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Cardiovascular risk in chronic autoimmune thyroiditis and subclinical hypothyroidism patients. A cluster analysis

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ABSTRACT

Background: Subclinical hypothyroidism (SCH) and chronic autoimmune thyroiditis (CAT) are linked to an increased risk of atherosclerosis and coronary heart disease (CHD). The aim of this study was to look for positive markers of CHD and correlations with thyroid blood tests in patients with SCH or CAT, but no symptoms of CHD, so as to identify CHD risk conditions that otherwise would likely be missed.
Methods: We measured a series of thyroid, clinical-metabolic and cardiovascular parameters in 30 consecutive endocrinology patients enrolled in our ambulatory endocrinological referral center of "Sapienza" University of Rome. (19 with CAT, 11 with SCH) from January 2015 to March 2015. 13 asymptomatic subjects were enrolled as controls. In each patient, we measured a series of 34 thyroid, clinical-metabolic and cardiovascular parameters.
Results: in the statistical analysis of collected data, the oblique principal components clustering procedure (OPC) revealed the presence of an interesting mixed cluster, composed of a thyroid parameter (TPO-Ab), a metabolic parameter (homocysteine level) and a cardiovascular parameter (MAPSE), in which we assessed the relationships between the single components. Our preliminary results indicate that in both groups of patients elevated TPO-Ab, when accompanied by reduced MAPSE and increased IMT and homocysteine values, may be taken to indicate the presence of clinically unrecognized CHD.
Conclusions: Confirmation of these results in larger series of patients could justify hormone therapy for prevention of CHD in these thyroid patients versus placebo treatment.

1. Background

Thyroid dysfunction is relatively common, and its prevalence increases with advancing age. Thyroid hormones have a significant influence on the cardiovascular system [1]. Hypothyroidism has been observed to increase the risk of coronary heart disease (CHD) and atherosclerosis [2,3].

Subclinical hypothyroidism (SCH) is a common endocrine disorder characterized by increased levels of thyroid stimulating hormone (TSH) with normal levels of free thyroid hormones. SCH has also been linked to an increased risk of atherosclerosis [4], and meta-analyses have shown that it is associated with a 20% increased risk of coronary heart disease (CHD) [5,6] and with increased blood pressure [7].

Hashimoto's thyroiditis, or chronic autoimmune thyroiditis (CAT), is the most common organ-specific autoimmune disorder. The thyroiditis is characterized by diffuse lymphocytic infiltration of the thyroid gland with clinical evidence of goiter or atrophic gland and frequent thyroid dysfunction [8,9]. CAT frequently presents as clinical hypothyroidism, which is also associated with cardiovascular disorders and has been shown to affect both left and right ventricular function [10]. The effect of therapy with L-thyroxine, shown to modulate the immune process in animal models, is not clear in the human disease, especially in euthyroid patients in whom the use of L-thyroxine therapy is debated [11,12,13].

The aim of this study was to look for positive markers of CHD and correlations with thyroid blood tests in patients with SCH or CAT, but no symptoms of CHD, so as to identify CHD risk conditions that otherwise would likely be missed.

2. Methods

A total of 43 Caucasian patients recruited from the *Endocrinology A* unit at the University Hospital Policlinico Umberto I, from January 2015 to March 2015. Patients were divided into three groups: Group A consisted of 19 patients with CAT (4 males and 15 females; mean age 37.47 ± 8.57 years), in an euthyroid state; Group B comprised 11 SCH patients (3 males and 8

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females, mean age 39 ± 8.85 years); Group C consisted of 13 asymptomatic and euthyroid subjects (4 males and 9 females, mean age 33.38 ± 8.59 years) enrolled during a cardiovascular screening project in the general population, serving as a control group. Written informed consent was obtained from all individual participants included in the study and the IRB approval was obtained by Ethical Committee of Policlinico Umberto I.

The exclusion criteria were: 1) smoking; 2) hypertension (diastolic BP >90 mm Hg and systolic BP >140 mm Hg); 3) diabetes mellitus; 4) body mass index (BMI) >30 kg/m^2; 5) a history of CHD; 6) use of drugs that can affect cardiac kinetics; 7) ongoing treatment for thyroid disease. No patient had any systemic illness (including renal, pulmonary and liver disease) or was on any drug regimen. All had a sedentary lifestyle and all were in sinus rhythm.

