Elevated serum concentrations of IGF-1 and IGF-1R in patients with thyroid cancers

Hanna Lawnicka[a#], Ewelina Motylewska[a#], Magdalena Borkowska[b], Krzysztof Kuzdak[b], Agnieszka Siejka[c], Jacek Swietoslawski[d], Henryk Stepien[a] , Tomasz Stepien[b]

Background. The rising incidence of thyroid cancer observed in the last few decades requires an improvement in diagnostic tools and management techniques for patients with thyroid nodules.

Aims. The aim of this study was to assess serum concentrations of IGF-1 and IGF-1R in patients diagnosed with thyroid cancers.

Methods. 36 patients diagnosed with papillary thyroid cancer (PTC), 11 subjects with follicular thyroid cancer (FTC), 9 patients with anaplastic thyroid cancer (ATC) and 19 subjects with multinodular nontoxic goiter (MNG) were enrolled to the study. The control group (CG) consisted of 20 healthy volunteers. Blood samples were collected one day before surgery. Serum IGF-1 and IGF-1R concentrations were measured using specific ELISA methods.

Results. Significantly higher concentrations of IGF-1 were found in patients with PTC as compared with controls but not that obtained from subjects diagnosed with MNG. The concentration of IGF-1R was significantly elevated in subjects with PTC and ATC as compared with healthy volunteers. Similarly, patients diagnosed with PTC or ATC presented significantly higher serum concentration of IGF-1R in comparison to the MNG group.

Conclusions. Our results show that the IGF-1 – IGF-1R axis plays a significant role in the development of PTC and ATC and imply that serum concentrations of both cytokines may be considered as additional markers for the differentiation of malignancies during the preoperative diagnosis of patients with thyroid gland tumors. These results indicate that IGF-1R serum concentrations allow us to differentiate between MNG and PTC or ATC. Moreover IGF-1R serum values appear to be better predictor of PTC and ATC than IGF-1 concentrations.

[a]Department of Immunoendocrinology, Chair of Endocrinology, Medical University of Lodz, Pomorska 251, 92-213, Lodz, Poland
[b]Clinic of Endocrinological and General Surgery, Chair of Endocrinology, Medical University of Lodz, Pabianicka 62, 93-513 Lodz, Poland
[c]Clinic of Endocrinology, Chair of Endocrinology, Medical University of Lodz, Pomorska 251, 92-213, Lodz, Poland
[d]Department of Neuroendocrinology, Interdepartmental Chair of Laboratory and Molecular Diagnostics, Medical University of Lodz, Pomorska 251, 92-213, Lodz, Poland
[#]These authors contributed equally and share first authorship
Corresponding author: Hanna Lawnicka, e-mail: hanna.lawnicka@umed.lodz.pl

INTRODUCTION

Thyroid cancer (TC) is the most common endocrine malignancy with a rapidly rising incidence rate that has been observed continuously all over the world over the last few decades[1,2]. Recently ~1.5% of new cancer cases in Europe have been diagnosed as TC (ref.[3]). Currently, according to the National Cancer Institute Surveillance, Epidemiology, and End Results Program, TC accounts for 3.4% of all cancers diagnosed yearly[4] and, in 2030, it is estimated to be the fourth leading cancer diagnosis in United States[5]. Much of this rise appears to be the result of progress in diagnostics, e.g. the increased use of thyroid ultrasound, which can detect small thyroid nodules that might not otherwise have been found in the past (www.cancer.org). Thus an improvement in reliable diagnostics and management techniques for patients diagnosed with certain types of TC is required in order to avoid overdiagnosis and overtreatment in low-risk patients and underdiagnosis and undertreatment in high-risk patients[6].

Histopathological classification differentiates TC derived from follicular thyroid cells as the most frequent papillary thyroid cancer (PTC), followed by follicular thyroid cancer (FTC), poorly differentiated thyroid cancer and rare anaplastic thyroid cancer (ATC). PTC and FTC subtypes are together classified as differentiated thyroid cancers (DTC) with good prognosis, while ATC is a lethal malignancy[7]. Thus, histopathological diagnosis of TC subtypes together with TNM staging and molecular markers (including *BRAF* and *RAS* point mutations; *RET/PTC* and *PAX8/PPARγ* genes rearrangements; MAPK; PI3K; p53; Wnt–beta catenin; HIF1α and NF-kappaB signaling pathways; and microRNA profiles) are critical for patient management and prognosis[7,8].

