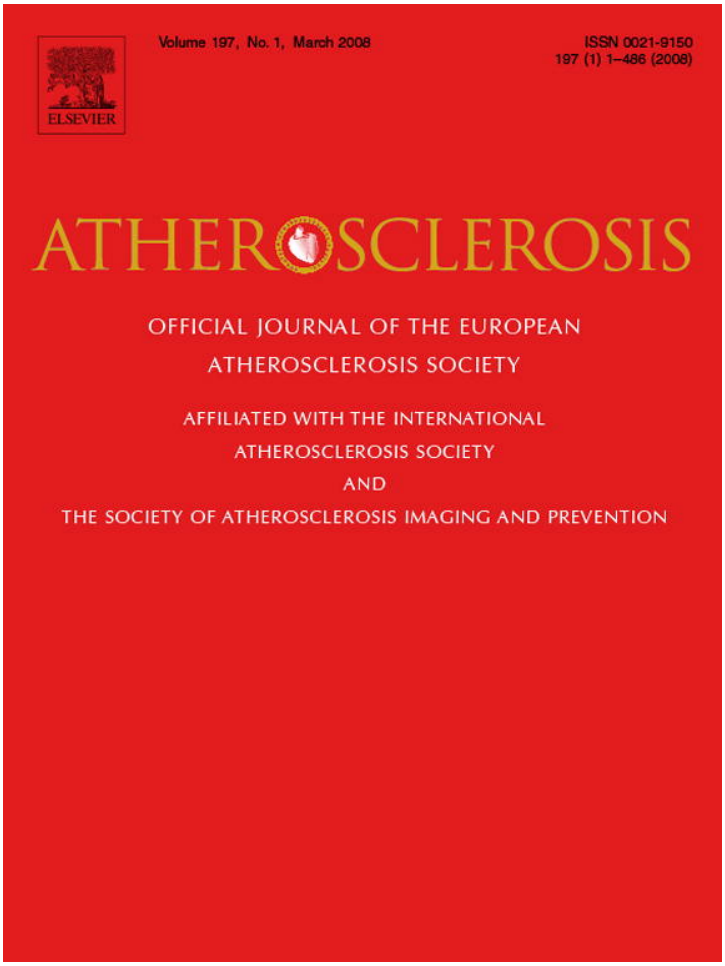

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Letter to the Editor

Lack of association between *NFKBIL1/LTA* polymorphisms and hypertension, myocardial infarct, unstable angina and stable angina in a large Irish population sample

Keywords: Lymphotoxin alpha; Acute coronary syndrome; Genetic risk

Dear Editor,

Genetic variation at the lymphotoxin alpha gene (*LTA*), which encodes a critical mediator of the immune and inflammatory response, was first identified as a risk factor for myocardial infarct (MI) in the Japanese population by Ozaki et al. [1]. The finding has been replicated in the same population [2]. Ozaki et al. also implicate variation in *NFKBIL1* (*IkBL*), which is located near *LTA* on a shared haplotype block and bears homology to the IkB family of NFkB regulators, which exert profound influence on the immune and inflammatory responses.

Attempts to replicate the association by case–control analyses in European populations have not yielded positive results, despite considerable statistical power [3,4]. However, transmission disequilibrium analysis of a large sample of European families has revealed excess transmission of the *LTA 804A*(*N26*) allele on the *LTA 10A/252G/804A*(*N26*) haplotype [5] and stratification of patients on the basis of the number of diseased arteries has revealed a role for the same polymorphism in the extent of atherosclerosis in male patients [6]. For this reason, the importance of this locus in different geographical regions is not fully resolved.

We have assayed genetic variation at *LTA 252A/G* and *NFKBIL1–62A/T*, as these two variants were reported to exert a functional effect on the gene expression levels of these genes [1]. Samples included 835 MI patients, 309 individuals with unstable angina (UA), 354 patients with stable angina (SA) and 105 patients with hypertension (HYP). Two control samples, one composed of bank workers (Control I, $N = 351$) and the other of blood donors (Control II, $N = 346$), were included in the analysis. The age structure and genotype frequencies of the populations analysed are outlined in Table 1. Both loci conformed to Hardy–Weinberg expectations in all populations, with the exception of *LTA 252* in the MI population, which showed a slight deficiency of heterozygotes (uncorrected $P = 0.016$). Correspondingly, both homozygotes were slightly more frequent than expected. However, this deviation is not statistically significant after Bonferroni correction for multiple testing.

