SHORT COMMUNICATION

Lipoprotein (a) levels, apolipoprotein (a) phenotypes and thyroid autoimmunity

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Abstract

It has been reported that euthyroid normolipidemic males and postmenopausal females exhibit significantly higher serum lipoprotein (a) (Lp(a)) levels compared with age- and sex-matched normolipidemic controls. However, it is well known that there is an inverse correlation between Lp(a) concentration and apolipoprotein (a) (apo(a)) isoform size. Thus, it is imperative to exclude differences in apo(a) isoform frequencies between subjects with or without thyroid autoimmunity in order to verify if there is an association between thyroid autoimmunity and increased Lp(a) concentration. To exclude such an effect of different apo(a) isoform frequencies, we determined apo(a) phenotypes in 22 patients (9 males and 13 postmenopausal females) with thyroid autoimmunity and in 64 (29 males and 35 females) age- and sex-matched individuals without thyroid autoimmunity (control group). There were no significant differences in the values of lipid parameters between the two groups, including Lp(a). We did not detect any significant differences in the apo(a) phenotype frequencies between the two groups. Additionally, in neither of the subgroups formed according to the presence of low molecular vs high molecular weight apo(a) isoforms were there any significant differences in median serum Lp(a) levels between patients with and without thyroid autoimmunity. Thus, our results contradict the previously reported association between thyroid autoimmunity and Lp(a) concentrations.

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Introduction

Lipoprotein (a) (Lp(a)) is a low density lipoprotein (LDL)-like particle in which apolipoprotein (a) (apo(a)) is linked to apoB100 by a disulfide bridge (1). Several studies have pointed out that high Lp(a) levels are associated with atherosclerotic disease, including myocardial infarction (2–4). Lp(a) concentrations may be altered by various environmental or hormonal factors as well as pathological situations (5–7). It has recently been reported that euthyroid males and postmenopausal females with thyroid autoimmunity present with increased serum Lp(a) levels, implying a role of thyroid autoimmunity in the Lp(a) metabolism (8). It should be mentioned, however, that the apolipoprotein (a) gene locus on chromosome 6q 2.6–2.7 is the major gene controlling Lp(a) serum concentrations (9, 10). Six different isoforms (designated F, B, S₁, S₂, S₃, S₄ according to different electrophoretic mobilities) that vary in size from ~400 kDa to >800 kDa were originally described. In the general population the size of these isoforms is inversely related to serum concentrations of

Lp(a). That is, individuals with low molecular weight (LMW) isoforms (F, B, S₁ and S₂) have, on average, high Lp(a) concentrations, whereas those with high molecular weight (HMW) isoforms (S₃, S₄) express low Lp(a) serum concentrations (9–11). Thus, it is necessary to exclude differences in apo(a) isoform frequencies between subjects with or without thyroid autoimmunity in order to verify if there is an association between thyroid autoimmunity and increased serum Lp(a) concentration.

We undertook the present study to examine the correlation of thyroid autoimmunity with lipoprotein (a) concentration and apolipoprotein (a) phenotypes.

Materials and methods

We studied 22 subjects (9 males and 13 postmenopausal females) aged 31–52 years with thyroid autoimmunity (increased titers of thyroperoxidase and/or thyroglobulin antibodies) as well as 64 subjects (29 males and 35 females) aged 30–51 years without

thyroid autoimmunity (control group), selected from individuals receiving medical check-up at our hospital free from any illness by history, physical examination, and routine laboratory data. Patients and controls with a known family history of primary hyperlipidemia, excessive alcohol consumption, diabetes mellitus, obesity (body mass index $>30 \text{ kg/m}^2$), liver disease, and systemic illnesses were excluded from the study. Furthermore, in all individuals thyroid function tests were within normal limits.