In each patient, we measured a series of parameters of three types: thyroid (T), clinical-metabolic (M) and cardiovascular (C) (Table 1). The reference (normal range) values for the thyroid parameters were: TSH, 0.4–4.0 μUI/ml; FT3, 1.8–4.20 pg/ml); FT4, 0.65–1.45 ng/dl; anti-thyroglobulin antibodies (Tg-Ab), 0–40 UI/ml; anti-thyroid peroxidase antibodies (TPO-Ab), 0–35 UI/ml. Thyroid hormones were measured with an Elecsys Analyzer using an electrochemiluminescence immunoassay (ECLIA) method; TPO-Ab and Tg-Ab were measured using an immunoluminometric assay (ILMA).

The clinical-metabolic parameters investigated were: BMI in kg/m^2; body surface area (BSA) in m^2; total cholesterol (normal range 70–200 mg/dl); LDL cholesterol (normal range < 130 mg/ dl); HDL cholesterol (normal values > 40 mg/dl); triglycerides (normal range 50–190 mg/dl); glycemia (normal values <100 mg/dl); homocysteine concentration (normal range 5–15 μmol/l).

The cardiovascular parameters measured were: systolic blood pressure (mmHg); diastolic blood pressure (mmHg); heart rate (b/m); intima media thickness (IMT) in mm; flow-mediated dilation (FMD) (%); left ventricular mass (g); indexed left ventricular mass (g/m^2); systolic diameter (mm); diastolic diameter (mm); ejection fraction (%); E wave velocity (cm/s); A wave velocity (cm/s); E wave/A wave ratio deceleration time (mm/s); maximum aortic velocity (cm/s); mitral annular plane systolic excursion (MAPSE) (mm); tricuspidal annular plane systolic excursion (TAPSE) (mm); E′ wave velocity (cm/s); A′ wave velocity (cm/s); E wave/E′ wave ratio; left ventricular outflow tract systolic acceleration time (Lvot Acc) (m/s).

All blood tests were run at the same laboratory. Similarly, ultrasound study of the thyroid using 7.5 and 10 MHz linear transducers (Toshiba Aplio XV) and a Samsung Doppler ultrasound machine with 4–8 MHz probes, for the cardiac tissue Doppler, was always performed at the same center. CAT was diagnosed on the basis of the positive values of TPO-Ab and/or Tg-Ab supported by ultrasound findings of thyroid parenchymal heterogeneity [14]. SCH was diagnosed in the presence of TSH values above 4.0 μUI/ml and <10 μUI/ml, and FT3 and FT4 values at or below the lower limit of the normal range, for at least six months prior to enrollment in the study.

Ultrasound examination of the carotid arteries, allowing assessment of IMT and therefore FMD, was performed at the Department of Surgical Sciences at the Policlinico Umberto I using 7.5 and 10 MHz linear transducers (Toshiba Aplio XV). Intake of drugs and caffeine in the 12 h prior to the examination was prohibited. The examination was conducted with the patient comfortably settled in a supine position, assumed at least five minutes beforehand, and with an ambient temperature of around 20–22 °C. To calculate IMT, an automatic system was used to take longitudinal section measurements: of the common carotid artery within 10 mm of the carotid bifurcation, and at the origin of the internal carotid artery; the average of these three measurements, automatically supplied, was taken as the reference value [15].

FMD was calculated as the percentage difference between the maximum post-ischemic diameter and the mean basal diameter of the brachial artery: FMD = post-hyperemia diameter − basal diameter/basal diameter × 100. In accordance with international guidelines [16,17], FMD values >10% were taken as normal.

A Toshiba Aplio XV device equipped with a 2.5 MHz electronic probe was used for one-/two-dimensional and Doppler ultrasound study of cardiac function, while tissue Doppler was performed using a Samsung UGEO ultrasound machine. All examinations were carried out by the same sonographer, always calculating the mean value of three beats. As recommended by the American Society of Echocardiography, ventricular size and wall thickness values were obtained using the M-mode technique applied to 2D images of the heart at the cross-section of the left parasternal long axis, immediately below the mitral valve leaflets [18].

For the statistical analysis of the data, we calculated, for each experimental group, the mean values and standard deviations of each parameter analyzed. The normality of data was examined by Shapiro-Wilk test. Oblique principal components (OPC) variable clustering procedure was used in order to generate classes of homogeneous (mutually correlated) variables [19]. Pearson's correlation coefficient (r) was used to identify relationships between variables within clusters obtained using OPC; significance was evaluated using Student's *t*-test applied to the value of r. One-way analysis of variance (ANOVA or general linear model, GLM) was used for variables analyzed within a cluster. Differences between the two disease groups were considered significant at $p < 0.05$.

