

Molecular-genetic mechanisms in development of degree of function and hyperplasia of thyroid gland of patients with nodular goiter with autoimmune thyroiditis and thyroid adenoma

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ABSTRACT

Introduction. Autoimmune thyroiditis affects in average 2% to 5% of the general population, with young adult females and the elderly being the most vulnerable patients. Hashimoto's thyroiditis causing hypothyroidism is the most prevalent etiology. Although genetics is well known to cause and influence the progression of autoimmune diseases in approximately 79%, other environmental factors are known to be involved in the development of autoimmune thyroid diseases: quantity of ingested iodine, stress, drugs, pregnancy, and changes in sexual hormones.

Autoimmune thyroiditis, as a background disease of nodular goiter, in which hypothyroidism usually develops, has been insufficiently studied.

Material and methods. The BCL-2 (rs17759659), CTLA-4 (rs231775), Fas (rs2234767) genes' polymorphism were studied by Real-Time Polymerase Chain Reaction in 95 patients with NGAIT, 30 patients with thyroid adenoma (TA) and 25 healthy individuals. The thyroid gland (TG) functional activity changes (normal function, subclinical and clinical hypothyroidism) and TG hyperplasia degrees (IB, II and III) were analyzed.

Results. TA and NGAIT are more common in the minor G-allele carriers (GA- and GG-genotypes) of the BCL-2 gene and in homozygous G allele patients (GG-genotype) of the Fas gene by 11.5 and 4.34 times ($p < 0.01$), with no significant interdependence between the CTLA4 gene's genotypes. TG hyperplasia in patients' general cohort as well as in those with NGAIT is associated with the wild A alleles of the CTLA-4 gene (AA- and AG-genotypes): the I and III degree hyperplasia occurred reliably more frequently in the AA genotype carriers, and II degree of the TG enlargement in the AG genotype patients.

Conclusions. Pathology of the thyroid gland has unreliable chances to be inherited depending on the polymorphism of BCL-2 (rs17759659), CTLA-4 (rs231775) and Fas (rs2234767) genes in Bukovina region (Western Ukraine). We did not find any difference between the relative incidences of the genotypes of the analyzed genes in the patients with NGAIT and those with TA or depending on the TG function (euthyroid goiter, subclinical and clinical hypothyroidism).

Keywords: nodular goiter with autoimmune thyroiditis, thyroid adenoma, functional state, hyperplasia, polymorphisms of APO-1 / FAS, CTLA-4 and BCL-2 genes

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Abbreviations

NGAIT – nodular goiter with autoimmune thyroiditis

AIT – autoimmune thyroiditis

TG – thyroid gland, TA - thyroid adenoma

INTRODUCTION

Autoimmune thyroiditis and nodular goiter represent very dynamic yet recent subjects in the field of interdisciplinary approach involving endocrinologists, oncologists, surgeons, geneticists, as well as pathologists (1-8). The clinical evaluation starts from the discovery of a thyroid nodule (4,5).

Nodular goiter is considered as a non-autoimmune thyroid disease, and there have been findings to support this hypothesis (4-6). However, several immunological alterations have been found in these patients, such as HLA-DR antigen expression in thyrocytes, the presence of growth-stimulating immunoglobulins, and an increase in dendritic cells and lymphocytes, which suggest the possibility of autoimmune problems (1,8-10). Although many of these findings may be an epiphenomenon of other primary defects in immunoregulation, the alteration of lymphocyte populations indicates a primary defect (6,7,10,12-18). Variations in several genes have been studied as possible risk factors for Hashimoto thyroiditis. Some of these genes are part of a family called the human leukocyte antigen (HLA) complex (8-10). The HLA complex helps the immune system distinguish the body's own proteins from proteins made by foreign invaders (such as viruses and bacteria). Other genes that have been associated with AIT help regulate the immune system or are involved in normal thyroid function. Most of the genetic variations that have been discovered are thought to have a small impact on a person's overall risk of developing this condition (14-18).