The recent search for preoperative serum biomarkers for thyroid carcinoma has resulted in the establishment

of a preoperative neutrophil-to-lymphocyte ratio that closely relates to the stage of PTC (ref.[9]), preoperative thyroglobulin antibodies as potential marker for PTC in patients with indeterminate thyroid nodules[10], serum SERPINE2/Protease Nexin-1 and Secretory Leukocyte Protease Inhibitor (SLPI) concentrations as preoperative PTC biomarkers[11], circulating miRNAs as a non-invasive biomarker for the diagnosis of PTC (ref.[12]) and preoperative serum thyroglobulin (Tg) and changes in serum Tg during thyroid stimulating hormone (TSH) suppression as independent predictors of FTC in thyroid nodules with a cytological diagnosis of follicular lesion[13]. Nevertheless, continuous search for specific growth factors involved in the pathogenesis and progression of differentiated and anaplastic thyroid cancers still remains of utmost importance for improving preoperative diagnostics and for designing new therapeutic approaches to dealing with TC.

The insulin-like growth factors (IGFs) family, comprised of insulin and two natural growth hormones, IGF-1 and IGF-2, is crucially involved in cellular growth, survival and development. IGFs directly regulate cellular functions by interacting with specific cell surface receptors, mainly type I (IGF-1R). IGF-1R belongs to the family of tyrosine kinase growth factor receptors which are involved in cell transformation and protection of transformed phenotypes[14]. It has been proved that the IGF1 – IGF-1R axis plays an essential role in cancer pathogenesis by suppressing apoptosis and promoting cell cycle progression, angiogenesis, and metastatic activities in various cancers[14]. This is achieved by direct activation of the PI3K/Akt signaling pathway initiated by IGF-1 – IGF-1R, leading to stimulation of mitogenesis, cell cycle progression, and protection against apoptotic stresses. The IGF-1 axis activates some other signaling pathways involved in cells growth such as MAPK and mTOR. Alterations of the IGF-IGFR system in lung cancer, gastrointestinal neoplasms (colorectal, gastric, pancreatic, and hepatocellular cancers), female and male reproductive system neoplasms (breast, ovarian, endometrial, cervical, and prostate cancers), skin malignancies (basal cell carcinoma and melanoma malignum), head and neck cancers, bone and soft tissues cancers, hematologic malignancies, central nervous system neoplasms, urinary tract neoplasms have been described (for review see Kasprzak et al.[15]). Thus, the components of the IGF system are first cytokines considered as diagnostic serous biomarkers and prognostic factors for various cancers, as well as targets for modern anti-tumor therapies[14,15].

The proved presence of IGF-1 in normal and thyroid cancer tissues with significantly higher values in malignant tissues than in normal tissues as well as the overexpression of IGF-1 receptors found in both thyroid cancer cell lines and specimens[16] resulted in the conception that the IGF system may be involved in thyroid cancer growth[17].

Regardless of the significant advances in TC preoperative diagnostics, including certain molecular and serum markers characteristic for DTC and some progress in understanding pathogenesis of ATC, there is still a great need for a better understanding of TC pathogenesis and improvement in preoperative diagnostics. Moreover, although the malignancy rate of incidental thyroid nodules in healthy populations is quite low (~5%), it depends on patients' selection and detection technique: in lesions found in FDG-PET the malignancy rate reaches 33-35% (ref.[18]), and in patients with non-thyroidal malignancies managed by USG or PET/CT, PTC proportion in identified nodules was 3.4% or 8.9% respectively[19]. Thus, the improvement in diagnostics of thyroid nodules and identification of those in need of surgical treatment is of utmost importance. Therefore, the aim of the present study was to identify whether IGF-1 and IGF-1R serum concentrations might be considered additional prognostic markers of malignancy in patients diagnosed with thyroid gland nodules.

MATERIAL AND METHODS

Study design and patient characteristics

Seventy-five euthyroid patients aged from 18 to 75 years (53.72 ± 12.62) (mean ± standard error of the mean) that underwent surgery in the Clinic of Endocrinological and General Surgery, Copernicus Memorial Hospital, Lodz, Poland were enrolled in the study. The examined group was composed of 36 subjects diagnosed with PTC (PTC group), 11 with FTC, 9 with ATC and 19 with multinodular nontoxic goiter (MNG).

Selected cases were diagnosed by fine-needle biopsy and confirmed by postoperative histopathologic examination. Other thyroid gland pathologies were excluded on the basis of familial and clinical history, clinical examination, ultrasonography and thyroid function tests (aTPO, aTG, TSH, fT3, fT4 and calcitonin serum concentrations). The control group (CG) included 20 healthy, age-matched volunteers with no history of any thyroid disease confirmed by clinical and hormonal tests, thyroid

Table 1. Demographic and clinical characterization of the studied groups.

