
Response

Genetic admixture in sickle cell disease

Genetic studies require careful thought about ancestry in the study design and analysis to avoid population stratification bias and to maximize the power of finding novel variants.1 If both the genetic marker and the phenotype vary with respect to ancestry, then a spurious association will occur between the genetic marker and phenotype if one does not adjust properly for ancestry. In this issue of Blood, Silva et al examine the level of admixture in a cohort of Brazilian sickle cell patients and find that patients with sickle cell disease have a wide range of African admixture (15%-85%).2 They correctly acknowledge that one must appropriately adjust for admixture to avoid false positive findings and that this population may be useful for admixture mapping. However, admixture mapping is only useful if the phenotype of interest also varies with admixture and further research is required to establish this relation in this population. In a cohort of African Americans with sickle cell disease we did not find a significant association between admixture (measured by the first principal component from a principal component analysis) and fetal hemoglobin.3 However, one should note that the African Americans in this study on average did not have high levels of Caucasian admixture.4 Admixture mapping has been successfully used to find novel genetic variants and regions for other phenotypes that were related to admixture such as white cell counts and prostate cancer.5,6 Furthermore, examining ancestry in genetic studies of sickle cell disease could lead to novel loci that are either more prevalent or specific to certain ethnic groups. For example, sickle cell patients from the Southwestern Province of Saudi Arabia have fetal hemoglobin (HbF) levels twice as high as African Americans despite having similar HBB haplotypes.7 Furthermore, sickle cell patients from the Eastern Province have even higher levels of HbF (mean [SD] 30.4 ± 6.9).8 These findings suggest that there are HbF-associated variants that are more prevalent or specific to the Saudi population and that leveraging on ancestry in the genetic analysis can help identify novel variants.

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References


To the editor:

Prothrombin 20210G>A genotype and C-reactive protein level

Thrombin is central not only in procoagulatory processes like fibrinogen or platelet activation but also in other systems that are related to inflammation control.1 Recently, Flick et al characterized inflammatory responses of transgenic prothrombin mutant (F2210G) mice in a collagen-induced arthritis (CIA) model.2 Mice carrying the F2210G transgene that has dramatically reduced procoagulatory effects on fibrinogen and protease-activated receptor 1 exhibited a significantly attenuated inflammatory joint disease in CIA.

Prothrombin 20210G>A (F2210G>A) is a gain-of-function variant resulting in increased prothrombin expression and elevated
risk of venous thromboembolic events (VTE).\textsuperscript{3} Based on the data of Flick et al\textsuperscript{2} we hypothesized that F2\textsubscript{20210G}>A is associated with increased inflammatory responses, and we tested this hypothesis in a cohort with longitudinal measurements of C-reactive protein (CRP) levels. Study population consisted of 565 women [age, median (interquartile range), 32 years (28-36)] with 2,258 observations [observations per patient: 2 (n = 153), 3 (n = 109), 4 (n = 98), 5 (n = 86), > 5 (n = 119)] followed during pregnancy predominantly because of history of fetal loss, placental dysfunction or VTE. Pregnancy is known as a mild proinflammatory condition with moderately elevated CRP levels.\textsuperscript{4} These are increased further in case of disturbed placentation or other pregnancy complications.\textsuperscript{5} An association between CRP levels and risk of VTE has been described previously, as well.\textsuperscript{6} To adjust for intra-individual CRP level variations evaluation was restricted to patients with CRP levels of at least 2 independent times of presentation.\textsuperscript{7} Therefore, for univariate and multivariate analyses as well as nonparametric comparisons robust clustered estimates of variances were calculated to allow for intracluster correlation and to relax requirement for independent observations (Stata 10.1 Macintosh, StataCorp). F2\textsubscript{20210G}>A genotypes were characterized as described,\textsuperscript{8} plasma CRP levels were quantified by an immunoturbidimetric method standardized according to International Federation of Clinical Chemistry and Laboratory Medicine (Roche Diagnostics). The study was approved by the local ethics committee. All individuals were included in this study, after informed consent had been provided.

Carrier frequency of F2\textsubscript{20210A} was 5.5%. Carriers of the F2\textsubscript{20210A} variant exhibited significantly more frequently CRP elevations (\geq 5 mg/L, 47.5% [65/137 observations], \geq 10 mg/L, 25.6% [35 of 137 observations], \geq 15 mg/L, 11.7% [16 of 137 observations]) compared with F2\textsubscript{20210G} wild-type individuals (\geq 5 mg/L, 30.7% [650/2121 observations], \geq 10 mg/L, 10.3% [219 of 2121 observations], \geq 15 mg/L, 4.8% [101 of 2121 observations]).

The F2\textsubscript{20210G}>A genotype is known to influence prothrombin expression.\textsuperscript{9} In our study population prothrombin levels (mean prothrombin activity [95% CI]) were assessed in 122 patients and were significantly increased in F2\textsubscript{20210A} carriers compared with F2\textsubscript{20210G} wild-types F2\textsubscript{20210G}, 112% [109-115], F2\textsubscript{20210A}, 138% [111-166], t test, P = .0005). Thus, our finding that the gain-of-function variant F2\textsubscript{20210A} is associated with increased CRP levels further corroborates the assumption of Flick et al that (pro)thrombin in positively involved in inflammatory processes and that targeting (pro)thrombin activity could have the potential to modulate inflammatory disease processes.\textsuperscript{2}

### References

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