AIT thought to result from a combination of genetic and environmental factors (1-3, 8-18). Some of these factors have been identified, but many remain unknown (19-24).

AIT is an autoimmune disorder, one of a large group of conditions that occur when the immune system attacks the body's own tissues and organs (23). This shortage of thyroid hormones underlies the signs and symptoms of Hashimoto thyroiditis. However, some individuals with thyroid antibodies never develop hypothyroidism or experience any related signs or symptoms (24).

Other, nongenetic factors also play a role in Hashimoto thyroiditis. These factors may trigger the condition in people who are at risk, although

the mechanism is unclear. Potential triggers include changes in sex hormones (particularly in women), viral infections, certain medications, exposure to ionizing radiation, and excess consumption of iodine, a substance involved in thyroid hormone production (2,24).

In Ukraine, the polymorphism of genes associated with apoptosis of thyrocytes, lymphoid cells, or regulation of T cell activation (Bcl-2, Fas (APO-1), CTLA-4) in NGAIT and TA had not been studied previously, while there may be several national and local peculiarities.

MATERIAL AND METHODS

Ninety-five women with NGAIT underwent examination during 2013 to 2016, based on Chernivtsi Regional Hospital, Ukraine. The age of patients ranged from 23 to 72 years. Diagnosis is exposed based on clinical and laboratory examinations (thyroid peroxidase antibodies (ATPO) – 60-250 U/ml thyroglobulin antibody (ATTH) – 60-500 U/ml; thyroid-stimulating hormone (TSH) – 4.10 IU/l), and thyroid sonography and histologic confirmation after surgical operation.

The group of 30 women passed selection based on ultrasound, fine needle aspiration puncture biopsy and thyroid histological conclusion after surgery diagnosed with thyroid adenoma. We have identified this group because this pathology is one of the most common forms of nodular goiter. Control group included 25 healthy donors.

Genetic studies performed in the laboratory of genetics at the "Nicolae Testemițanu" State University of Medicine and Pharmacy Chisinau (Republic of Moldova). DNA was extracted from whole venous blood lymphocytes. Venous blood was stored in test tubes, stabilized with K2-EDTA. Isolation and purification of DNA from the material obtained was performed according to methodological guidance of Thermo Scientific GeneJET Genomic DNA Purification kit (#K0721, Thermo Fisher Scientific).

Quantitative Real-Time PCR (RT-PCR)

Polymerase chain reaction (PCR) was performed in real-time (RT-PCR) using Taq-DNA polymerase and specific primers on QuantStudio 6 equipment, Applied Biosystems (USA), which allowed us to obtain amplicons to determine their number in "real time" and reduce the likelihood of diagnostic error. Analysis of the data was performed using the Quant Studio Real Time Software (Fig. 1-3).