Group	Number of patients (Gender F/M)	Age (years) Mean ± SEM
Control	20 (10/10)	55.15 ± 9.80
MNG	19 (10/9)	54.71 ± 10.23
PTC	36 (20/16)	52.73 ± 11.20
FTC	11 (6/5)	52.42 ± 9.11
ATC	9 (4/5)	57.12 ± 12.36

PTC – papillary thyroid cancer, FTC – follicular thyroid cancer, ATC – anaplastic thyroid cancer, MNG – multinodular nontoxic goiter, C – healthy controls

Table 2. Histopathological diagnosis and clinical staging of thyroid cancer patients recommended by the 2010 TNM system edition by the Union for International Cancer Control.

	STAGE		
I	II	III	IVA
PTC (n=36)			
12	12	10	2
FTC (n=11)			
5	4	1	1
ATC (n=9)			
0	0	4	5

PTC – papillary thyroid cancer, FTC – follicular thyroid cancer, ATC – anaplastic thyroid cancer

Table 3. Mean serum concentration ± SD of IGF-1 (ng/mL) and IGF-1R (ng/mL) in patients diagnosed with papillary thyroid cancer, follicular thyroid cancer, anaplastic thyroid cancer, multinodular goiter and healthy volunteers.

Mean±SD	Control	MNG	PTC	FTC	ATC
IGF-1 [ng/mL]	216.0±123.4	242.1±129.0	300.6±82.2	265.3±104.8	190.3±53.7
IGF-1R [ng/mL]	16.7±13.4	18.4±10.9	39.7±22.8	29.3±10.1	36.3±24.6

C – healthy controls, MNG – multinodular nontoxic goiter, PTC – papillary thyroid cancer, FTC – follicular thyroid cancer, ATC – anaplastic thyroid cancer

ultrasound scan, and the presence of thyroid autoantibodies. Demographic and clinical characteristics of the examined groups and healthy controls are presented in Table 1. All patients diagnosed with thyroid cancer (PTC, FTC or ATC) were treated with total thyroidectomy, and therapeutic neck dissection was performed with standard indications. The MNG patients group was treated with bilateral subtotal thyroidectomy. Histopathological diagnosis and clinical staging of patients included in the study is presented in Table 2. The multinodular nontoxic goiter group was comprised of 19 cases with benign postoperative histopathological diagnoses, including 8 follicular adenomas and 11 adenomatous nodules.

The project was approved by the Bioethics Committee of the Medical University of Lodz.

Measurements of IGF-1 and IGF-1R serum levels by ELISA

Blood samples were collected from the antecubital vein between 7:00 and 8:00 am after an overnight fast, one day before surgery. Blood samples were processed within one hour after collection, and the serum was aliquoted and stored at -80 °C until analysis. Determination of IGF-1 (Mediagnost GmbH) and IGF-1R (Shanghai Sunred Biological Technology Ltd.) concentrations were evaluated using enzyme-linked immunosorbent assay (ELISA) kits, following the manufacturer's instructions. All measurements were taken in duplicate and averaged.

Statistical analysis

Results were presented as mean ± standard error of the mean (SEM) in the figures and text and as mean ± standard deviation (SD) in the table. The Shapiro-Wilk test was applied to analyze the data distribution. ANOVA fol-

lowed by Fisher's protected Least Significant Difference was used to calculate differences between investigated groups; $P<0.05$ was considered significant. All statistical analyses were performed using StatSoft statistical software v.10.0. (Statistica PL).

RESULTS

The quantitative determination of the IGF-1 and IGF-1R concentration in the serum and the statistical evaluation of these results are presented in Fig. 1. and 2. and Table 3. The mean serum concentration of IGF-1 in PTC patients (300.6 ± 13.7 ng/mL) was significantly higher than that obtained in controls (216.0 ± 27.6 ng/mL; $P<0.01$) (Fig. 1, Table 3). The mean IGF-1R serum concentration was significantly higher in patients diagnosed with PTC (39.7 ± 3.8 ng/mL) than in healthy volunteers (16.7 ± 3.0 ng/mL; $P<0.01$) or in the MNG group (18.4 ± 2.5 ng/mL; $P<0.05$) (Fig. 2, Table 3). Similarly, the mean IGF-1R serum concentration obtained from patients diagnosed with ATC (36.3 ± 8.2 ng/mL) was significantly higher than that observed in controls (16.7 ± 3.0 ng/mL; $P<0.05$) or in MNG subjects (18.4 ± 2.5 ng/mL; $P<0.05$) (Fig. 2, Table 3).