There was no association between *LTA* or *NFKBIL1* variation and any of the coronary artery disease (CAD) phenotypes, either analysed separately or pooled (all CAD). This was the case for allele frequency comparison by χ^2 test and analysis based on the high-risk genotypes identified in previous studies (*NFKBIL1–62* genotype AA, *LTA 252* genotype GG). Haplotype analyses using HAPLO.SCORE statistics [7], adjusting for the effects of covariates age and sex, showed no association with either MI (haplotype score global statistic = 0.75731, $P = 0.686$), hypertension (global statistic = 0.03917, $P = 0.983$), unstable angina (global statistic = 2.61302, $P = 0.266$) or stable angina (global statistic = 1.54227, $P = 0.464$).

While case–control studies have not shown an effect in the European population, Laxton et al. [6] reported association between *LTA N26* homozygosity and the extent of atherosclerosis (1 versus 2 or 3 diseased arteries) in a cohort of British men, but not women. It was not possible to perform a similar stratification analysis on this case–control study, as angiography data are not available for controls, but HAPLO.SCORE analysis, adjusting for covariates age and sex, showed no effect. The PROCARDIS Consortium [5] showed an influence of the *LTA N26* allele with MI in 447 European families. However, study subjects were selected from a large European database on the basis of the availability of parental genotypes. The authors pointed out that their study was retrospective and the *LTA N26* haplotype may be a risk-factor only in survivors of MI and/or CAD.

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Table 1
Population characteristics and genotype frequencies

Count (%)	Control I	Control II	HYP	MI	SA	UA
N	351	346	105	835	354	309
Age (mean)	52.4815	–	65.1524	61.9449	65.8785	62
S.D.	9.9854	–	11.7103	8.5375	7.4468	7.7531
NFKBIL1 (IkBL)–62						
TT	143 (41.21)	125 (36.34)	44 (43.56)	317 (39.23)	127 (38.14)	109 (35.97)
TA	156 (44.96)	172 (50.00)	41 (40.59)	364 (45.05)	157 (47.15)	154 (50.83)
AA	48 (13.83)	47 (13.66)	16 (15.84)	127 (15.72)	49 (14.71)	40 (13.20)
Total	347	344	101	808	333	303
LTA 252						
AA	139 (40.29)	130 (37.57)	39 (43.82)	301 (40.46)	119 (37.19)	96 (34.41)
GA	157 (45.51)	169 (48.84)	38 (42.70)	320 (43.01)	151 (47.19)	144 (51.61)
GG	49 (14.20)	47 (13.58)	12 (13.48)	123 (16.53)	50 (15.63)	39 (13.98)
Total	345	346	89	744	320	279

The PROCARDIS Consortium [5] reported genotype risk ratios of 1.40 (1.12–1.77) for the *LTA N26* allele and 1.96 (1.25–3.13) for *LTA N26* homozygotes. This is comparable to Ozaki et al.'s odds ratio of 1.78 (1.39–2.27) for the Japanese population [1]. Our power calculations using the Genetic Power Calculator [8] show that we have >85% power to detect an effect of the magnitude indicated above, for 835 MI patients and 351 controls, at the 1% level of statistical significance.

Therefore, while the association between *LTA* polymorphism and MI has been successfully replicated, this is not the case for case–control studies of European populations, where the present study and two other high-powered case–control studies have not replicated the association [3,4]. Any effect of *LTA* polymorphism in European populations appears to be limited to specific subsets of patients (males with multiple vessel disease [6], or MI patients who have parents/siblings available for genotyping [5]). Thus, notwithstanding the fact that inflammatory effects are widely accepted as playing a role in CAD, the results point to differing underlying genetic and environmental effects in susceptibility to MI and other forms of CAD in diverse human populations.

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