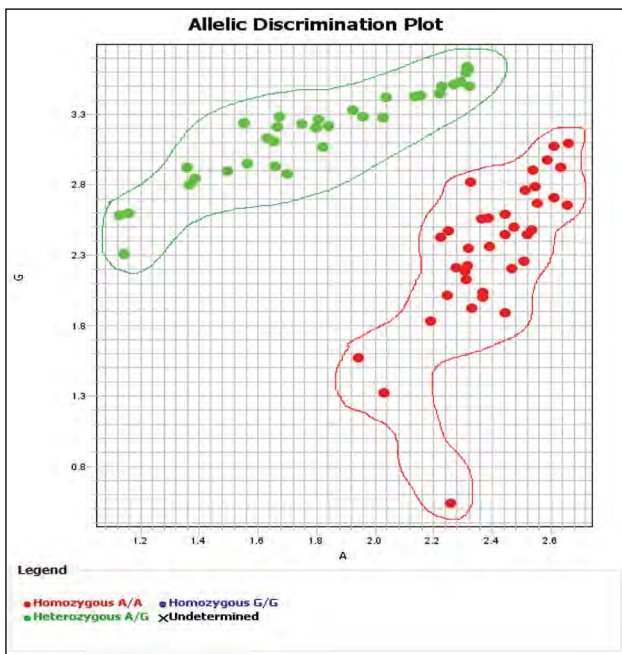


FIGURE 1. CTLA4 gene polymorphism (rs 231775) alleles discrimination

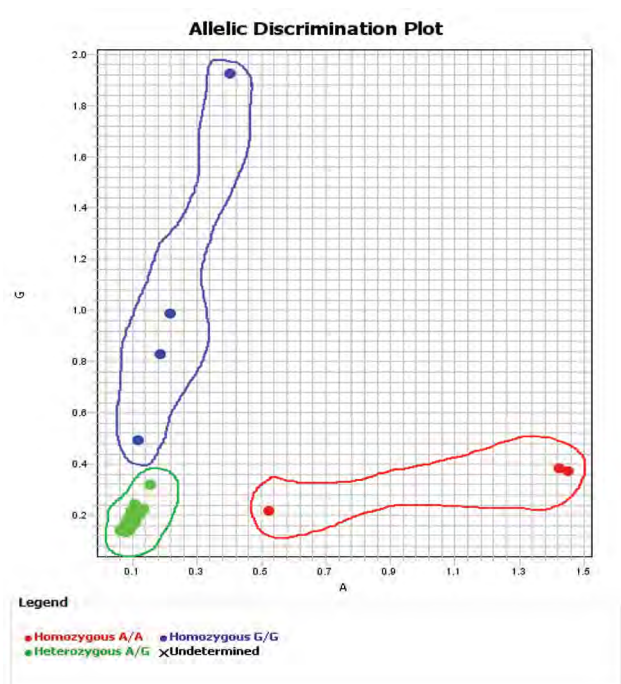


FIGURE 2. BCL-2 gene polymorphism (17759659) alleles discrimination

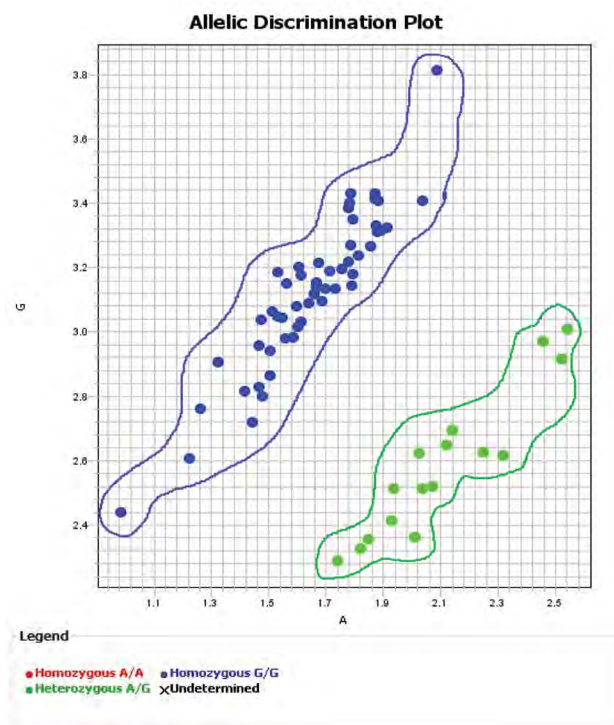


FIGURE 3. FAS gene polymorphism (rs 2234767) alleles discrimination

STATISTICAL ANALYSIS

Statistical analysis was performed using Statistica 7.0 (Stat Soft Inc., USA) software. Nominal data presented in the form of quantitative and

percentages. For the genotypes distribution comparison used Pearson’s criterion (c^2). Analysis of qualitative data (categorical variables), risk of thyroid pathology development was assessed using a binary logistic regression model using the relative risk (RelR), risk ratio (RR) and odds ratio (OR) with 95% confidence interval [95% CI], chi-square test (χ^2) (df=1). The difference was considered reliable at $p < 0.05$.

