Author's response to reviews

Title: Single nucleotide polymorphisms in the apolipoprotein B and low density lipoprotein receptor genes affect response to antihypertensive treatment

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

In response to the comments by the Reviewers, we have now revised our manuscript “Single nucleotide polymorphisms in the apolipoprotein B and low density lipoprotein receptor genes affect response to antihypertensive treatment” 1966975485387473. Our responses to the two Reviewers are detailed point-by-point below. We hope that you will find our revised manuscript acceptable for BMC Cardiovascular Disorders.

With best regards,
Ulrika Liljedahl

Reviewer 1: Martin C. C. Michel

Major compulsory revisions
1. The first issue raised by this reviewer is regarding the important issue risk of false positive results in genetic association studies and the need for multiple testing corrections in the statistical analyses. To clarify our earlier studies in the SILVHIA sample set, we have added a short paragraph with information concerning these SNP analyses to the Results and Discussion section on page 9. The paragraph states that a set of 74 SNPs were tested in an early, exploratory study of the SILVHIA samples using multiple regression analysis. The major aim of this initial study was to establish the microarray based genotyping system. As stated in the Materials and Methods section, nine of the ten SNPs in lipid genes analyzed in the current study belonged to this original set. In addition, 28 other of the original SNPs, located in genes from the RAAS system, have been analyzed individually in the SILVHIA samples. The SILVHIA study was a multicenter study, and we are aware of the fact that additional genetic association studies of SNPs not included in our microarray panel have been performed in the SILVHIA samples, both regarding blood pressure reduction and reduction in left ventricular mass. We think, however that it is not reasonable to consider SNPs from previous studies when performing correction for multiple testing of the ten SNPs analyzed in the current study that focuses strictly on lipid-related genes. We have also added a final sentence to Results and Discussion stating that our results need to be replicated in another sample set to indicate that the study can be considered exploratory.

2. In response to the concern on the small size of the SILVHIA samples, we have added two sentences discussing the limitations set by a small sample size to the section in Results and Discussion on page 9.

3. In response to the third major suggestion by this referee, we have added the genotype frequencies for all tested SNPs in both treatment groups to Figure 1, instead of including a table with the allele frequencies of all tested SNPs. In adding this information to Figure 1, we followed the suggestion by the second referee.

Minor essential revisions:
1. In the 2nd paragraph of the Results and Discussion section we state that the pattern of the blood pressure response for the SNP C711T in the apolipoprotein B gene was similar
for both SBP and DBP in the irbesartan treatment group. Additionally, we state that the pattern is NOT similar for the SBP and DBP response for the SNP C16730T in the low density lipoprotein gene in the atenolol treatment group. The latter sentence has probably been misread by the reviewer.

2. The layout of Figure 1 has been changed so that the SBP and DBP data are within the same row for each SNP, which makes it easier to follow, as suggested by the reviewer.

3. As indicated by the reviewer, we had included Figure 2 to be able to include the genotype frequencies for the two genotype patterns being significantly associated with blood pressure reduction in a more readable font. The panels in Figure 1 have been enlarged, Figure 2 has been deleted and the genotype frequencies have instead been added to all panels in Figure 1, see point 3 in Major compulsory revisions.

**Discretionary revisions**

1. The Abstract has been revised by adding the number of subjects analysed and the doses for the two drugs. Additionally, the findings have been specified by adding the SNPs and their effect on blood pressure reduction.

2. As suggested, we have included the more recent reference Koopmans et al. 2003 to the first paragraph of the Background, in the sentence concerning markers for antihypertensive treatment.

3. One sentence and a reference on a suggested phenotypic effect of the APOB C711T SNP on the APOB protein have been added to the third paragraph of the Results and Discussion section. The C16730T SNP in the LDLR gene is synonymous, and no information has been found on SNPs in linkage disequilibrium with functional importance.

4. Information on the average reduction in mmHg of the subjects carrying the C-allele of the SNP APOB C711T is given in the second paragraph of section “Results and discussion”. We are hesitant about calculating the fraction of total blood pressure reduction that can be attributed to the identified SNPs, because such a calculation gives very high effects that seem unrealistic given the multifactorial nature of blood pressure response.

**Reviewer 2: Markku Savolainen**

**Major compulsory revisions**

The number of subjects of each genotype has been added for each SNP in Figure 1. See also point three of Major compulsory revisions for Reviewer 1.