The mean serum concentration of IGF-1 in FTC (265.3 ± 33.1 ng/mL), ATC (190.3 ± 17.9 ng/mL) and MNG (242.1 ± 29.6 ng/mL) patients did not significantly differ from that of the control group (216.0 ± 27.6 ng/mL). Similarly, the mean serum concentration of IGF-1R in FTC patients (29.3 ± 3.0 ng/mL) did not significantly differ from that of the healthy volunteers (16.7 ± 3.0 ng/mL) or the MNG group (18.4 ± 2.5 ng/mL).
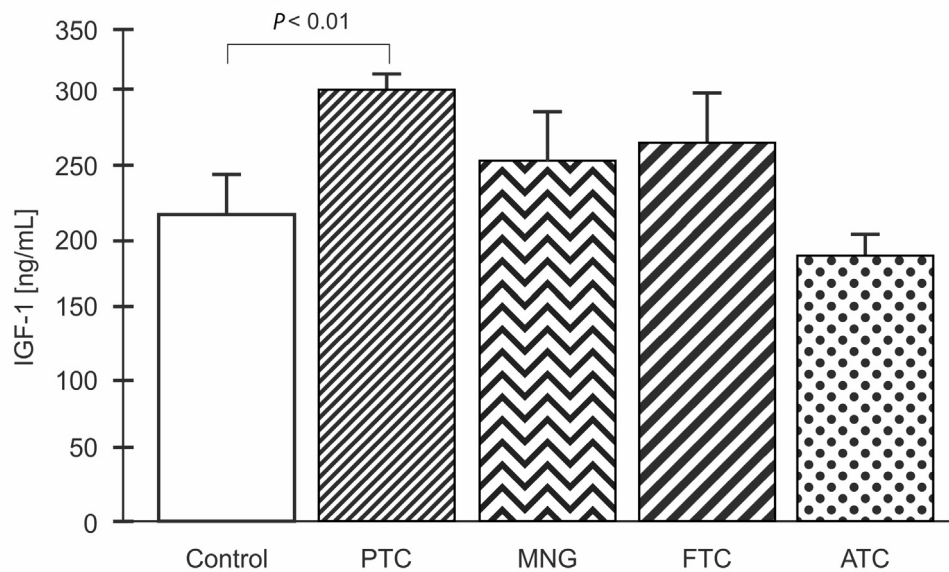
Fig. 1. Mean serum concentration of IGF-1 (ng/mL) in patients diagnosed with papillary thyroid cancer, follicular thyroid cancer, anaplastic thyroid cancer, multinodular goiter and in healthy volunteers. Bars represent mean ± SEM

C – healthy controls, PTC – papillary thyroid cancer, MNG – multinodular nontoxic goiter, FTC – follicular thyroid cancer, ATC – anaplastic thyroid cancer
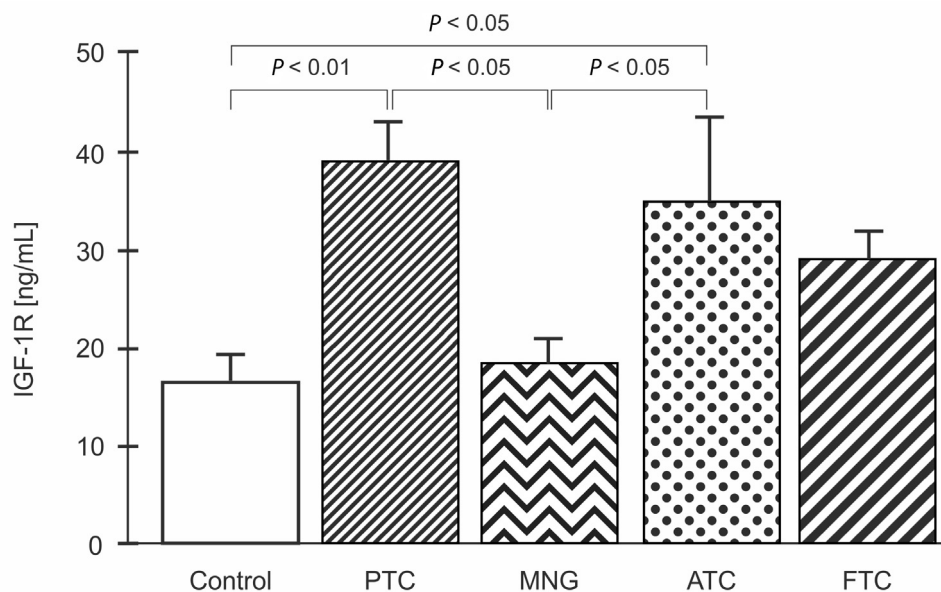

Fig. 2. Mean serum concentration of IGF-1R (ng/mL) in patients diagnosed with papillary thyroid cancer, follicular thyroid cancer, anaplastic thyroid cancer, multinodular goiter and healthy volunteers. Bars represent mean ± SEM