RESULTS AND DISCUSSIONS

Analysis of the incidence of conditions, considering the thyroid function, (euthyroid goiter, subclinical and clinical hypothyroidism) did not reveal a reliable difference depending on the genotype of the CTLA-4 (rs231775), BCL-2 (rs17759659) and APO-1 / Fas (rs2234767) genes (tab. 1). At the same time, we established the dominance of BCL-2 gene’s GA-genotype over AA- and GG-genotypes by 6.5 and 25.97 times ($p < 0.001$) regardless of the thyroid function. A-allele of the CTLA-4 gene dominated over the G-allele 2.65 and 2.30 times ($p < 0.001$) in patients with NGAIT and TA with euthyroid goiter and subclinical hypothyroidism. GG-genotype of the CTLA-4 gene was observed in patients with clinical hypothyroidism. In all patients regardless of the thyroid function, GG-genotype of the Fas gene predominant the AG-genotype by 3.18-10.5 times ($p < 0.001$), respectively.

TABLE 1. The distribution of the polymorphic variants of the BCL-2 (rs17759659), CTLA-4 (rs231775) and APO-1/Fas (rs2234767) genes in patients with thyroid pathology considering its function

The genes under study, n (%)		Control, n=25 (%)	The thyroid gland function			c ² p
			Euthyroid goiter, n=31	Subclinical hypothyroidism, n=71	Clinical hypothyroidism, n=23	
BCL-2 (A/G), n (%)	AA	3 (12.0)	4 (12.90)	4 (5.63)	2 (8.70)	c ² =1.57 p>0.05
	GA	21 (84.0)	26 (83.87)	65 (91.55)	19 (82.61)	c ² =1.98 p>0.05
	GG	1 (4.0)	1 (3.23)	2 (2.82)	2 (8.70)	c ² =1.63 p>0.05
c ² p		c ² =64,80 p<0,001	c ² =54.10 p<0.001	c ² =162.55 p<0.001	c ² =37.70 p<0.001	–
CTLA-4 (+49G/A), n (%)	AA	15 (60.0)	15 (48.39)	31 (43.66)	13 (56.52)	c ² =1.18 p>0.05
	AG	9 (36.0)	15 (48.39)	37 (52.11)	10 (43.48)	c ² <1.0 p>0.05
	GG	1 (4.0)	1 (3.23)	3 (4.23)	0	p>0.05
c ² p		c ² =36,48 p<0,001	c ² =18.67 p<0.001	c ² =41.75 p<0.001	c ² <1.0 p>0.05	–
Fas (-1377G/A), n (%)	GA	6 (24.0)	4 (12.90)	17 (23.94)	2 (8.70)	c ² =3.52 p>0.05
	GG	19 (76.0)	27 (87.10)	54 (76.06)	21 (91.30)	
c ² p		c ² =72,20 p<0,001	c ² =34.13 p<0.001	c ² =38.56 p<0.001	c ² =31.39 p<0.001	–

TABLE 2. The distribution of polymorphic variants of the BCL-2 (rs17759659), CTLA-4 (rs231775) and APO-1 / Fas (rs2234767) genes in patients with thyroid adenoma, considering its features

The genes under study, n (%)		Control, n=25 (%)	The thyroid gland function, n (%)			c ² p
			Euthyroid goiter n=10	Subclinical hypothyroidism, n=13	Clinical hypothyroidism, n=7	
BCL-2 (A/G), n (%)	AA	3 (12.0)	0	1 (7.69)	0	–
	GA	21 (84.0)	10 (100.0)	12 (92.31)	7 (100.0)	p>0.05
	GG	1 (4.0)	0	0	0	0
c ² p		c ² =64,80 p<0,001	–	c ² =18.62 p<0.001	–	–
CTLA-4 (+49G/A), n (%)	AA	15 (60.0)	5 (50.0)	3 (23.08)	4 (57.14)	c ² =2.82 p>0.05
	AG	9 (36.0)	5 (50.0)	10 (76.92)	3 (42.86)	
	GG	1 (4.0)	0	0	0	–
c ² p		c ² =36,48 p<0,001	–	c ² =7.54 p=0.006	c ² <1.0 p>0.05	–
Fas (-1377G/A), n (%)	GA	6 (24.0)	0	4 (30.77)	1 (14.29)	c ² =3.89 p>0.05
	GG	19 (76.0)	10 (100.0)	9 (69.23)	6 (85.71)	
c ² p		c ² =72,20 p<0,001	–	c ² =3.85 p=0.049	p=0.015	–