In patients and controls blood samples were taken after a 14-h overnight fast for the determination of lipid parameters, thyroid function tests (total and free thyroxine (T_4), total tri-iodothyronine (T_3), and thyrotropin), and thyroid autoantibodies (thyroperoxidase and thyroglobulin antibodies). The measurement of serum lipid parameters was performed by standard commercially available techniques as previously described (12). Lp(a) was measured using a monoclonal anti-Lp(a) antibody technique by the enzyme immunoassay Macra Lp(a) (Terumo Medical Corporation Diagnostic Division, Elkton, MD, USA). The lower limit of detectability was 0.8 mg/dl. In cases of Lp(a) levels less than 0.8 mg/dl, the value of 0.8 mg/dl was used for statistical reasons. The intra-assay and the interassay coefficients of variation were less than 6% and 10.3% respectively. In a recent report using this monoclonal antibody no cross reactivity with plasminogen, LDL, very low density lipoprotein (VLDL), or high density lipoprotein (HDL) was observed (13). Thyroid function tests were measured by immunoassay on an AxSYM analyzer (Abbott Laboratory, Abbott Park, IL, USA). Antiperoxidase and thyroglobulin antibodies were detected by hemagglutination. Positive samples were regarded as those which provided agglutination to a dilution of at least 1/400 and 1/640 respectively.

Apo(a) isoform size was then determined by agarose gel electrophoresis followed by immunoblotting as described previously (12). A standard solution (Immuno-France, S.A.R.L., Rungis, France) containing six different isoforms corresponding to the F, B, S_1 , S_2 , S_3 , S_4 isoforms in the nomenclature of Utermann *et al.* (11) was used in order to characterize our samples.

Statistical analysis

Lipoprotein (a) values were expressed in terms of median and range. A comparison of continuous variables was performed by an unpaired two tailed Student's *t*-test. Because of the highly skewed distribution of Lp(a) the non parametric Mann-Whitney U test was applied to discriminate for differences in Lp(a) levels between groups. Pearson's χ^2 test was applied to compare frequencies between groups. The LMW group includes all subjects with at least one of the isoforms F, B, S_1 , or S_2 , while the HMW group comprised all subjects with only S_3 or S_4 isoforms or with null type. Significance levels were set at $P < 0.05$ in all cases.

Results

There were no significant differences in the values of lipid parameters between the two groups, including Lp(a) (8.6 mg/dl (0.8–84 mg/dl) vs 9.4 mg/dl (0.8–108 mg/dl), $P = \text{not significant}$). We did not detect any significant differences in the apo(a) phenotype frequencies between the two groups (Table 1). Therefore, the absence of difference in serum Lp(a) concentration is a true finding and not biased by apo(a) type frequency differences. Additionally, in neither of the subgroups formed according to the presence of low molecular weight vs high molecular weight apo(a) isoforms were there any significant differences in median serum Lp(a) levels between patients with and without thyroid autoimmunity (Table 1).

Discussion

In our study no association was found between thyroid autoimmunity and increased serum Lp(a) concentration when apo(a) phenotypes were taken into account. In other words, our results do not seem to confirm previously published data which showed an association between thyroid autoimmunity and increased Lp(a) levels in normolipidemic healthy individuals (males and postmenopausal females) (8). Since the Lp(a) concentration has been shown to be elevated in inflammatory

Table 1 Distribution of apo(a) phenotypes and Lp(a) concentrations in the study population.

Phenotype	Patients with thyroid autoimmunity (n = 22)			Patients without thyroid autoimmunity (n = 64)		
	Number of patients	%	Median value (range) mg/dl	Number of patients	%	Median value (range) mg/dl
HMW ^a	15	68.2	4 (0.8–28)	45	70.3	4.5 (0.8–32)
LMW ^b	7	31.8	20 (0.8–84)	19	29.7	22 (0.8–108)

^a HMW, high molecular weight. All subjects with only S_3 or S_4 isoforms or with null type; ^b LMW, low molecular weight. All subjects with either of the isoforms F, B, S_1 or S_2 .

or immune processes and the role of an inflammatory process in the atherosclerotic plaque formation and rupture is prominent, the authors proposed that an autoimmune process, triggered for example by a concomitant intracellular infection, may occur in patients with high Lp(a) levels (8, 14).

The reasons for the observed discrepancies are not entirely clear-cut but may be related to the selection of patients (Lotz and Salabe (8) selected normolipidemic subjects and patients, while in our cohort the study population was randomly selected), to ethnic differences or to other poorly understood factors. However, the lack of apo(a) phenotypes in the study of Lotz and Salabe make their results problematic (8). Further studies with an adequate number of individuals of different ethnic backgrounds, including additional markers of autoimmunity are needed further to clarify this interesting issue.

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