C – healthy controls, PTC – papillary thyroid cancer, MNG – multinodular nontoxic goiter, FTC – follicular thyroid cancer, ATC – anaplastic thyroid cancer

DISCUSSION

Insulin and IGF-1 have been shown to be important growth factors that, together with TSH, interact in the regulation of thyroid cells proliferation and differentiation[20]. The expression of IGF-1 and IGF-1R in thyroid malignancy has been documented[21,22], but the role of the IGF-1-IGF-1R system in TC, especially in DTC, has not been widely explored. IGF-1 and IGF-1R or IGF-1R mRNA were measured in thyroid cancer cell lines and tissue specimens from thyroid epithelial tumours showing overexpression of IGF-1 and IGF-1 receptors in malignant tissues. However, data concerning *in vivo* IGF-1 expression in TC and its relevance as a prognostic factor are limited[20].

IGF-1 and IGF-1R were detected in both PTC and FTC subtypes in children and adolescents. IGF-1 and IGF-1R expression was found more often in PTC than in normal surrounding thyroid tissue and intense IGF-1R staining was connected with more aggressive clinical features[23]. Although the relationship between TC and nodular goiter is still debated, IGF-1 expression in tissues obtained from patients who had undergone total thyroidectomy for nodular thyroidal disease was higher in benign nodules and papillary carcinomas than in extranodular healthy tissues in the same patients and was higher in papillary carcinomas than in benign nodules[24]. Interestingly, IGF-1 levels in tissue were higher in nodules than in healthy tissues in patients with papillary cancer. In our study IGF-1R serum concentration in patients diagnosed with MNG did not significantly differ from values obtained in healthy subjects. In other studies on thyroid nodules, IGF-1 and IGF-1R expression as well as IGF-1 mRNA and IGF-1R mRNA expression were shown to be significantly higher in nodular goiters, follicular adenomas, and PTC than in normal thyroid tissues. PTC tissues showed significantly higher IGF-1 and IGF-1R protein and mRNA expression compared with follicular adenomas and nodular goiters[25]. These findings indicate that IGF-1 plays a role in the development of benign and malignant thyroid nodules and suggest a link between IGF-1 and papillary cancer that may be equally or more intense than that between IGF-1 and benign nodules[24]. Other types of thyroid cancers do not seem to have such a clear correlation with the tissue levels of IGF-1 as that postulated in the case of patients diagnosed with PTC. In fact, Chakravarty et al.[26] reported significantly higher expression of phosphorylated IGF-1R in highly differentiated than in poorly differentiated thyroid cancers. Our results showed higher serum concentrations of IGF-1 in PTC patients than in controls and higher serum IGF-1R concentrations in those diagnosed with PTC, than in healthy volunteers and multinodular nontoxic goiter subjects. Our data is coherent with the findings from samples of healthy thyroid, nodular goiters and PTC tissues and support the role of the IGF-1- IGF-1R pathway dysfunction in pathogenesis of DTC. In fact, increased expression of phosphorylated p70S6 and Akt, which both are kinases phosphorylated and activated by growth factors including IGF-1, found in papillary thyroid cancer tissues[27] further supports the role of the IGF-1 axis in PTC pathogenesis and development. However, in contrast to the mentioned studies on poorly differentiated TC tissues samples, we observed a higher IGF-1R serum concentration in the group of ATC patients than in controls or MNG subjects. It must be stressed that our results concerning higher serum IGF-1R concentrations in anaplastic cancer patients in *in vivo* conditions are supported by the studies of Wang et al.[28] who demonstrated IGF-1R expression on ATC tissue samples and the inhibitory effect of IGF-1 antibody on cell proliferation and angiogenesis as well as its proapoptotic activity in an orthotopic nude mouse model of anaplastic thyroid cancer *in vivo*.

In addition, a positive association between IGF-1 concentrations and risk of differentiated thyroid cancer was documented in the prospective multicenter study with a group of 345 incident cases of DTC who were individually matched to 735 controls by study center, sex and age, date, time, fasting status at blood collection, follow-up duration, and menopausal status, use of exogenous hormones, and phase of menstrual cycle at blood collection for women[29]. This can be confirmed by uncontrolled hyperactivity of the GH-IGF-1 axis in the development of PTC rather than the *BRAFV600E* mutation in acromegalic patients[30] and by the observation that thyroid cancer is the most common cancer associated with acromegaly[31].

In contrast to PTC diagnosed patients, the lack of significant changes in IGF-1 or IGF-1R serum concentrations in FTC subjects observed in our study was in agreement with Schmidt et al.[29].

