Preoperative serum inflammation-based scores in medullary thyroid cancer

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Abstract

Introduction: Neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR) and systemic immune-inflammation index (SII) are prognostic factors in several tumours, however little is known in medullary thyroid cancer (MTC).

Objective: To evaluate the association between preoperative NLR, PLR and SII with MTC clinicopathological and molecular features, and their predictive value for lymph-node and distant metastasis.

Methods: We retrospectively analysed 75 patients with MTC who underwent surgery at our institution.

Results: In our cohort, 56% were females, the median age at diagnosis was 57 years (44–69), the median tumour diameter was 25mm (15–50); 21.3% were multifocal and 34.7% had extrathyroidal extension. Fibrosis was present in 30 of the 37 analysed samples; *RET* somatic status was assessed in 35 cases and 21 harboured a mutation. Lymph-node and distant metastasis were observed in 36 (48.0%) and 8 (10.7%), respectively. Higher NLR was associated with preoperative calcitonin, angioinvasion, extrathyroidal extension, moderate/severe fibrosis; higher PLR was associated to extrathyroidal extension and advanced T stages; lower SII and NLR were associated with biochemical cure after surgery. Increased PLR, NLR and SII were associated with advanced MTC stages. In the univariate analysis, only NLR was associated with lymph-node metastasis (odds ratio (OR) = 2.69, 95% confidence interval (CI): 1.50–5.84; p = 0.004); however, in the multivariate model, NLR was no longer a predictive factor for lymph-node metastasis.

Conclusion: None of these serum inflammatory markers predicted the occurrence of distant metastasis. In conclusion, NLR, PLR and SII may indicate aggressive MTC disease, but do not predict lymph-node or distant metastasis.

Introduction

Medullary thyroid cancer (MTC) arises from the parafollicular cells of thyroid parenchyma, and it can occur sporadically, or be hereditary in case of germline *RET* proto-oncogene mutations [1]. MTC represents less than 5% of thyroid malignancies, but is responsible for 13.4% of thyroid cancer-related deaths [2]. MTC is more often associated with lymph-node and distant metastasis in comparison to differentiated thyroid cancer, and as many as 70% and 10 % of patients with MTC present, respectively, with nodal or distant metastasis at diagnosis [3]. Surgery is the treatment of choice for loco-regional disease, consisting of at least total thyroidectomy with central lymph node dissection [4].

Tumourigenesis results from the imbalance between cancer promoting and cancer-inhibiting molecular pathways. Inflammation plays an important role in tumour biology, not only in the local tumour microenvironment, but also systemically, influencing tumour progression or recurrence, and promoting tumour cell proliferation, angiogenesis, invasion and metastasis [5, 6]. Responses to systemic

inflammation include alterations in the haematopoiesis and in the secretion of acute-phase proteins, cytokines, growth factors and hormones. Different serum inflammation-based scores, such as the neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR) and systemic immune-inflammation index (SII), can reflect the degree of systemic inflammation and predict clinical outcomes and prognosis of patients with cancer [7–10].

Several studies have reported a critical role for neutrophils in terms of tumour progression, through the release of cytokines and angiogenic factors. Also lymphocytes can secrete large amounts of cytokines, which can inhibit the proliferation of cancer cells [9]. Granulocyte-colony stimulating factor and other cytokines, secreted by the tumour cells or non-neoplastic cells within the tumour microenvironment, induce neutrophils proliferation and myeloid suppressor cell production by bone marrow, and inhibits lymphocyte proliferation, which result in an elevation of NLR, PLR and SII [11]. Hence, in general, high NLR, PLR and SII are associated with poorer cancer outcomes. There are a large number of studies analysing the clinical usefulness of NLR and PLR in different cancers [7, 8, 11, 12], including in endocrine-related cancers [13–16], but little is known in MTC [17–19].

Recently, the biomarker SII has been also shown to predict clinical outcomes in a variety of cancers [20–24], with some studies highlighting its superiority over other inflammatory markers, including PLR and NLR, as it combines lymphocyte, neutrophil and platelet counts, and therefore reflects better the balance between the inflammatory and the immunity status [21, 23, 25]. There are no studies investigating the role of SII in patients with MTC.

Thus, we aimed to evaluate the association of the preoperative serum NLR, PLR and SII with MTC clinicopathological and molecular features at diagnosis, and to determine their usefulness in predicting lymph-node and distant metastasis.

