Reviewer's report

Title: In vivo antimalarial activity of the crude leaf extract and solvent fractions of Croton macrostachyus (Euphorbiaceae) against Plasmodium berghei in mice

Version: 2 Date: 12 December 2013

Reviewer: Ronan Batista

Reviewer's report:

I found a manuscript that describes reasonable results and, for this reason, appears to be a literature that will attract limited interest from readers if accepted for publication. There is a serious problem concerning the methodology of fractionation used for the crude extract. Also, I often found a super estimated and not supported discussion from the authors concerning their results. In my opinion, this manuscript is not suitable for publication and deserves to be restructured and improved substantially by the authors before being reconsidered for publication.

1. Major Compulsory Revisions

1.1. Introduction should be substantially improved by presenting an exhaustive survey including additional and succinct information on chemical constitution and biological activities already described for Croton macrostachyus. I recommend that additional references are taken into account. Please consider the three suggestions below as a possible starting point.

1.2. The last phrase of the Introduction section is completely wrong. This manuscript only describes the evaluation of in vivo antimalarial activity for crude extract and fractions. It does not report any bioguided fractionation.

1.3. The methodology employed for “solvent fractionation” of the crude hydromethanolic extract does not make any sense and it is not supported by high standard chemical literature. The use of soxhlet apparatus is not appropriate to fractionate crude extracts. As the crude hydromethanolic extract was shown to exhibit the most interesting antimalarial activity at all, I recommend the authors to reformulate their manuscript by removing all methodology and results concerning chloroformic, methanolic and aqueous fractions, in order to describe only results concerning the crude extract.

1.4. In the Results section, item 3.3.1, the first phrase: why do the authors believe that the crude extract displayed a very good chemosuppressive activity against P. berghei? Which data comparison support this conclusion? The
majority of articles reporting the in vivo antimalarial activity of crude extracts usually describe the equivalent (or better) chemossuppression at lower dosis. Please comment.

1.5. In the Results section, item 3.3.1, the last phrase of the first paragraph: “… no apparent difference was noted in survival time between CM600 and CQ25.” I do not agree: Table 2 shows a very significant difference between these data.

1.6. In the Results section, item 3.4.2, the first phrase of the first paragraph: Why do the authors believe that chloroform fraction showed the highest antimalarial activity in the 4 day suppressive test? Results in Table 2 a different conclusion.

1.7. In the Discussion section, item 4, third paragraph: “The crude extract of Croton macrostachys (91%) appears to be more effective than other species of the same genus such as Croton zambesicus… 80% [34].” Again the authors seem to state a mistake. Ref. 34 shows that C. zambesicus presented a 80% chemossuppression at the dose of 200 mg/Kg. When the performance of C. macrostachys (91%) is considered, the dose of 600 mg/Kg should be taken into consideration. So, I do not see any reason to the authors consider their results as more effective than that observed for C. zambesicus.

1.8. Conclusions (item 5) are not supported by results. This section must be completely reformulated.

2. Minor Essential Revisions

2.1. Labels on Tables should be revised, corrected and extensively improved.

2.2. A thorough English revision is also necessary for the entire manuscript.

**Level of interest:** An article of limited interest

**Quality of written English:** Not suitable for publication unless extensively edited

**Statistical review:** Yes, but I do not feel adequately qualified to assess the statistics.

**Declaration of competing interests:**

I declare that I have no competing interests.