Lack of Association Between Human TGF-β1 Gene Variants and Primary Hypertension

Epidemiologic, clinical, and experimental evidence has suggested that upregulation of transforming growth factor-β1 (TGF-β1) may play a role in hypertensive disease. TGF-β1 is a multifunctional cytokine that regulates cell growth and differentiation, modulation of extracellular matrix and repair, which in turn regulate inflammatory and immune responses and can be involved in vascular remodelling.

Among the seven polymorphisms that have been identified at the TGF-β1 gene locus, the TGF-β1*25C (Pro25) allele has been found to be associated with lower systolic blood pressure and lower risk of essential hypertension (EHT), but with increased risk of myocardial infarction in sample populations from Northern Ireland and France. The authors, together with an accompanying editorial, offered a cautious interpretation of their data, which therefore needs further probing in other populations. Furthermore, that study did not address the issue of quantitative phenotypes related to TGF-β1 production in relation to EHT. This approach became available through the findings that a polymorphism at position 75 in the signal peptide sequence, which changes codon 25 (Arg→Pro), is associated with interindividual variation in TGF-β1 production. The Arg-to-Pro at codon 25 is in the signal sequence of the precursor, and the mutation results in the substitution of a large polar amino acid with a small nonpolar residue. No study has so far been conducted to determine whether the substitution has functional and biologic importance, but it could affect protein transport into the endoplasmic reticulum.

As data from genetically isolated populations are of utmost importance to resolve issues of contention related to studies based on the concept of linkage disequilibrium, we carried out an association study on unrelated normotensive and hypertensive subjects the distributions of genotypes and alleles of the two following two-allele polymorphisms: a T-to-C transition in exon 10 of the TGF-β1 gene (TGF-β1*10[T→C]), which leads to a Leu-to-Pro mutation; and the G-to-C transversion in exon 25 of TGF-β1 (TGF-β1*25[G→C]). This project was approved by the Research Ethics Committee of the Faculty of Medicine and Health Sciences (UAE University, Al Ain, UAE).

The sample population of 144 unrelated subjects (75 men, 69 women) had a mean age (± standard deviation) of 52.1 ± 12.3 years and was composed of the following two groups: 72 patients with EHT (mean age, 52.8 ± 12.1 years), and 72 controls used as a comparison group (mean age, 51.7 ± 11.5 years). Hypertensive and normotensive subjects have been described before. They were matched for age and gender, and there was no difference in body mass index values among the two groups. Total serum cholesterol levels were significantly higher in the group of hypertensives.

TGF-β1*10C allele frequencies were 0.46 ± 0.04 and 0.41 ± 0.04 among normotensives and hypertensives, respectively; TGF-β1*25C allele frequencies were 0.07 ± 0.02 and 0.08 ± 0.02 (data not shown). There was no statistically significant difference in the distributions of genotypes of the two individual markers according to clinical phenotype (data not shown). The overall allelic frequencies of both dimorphisms were similar to what has been found in a European population. Thus, the TGF-β1*10C allele frequency was 0.41 to 0.46 ± 0.04 among Emirati, compared with 0.35 ± 0.03 in a UK sample population and 0.41 to 0.43 in other European populations. TGF-β1*25C allele frequencies were 0.07 to 0.08 ± 0.02 among Emirati, 010 ± 0.02 among UK subjects, and 0.07 to 0.11 among Irish and French subjects from different Centers.

Haplotypes combining both sites were constructed and linkage disequilibrium values (gametic diallelic pairwise disequilibrium, D) for the pair of dimorphisms (10T→C and 25G→C) were measured by calculating the deviation of the observed frequency of the haplotype from that expected from multiplication of the individual allele frequencies. In the case of double heterozygotes (10T/10C with 25G/25C), the haplotype phase was estimated by the maximum-likelihood procedure described by Hill. The significance of the difference of D = D/D max from 0 (Dmax is the maximum possible disequilibrium for a given pair of allele frequencies) was calculated as χ² with 1 degree of freedom (df) using a method described elsewhere. The resulting values were as follows: for
the normotensive group, the 71 studied pairs of sites were associated with D = 0.0028, D_m = 0.0378, D* = 0.074, \( \chi^2 = 0.03 \) (not significant); in the case of the EHT group, D = 0.0032, D_m = -0.0392, D* = 0.082, \( \chi^2 = 0.04 \) (70 pairs of sites; not significant). There was thus no significant departure from equilibrium. The fact that the two markers are in linkage equilibrium should increase the chance of detecting an association with the clinical phenotype under study (if such an association exists). Yet the distribution and frequencies of the four haplotypes (Table 1) indicate no statistically significant difference between normotensive and hypertensive UAE subjects (\( \chi^2 = 0.61, 3 \text{ df}, P = .74 \)).

Therefore, our results indicate a lack of association between TGF-\( \beta \)-10(T\( \rightarrow \)C) and TGF-\( \beta \)-25(G\( \rightarrow \)C) markers and clinical diagnosis of EHT in the population under study. These data therefore suggest that changes in the signal sequence due to TGF-\( \beta \)-10(T\( \rightarrow \)C) and TGF-\( \beta \)-25(G\( \rightarrow \)C) dimorphisms do not exert a major effect on the pathogenesis of HHT among Emiratis; and putative TGF-\( \beta \) variants whose DNA variations might be in linkage disequilibrium with TGF-\( \beta \)-10(T\( \rightarrow \)C) and TGF-\( \beta \)-25(G\( \rightarrow \)C) dimorphisms do not play a significant role in the determination of hypertension among Emiratis.

It could be argued, however, that the relatively small number of subjects (144) could give a low probability of detecting a small effect of the polymorphism (a small gene effect is expected in the case of a disease as complex as hypertension). This is why we cannot exclude the possibility that the TGF-\( \beta \) gene is somehow involved in the pathogenesis of hypertension. Nonetheless, such a putative involvement is not reflected by the TGF-\( \beta \)-10(T\( \rightarrow \)C) and TGF-\( \beta \)-25(G\( \rightarrow \)C) changes of the signal sequence of TGF-\( \beta \) in the Emirati population studied here.

**REFERENCES**


