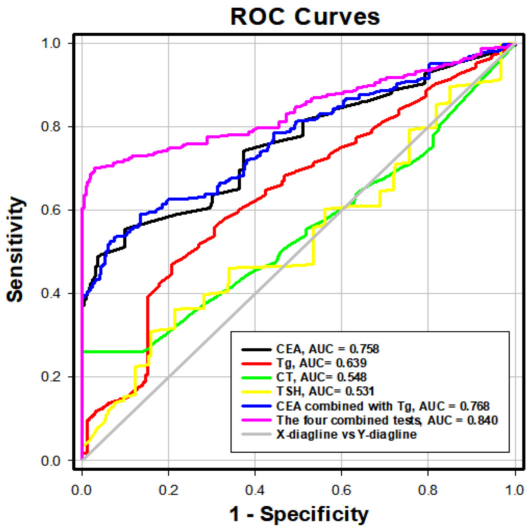

Figure 2. ROC curves for the diagnostic value of thyroid cancer. ROC curve analysis revealed the area under the curve (AUC) values for CEA, thyroglobulin (Tg), calcitonin (CT), thyroid-stimulating hormone (TSH), CEA combined with Tg, and the four combined tests for diagnosing thyroid cancer were 0.758, 0.639, 0.548, 0.531, 0.768, and 0.840, respectively.

inative ability, while Tg yielded a moderate AUC of 0.639. The combination of CEA and Tg further improved diagnostic accuracy (AUC = 0.768). The four-biomarker combination (CEA, Tg, CT, and TSH) produced the highest diagnostic value (AUC = 0.840).

Statistical comparison (DeLong test) showed that the AUCs of the combined test were significantly higher than those of all single markers ($P<0.001$), indicating complementary diagnostic roles among the biomarkers. Detailed results are shown in **Table 6**.

Multivariate logistic analysis of TC

Multivariate analysis showed that CEA, Tg, CT, TSH, and ultrasound rating as independent influencing factors for TC. Substituting the regression coefficients from **Table 7** into the equation produced the following predictive model: P=1/(1+e^)[X=7.679-0.999×(x1)-0.133×(x2)-0.297×(x3)-0.472×(4)-3.736×(5)]. (See **Table 7** for parameter details).

Overall, ROC curve analysis confirmed that the combined detection of CEA, Tg, CT, and TSH provided the highest diagnostic accuracy for TC, representing the most valuable approach for clinical diagnosis and intervention (**Table 8**).

Discussion

Serological biomarkers play a crucial role in the diagnosis and monitoring of thyroid carcinoma, with Tg, CT, CEA, and TSH being the most frequently used indicators [12, 13, 16, 17]. Accurate and early diagnosis remains critical for improving prognosis. Serological biomarkers are increasingly recognized as convenient, non-invasive adjuncts to imaging in TC detection. This study evaluated the diagnostic performance of four serum biomarkers individually and in combination, aiming to enhance diagnostic accuracy and clinical applicability.

Our results demonstrated that the combined assay achieved superior diagnostic performance compared with single-marker detection, with an AUC of 0.840, exceeding that of Doppler ultrasonography (AUC = 0.804) [18]. Among different TC subtypes, CT was the most frequently elevated marker, showing positivity rates of 43.68%, 45.95%, and 52.63% in papillary, follicular, and medullary carcinoma, respectively. The positivity rate of the combined test reached 75.63% overall and 83.33% among TC subtypes, significantly higher than any single marker. These findings indicate that the four markers complement each other and provide a more comprehensive reflection of tumor activity and functional disturbance.

In contrast, the control group exhibited lower positivity rates, whereas the benign disease group showed moderately higher levels than controls, possibly due to benign disease-related stimulation enhancing marker expression [18-20]. Among the single markers, CEA exhibited the best discriminative ability (AUC = 0.758), followed by Tg (AUC = 0.639), while CT and TSH alone showed limited diagnostic value (AUCs 0.548 and 0.531, respectively). Elevated Tg levels in TC compared with benign thyroid disease and healthy controls are consistent with previous studies [21-23]. However, Tg alone is not specific, as it may also increase in autoimmune thyroiditis, adenoma, or subacute thyroiditis [24]. CEA was most elevated in follicular TC, whereas CT was highest in medullary TC, in line with its biological role as a calcitonin-secreting tumor marker [25]. These subtype-specific differences underscore the necessity of multi-marker detection to capture the heterogeneous biological features of TC.

Table 6. Efficiency of the four biomarkers in diagnosing thyroid cancer by single and combined tests

Biomarkers	AUC	Truncation value	Sensitivity	Specificity	Accuracy	PPV	NPV
CEA	0.758 (0.673-0.843)	5.47 ng/ml	43.44%	96.25%	66.07%	93.92%	56.07%
Tg	0.639 (0.545-0.733)	28.91 ng/ml	20.94%	87.08%	49.29%	68.37%	45.24%
CT	0.548 (0.451-0.645)	29.15 pg/ml	27.19%	86.25%	52.50%	72.50%	47.05%
TSH	0.531 (0.434-0.628)	3.46 ug/ml	15.00%	91.67%	47.86%	70.59%	44.72%
CEA combined with Tg	0.768 (0.685-0.851)		51.56%	85.83%	66.25%	82.91%	57.06%
The four combined tests	0.840 (0.769-0.911)		75.63%	75.42%	75.54%	80.40%	69.88%

CEA: Carcinoembryonic Antigen; Tg: thyroglobulin (Tg); CT: calcitonin (CT); TSH: thyroid-stimulating hormone.