In the patients with TA (table 2) and NGAIT (table 3), the distribution, taking into account the thyroid function, showed no statistically significant difference in the incidence of polymorphic variants of the genes analyzed.

I and III degree thyroid hyperplasia occurred much more frequently in individuals with AA-genotype CTLA-4 gene by 30.13% and 26.35% (c²=9.26; p=0.01), whereas the II degree of the thyroid gland

enlargement was more frequently recorded in the patients with AG-genotype by 33.52% and 34.04% (c²=12.34; p=0.002) respectively (tab. 4).

We have not found the reliable dependence of the thyroid hyperplasia degree on the polymorphic variants of the analyzed genes in the patients with TA (table 5).

In NGAIT patients, I and III degree hyperplasia was more often found in those having AA-geno-

type of CTLA-4 gene than AG- and GG-genotype by 28.21% and 56.41% ($c^2=27.92$; $p<0.001$) and 15.39% and 46.16% ($c^2=12.92$; $p<0.001$), respectively, as well as the II degree of the thyroid gland enlargement – by 31,54% and 23,85% ($c^2=7,02$; $p=0,03$) (tab. 6). On the other hand, the II degree hyperplasia was more frequently diagnosed in the individuals with AG-genotype, than in those with AA – by 40.0% ($c^2=9.60$; $p=0.002$), as well as more frequently than I and III degree thyroid hyperplasia by 36.67% and 31.54% ($c^2=10.06$; $p=0.007$).

In patients with euthyroid goiter I degree of thyroid hyperplasia was more common: in those with

wild A-allele of BCL-2 gene by 11,49-52,45% ($r\leq 0,029-0,001$), A-allele of CTLA-4 gene – by 25.86-35.35% ($c^2=10.14-11.58$; $p=0.003-0.006$), and favorable GG-genotype of the Fas gene by 46.43% and 60.03% ($c^2=25.39$; $p<0.001$) (tab. 7).

Patients with subclinical hypothyroidism were more often diagnosed with the II degree hyperplasia of the thyroid gland: in the carriers of intermediate genotype (AG) of BCL-2 gene and CTLA-4 gene, by 25.53% and 37.78% ($r\leq 0.001$) (tab. 7-8) and in the owners of favorable GG-genotype of Fas gene by 25.53% and 26.51% ($c^2=11.33$; $p=0.003$) (tab. 8).

TABLE 3. The distribution of polymorphic variants of the BCL-2 (rs17759659), CTLA-4 (rs231775) and APO-1 / Fas (rs2234767) genes in patients with nodular forms of goiter against the background of autoimmune thyroiditis, considering the thyroid function

