Reviewer's report

Title: HLA HAPLOTYPES ASSOCIATED WITH HEMOCHROMATOSIS MUTATIONS IN THE SPANISH POPULATION

Version: 2 Date: 30 July 2004

Reviewer: Graca Porto

Reviewer's report:

The authors have generally answered to the referees’ comments. Some details, however, should be still corrected:

1. I understand that the authors have used blood samples which have been probably referred for diagnosis (probands?) but from whom they do not have much clinical information. For the purpose of this study, whose objective is to test a large sample of homozygous for the mutations, it is not essential to have a full clinical description, however, a clear definition of the sample is needed. I would suggest to change this paragraph as follows:

“A total of 100 unrelated individuals homozygous for the C282Y mutation, 138 unrelated individuals homozygous for the H63D mutation and 17 individuals heterozygous for the S65C mutation were selected for this study. These were subjects in whom HFE genotyping has been previously performed on a medical care basis because of a presumptive diagnosis of hemochromatosis. In addition, 1113 unrelated, apparently healthy subjects were used as controls for the study. HFE genotyping was performed in 230 individuals from the control group.” (cut this phrase from the next section).

At this point I am confused. The authors refer that they genotyped 230 controls. In the results section they refer to 230 individuals without HFE mutations. Were all the genotyped subjects wild type? This is not possible given the frequency of the mutations in the normal Caucasian population. Please clarify.

2. The first paragraph of the discussion on: H63D and HLA does not provide a correct interpretation of the cited papers. I would suggest, instead of “and between HLA-B44 and the same clinical form”: “and a strong linkage disequilibrium between H63D and all A29 containing haplotypes assigned in a large population of normal Portuguese families(22)".

I think this point is relevant. First because reference 22 is not a study of individual associations but it is a study in 587 normal subjects whose haplotypes were assigned by family segregation and not by linkage disequilibrium. Secondly, as already pointed out by M.D. de Juan, this finding is confirmed with success with this study, and not contradicted. The major difference between the two studies is the significance of the association with the haplotype A29-B44, but this could be due to differences in the method of assigning haplotypes. In conclusion, I would also suggest modifying the discussion in this chapter on line 5 of the second paragraph. Instead of: “with lesser levels of significance” to substitute for:

“confirming the results reported by Cardoso et al (22) in the normal Portuguese population.”

What next?: Accept after minor essential revisions
**Level of interest:** An article of importance in its field

**Quality of written English:** Acceptable

**Statistical review:** No

**Declaration of competing interests:**

None