Methods

Study population

Patients diagnosed with MTC and submitted to surgery between 1990 and 2016 at our institution were retrospectively analysed. Patients with first surgery performed at another hospital, with unresectable tumour at the time of referral and those who received any other form of treatment before the operation (such as radiotherapy, chemotherapy or targeted therapy with tyrosine kinase inhibitors) were excluded from the study. Patients with chronic medical disorders affecting full blood count, Hashimoto's thyroiditis, ectopic Cushing's syndrome, history of other active malignancy and infectious/inflammatory diseases or patients with acute myocardial infarction in the past 6 months, as well as patients having immunosuppressive therapy or corticosteroids, were also excluded. The study population consisted of a total of 75 MTC patients.

Clinico-pathological Features And Definition Of The Study Subgroups

Patients' demographic data, preoperative levels of calcitonin, histological characteristics of the tumour, such as size, multifocality, fibrosis, angioinvasion, microscopic extrathyroidal extension, as well lymph-node and distant metastasis at diagnosis, and somatic *RET* 918 and 883 codons mutation status were obtained. Biochemical cure of the disease was defined as undetectable postoperative serum calcitonin levels during the follow-up. The grade of fibrosis was recorded as being negative (absent), low (+), moderate (++), or intense (+++) [26]. Patients were divided in two groups: group 1 (absent and mild fibrosis) and group 2 (moderate and severe fibrosis). *RET* and *RAS* molecular characterisation of the series has been previously reported [27, 28].

The Tumour-Node-Metastasis (TNM) classification of all tumour specimens and stage grouping were performed according to the criteria described in the American Joint Committee on Cancer (AJCC) TNM classification of MTC [29], and two subgroups including T1/T2 patients and T3/T4 patients were formed and subject to comparative analysis. Regarding the AJCC stage, two subgroups were also formed, one including stage I and II patients and the other including patients staged III and IV.

Preoperative haematological data collection and calculation of serum inflammatory ratios

Blood samples were routinely taken 3 days prior to surgery and analysed at our institution's certified laboratory in a standardised manner on automated counters. The preoperative NLR was calculated dividing the absolute neutrophil count by the absolute lymphocyte count. PLR was calculated by dividing the absolute platelet count by the absolute number of lymphocytes. SII was defined as the absolute neutrophil count multiplied by the absolute platelet count divided by the absolute lymphocyte count.

Statistical analysis

Categorical variables are shown as absolute number and percentage, while the continuous non-normally distributed (as determined by Gaussian distribution with the Shapiro-Wilk test) variables data are shown as median and interquartile range (IQR). Non-parametric data were further analysed with Mann-Whitney U test. Correlations between continuous variables were determined by Spearman's correlation coefficient (rho) for non-normally distributed variables. To evaluate if NLR, PLR and SII represent independent risk factors for lymph-node and distant metastasis, we used multivariable linear regression analysis. A 95% confidence interval (CI) was used to estimate the precision of the odds ratio (OR). Statistical analyses were carried out using the SPSS software version 25.0 (IBM, USA). A p-value < 0.05 was considered statistically significant.

Results

The study cohort included 75 patients, 42 were women (56%), with a median age at MTC diagnosis of 57 years (44–69). Median NLR was 2.2 (1.7–3.3), median PLR was 141.5 (108.8-192.6) and median SII was 615.4 (384.0-931.2). The main characteristics of our cohort of MTC patients are shown in Table 1. Concerning histopathological characteristics, the median tumour diameter was 25 mm (15–50), multifocality was seen in 16 (21.3%) patients, 26 (34.7%) had extrathyroidal extension and 43 (57.3%) angioinvasion. Regarding TNM classification, 22 (29.3%) patients were classified as pT1, 13 (17.3%) as pT2, 18 (24.0%) as pT3, 17 (22.7%) as pT4a and 5 (6.7%) as pT4b (Table 1). Presence of fibrosis was assessed in 37 samples: in 7 (18.9%) fibrosis was absent, in 12 (32.4%) was mild, in 11 (29.7%) was moderate and in 7 (18.9%) was severe. *RET* somatic status was analysed in 35 patients, and 21 harboured a *RET* somatic mutation (Table 2).