3. Results

Before beginning the inferential analysis proper, conducted to assess a possible effect of the disease states considered in the study, we studied the reciprocal relationships between the variables belonging to each of

Table 1
Thyroid hormone parameters (T), Clinical-metabolic parameters (M) and Cardiovascular parameters (C). Mean values ± standard deviation.

	Parameters	CAT	SCH	Controls
T	TSH (μIU/ml)	1.88 ± 0.82	6.52 ± 1.64	1.99 ± 0.68
	FT3 (pg/ml)	3.21 ± 0.37	3.25 ± 0.45	3.08 ± 0.78
	FT4 (ng/dl)	1.05 ± 0.16	0.98 ± 0.18	1.26 ± 0.13
	TG-Ab (IU/ml)	126.2 ± 239.5	83.65 ± 113.70	20.15 ± 6.43
	TPO-Ab (IU/ml)	115.8 ± 84.9	259.25 ± 414	20.69 ± 7.06
M	Age (yrs)	37.47 ± 8.57	39 ± 8.85	33.38 ± 8.59
	BMI (kg/m^2)	22.14 ± 3.31	24.32 ± 2.94	24.48 ± 1.29
	BSA (m^2)	1.70 ± 0.16	1.75 ± 0.19	1.91 ± 0.15
	Glycemia (mg/dl)	86.79 ± 9.70	83.91 ± 9.12	82.23 ± 5.37
	Total cholesterol (mg/dl)	192.05 ± 38.70	197.6 ± 23.6	173.31 ± 20.4
	HDL cholesterol (mg/dl)	61.68 ± 13.88	57.2 ± 9.56	66.9 ± 11.97
	LDL cholesterol (mg/dl)	110.40 ± 28.01	119.86 ± 13.2	89.7 ± 19.7
	Triglycerides (mg/dl)	101.58 ± 47.39	105.18 ± 32.01	87.75 ± 14.03
	Homocysteine (μmol/l)	8.83 ± 2.86	11.75 ± 6.23	7.03 ± 0.91
C	Diastolic diameter (mm)	46.04 ± 3.85	42.62 ± 5.65	46 ± 4.92
	Systolic diameter (mm)	28.77 ± 4.02	23.93 ± 6.58	29.23 ± 2.68
	Ejection fraction (%)	66.95 ± 7.01	71.73 ± 6.96	67.62 ± 4.19
	E wave (cm/s)	87.99 ± 14.79	90.93 ± 11.84	93.69 ± 7.59
	A wave (cm/s)	60.59 ± 14.68	67.36 ± 14.96	63.23 ± 9.67
	E wave/A wave ratio	1.56 ± 0.57	1.41 ± 0.49	1.52 ± 0.31
	Deceleration time (m/s)	0.19 ± 0.04	0.15 ± 0.03	0.18 ± 0.02
	Max. aortic velocity	108.95 ± 13.19	117.55 ± 10.21	120.85 ± 9.33
	Lvot Acc (m/s)	12.54 ± 2.43	12.61 ± 1.46	13.15 ± 1.21
	MAPSE (mm)	16.53 ± 2.01	16.55 ± 1.29	17.62 ± 1.26
	TAPSE (mm)	24.79 ± 3.57	22.45 ± 3.17	24.85 ± 2.70
	E′ wave (cm/s)	19.39 ± 4.08	22.09 ± 8.85	33.38 ± 9.79
	A′ wave (cm/s)	33.38 ± 9.79	10.59 ± 3.34	16.54 ± 8.93
	E/E′	4.72 ± 1.31	4.57 ± 1.44	3.13 ± 1.14
	Left ventricular mass	119.50 ± 34.72	127.73 ± 36.47	123.62 ± 31.21
	Indexed left ventricular mass	76.47 ± 23.11	75.19 ± 22.62	82.93 ± 34.66
	IMT (mm)	0.81 ± 0.20	0.96 ± 0.22	0.67 ± 0.12
	FMD (%)	24.52 ± 12.18	15.46 ± 12.17	24.39 ± 6.29
	Systolic bp (mm/Hg)	108.68 ± 10.39	115.91 ± 10.44	113.85 ± 11.2
	Diastolic bp (mm/Hg)	67.89 ± 7.87	75.45 ± 7.23	68.46 ± 6.25
	Heart rate (bpm)	75.26 ± 7.29	73.45 ± 7.09	76.85 ± 5.27

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