Author's response to reviews

Title: Familial hypercholesterolemia in St.-Petersburg: the known and novel mutations found in the LDL receptor gene.

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Author’s response to reviews:

Dear Editors,

Thank you for careful reading of our manuscript (MS: 8639351504823171) and very helpful critical comments of the referees. Enclosed is the revised form of our paper "Familial hypercholesterolemia in St.-Petersburg: the known and novel mutations found in the LDL receptor gene". In the revised manuscript we have considered all the drawbacks pointed out by the referees and made changes in the text and figures according to their suggestions. Below, please, find the list of our answers to the referees' comments and modifications of the manuscript.

We hope that you will find our revised manuscript suitable for publication in the BMC Medical Genetics. We shall ready to answer to any questions concerning our manuscript, if they will arise.

Reviewer: Kimmo Kontula.

1. We have added several sentences about genes involved into monogenic hypercholesterolemias, namely ARH, PCSK9 and ABCG5/ABCG8 transporters into Introduction.

2. Phenotypic details of all the patients are summarized in Table 2, containing all lipid data of patients with specific mutations and without, i.e. the full data of patients recruited for the study.

3. Sentence about the necessity of expression studies to confirm the role of new-missense mutations D601N and C646S was added into Discussion.

4. Ethnic origin of patients was considered in the Discussion, when data were available.

5. Figure 2 was deleted.

6. Data of all previous mutations found in St.-Petersburg were listed in Table 3, helping to summarize these scattered data for English-reading Society. Also two sentences about distribution of LDL receptor gene
mutations in different regions of Russia and also giving total number of mutations in Russia (47) is given in the end of Discussion.

Reviewer: Anne Soutar.

1. Since we have used only slightly modified primer set suggested by Hobbs (1992) most of exon-intronic boundaries were not analyzed. We have stated now clearly that only 14 out of 34 intronic boundaries were analyzed what could lead to missing of some mutations in our FH group.

2. SSCP was considered not as the best method for identification of point mutations, despite in our hands and in the previous work of the Danish laboratory, the results of automatic SSCP analysis prior to sequencing and direct DNA sequencing gave similar results in recovering of the mutations. This item is considered in details in the text.

3. Large deletions were considered as a minor contributing factor to FH in St.-Petersburg population due to experimental work performed previously in St.-Petersburg (References included, e.g. Mandelshtam, 2003) and considerations of small contributions of this type of mutations in outbreed populations were extended.

4. PCSK9 gene was neither sequenced, nor searched for specific mutations in the current study what is now clearly stated. Due to European data one cannot expect of high impact of these mutation into FH morbidity. Because of many mutations in the LDL receptor gene an be still missed when using our mutation screening technology (see items 1 and 2), it is impossible to determine how many out of patients SURELY have NO mutations in he LDL receptor gene.

5. This was slipping, of course it was P518P, and not P548P. It was corrected with thanks.

6. Possible effect of mutations V806I and V776M was considered in Discussion, as suggested.

Minor revisions:

1. Word "invalid" was changed to another.

Discretionary Revisions:

1. Probable importance of linkage of I705 allele with other mutations for genesis of FH was considered.

2. Patients were selected in the Lipid Clinics of the Institute of Human Brain due to tradition of cooperation; we have not answered this item in the text, since it is of poor interest to the FH society.

We have also included several new References and thanks to the Reviewers into the manuscript.

Sincerely yours,

Faina M. Zakharova