Table 7. Multivariate logistic analysis of thyroid cancer

Variable	β	OR	95% CI	P
CEA (x1)	0.999	1.978	1.011-3.932	0.021
Tg (x2)	0.133	1.657	1.110-3.847	0.037
CT (x3)	0.297	1.680	1.436-2.812	0.010
TSH (x4)	0.472	1.766	1.172-3.704	0.003
TI-RADS Grading (IV) (x5)	3.736	17.318	2.772-22.358	<0.001
Constant	7.679	4.660	3.001-11.550	0.032

CEA: Carcinoembryonic Antigen; Tg: thyroglobulin; CT: calcitonin; TSH: thyroid-stimulating hormone; TI-RADS: Thyroid Imaging, Reporting and Data System.

Table 8. Comparison of diagnostic efficacy between combined serological markers and individual markers using Delong test

Model	AUC	95% CI	Comparison	Z-value	*p*-value
Combined Model	0.840	0.792-0.894	-	-	-
CEA	0.758	0.654-0.770	vs. Combined	-5.322	<0.001
Tg	0.639	0.625-0.745	vs. Combined	-6.179	<0.001
CT	0.548	0.452-0.680	vs. Combined	-4.027	<0.001
TSH	0.534	0.468-0.697	vs. Combined	-7.298	<0.001

CEA: Carcinoembryonic Antigen; Tg: thyroglobulin; CT: calcitonin; TSH: thyroid-stimulating hormone.

The four-marker combination demonstrated the highest sensitivity, accuracy, and NPV, confirming its clinical utility for early detection and screening. Although specificity decreased compared with single tests, this trade-off is acceptable in screening contexts, where sensitivity and NPV are prioritized to minimize missed diagnoses. A negative combined test could reliably exclude malignancy, serving as an effective rule-out tool. Positive cases, in contrast, should undergo confirmatory imaging or biopsy to prevent overtreatment.

In patients with lymph node or distant metastasis, combined testing yielded higher positivity rates than in localized disease, suggesting that simultaneous elevation of multiple biomarkers may reflect higher tumor burden or systemic dissemination. Thus, this multimarker model could also serve as a surrogate indicator for disease aggressiveness and metastatic potential [26-28].

ROC analysis showed that the diagnostic value of combined biomarkers (AUC = 0.840) approached that of contrast-enhanced or elastographic ultrasound (AUC = 0.85-0.90) [29]. This supports the concept of multimodal precision diagnosis - integrating serological and imaging data for optimal clinical decision-making. In particular, combining the high sensitivity of serum markers with the anatomical accuracy of ultrasound could enable more refined patient stratification and reduce unnecessary invasive procedures [30, 31].

Serological testing is noninvasive, cost-effective, and easily standardized, making it well-suited for large-scale screening or postoperative surveillance. The observed improvements in sensitivity and early-stage detection suggest that multi-marker assays may facilitate timely intervention and better prognostication. Moreover, incorporating CEA, Tg, CT, and TSH into clinical workflows could enhance diagnostic confidence, especially in resource-limited settings where advanced imaging is not universally available.

The findings also have potential implications for personalized management. For instance, elevated CT may guide suspicion toward medullary TC and prompt genetic screening for RET mutations, while CEA and Tg trends could assist

in postoperative monitoring for recurrence or residual disease. Such marker-guided algorithms align with current trends toward precision endocrinology and individualized patient care.

Several limitations warrant consideration. First, the small number of undifferentiated TC cases limited statistical power for subtype-specific conclusions; results for this group should be interpreted cautiously. Second, this was a single-center retrospective study, which may introduce selection bias and limit generalizability. Third, although chemiluminescence-based assays offer high reproducibility, inter-laboratory variability in cutoff values and kit performance remains a potential confounder. Finally, no external validation cohort was available to confirm the robustness of the predictive model.

Future research should focus on multi-center prospective validation with larger and more diverse populations. Integration of serological and radiomic features, as well as machine learning-based fusion models, could further refine diagnostic accuracy. Additionally, exploring longitudinal marker dynamics before and after surgery may elucidate their prognostic and surveillance roles in TC management.

In conclusion, combined chemiluminescence detection of CEA, Tg, CT, and TSH significantly enhances the diagnostic accuracy of TC compared with individual markers. This multi-marker strategy provides a sensitive, noninvasive adjunct to imaging, offering clinical value in early detection, subtype differentiation, and disease monitoring. With further validation, this approach may represent an important step toward more precise and individualized diagnosis of TC.

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Disclosure of conflict of interest

None.

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