Author's response to reviews

Title: Effect of Ambrotose AO(R) on resting and exercise-induced antioxidant capacity and oxidative stress in healthy adults

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Author's response to reviews: see over
Reviewer’s report 1

Title: Effect of Ambrotose AO(R) on resting and exercise-induced antioxidant capacity and oxidative stress in healthy adults

GENERAL COMMENTS
This paper addresses great information about an important issue. Overall, I think that it is well written and it should be published. However, I have some comments to make.

We thank the reviewer for the detailed and constructive comments. Changes are highlighted in yellow throughout the manuscript and our responses are below each comment.

SPECIFIC COMMENTS
There are some minor essential revisions to be made

BACKGROUND
The authors state that the use of supplemental antioxidant nutrient intake in order to minimize oxidative stress levels is controversial. Could the reasons for this affirmation be explained?

Yes, we now provide additional information related to this statement.

Please, provide more information about the unpublished study, such as where or who carried it out.

One of these studies has since been published. We now provide the reference data.

Some information is missing concerning the reasons that justify the type of physical exercise chosen for the research as a stressor. In this line, it might be useful to explain a little bit more the relationship between aerobic and anaerobic exercise and why it was decided to carry out a progressive test.

Excellent point. We have now included this information as requested.

METHODS
It is said that prior to the start of the study, trained subjects were asked to engage in regular exercise for a minimum of 4 hours per week. According to Table 1, these people were trained enough, so why was this necessary? Please, describe in detail, if possible, the training program carried out (how many weeks it lasted, total volume, modality of exercise performed, training supervision...).
We apologize for the miscommunication. What we meant was that subjects were already exercising regularly on their own before being recruited to participate. Table 1 shows the amount of exercise they were engaged in. We did not instruct them to start exercising prior to beginning the study. We have attempted to clarify our writing related to this.

From my point of view, free weight resistance training, although it is mainly an anaerobic exercise, can be aerobic too, depending on the total amount of weight, resting time between sets and repetitions or execution speed. Because of that, I think that this affirmation should be revised.

We agree to some extent. Depending on rest interval length and intensity level, it is possible to gain an aerobic benefit from resistance exercise. We have attempted to clarify this statement.

Regarding the sentence “Please see Table 1”, there is no need to use the word “Please”.

Corrected as suggested.

I would introduce a “measurements” subsection, in order to describe how all the measurements were taken as well as the tools used (questionnaires, heart-rate monitor..) and I think that even the explanation of the GXT should be written here too.

Done as suggested. Sections have been re-ordered appropriately.

Although it can be obvious for most of the readers, it could be appropriate to explain the objective of the physical test and what the percentage of each interval means.

Understood and modified as suggested to provide some general idea of the effort involved in the test stages. This obviously varies by subjects, but the info now provided in the text is accurate for most subjects.

“Exercise Test Data”
I do not understand the following paragraph “No condition differences were noted for GXT time to exhaustion (p>0.05). However, a difference was noted between men and women and between trained and untrained subjects (p<0.05). If there were no differences in time to exhaustion, this means that trained people and untrained people showed the same anaerobic performance.” Please, explain this data again.

The “condition” refers to placebo or supplement. There was indeed a difference between trained and untrained subjects, as we have indicated in the paragraph.
above. There was simply no difference between placebo and supplement (when pooling all trained and untrained subjects together).

There is no need to write (\(p>0.05\)) regarding the absence of significant differences.

*We understood and believe this is a matter of personal preference. However, we prefer to leave this as is, knowing that some statisticians prefer to see this written as we have it. Thank you for understanding.*

DISCUSSION
I advise the authors to include their comments for future studies at the end of this section, rather than in the conclusion.

*Understood and corrected as suggested.*

CONCLUSION
This section is too long, it must be shortened.

*This has been done.*

TABLES AND FIGURES
There are too many tables and figures. If possible, the authors should find a way to reduce them down to 5-6 each.

*We understand the concern here and we have reduced the number of figures by using multi-panel figures (from 11 figures to 5 figures). However, we would like to retain all 6 tables, as we believe this is important information to include and allows for a complete presentation of all results. We spent considerable time and expense to collect these data and would like to present all. Moreover, Nutrition Journal is published online and page restrictions are not a concern. Thank you for understanding.*
Reviewer's report 2

Title: Effect of Ambrotose AO(R) on resting and exercise-induced antioxidant capacity and oxidative stress in healthy adults

We thank the reviewer for the detailed and constructive comments. Changes are highlighted in green throughout the manuscript and our responses are below each comment.

Reviewer's report:

Major Compulsory Revision
1 - I would want to have you increase information on the exercise protocol (did you calculate the relationship between work rate and double product, before and after the period of supplementation?)

As requested, we included more information on the exercise test (much of this is in yellow text). However, we did not calculate double product during exercise. The purpose of the exercise test was to increase ROS production and to determine if antioxidant supplementation could attenuate this rise in ROS. We were not concerned with the hemodynamic response to exercise in relation to the antioxidant intake. However, this may have been of interest.

Discretionary Revision
1 - It would have been interesting to dose the specific markers of oxidative stress (enzymes and protein carbonilation) instead of total antioxidant capacity of serum.

You are correct and this may be the focus of future work related to this blended antioxidant. In this study we simply focused on a more global marker of antioxidant status, basing our design on prior work using this same antioxidant to increase ORAC. We mentioned your suggestion within the discussion.

2 - It would have been interesting to use an exercise protocol including the calculation of VO2 max.

This is true, however we are unaware of many studies reporting an increase in VO2max following antioxidant supplementation. As with double product, our failure to measure VO2max may be considered a limitation of this work. The purpose of the exercise test was to increase ROS production and to determine if antioxidant supplementation could attenuate this rise in ROS. We did not expect to observe an increase in VO2max.