The genes under study, n (%)		Control, n=25 (%)	The thyroid gland function, n (%)			c ² p
			Euthyroid goiter, n=21	Subclinical hypothyroidism, n=58	Clinical hypothyroidism, n=16	
BCL-2 (A/G), n (%)	AA	3 (12.0)	4 (19.05)	3 (5.17)	2 (12.50)	c ² =3.67 p>0.05
	GA	21 (84.0)	16 (76.19)	53 (91.38)	12 (75.0)	c ² =4.44 p>0.05
	GG	1 (4.0)	1 (4.76)	2 (3.45)	2 (12.50)	c ² =2.11 p>0.05
c ² p		c ² =64.80 p<0.001	c ² =27.0 p<0.001	c ² =131.9 p<0.001	c ² =18.75 p<0.001	–
CTLA-4 (+49G/A), n (%)	AA	15 (60.0)	10 (47.62)	28 (48.28)	9 (56.25)	c ² <1.0 p>0.05
	AG	9 (36.0)	10 (47.62)	27 (46.55)	7 (43.75)	c ² <1.0 p>0.05
	GG	1 (4.0)	1 (4.76)	3 (5.17)	0	c ² <1.0 p>0.05
c ² p		c ² =36.48 p<0.001	c ² =11.57 p=0.003	c ² =31.09 p<0.001	c ² <1.0 p>0.05	–
Fas (-1377G/A), n (%)	GA	6 (24.0)	4 (19.05)	13 (22.41)	1 (6.25)	c ² =2.13 p>0.05
	GG	19 (76.0)	17 (80.95)	45 (77.59)	15 (93.75)	
c ² p		c ² =72.20 p<0.001	c ² =16.10 p<0.001	c ² =35.31 p<0.001	c ² =24.50 p<0.001	–

TABLE 4. The distribution of polymorphic variants of the BCL-2 (rs17759659), CTLA-4 (rs231775) and APO-1 / Fas (rs2234767) genes in patients with thyroid disorders, taking into account the degree of its enlargement

The genes under study, n (%)		Control, n=25 (%)	The thyroid gland hyperplasia, n (%)			c ² p
			I degree, n=59	II degree, n=40	III degree, n=26	
BCL-2 (A/G), n (%)	AA	3 (12.0)	5 (8.47)	1 (2.50)	4 (15.38)	c ² =3.59 p>0.05
	GA	21 (84.0)	52 (88.14)	38 (95.0)	20 (76.92)	c ² =3.59 p>0.05
	GG	1 (4.0)	2 (3.39)	1 (2.50)	2 (7.69)	c ² =1.21 p>0.05
c ² p		c ² =64.80 p<0.001	c ² =119.9 p<0.001	c ² =102.7 p<0.001	c ² =33.69 p<0.001	–
CTLA-4 (+49G/A), n (%)	AA	15 (60.0)	34 (57.63)	11 (27.50)	14 (53.85)	c ² =9.26 p=0.01
	AG	9 (36.0)	23 (38.98)	29 (72.50)	10 (38.46)	c ² =12.34 p=0.002
	GG	1 (4.0)	2 (3.39)	0	2 (7.69)	p>0.05
c ² p		c ² =36.48 p<0.001	c ² =40.32 p<0.001	c ² =16.20 p<0.001	c ² =12.92 p=0.002	–
Fas (-1377G/A), n (%)	GA	6 (24.0)	9 (15.25)	10 (25.0)	4 (15.38)	c ² =1,71 p>0,05
	GG	19 (76.0)	50 (84.75)	30 (75.0)	22 (84.62)	
c ² p		c ² =72.20 p<0.001	c ² =56.98 p<0.001	c ² =20.0 p<0.001	c ² =24.92 p<0.001	–

TABLE 5. The distribution of polymorphic variants of the BCL-2 (rs17759659), CTLA-4 (rs231775) and APO-1 / Fas (rs2234767) genes in patients with adenoma of the thyroid gland, considering the degree of its enlargement

The genes under study, n (%)		Control, n=25 (%)	The thyroid gland hyperplasia, n (%)		c ² p
			I degree, n=20	II degree, n=10	
BCL-2 (A/G), n (%)	AA	3 (12.0)	1 (5.0)	0	p>0.05
	GA	21 (84.0)	19 (95.0)	10 (100.0)	
	GG	1 (4.0)	0	0	
c ² p		c ² =64.80 p<0.001	–	–	–
CTLA-4 (+49G/A), n (%)	AA	15 (60.0)	10 (50.0)	2 (20.0)	c ² <1,0 p>0,05
	AG	9 (36.0)	10 (50.0)	8 (80.0)	
	GG	1 (4.0)	0	0	
c ² p		c ² =36.48 p<0.001	–	c ² =7.20 p=0.007	–
Fas (-1377G/A), n (%)	GA	6 (24.0)	4 (20.0)	1 (10.0)	c ² <1,0 p>0,05
	GG	19 (76.0)	16 (80.0)	9 (90.0)	
c ² p		c ² =72.20 p<0.001	c ² =14.40 p<0.001	c ² =12.80 p<0.001	–