Our results expand and complement the mentioned studies on DTC and additionally suggest the role of the IGF-1-IGF-1R system in anaplastic thyroid cancer development and progression as well. The latter is particularly interesting since TSH is known to be the major growth factor for differentiated thyroid cancer, while TSH independent cancer growth and metastatic spread become important in cases with loss of differentiation. This means the efficacy of TSH suppression in poorly differentiated or undifferentiated TCs is limited[20]. Our results indicate the potential role of the IGF-1 – IGF-1R system in those cases and might suggest the beneficial blockade of IGF-1R in the management of patients diagnosed with ATC. This can be supported by the aforementioned study of Wang et al.[28] who demonstrated the inhibition of cell proliferation and antiangiogenic and proapoptotic activity of the IGF-1 antibody on ATC animal model *in vivo*. Moreover, as it was shown in thyroid cell cultures, the mitogenic effects of TSH may be insignificant without other growth factors, although they are greatly enhanced by the presence of insulin or IGF-1 (ref.[20,32]).

The studies concerning serum biomarkers of thyroid cancer make evident the fact that development of TC is a complex process that involves multiple metabolic and growth factors pathways' dysregulation. In our previous studies on thyroid cancer, PTC, FTC and ATC patients had significantly lower concentrations of βKlotho as compared with the MNG group and controls[33]. Interestingly, secreted Klotho is a circulating protein with anti-aging and anti-oncogenic properties that modulates the activity of multiple growth factors receptors, including the IGF-1R axis and is downregulated in several neoplasms[34,35]. Together with the results of the present paper, the interplay between circulating βKlotho and IGF-1R axis in the development of PTC and ATC might be considered.

CONCLUSIONS

Our findings support the role of the IGF-1 – IGF-1R axis in the development of papillary cancer and suggest the involvement of overexpression of this pathway in anaplastic cancer as well.

These results indicate that only IGF-1R values are able significantly differentiate between MNG and PTC or ATC (but not FTC) in this study with respect to lim-

ited number of patients. Moreover it seems, that IGF-1R is a better predictor of PTC and ATC than IGF-1. Thus IGF-1R serum concentration can be considered as additional biomarker for improving preoperative diagnosis of patients with thyroid nodules and with special value for papillary and anaplastic thyroid cancers diagnosis.

ABBREVIATIONS

ATC, Anaplastic thyroid cancer; CG, Control group; DTC, Differentiated thyroid cancers; FTC, Follicular thyroid cancer; GH, Growth hormone; IGF-1, Insulin-like growth factor type 1; IGF-1R, Insulin-like growth factor type 1 Receptor; IGF-2, Insulin-like growth factor type 2; IGFs, Insulin-like growth factors family; MAPK, Mitogen-activated protein kinases; MNG, Multinodular nontoxic goiter; mTOR, Mammalian target of rapamycin kinase; PTC, Papillary thyroid cancer; SLPI, Secretory Leukocyte Protease Inhibitor; TC, Thyroid cancer; Tg, Thyroglobulin; TSH, Thyroid stimulating hormone.

REFERENCES

1. Pellegriti G, Frasca F, Regalbuto C, Squatrito S, Vigneri R. Worldwide increasing incidence of thyroid cancer: update on epidemiology and risk factors. J Cancer Epidemiol 2013; 2013:965212. doi: 10.1155/2013/965212

2. Sanabria A, Kowalski LP, Shah JP, Nixon IJ, Angelos P, Williams MD, Rinaldo A, Ferlito A. Growing incidence of thyroid carcinoma in recent years: Factors underlying overdiagnosis. Head Neck 2018;40(4):855-66. doi: 10.1002/hed.25029

3. Ferlay J, Steliarova-Foucher E, Lortet-Tieulent J, Rosso S, Coebergh JWW, Comber H, Forman D, Bray F. Cancer incidence and mortality patterns in Europe: Estimates for 40 countries in 2012. Eur J Cancer 2013;49(6):1374-403. doi: 10.1016/j.ejca.2012.12.027

4. National Cancer Institute Surveillance, Epidemiology, and End Results Program. Available from: https://seer.cancer.gov/statfacts/html/thyro.html (accessed on 25 April 2017).

5. Rahib L, Smith BD, Aizenberg R, Rosenzweig AB, Fleshman JM, Matrisian LM. Projecting cancer incidence and deaths to 2030: the unexpected burden of thyroid, liver, and pancreas cancers in the United States. Cancer Res 2014;74(11):2913-21. doi: 10.1158/0008-5472.CAN-14-0155

6. Rusinek D, Chmielik E, Krajewska J, Jarzab M, Oczko-Wojciechowska M, Czarniecka A, Jarzab B. Current Advances in Thyroid Cancer Management. Are We Ready for the Epidemic Rise of Diagnoses? Int J Mol Sci 2017;18(8):pii: E1817. doi: 10.3390/ijms18081817

7. Omur O, Baran Y. An update on molecular biology of thyroid cancers. Crit Rev Oncol Hematol 2014;90(3):233-52. doi: 10.1016/j.critrevonc.2013.12.007

8. Glikson E, Alon E, Bedrin L, Talmi YP. Prognostic Factors in Differentiated Thyroid Cancer Revisited. Isr Med Assoc J 2017;19(2):114-8.

