Reviewer's report

Title: Genetic polymorphisms in MDR1, CYP3A4 and CYP3A5 genes and its impact on Ivermectin Therapy in Onchocerciasis

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Reviewer: Anne Slavotinek

Reviewer's report:

Major compulsory revisions:

The authors need to mention the limitations of their study - an extremely small sample size (21 suboptimal responders only), no exclusion of stratification of the different responder populations, and no replication is provided. In fact, the numbers are so small that it is still possible that there are host factors that are important. What power does the study have to detect a significant difference between genotypes with the low numbers?

The authors should make very clear any links between statistically significant genotypes and enzyme function in the discussion. If none of the statistically significant different genotypes have functional consequences, they should state this. Are any drug levels available in responders and non-responders?

How were responder and suboptimal responders defined?

You state that host factors are unlikely to be important in suboptimal response to Ivermectin in both the abstract and the discussion, and yet you point out a statistically significant difference between responder and suboptimal responder genotypes: “CYP3A5*1/CYP3A5*1 and CYP3A5*1/CYP3A5*3 genotypes were found to be significantly different for responders and SORs. Haplotype (*1/*1/*3/*1) was determined to be significantly different between responders and SORs”. How do you explain these results if you deem that there are no significant host factors?

“Some of these variants alleles are being reported for the first time among the Ghanaian population”. – this sentence is too loose – you need to state which variant alleles.

For tables 4 onwards, all p values need to be included with the data.

Table 1 primers have all been published and should be supplementary data; similarly Table 2 could also be a supplementary table.

Table 3: Are the genotypes combined for all study subjects in this table and are those for the responders and SORs listed separately in table 4? – this should be clarified.

At the start of the results section, you should redefine the subject population. How do your MDR1 and CYP frequencies in subjects compared with dbSNP?
Minor essential revisions:

There are numerous minor points and the manuscript needs further proof reading.

Background – of the MDR1 gene
energy dependent
where it acts at first level as a protective barrier
can you say more about the MDR1 haplotypes that have been associated with functional differences?

P4: which leads to an amino acid change
which leads to the amino acid change
Several SNPs have also been reported for the CYP3A5 gene, which impact enzyme function.

P5: SOR phenotypes observed by Awadzi and colleagues in the use of IVM for the control of onchocerciasis – this sentence needs a reference.

At the start of the results section, you should redefine the subject population. How do your MDR1 and CYP frequencies in subjects compared with dbSNP?

Haplotypes for MDR1 do not mean much if there is only one SNP with different alleles

Discussion

P9: loci 12, 21 and 26 – do you mean SNPs in exons 12, 21 and 26 rather than loci?

P9: “The CYP3A gene has been extensively studied in industrialised populations because of its involvement in metabolism of many pharmaceutical and recreational drugs. The CYP3A4 and CYP3A5 enzymes are the two predominant isoforms of the CYP3A subfamily expressed in the human liver and small intestine (14). Both CYP3A4 and CYP3A5 genes are polymorphic with variant alleles generally occurring at low frequencies in different ethnic groups.” – this is just repetition and could be omitted.

P11: The SORs were twice more as likely to be carriers of the 3435T variant allele.

The combined effect
in linkage disequilibrium with the…” and same for subsequent sentence