HAEMOLYSIS IN SICKLE CELL ANAEMIA: A GENOTYPE/PHENOTYPE ASSOCIATION STUDY

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INTRODUCTION & OBJECTIVES

Sickle-cell anaemia (SCA) is a clinically heterogeneous autosomal recessive monogenic chronic anaemia characterized by recurrent episodes of severe vaso-occlusion, haemolysis and infection. Several genetic and environmental modifiers have been suggested to modulate the onset and course of SCA (1).

As part of a wider research on the development and validation of vaso-occlusion early predictors in SCA, we have studied the association between three haemolysis biomarkers and the inheritance of genetic variants of eleven candidate genes in a series of paediatric SCA patients.

METHODS

Subjects: 99 paediatric SCA (SS) patients (median current age of 9.9 years) followed-up in two general hospitals in Greater Lisbon area (median follow-up/patient of 5.0 years).

Haemolysis biomarkers: LDH and total bilirrubin level and reticulocyte count.

Candidate gene genotyping: *BCL11A, CD36, EDN1, HBA, HBB* cluster (including *HBG), HBS1L-MYB*, *ITGA4, HMOX1, NOS3*, *THBS1* and *VCAM1.*

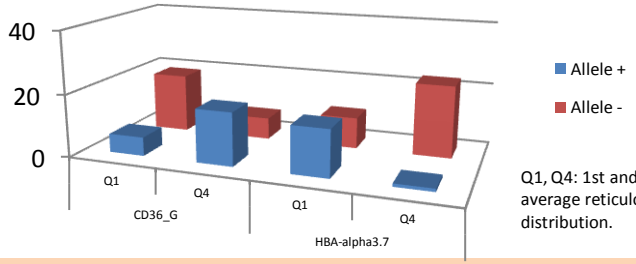
Statistical analysis: Association studies between candidate genotypes and haemolysis biomarkers using T test ANOVA parametric tests (LDH, total bilirrubin) or Mann-Whitney/Kuskal-Wallis non-parametric tests (reticulocyte count), all performed with SPSS v20.0 software, with subsequent correction for multiple testing (false discovery rate).

RESULTS

Although in a large number of tests a seemingly significant (i.e., $p<0.05$) association was observed, only the following ones were confirmed upon correction for the false discovery rate:

- An elevated LDH was associated to haplotype 7 within *VCAM1*.
- A lower total bilirrubin was associated to the 3.7kb deletion at *HBA*, rs2070744_T allele and haplotypes 3 and 4 at *NOS3* and haplotype 9 within *VCAM1* and rs3783598_G and rs3917024_T alleles at *VCAM1* promoter.
- A diminished reticulocyte count was associated to the 3.7kb deletion at *HBA* , whereas an elevated count was associated to rs1984112_G allele at *CD36* (see figure).

Furthermore, at the phenotypic level all three haemolysis biomarkers were positively associated to left ventricle dilation, a common chronic complication of SCA.



Q1, Q4: 1st and 4th quartile average reticulocyte count distribution.

CONCLUSION

On the whole, our findings suggest a complex genetic architecture for the haemolytic endophenotype in SCA involving multiple pathways, namely control of erythrocyte volume and haemoglobinisation, vascular cell adhesion, NO synthesis and lipid metabolism. Further mechanistic studies are needed to explore these avenues leading to a better understanding of the inter- and intra-individual clinical variability of SCA.

Reference: (1) Steinberg MH and Sebastiani P. Genetic modifiers of sickle cell disease. Am J Hematol 87:795-803, 2012.

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