9. Gong W, Yang S, Yang X, Guo F. Blood preoperative neutrophil-to-lymphocyte ratio is correlated with TNM stage in patients with papillary thyroid cancer. Clinics (Sao Paulo) 2016;71(6):311-4. doi: 10.6061/clinics/2016(06)04

10. Karatzas T, Vasileiadis I., Zapanti E, Charitoudis G, Karakostas E, Boutzios G. Thyroglobulin antibodies as a potential predictive marker of papillary thyroid carcinoma in patients with indeterminate cytology. Am J Surg 2016;212(5):946-52. doi: 10.1016/j.amjsurg.2015.12.030

11. Stępień T, Brożyna M, Kuzdak K, Motylewska E, Komorowski J, Stępień H., Ławnicka H. Elevated Concentrations of SERPINE2/Protease Nexin-1 and Secretory Leukocyte Protease Inhibitor in the Serum of Patients with Papillary Thyroid Cancer. Dis Markers 2017; 2017:4962137. doi: 10.1155/2017/4962137

12. Zhang Y, Xu D, Pan J, Yang Z, Chen M, Han J, Zhang S, Sun L, Qiao H. Dynamic monitoring of circulating microRNAs as a predictive biomarker for the diagnosis and recurrence of papillary thyroid carcinoma. Oncol Lett 2017;13(6):4252-66. doi: 10.3892/ol.2017.6028

13. Kim HJ, Mok JO, Kim CH, Kim YJ, Kim SJ, Park HK, Byun DW, Suh K, Yoo MH. Preoperative serum thyroglobulin and changes in serum thyroglobulin during TSH suppression independently predict follicular thyroid carcinoma in thyroid nodules with a cytological diagnosis of follicular lesion. Endocr Res 2017;42(2):154-62 doi: 10.1080/07435800.2016.1262395

14. Brahmkhatri VP, Prasanna C, Atreya HS. Insulin-like growth factor system in cancer: novel targeted therapies. Biomed Res Int 2015;2015:538019.

15. Kasprzak A, Kwasniewski W, Adamek A, Gozdzicka-Jozefiak A. Insulin-like growth factor (IGF) axis in cancerogenesis. Mutat Res Rev Mutat Res 2017;772:78-104. doi: 10.1016/j.mrrev.2016.08.007

16. Belfiore A, Pandini G, Vella V, Squatrito S, Vigneri R. Insulin/IGF-I hybrid receptors play a major role in IGF-I signaling in thyroid cancer. Biochimie 1999;81(4):403-7.

17. Vella V, Sciacca L, Pandini G, Mineo R, Squatrito S, Vigneri R, Belfiore A. The IGF system in thyroid cancer: new concepts. Mol Pathol 2001; 4(3):121-4.

18. Hoang JK, Langer JE, Middleton WD, Wu CC, Hammers LW, Cronan JJ, Tessler FN, Grant EG, Berland LL. Managing incidental thyroid nodules detected on imaging: White paper of the ACR Incidental Thyroid Findings Committee. J Am Coll Radiol 2015;12:143-50. doi: 10.1016/j.jacr.2014.09.038

19. Cansu GB, Yılmaz N, Toru S, Sarı R, Gökhan Ocak G, Arıcı C, Altunbaş HA, Balcı MK. Evaluation of Incidental Thyroid Nodules in Cancer Patients. J Natl Med Assoc 2017;109(4):299-306. doi: 10.1016/j.jnma.2017.02.011

20. Ciampolillo A, De Tullio C, Perlino E, Maiorano E. The IGF-I axis in thyroid carcinoma. Curr Pharm Des 2007;13(7):729-35.

21. Maiorano E, Ciampolillo A, Viale G, Maisonneuve P, Ambrosi A, Triggiani V, Marra E, Perlino E. Insulin-like growth factor 1 expression in thyroid tumors. Appl Immunohistochem Mol Morphol 2000;8(2):110-9.

22. Ciampolillo A, De Tullio C, Giorgino F. The IGF-I/IGF-I receptor pathway: Implications in the Pathophysiology of Thyroid Cancer. Curr Med Chem 2005;12(24):2881-91.

