

## Response to reviewers' comments

Thank you and the reviewers for handling the manuscript (manuscript number: amt-2021-407). Responses to reviewers are in *italics*. The changes in the manuscript have been marked in **blue**. Please refer to the point-by-point response to the reviewers' comments and concerns.

Thank you again and the reviewers for such detailed suggestions for revision.

### **Comment on amt-2021-407**

#### **Anonymous Referee #2**

The authors have addressed most of my comments except my 1st major comment regarding the initial DTT concentration. I don't think the authors have understand it clearly. The authors' responses are focused on the changes of DTT concentrations over time while my comment is on the INITIAL DTT concentration. A previous study (Lin et al., 2019, see below) has shown that initial DTT concentration matters. Studies using different initial DTT concentrations should NOT be compared directly. The authors were using a different initial DTT concentration than Fang et al. (2015) and Puthussery et al. (2018) while made conclusions based on the comparison. The authors need to justify why they used a different concentration and be cautious about comparing their results to other studies.

***Response:** Thanks for the question, the initial concentration of DTT in our experiment is different from that of Fang et al. (2015) and Puthussery et al. (2018). The study by Lin et al. found that the initial concentration of DTT has an effect on the rate of DTT consumption, and our comparison may be affected by the initial concentration. Therefore, direct comparisons with Fang et al. (2015) and Puthussery et al. (2018) have been removed in the paper. In addition, the condition that the initial concentration needs to be considered is added in the paper. However, in this experiment, the DTT concentration is always kept at 1mM, so it does not affect the judgment of the daily change of ROS content. In addition, due to the limitations of the instrument and sample concentration, we adopted a DTT concentration of 1 mM. In our experiment, except for the initial concentration and volume of addition of DTT, the concentrations and volumes of addition of other substances are also different. We are similar to the method of Cho et al. by calculating the final concentration after the potassium phosphate buffer mixed solution. We add a new text to line 150 in the revised manuscript:*

*“To adapt to the experimental process, the concentration and addition volume of each substance are changed in the experiment, but the concentration of each substance in the mixed solution is similar to that of Cho et al. (Cho et al., 2005). Lin et al. found that the initial DTT concentration will affect the final DTT<sub>v</sub> value (Lin et al., 2019). During the experiment, the initial DTT concentration was always kept at 1 mM, so it did not affect the judgment of the daily change of ROS content.”*