Table 1
Baseline clinicopathological characteristics of the studied population

Age, median (IQR), years	**57 (44–69)**
Female, n (%)	42 (56.0)
Serum calcitonin at diagnosis, median (IQR), pg/mL	1699 (7232)
Largest tumour diameter, median (IQR), mm	25 (15–50)
Multifocality, n (%)	16 (21.3)
Extrathyroidal extension, n (%)	26 (34.7)
Angioinvasion, n (%)	43 (57.3)
TNM classification – Primary tumour (T), n (%)	22 (29.3)
T1	13 (17.3)
T2	18 (24.0)
T3	17 (22.7)
T4a	5 (6.7)
T4b	
Lymph-node metastasis at diagnosis, n (%)	36 (48.0)
Central	5 (13.9)
Lateral and Central	31 (86.1)
Distant metastasis at diagnosis, n (%)	8 (10.7)
AJCC staging, n (%)	18 (24.0)
I	15 (20.0)
II	5 (6.7)
III	25 (33.3)
IVA	4 (5.3)
IVB	8 (10.7)
IVC	
Biochemical cure after surgery, n (%)	28 (37.3)
Follow-up, median (IQR), months	72 (36–180)

AJCC, American Joint Committee on cancer; CEA, carcinoembryonic antigen; IQR, interquartile range; NLR, neutrophil-to-lymphocyte ratio; PLR platelet-to-lymphocyte ratio; SD, standard-deviation SII, systemic immune-inflammatory index; TNM, Tumour-Node-Metastasis.

Age, median (IQR), years	**57 (44–69)**
NLR, median (IQR)	2.2 (1.7–3.3)
PLR, median (IQR)	141.5 (108.8-192.6)
SII, median (IQR)	615.4 (384.0-931.2)
AJCC, American Joint Committee on cancer; CEA, carcinoembryonic antigen; IQR, interquartile range; NLR, neutrophil-to-lymphocyte ratio; PLR platelet-to-lymphocyte ratio; SD, standard-deviation SII, systemic immune-inflammatory index; TNM, Tumour-Node-Metastasis.	

Table 2
List of *RET* somatic mutations identified in our cohort of 35 medullary thyroid cancer patients after molecular analysis

***RET* somatic mutation**	**Number of patients with *RET* somatic mutation (n = 21)**
c.1852T > C p.(Cys618Arg)	1
c.1858T > C p.(Cys620Arg)	1
c.1859G > C p.(Cys620Ser)	1
c.1888T > G p.(Cys630Gly)	1
c.1888T > C p.(Cys630Arg)	1
c.1900T > C p.(Cys634Arg)	1
c.1901G > A p.(Cys634Tyr)	1
c.2646_2648delinsTTT p.(Ala883Phe)	1
c.2647_2648delinsTT p.(Ala883Phe)	2
c.2753T > C p.(Met918Thr)	10
c.1892_1893delinsCG(;)1894_1914del p.(Asp631Ala)(;)(Glu632_Ile638del)	1
RET, Rearranged during Transfection.	

At diagnosis, lymph-node metastasis were observed in 36 (48.0%) patients, 31 (86.1%) of whom had lateral and central compartment and 5 (13.9%) had only central compartment lymph-node metastasis (Table 1). Regarding distant metastasis, only 8 (10.7%) patients presented them at diagnosis. Biochemical cure after surgery was observed in 28 (37.3%) patients, with a median follow-up of 72 months (36–180).

The associations between the inflammation-based scores NLR, PLR and SII and MTC clinicopathological features are shown in the Tables 3 and 4. An increased NLR was associated with the presence of angioinvasion (2.7 (1.8–4.2) vs 2.1 (1.6–2.7); p = 0.024), extrathyroidal extension (2.8 (2.1–4.5) vs 2.1

(1.7–2.9); p = 0.020), higher grades of fibrosis (group 1: 2.1 (1.8–2.9) vs group 2: 3.7 (2.3–4.9); p = 0.019) and advanced AJCC stages (stages I + II: 1.9 (1.6-3.0) vs stages III + IV: 2.8 (2.0-4.5); p < 0.001). Contrariwise, a low NLR was associated with biochemical cure after surgery (1.8 (1.5–2.2) vs 2.8 (2.1–4.3); p < 0.001) (Table 4). There was also a positive correlation between NLR and both serum calcitonin levels at diagnosis (rho = 0.283; p = 0.027) and number of lymph-node metastasis (rho = 0.409; p = 0.003) (Fig. 1 and Table 3).

