Author's response to reviews

Title: Association of soluble endothelial protein C receptor plasma levels and PROCR rs867186 with cardiovascular risk factors and cardiovascular events in coronary artery disease patients: The AtheroGene Study

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Author's response to reviews: see over
Dear Editor,

We thank you for your continued interest in our manuscript entitled "Association of soluble endothelial protein C receptor plasma levels and PROCR rs867186 with cardiovascular risk factors and cardiovascular events in coronary artery disease patients: The AtheroGene Study".

We are pleased to send you a revised version of this manuscript that now incorporates a more detailed analysis of the PROCR SNPs as requested by reviewer 2.

We hope you would find this manuscript suitable for publication in BMC Medical Genetics

Sincerely yours

Trégouët David-Alexandre

Referee 1

"there is no major revision, but may be minor revision is required for correlation analysis. Associations between individual PROCR Ser219Gly genotypes and baseline quantitative phenotypes were assessed using multiple linear regression models. All regression models were minimally adjusted for age and sex. To reduce the influence of environmental variation, regression models were additionally adjusted for cholesterol and triglycerides. Associations between individual PROCR Ser219Gly genotypes and risk of incident CVD events or mortality during follow-up were assessed using Cox regression, adjusted for major clinical risk factors (age, sex, diabetes, hypertension, clinical CVD status, smoking, and serum creatinine)."

We are sorry but we were not able to figure out in these comments what minor corrections, if any, the referee wanted us to address. As such, no specific corrections were brought.

Referee 2

Major points
1) It would be of great interest to know whether other polymorphisms are associated with sEPCR plasma levels. Genotyping was performed on the Affy 6.0 array. The authors must have the genotypes for other SNPs within and near the PROCR gene. Are there any other polymorphisms in this gene that influence the sEPCR plasma levels? A LD plot of this locus would be useful.

We thank the reviewer for suggesting to assess whether other PROCR SNPs could modulate sEPCR levels. Four other PROCR SNPs were typed on the Affy 6.0 array and were thus analyzed in the current work. Linkage disequilibrium (new Table 4) and haplotype (new Table 6) analyses were also conducted. Only the haplotype carrying the rs861716-G allele was associated with sEPCR levels.
Minor points

1) P values testing differences between the two groups in Table 1 should be presented.

This has been done.

2) The median follow-up time is 3.7 years. The number of events during that time is 136. The authors should provide power calculation and indicate whether larger sample size or longer follow-up period is needed.

This point was now addressed in the discussion.
"It could also be argued that the low number of events observed during the follow-up with median time of 3.7 years may have limited our power to detect any association of sEPCR with future CVE, especially if sECPR effects, if any, exert at a later time period. Nevertheless, our study was large enough to detect the association of several biomarkers, including parameters characterizing the renal function, with the risk of future CVE."

However, we decided not to include exact power calculations as they would not have been straightforward. They would depend on different arbitrary values including time to follow-up and anticipated effects of sEPCR.

3) Limitations should be discussed.

We have modified the discussion to mention some limitations related to the size of our study and the modest follow-up time.
"It could also be argued that the low number of events observed during the follow-up with median time of 3.7 years may have limited our power to detect any association of sEPCR with future CVE, especially if sECPR effects, if any, exert at a later time period. Nevertheless, our study was large enough to detect the association of several biomarkers, including parameters characterizing the renal function, with the risk of future CVE."

"This is unlikely due to a loss power since the same allele frequencies were observed in both groups of patients with or without future CVE. ..... Nevertheless, it would be highly interesting to investigate whether the trend of association observed between sEPCR and CVE risk in rs867186-G carriers only could replicate in a much larger cohort with a longer follow-up."