Letters to the Editor

Role of endothelial nitric oxide synthase (eNOS) polymorphisms in cardiovascular disease and rheumatoid arthritis

Sirs,

We read with much interest the paper by Gonzalez-Gay and colleagues (1) in which the authors assessed the contribution of inducible and endothelial nitric oxide synthase (NOS2A and NOS3) gene polymorphisms to cardiovascular (CV) events in a cohort of patients with rheumatoid arthritis. The results suggested NOS2A or NOS3 gene polymorphisms do not have a direct effect in CV risk in RA, but an increased frequency of CV events was observed in RA patients who carried the HLA-DRB1*0404 allele and were homozygous for the presence of long NOS2A alleles or for the NOS3 (-786) TT genotype. We would like to raise an important issue concerning to the last finding. In genetic association studies, candidate polymorphisms should be related with an observable influence in the expression and functional activity of a specific gene product (2). That is the case for some eNOS polymorphisms, such as T-786C in the promoter, Glu298Asp variant in the exon 7 and the variable number of tandem repeats (VNTRs) in intron 4. In respect of the former, as assessed by luciferase reporter gene assays, the T-786C mutation resulted in a significant reduction in eNOS gene promoter activity, leading to a reduction in the endothelial NO synthesis (3). This altered eNOS expression and the resulting lack of NO availability in the vessel contribute for the endothelial dysfunction, which is directly related with higher risks for atherosclerosis (4). In the light of that, it would be more logical to conclude that the C variant is additional. Clinical data from the GENICA (Genetic and Environmental factors In Coronary Atherosclerosis) study had shown C allele was associated with a higher risk of multivessel coronary artery disease in Caucasians (5). Taken together, these data contradict the conclusion that the TT genotype may interact with the HLA-DRB1 and play a relevant role in increasing the risk for CV events in RA.

Nevertheless, we agree with the authors that endothelial dysfunction is an important feature of RA and is influenced by genetic factors as reported in a previous work (6). In this regard, we believe that eNOS polymorphisms might be implicated not only in a higher CV risk, but also in susceptibility for RA and clinical expression of disease, as the production of NO in the endothelium may offer a protective or antiinflammatory effect by preventing the adhesion of leukocytes and the release of oxidants by activated neutrophils in the microvasculature (7). The potential role of the T-786C polymorphism in susceptibility for RA was assessed in two previous studies with conflicting results. In 2004, Gonzalez-Gay *et al*. did not find significant differences between 200 RA patients and 251 ethnically matched Caucasoid controls in genotype or allele frequencies (8). Two years later, Melchers *et al*. found a higher frequency of the C/C genotype in 596 RA patients as compared to the general German population (9). Interestingly, these same authors observed that endothelial cells of the C/C genotype, differently from cells of other genotypes, did not respond by increasing NOS3 expression after IL-10 induction, and suggested that this insensitivity to an antiinflammatory cytokine could represent a risk factor for rheumatoid arthritis. Recently, in a smaller case-control study, although no differences were observed in genotypic or in allelic frequencies between RA and control Brazilian individuals, the C/C genotype was significantly associated with extraarticular manifestations compared with the T/T and T/C genotypes taken together (OR=4.9, 95% CI=1.3–18.9), again suggesting a role of NOS3 on the pathophysiology of rheumatoid arthritis (10). It is known that several factors, such as the genetic background and ethnicity of the population analysed, could influence on complex disease susceptibility and therefore further studies are needed to elucidate the role of eNOS polymorphisms in the pathogenesis of clinical manifestations and CV disease in RA.

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References

1. GONZALEZ-GAY MA, LLORCA J, PALOMINO-MORALES R, GOMEZ-ACEBO I, GONZALEZ-JUANATEY C, MARTIN J: Influence of nitric oxide synthase gene polymorphisms on the risk of cardiovascular events in rheumatoid arthritis. *Clin Exp Rheumatol* 2009; 27: 116-9.
2. HINGORANI AD: Polymorphisms in endothelial nitric oxide synthase and atherogenesis: John French Lecture 2000. *Atherosclerosis* 2001; 154: 521-7.
3. NAKAYAMA M, YASUE H, YOSHIMURA M *et al*.: T-786-->C mutation in the 5'-flanking region of the endothelial nitric oxide synthase gene is associated with coronary spasm. *Circulation* 1999; 99: 2864-70.
4. SUWAIDI JA, HAMASAKI S, HIGANO ST et al., Long-term follow-up of patients with mild coronary artery disease and endothelial dysfunction. *Circulation* 2000; 101: 948-54.
5. ROSSI GP, CESARI M, ZANCHETTA M *et al*.: The T-786C endothelial nitric oxide synthase genotype is a novel risk factor for coronary artery disease in Caucasian patients of the GENICA study. *J Am Coll Cardiol* 2003; 41: 930-7.
6. GONZALEZ-JUANATEY C, TESTA A, GARCIA-CASTELO A *et al*.: HLA-DRB1 status affects endothelial function in treatedpatients with rheumatoid arthritis. *Am J Med* 2003; 114: 647-52.
7. ABRAMSON SB, AMIN AR, CLANCY RM, ATTUR M *et al*.: The role of nitric oxide in tissue destruction. *Best Pract Res Clin Rheumatol* 2001; 15: 831-45.
8. GONZALEZ-GAY MA, LLORCA J, SANCHEZ E *et al*.: Inducible but not endothelial nitric oxide synthasepolymorphism is associated with susceptibility to rheumatoid arthritis in northwest Spain. *Rheumatology* (Oxford) 2004; 43: 1182-5.
9. MELCHERS I, BLASCHKE S, HECKER M, CATTARUZZA M: The -786C/T single-nucleotide polymorphism in the promoter of thegene for endothelial nitric oxide synthase: insensitivity to physiologic stimuli as a risk factor for rheumatoid arthritis. *Arthritis Rheum* 2006; 54: 3144-51.
10. BRENOL CV, CHIES JA, BRENOL JC *et al*.: Endothelial nitric oxide synthase T-786C polymorphism inrheumatoid arthritis: association with extraarticular manifestations. *Clin Rheumatol* 2009; 28: 201-5.