Table 3
Correlation between NLR, PLR and SII and MTC clinicopathological features

	NLR	PLR	SII
Serum calcitonin levels at diagnosis	rho = 0.283; **p = 0.027**	rho = 0.127; p = 0.330	rho = 0.114; p = 0.383
Number of lymph-node metastasis	rho = 0.409; **p = 0.003**	rho = 0.251; p = 0.075	rho = 0.346; **p = 0.013**
Largest tumour diameter	rho = 0.194; p = 0.073	rho = 0.213; p = 0.082	rho = 0.411; **p < 0.001**

NLR, neutrophil-to-lymphocyte ratio; PLR, platelet-to-lymphocyte ratio; SII, systemic immune-inflammatory index.

Table 4
Association between NLR, PLR and SII and MTC clinicopathological features

	NLR median (IQR)	PLR median (IQR)	SII median (IQR)
Multifocality	2.3 (1.8–3.6)	141.5 (107.1-209.2)	615.4 (422.1-1236.3)
No	1.7 (1.8–2.9)	143.3 (114.7-178.9)	450.9 (360.1–869.0)
Yes	p = 0.643	p = 0.593	p = 0.245
Angioinvasion	2.1 (1.6–2.7)	133.1 (104.9-162.1)	515.9 (352.6–883.0)
No	2.7 (1.8–4.2)	152.4 (122.5-209.2)	678.6 (422.1-1236.3)
Yes	**p = 0.024**	p = 0.090	p = 0.161
Extrathyroidal extension	2.1 (1.7–2.9)	134.2 (98.9-172.8)	540.8 (366.8-909.1)
No	2.8 (2.1–4.5)	156.4 (125.5-210.7)	689.3 (491.6-1267.7)
Yes	**p = 0.020**	**p = 0.040**	p = 0.161
Primary tumour (T)	2.1 (1.6–2.7)	122.6 (91.1-154.9)	511.0 (373.3-914.7)
Groups T1 + T2	2.5 (1.8–3.7)	152.6 (123.4-209.4)	650.3 (406.9-1029.9)
Groups T3 + T4	p = 0.100	**p = 0.016**	p = 0.486
AJCC staging	1.9 (1.6-3-0)	131.9 (99.4-153.2)	477.7 (323.4–798.0)
Stages I + II	2.8 (2.0-4.5)	155.9 (121.1-210.7)	689.3 (496.3-1240.9)
Stages III + IV	**p < 0.001**	**p = 0.032**	**p = 0.012**
Fibrosis	2.1 (1.8–2.9)	141.5 (109.4–184.0)	566.4 (379.6-919.8)
Group 1 (absent + mild)	3.7 (2.3–4.9)	154.2 (125.2-213.2)	920.1 (530.4-1319.1)
Group 2 (moderate + severe)	**p = 0.019**	p = 0.627	p = 0.145

AJCC, American Joint Committee on Cancer; IQR, interquartile range; MTC, medullary thyroid cancer; NLR, neutrophil-to-lymphocyte ratio; PLR, platelet-to-lymphocyte ratio; *RET*, Rearranged during Transfection; SII, systemic immune-inflammatory index.

	NLR median (IQR)	PLR median (IQR)	SII median (IQR)
Somatic RET **918 or 883 codon mutation**	2.5 (1.9–4.5)	144.8 (119.2-182.7)	724.8 (518.2-1266.8)
No	3.6 (2.1–4.4)	149.7 (104.9-214.3)	714.1 (389.3-1280.5)
Yes	p = 0.432	p = 0.864	p = 0.785
Distant metastases	2.2 (1.7-3.0)	136.7 (107.8–184.0)	566.4 (364.8-1605.7)
No	3.2 (1.8–6.6)	169.7 (142.8-218.6)	817.4 (535.5-2102.7)
Yes	p = 0.159	p = 0.127	p = 0.099
Biochemical cure after surgery	2.8 (2.1–4.3)	151.1 (113.6-207.3)	690.0 (419.1-1155.5)
No	1.8 (1.5–2.2)	129.1 (102.2–153.0)	481.8 (317.9-777.6)
Yes	**p < 0.001**	p = 0.095	**p = 0.019**

AJCC, American Joint Committee on Cancer; IQR, interquartile range; MTC, medullary thyroid cancer; NLR, neutrophil-to-lymphocyte ratio; PLR, platelet-to-lymphocyte ratio; *RET*, Rearranged during Transfection; SII, systemic immune-inflammatory index.

