Author's response to reviews

Title: Detection of allele specific difference of IL28B mRNA expression

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

BMC Medical Genetics

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Dear Editor,

Please find enclosed the manuscript of my paper entitled ‘Detection of allele specific difference in \textit{IL28B} mRNA expression” which we wish to submit for publication in BMC Medical Genetics.

This manuscript describes the results of my recent research project which we carry out within a European frame work 7 consortium “HepaCute” (grant number 26084) which aims to study the genetic factors involved in acute Hepatitis C virus (HCV) infection, with the help of our collaborating Egyptian research partners, for whom HCV infection is a major health problem with 10-15% of the Egyptian population being infected. As consortium partners at Imperial College we are responsible to contribute towards the genetics work package within the consortium. With our strong background in host genetics in HCV infection, we wanted to make a contribution towards a better understanding of the functional effect of the genetic polymorphism in the \textit{Interleukin-28} (\textit{IL28B}) gene, which has been reported to be associated with spontaneous as well as treatment induced clearance of HCV infection. But despite the use of \textit{IL28B} polymorphisms as predictive markers in clinical trials, little progress has been made towards allocating a function to these polymorphisms. Progress is hindered by the strong linkage disequilibrium of several markers in the area, making the identification of the functional polymorphism difficult, and by the 96% DNA sequence homology between \textit{IL28B} and the adjacent \textit{IL28A} gene, which makes it difficult to design \textit{IL28B} specific and \textit{IL28B} allele specific assays. We overcame this latter difficulty by establishing a relative quantification RT-PCR assay which is able to compare the relative levels of allele specific \textit{IL28B} transcript in cell lines heterozygous for the \textit{IL28B.rs4803217} polymorphism. Using this assay, we are able to detect a 2-fold increased expression of the C allele, which is in linkage disequilibrium with the C allele of \textit{IL28B.rs12979860} which is associated with clearance and treatment response in HCV infected patients. At the same time we detect variable ratios of C to T allele expression in 6 healthy donors, indicating that the allele specific expression varies between individuals. The data presented reveal that allele specific differences in \textit{IL28B} mRNA expression levels may at least in part contribute to the variable outcome and treatment
response of HCV infected patients, depending on their *IL28B* genotype. Our assay is a tool to further evaluate the allele specific expression level of the *IL28B* gene in patient cohorts.

I can confirm that this work has not previously been published, and that the manuscript has been seen and was approved by all listed authors.

Yours sincerely,

Susanne Knapp