TABLE 6. The distribution of polymorphic variants of the BCL-2 (rs17759659), CTLA-4 (rs231775) and APO-1 / Fas (rs2234767) genes in patients with nodular form of goiter against the background of autoimmune thyroiditis depending on the degree of the thyroid gland hyperplasia

The genes under study, n (%)		Control, n=25 (%)	The thyroid gland hyperplasia, n (%)			c ² p
			I degree, n=39	II degree, n=30	III degree, n=26	
BCL-2 (A/G), n (%)	AA	3 (12.0)	4 (10.26)	1 (3.33)	4 (15.38)	c ² =2.41 p>0.05
	GA	21 (84.0)	33 (84.62)	28 (93.33)	20 (76.92)	c ² =3.01 p>0.05
	GG	1 (4.0)	2 (5.13)	1 (3.33)	2 (7.69)	c ² <1.0 p>0.05
c ² p		c ² =64,80 p<0,001	c ² =69.46 p<0.001	c ² =72.90 p<0.001	c ² =33.69 p<0.001	–
CTLA-4 (+49G/A), n (%)	AA	15 (60.0)	24 (61.54)	9 (30.0)	14 (53.85)	c ² =7.02 p=0.03
	AG	9 (36.0)	13 (33.33)	21 (70.0)	10 (38.46)	c ² =10.06 p=0.007
	GG	1 (4.0)	2 (5.13)	0	2 (7.69)	p>0.05
c ² p		c ² =36,48 p<0,001	c ² =27.92 p<0.001	c ² =9.60 p=0.002	c ² =12.92 p=0.002	–
Fas (-1377G/A), n (%)	GA	6 (24.0)	5 (12.82)	9 (30.0)	4 (15.38)	c ² =3.55 p>0.05
	GG	19 (76.0)	34 (87.18)	21 (70.0)	22 (84.62)	
c ² p		c ² =72,20 p<0,001	c ² =43.13 p<0.001	c ² =9.60 p=0.002	c ² =24.92 p<0.001	–

TABLE 7. Association of polymorphic variants of the BCL-2 (rs17759659) gene with the degree of the thyroid gland hyperplasia and its function

The genes under study, n (%)			The thyroid gland function, n (%)			c ² p
			Euthyroid goiter, n=31	Subclinical hypothyroidism, n=71	Clinical hypothyroidism, n=23	
BCL-2 (A/G), n (%)	AA, n=10	I degree, n=5	4 (12.90)	1 (1.41)	0	p=0.029
		II degree, n=1	0	1 (1.41)	0	–
		III degree, n=4	0	2 (2.82)	2 (8.70)	p>0.05
	GA, n=110	I degree, n=52	23 (74.19)	24 (33.80)	5 (21.74)	c ² =19.07 p<0.001
		II degree, n=38	3 (9.68)	33 (46.48)	2 (8.70)	c ² =20.09 p<0.001
		III degree, n=20	0	8 (11.27)	12 (52.17)	p<0.001
	GG, n=5	I degree, n=2	1 (3.22)	1 (1.41)	0	p>0.05
		II degree, n=1	0	1 (1.41)	0	–
		III degree, n=2	0	0	2 (8.70)	–

The patients with clinical hypothyroidism were more often recorded with the third degree hyperplasia of the thyroid gland, including those with predominant heterozygous AG-genotype of BCL-2

and CTLA-4 genes by 40.90% ($p < 0.001$) and 26.21% ($p = 0.002$), respectively; GG-genotype of Fas gene – by 55.36% ($c^2 = 29.70$; $p < 0.001$) (tab. 7-8).