MTC patients with extrathyroidal extension had higher median PLR than those without (156.4 (125.5-210.7) vs 134.2 (98.9-172.8); p = 0.040), and the median PLR significantly differed between the TNM subgroups and MTC stages (Table 4).

Regarding SII, there was a significant association between higher SII and advanced AJCC stages (stage I + II: 477.7 (323.4–798.0) vs stage III + IV: 689.3 (496.3-1240.9); p = 0.012) and lower SII and biochemical cure after surgery (481.8 (317.9-777.6) vs 690.0 (419.1-1155.5); p = 0.019) (Tables 3 and 4). A positive correlation was found between SII and tumour diameter (rho = 0.411; p < 0.001) and between SII and number of lymph-node metastasis (rho = 0.346; p = 0.013) (Fig. 2 and Table 3).

On the univariate analysis, only NLR was a predictor of lymph-node metastasis (OR = 2.69, 95% CI: 1.50–5.84; p = 0.004). However, in the multivariate model, when adjusted for the other clinicopathological variables (variables included in Table 4), NLR no longer predicted lymph-node metastasis at diagnosis (OR = 1.14, CI 95%: 0.65–2.01; p = 0.649) (Table 5). None of the three serum inflammation-based scores had a statistical significant association with the presence of distant metastasis at diagnosis (data not shown).

Table 5
Univariate and multivariate analysis assessing the roles of NLR, PLR and SII in predicting lymph-node metastasis

	Univariate analysis	Multivariate analysis*
NLR	OR = 2.69, 95% CI: 1.5 – 5.84; **p = 0.004**	OR = 1.14; CI 95% 0.65–2.01; p = 0.649
PLR	OR = 1.00 95% CI: 0.99–1.01; p = 0.204	OR = 1.00; CI 95% 0.99–1.01; p = 0.628
SII	OR = 1.00 95% CI: 1.000-1.001; p = 0.630	OR = 1.00, 95% CI: 0.99 – 1.00; p = 0.501

CI, confidence interval; NLR, neutrophil-to-lymphocyte ratio; OR, odds ratio; PLR platelet-to-lymphocyte ratio; SII, systemic immune-inflammatory index. *The variables included in the multivariate model were those considered in the Table 4.

Discussion

In our cohort of MTC patients, we evaluated the association of clinicopathological characteristics with the serum inflammation-based scores NLR, PLR and SII. A number of studies have been carried out to investigate the role of NLR and PLR on thyroid cancer [13, 14, 30], however there are only three studies focused on MTC [17–19] (summarised in the Table 6); in none of these the usefulness of SII was assessed.

Table 6
Summary of the main findings from published studies investigating the role of serum inflammation-based scores in patients with medullary thyroid cancer

Study	Number of patients	Preoperative NLR or PLR as predicting lymph-node metastasis?	Association of NLR or PLR with other prognostic factors?
Jiang *et al.* 2016	70	- NLR was not an independent predictive factor for lymph-node involvement - PLR > 142.1 (OR = 3.452, 95%CI 1.0-11.8) was an independent predictor for lateral lymph-node metastasis by univariate and multivariate analysis	- PLR > 102.5 associated with larger tumours (p = 0.031), higher number of lymph-node metastasis (p = 0.019) and tended to recur more often (p = 0.088) - NLR > 1.9 was associated with multifocality (p = 0.001) and bilaterality (p = 0.001)
Jiang *et al.* 2017	78	- PLR was predictive of lymph-node metastasis (AUC of 0.644; p = 0.022) by univariate analysis	- PLR was predictive of capsule invasion (AUC of 0.666; p = 0.007) and advanced tumour stages (AUC of 0.657; p = 0.011) - PLR was predictive of recurrence on Kaplan-Meier and Cox regression analysis, with an AUC of 0.703 (95%CI 0.589–0.801; p = 0.002) - NLR showed no significant associations with other prognostic factors
Xu *et al.* 2018	61	- NLR was associated with lymph-node and distant metastasis (OR = 5.918, 95%CI 1.147–30.541; p = 0.034) by univariate and multivariate analysis - Best predictive cut-off NLR value estimated at 1.784 (AUC of 0.717, sensitivity 68.3%, specificity 80%) - PLR was not evaluated in the study	- Not evaluated in the study

AUC, area under the curve; CI, confidence interval; MTC, medullary thyroid carcinoma, NLR, neutrophil-to-lymphocyte ratio, OR, odds ratio; PLR, platelet-to-lymphocyte ratio.

