Author’s response to reviews

Title: A Prevalence Survey of Enteral Parasites in Preschool Children in the Mangochi District of Malawi

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Author’s response to reviews:

Dear Dr Nausch

Many thanks for your response and constructive feedback. We have taken time to scrutinise your comments and have amended the manuscript. In reference to the particular points which were raised please refer to our responses below:

- Authors refer to the MORDOR study, however, what samples were used for the present study remains unclear. Are participants included into the MORDOR study or analysed in addition (in particular since the MORDOR study protocol also includes tests groups comprising 40 children/village for extended analysis)? Even if participants were not directly part of the mentioned study, the treatment status might be unknown (as village status was not unblinded for the current study). Hence, the current study/manuscript needs an completely clear demarcation to the MORDOR study.

We have made changes to the manuscript to clarify that children were recruited as part of the MORDOR-Malawi study. Stool samples were collected from recruited children. Half of these stool sampled from each individual was used for analysis of helminth and protozoal carriage in this study, while the other half of the sample was frozen and analysed for microbiology and for AMR as part of the MORDOR study. Anthropometric data and haemoglobin measurements were used in both MORDOR-Malawi and this study’s analysis.
- Authors frequently argue/interpret that treatment has reduced the prevalence of helminths in the study area. However, they only refer vaguely to ongoing deworming programs. There is no information of previous treatments of participants in included villages. Hence some of the interpretations/conclusions may not be justified.

There is little available data on the treatment program up take or the availability of treatment at the time of the study in these specific villages. This is an issue which has been reported in previous helminth studies in Malawi. We have added a paragraph to clarify this and discuss its impact on our interpretations.

- Originally the study aimed to obtained samples/data from 320 children. However only 140 ended up being included. Here the question is, if a sample size calculation has been performed and if the final sample size actually is sufficient for aimed analyses – in particular for association between helminths and growth restrictions (since authors argue prevalence is low). The reason of excluding ‘50’ samples is also a little obscure.

The sample size calculations for estimating prevalence were based upon a predicted prevalence of 8%, such a prevalence was calculated to require 179 samples. The WHO recommends sample sizes of 200-250 to estimate prevalence in an ecologically homogenous environment. Based upon this information we attempted to obtain 200 samples. The 193 Samples collected were analysed for helminth prevalence all of which are reported in table 2. Due to issues with linking samples to collected anthropometric data, only 140 samples could be matched. As a result only 140 individuals were included in analysis of association with stunting. We have attempted to clarify this with a flow-diagram to clarify the reason for discrepancy in numbers used in analysis.

- A major limitation is also that only one stool sample/participant has been analysed – this critical for helminth diagnosis, in particular in groups where low infection levels are expected.

We have added a further paragraph to highlight this point more extensively, and to explain how the design attempted to mitigate this issue. We have explained the effect that this has on our results in relation to the true cohort prevalence. We have also explored how this compares to previous studies in Malawi where all published data has had this shortcoming.

- Some of the points mentioned also influence the interpretation of the results and conclusions which should be drown much more carefully.

In light of the above comments we have adjusted the discussion to reflect this.
We hope that you feel that these amendments are sufficient, if there is anything which lacks further clarity please do not hesitate to let us know.

Many thanks