TABLE 8. Association of polymorphic variants of the CTLA-4 (rs231775) and APO-1 / Fas (rs2234767) genes with the degree of the thyroid gland hyperplasia and its function

The genes under study, n (%)			The thyroid gland function, n (%)			c ² p
			Euthyroid goiter, n=31 (%)	Subclinical hypothyroidism, n=71 (%)	Clinical hypothyroidism, n=23 (%)	
CTLA-4 (+49G/A), n (%)	AA, n=59	I degree, n=34	15 (48.39)	16 (22.53)	3 (13.04)	c ² =10.14 p=0.006
		II degree, n=11	0	10 (14.08)	1 (4.35)	p>0.05
		III degree, n=14	0	5 (7.04)	9 (39.13)	p<0.001
	AG, n=62	I degree, n=23	12 (38.71)	9 (12.68)	2 (8.70)	c ² =11.58 p=0.003
		II degree, n=29	3 (9.68)	25 (35.21)	1 (4.35)	c ² =13.52 p=0.001
		III degree, n=10	0	3 (4.22)	7 (30.43)	p=0.002
	GG, n=4	I degree, n=2	1 (3.22)	1 (1.41)	0	p>0.05
		II degree, n=0	0	0	0	–
		III degree, n=2	0	2 (2.82)	0	–
Fas (-1377 G/A), n (%)	GA, n=23	I degree, n=9	4 (12.90)	4 (5.63)	1 (4.35)	c ² =2.06 p>0.05
		II degree, n=10	0	10 (14.08)	0	–
		III degree, n=4	0	3 (4.22)	1 (4.35)	p>0.05
	GG, n=102	I degree, n=50	24 (77.42)	22 (30.99)	4 (17.39)	c ² =25.39 p<0.001
		II degree, n=30	3 (9.68)	25 (35.21)	2 (8.70)	c ² =11.33 p=0.003
		III degree, n=22	0	7 (9.86)	15 (65.22)	c ² =29.70 p<0.001

CONCLUSIONS

TA and NGAIT are more common in the carriers of the minor G-allele (GA- and GG-genotypes) of the BCL-2 gene and in homozygous ones having the main G allele (GG-genotype) of the Fas gene by 11.5 and 4.34 times ($p < 0.001$), with no significant interdependence between the genotypes of the CTLA4 gene. We did not find any difference between the relative incidences of the genotypes of the analyzed genes in the patients with NGAIT and those with TA or depending on the TG function (euthyroid goiter, subclinical and clinical hypothyroidism). Hyperplasia of the TG in the patients in general as well as in those with NGAIT is associated with the wild A-alleles of the CTLA-4 gene (AA- and AG-genotypes): the I and III degree hyperplasia occurred reliably more frequently in carriers of the AA genotype by 30.13% and 26.35% ($c^2 = 9.26$; $p = 0.01$), and II degree of the TG enlargement in the patients with AG genotype by 33.52% and 34.04% ($c^2 = 12.34$; $p = 0.002$), respectively. In patients with NGAIT or TA, the TG function is asso-

ciated with its hyperplasia and with polymorphic sites of the genes under study. In patients with euthyroid goiter the I degree thyroid hyperplasia is more common: by 11.49-52.45% ($p \leq 0.029-0.001$) in the carriers of the wild A-allele of the BCL-2 gene and by 11.49-52.45% ($p \leq 0.029-0.001$) in those with A-allele of the CTLA-4 gene and by 46.43% and 60.03% ($p < 0.001$) with favorable GG-genotype of the Fas gene. In patients with clinical hypothyroidism, the III degree thyroid hyperplasia is more common: the carriers of the heterozygous AG-genotypes of the BCL-2 and CTLA-4 genes prevail by 40.90% ($p < 0.001$) and 26.21% ($p = 0.002$) respectively, and GG-genotype of the Fas gene by 55.36% ($p < 0.001$).

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