In two of these studies, PLR was found as an independent predictor of outcomes among MTC patients, with higher preoperative PLR correlating with presence of lymph-node metastasis, higher postoperative recurrence rates and lower disease-free survival [17, 18]. The optimal PLR cut-off values in predicting recurrence were estimated at 129.8 [17] or 128.9 [18], values above which recurrence was more likely; PLR > 105.3 was also found as independent predictor for lymph-node metastasis [17]. On the other hand, NLR did not associate with recurrence or clinical outcomes in the setting of MTC [18]. Such findings suggest that PLR may be superior than other serum inflammation-based scores in MTC patients, and may well be an useful tool in identifying high-risk patients and perhaps in guiding clinicians towards a more intensive follow-up and/or treatment approach [17, 18]. However, a more recent study from Xu and

colleagues described an association between preoperative NLR and lymph-node and distant metastasis, with a NLR > 1.784 predicting the occurrence of metastasis [19]. The potential usefulness of another inflammatory score in MTC, the Prognostic Nutrition Index, has also been reported [18].

Taking in consideration these conflicting observations regarding NLR and PLR from previous studies [17–19] and the lack of data about SII in patients with MTC, we further investigated the usefulness of NLR and PLR, as well as SII, in predicting more aggressive disease and clinical outcomes in this setting of patients. We observed an association between high NLR and aggressive histological characteristics of the tumour such as angioinvasion, extrathyroidal extension and number of lymph-node metastasis, features with recognised prognostic value in MTC [31]. PLR was associated only with extrathyroidal extension and the SII correlated positively with the tumour size and number of lymph-node metastasis. Ceylan *et al.* also studied the correlation of NLR and PLR with clinicopathological features in 201 PTC patients and found that higher NLR was correlated with tumour size and extra-thyroidal extension, whereas PLR was not associated with any of the clinicopathological characteristics studied [32]. Concerning the TNM classification, higher PLR, but not NLR, was significantly associated with advanced T stages, which may potentially reflect a poorer overall survival and cancer-specific survival [33]. NLR, PLR and SII were associated with advanced AJCC staging, but neither of them were found as independent predictive markers for lymph-node or distant metastasis. A high NLR was associated with lymph-node metastasis at diagnosis, but significance was lost on the multivariate analysis model, probably because histological findings, such as angioinvasion, may be more determinant for metastasis (whether regional or distant) than the inflammation itself. For this reason, we did not perform Receiver Operating Characteristic curves analysis for lymph-node metastasis. Also, none of the inflammatory markers showed a significant association with the presence of distant metastasis at diagnosis.

A low preoperatively NLR and low SII were associated with biochemical cure after surgery, which emphasizes the value of these markers as potential tools to predict clinical outcomes in MTC patients who undergo surgery. We also found a significant positive correlation between NLR and serum levels of calcitonin at diagnosis.

The median NLR was higher in the group of patients with moderate and severe fibrosis, a factor that has been considered correlated with poor prognosis in several cancers [34], including in PTC [35]. Somatic *RET* mutations involving the codons 918 or 883, which may confer increased aggressiveness [28], were not associated with NLR, PLR or SII. The mutational status was assessed in a small number of patients which can explain, at least in part, this lack of association. Several lines of evidence support a role for inflammation in the development and progression of thyroid malignancies: i) alteration of the expression of immune-related genes in PTC [36]; ii) activation of oncogenes involved in PTC (such as *RET/PTC*, *RAS*, *BRAF*) upregulates a pro-inflammatory profile in normal thyrocytes and results in malignant behavior [37, 38]; iii) thyroid tumour cells are an active source of cytokines and chemokines that may modulate the local tumour microenvironment as well as may have systemic pro-inflammatory implications [39].