23. Gydee H, O'Neill JT, Patel A, Bauer AJ, Tuttle RM, Francis GL. Differentiated thyroid carcinomas from children and adolescents express IGF-I and the IGF-I receptor (IGF-I-R). Cancers with the most intense IGF-I-R expression may be more aggressive. Pediatr Res 2004; 55(4):709-15.

24. Baştürk E, Kement M, Yavuzer D, Vural S, Gezen C, Gözü HI, Karadayı A, Oncel M. The role of insulin-like growth factor 1 in the development of benign and malignant thyroid nodules. Balkan Med J 2012; 29(2):133-8. doi: 10.5152/balkanmedj.2011.034

25. Liu YJ, Qiang W, Shi J, Lv SQ, Ji M, Shi BY. Expression and significance of IGF-1 and IGF-1R in thyroid nodules. Endocrine 2013;44(1):158-64. doi: 10.1007/s12020-012-9864-z

26. Chakravarty G, Santillan AA, Galer C, Adams HP, El-Naggar AK, Jasser SA, Mohsin S, Mondal D, Clayman GL, Myers JN. Phosphorylated insulin like growth factor-I receptor expression and its clinico-pathological significance in histologic subtypes of human thyroid cancer. Exp Biol Med (Maywood) 2009;234(4):372-86. doi: 10.3181/0809-RM-284

27. Miyakawa M, Tsushima T, Murakami H, Wakai K, Isozaki O, Takano K. Increased expression of phosphorylated p70S6 kinase and Akt in papillary thyroid cancer tissues. Endocr J 2003;50(1):77-83.

28. Wang Z, Chakravarty G, Kim S, Yazici YD, Younes MN, Jasser SA,

Santillan AA, Bucana CD, El-Naggar AK, Myers JN. Growth-inhibitory effects of human anti-insulin-like growth factor-I receptor antibody (A12) in an orthotopic nude mouse model of anaplastic thyroid carcinoma. Clin Cancer Res 2006;12(15):4755-65.

29. Schmidt JA, Allen NE, Almquist M, Franceschi S, Rinaldi S, Tipper SJ, Tsilidis KK, Weiderpass E, Overvad K, Tjønneland A, Boutron-Ruault MC, Dossus L, Mesrine S, Kaaks R, Lukanova A, Boeing H, Lagiou P, Trichopoulos D, Trichopoulou A, Palli D, Krogh V, Panico S, Tumino R, Zanetti R, Bueno-de-Mesquita HB, Peeters PH, Lund E, Menéndez V, Agudo A, Sánchez MJ, Chirlaque MD, Ardanaz E, Larrañaga N, Hennings J, Sandström M, Khaw KT, Wareham N, Romieu I, Gunter MJ, Riboli E, Key TJ, Travis RC. Insulin-like growth factor-i and risk of differentiated thyroid carcinoma in the European prospective investigation into cancer and nutrition. Cancer Epidemiol Biomarkers Prev 2014;23(6):976-85. doi: 10.1158/1055-9965.EPI-13-1210-T

30. Kim HK, Lee JS, Park MH, Cho JS, Yoon JH, Kim SJ, Kang HC. Tumorigenesis of papillary thyroid cancer is not BRAF-dependent in patients with acromegaly. PLoS One 2014; 9(10):e110241. doi: 10.1371/journal.pone.0110241

31. Gullu BE, Celik O, Gazioglu N, Kadioglu P. Thyroid cancer is the most common cancer associated with acromegaly. Pituitary 2010;13(3):242-8. doi: 10.1007/s11102-010-0224-9

32. Kimura T, Van Keymeulen A, Golstein J, Fusco A, Dumont JE, Roger PP. Regulation of thyroid cell proliferation by TSH and other factors: a critical evaluation of in vitro models. Endocr Rev 2001;22(5):631-56.

33. Motylewska E, Stępień T, Borkowska M, Kuzdak K, Siejka A, Komorowski J, Stępień H, Ławnicka H. Alteration in the serum concentrations of FGF19, FGFR4 and βKlotho in patients with thyroid cancer. Cytokine 2018;105:32-6. doi: 10.1016/j.cyto.2018.02.013

34. Zhou X, Wang X. Klotho. A novel biomarker for cancer. J Cancer Res Clin Oncol 2015;141:961-9. doi: 10.1007/s00432-014-1788-y

35. Xiangxiang Z, Xiaosheng F, Yujie J, Lingyun G, Xinyu L, Ying L, Kang L, Peipei L, Xiao L, Xin W. Klotho. An anti-aging gene, acts as a tumor suppressor and inhibitor of IGF-1R signaling in diffuse large B cell lymphoma. J Hematol Oncol 2017;10(1):37. doi: 10.1186/s13045-017-0391-5