There are some limitations to this study that are mainly inherent to its retrospective nature. Other limitations of our study include: i) single-center study, mainly including a population of Portuguese patients, hence limiting the generalisation of our findings to other populations or ethnicities; ii) we also cannot exclude that some patients would have unknown/ unreported concomitant diseases or medications capable of influencing haematopoiesis or systemic inflammation; iii) *RET* genetic analysis was only available for approximately half of the patients; iv) our study population is relatively small which limits the assessment of the studied serum inflammation-based scores as it results on small subgroups of patients and provides insufficient statistical power to detect significant differences, particularly after applying a multivariate analytical model, especially if taking in consideration that such haematological parameters have substantial inter- and intra-individual variability. Nevertheless, this is the second largest study evaluating the role of NLR and PLR in patients with MTC, as well as the first study assessing the usefulness of SII in this setting of patients.

In conclusion, NLR, PLR and SII may be useful biomarkers to indicate increased clinico-pathological aggressiveness in the MTC setting, but their usefulness in independently predicting lymph-node or distant metastasis appears to be limited. Further studies involving larger cohorts of patients, ideally multi-centric and involving multi-ethnical populations, are needed to confirm some of the observations from this exploratory study.

Declarations

Statement of Ethics

The research was carried out according to the principles of the Declaration of Helsinki. The local Ethics Review Committee of Instituto Português de Oncologia de Lisboa approved the study protocol.

Declaration of interest

The authors have no conflicts of interest to declare.

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Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Author contribution statement

A.A.F, J.S.-P., P.M. and V.L.: conceptualization. A.A.F., J.S.-P., M.M.M.: data processing. A.A.F, J.S.-P and S.E.: statistics. A.A.F.: writing of the original draft. J.S.-P., P.M., M.M.M., V.L.: writing, review, and editing.

V.L.: supervision.

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Figures

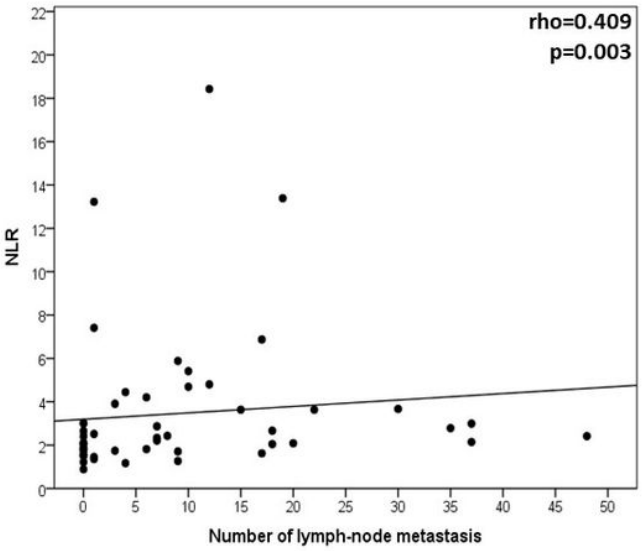

Figure 1

Correlation between the number of lymph-node metastasis and neutrophil-to-lymphocyte ratio (NLR)

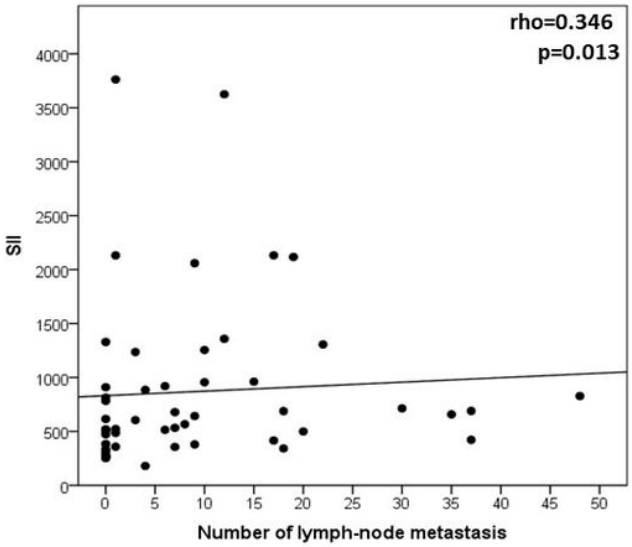

Figure 2

Correlation between the number of lymph-node metastasis and systemic immune-inflammatory index (SII)