To: The Editor-in-Chief, Date: 25 September 2023

Entomologia Experimentalis et Applicata

Subject: Revised manuscript submission

Dear Professor Beukeboom, dear Leo

We are pleased to submit the revised version of our manuscript titled “Effects of frass from black soldier fly (*Hermetia illucens*) larvae and yellow mealworms (*Tenebrio molitor*) on growth and resistance to insect herbivores of field mustard (*Brassica rapa*): differences between insect species and frass treatments, following a decision of “minor revision”.

We appreciate the insightful evaluation provided by the reviewers, and we are encouraged by their positive feedback on our work, acknowledging our strong grasp of the research topic and the uniqueness of our idea. We have carefully addressed the reviewers' comments to improve the manuscript's readability and align it with scientific writing and journal standards.

Below, please find our detailed responses to the reviewers' comments and additional comments by the Editor.

We hope that with this revision the manuscript meets the publication standard in Entomologia Experimentalis et Applicata.

Thank you for your consideration!

Sincerely,

Shaphan Chia, Joop van Loon and Marcel Dicke

**RESPONSE TO REVIEWER 1 COMMENTS**

GENERAL ASPECTS:

**Reviewer comment:**  
- Both the Introduction and the Discussion manifest an excellent domain of the research topic.  
- A very interesting, novel and original document. Congratulations for the great work done.  
**Author response:**

Thank you very much for your kind words and positive feedback. We greatly appreciate your assessment of our work.

KEYWORDS:

**Reviewer comment:**  
- Reduce the number of keywords by half. Now it is excessive.

**Author response:**

We have reduced the number of keywords as advised.  
  
ABSTRACT:

**Reviewer comment:**  
- Very well explained and complete.

**Author response:**

Thank you

INTRODUCTION:

**Reviewer comment:**  
- L61: why are only bacteria considered and not also fungi?

**Author response:**

We have added information about fungal stimulation through frass application

**Reviewer comment:**

- L64-73: Rewriting information. It's confusing right now.

**Author response:**

We have simplified the text by breaking down complex sentences and providing a clear flow of information. The revised text emphasizes the relationship between frass, soil, beneficial microorganisms, and plant resistance to insect herbivores.

**Reviewer comment:**  
- L74-75: order cites conologically.

**Author response:**

The current format adheres to alphabetical ordering of authors' names, which is consistent with the manuscript's style throughout.

**Reviewer comment:**  
- L94-96: add updated production data according to FAOSTAT.

**Author response:**

We have updated the production data for field mustard in the paragraph, using the latest available information.

**Reviewer comment:**

- L98-99: repeated citation.

**Author response:**

We have removed the repeated citation on lines 98-99 as suggested.

MATERIALS AND METHODS:

**Reviewer comment:**

- L139, 149, 150, 170, 171, 179, 180, 186, 187, 188 y 244: The full scientific name has already been written above. Reduce.

**Author response:**

We have abbreviated the scientific names consistently throughout the manuscript to reduce redundancy and align with best practices in scientific writing.

**Reviewer comment:**

- L142-143: Is the chemical analysis of the soil known? The nutritional one would be very interesting and could be added to the paper as supplementary material.

**Author response:**

We appreciate your observation and suggestion. We have cited a previous study for the soil's physical and chemical properties in our revised manuscript.  
  
RESULTS:  
**Reviewer comment:**

- L332, 366, 384, 393, 406, 411, 418, 427  and inside the figures: The full scientific name has already been written above. Reduce.

**Author response:**

We have removed the redundancy by omitting the full scientific names in the specified locations, as they were previously mentioned above.

**Reviewer comment:**

- Throughout the section, it is not considered necessary to put so much information from the statistical analysis in the text. However, it is very positive to put information of mean data between different treatments, which help the fluent understanding of the text and the study.

**Author response:**

We have reduced the statistical information in the text and, emphasized data visualization through figures. This should help streamline the text and enhance the reader's understanding of the study.

**Reviewer comment:**

- L468-469: spacing problem.

**Author response:**

Thank you for your observation. We have corrected the spacing problem.  
  
DISCUSSION:  
**Reviewer comment:**

- L538-539: The full scientific name has already been written above. Reduce.

**Author response:**

We have abbreviated the full scientific name for conciseness and clarity.

**Reviewer comment:**  
- L526-527: scientific names in italics.

**Author response:**

Done. Revised text (L526-527): *Clostridium perfringens*

**RESPONSE TO REVIEWER 2 COMMENTS**

**Reviewer comment:**  
Despite the amount of work that has been done already in the case of insect frass, the overall idea is unique and the methodological approach very interesting.

The ms can be accepted for publication, but there are some parts that need to be revised.

**Author response:**

Thank you for your positive feedback and enthusiasm about our manuscript. We have carefully reviewed your comments and have made the necessary revisions to address the identified issues.

**Reviewer comment:**  
97-8 and elsewhere. Use authorities in full in the first time that a scientific name appears on the ms.

**Author response:**

We have now used authorities in full for all scientific names throughout the manuscript at the first instance a scientific name was used.

**Reviewer comment:**  
109. There are additional data gaps that are not addressed by Wantulla et al. (2022), which can be illustrated here.

**Author response:**

We acknowledge the presence of additional knowledge gaps beyond the scope of Wantulla et al. (2022) that we have identified and addressed in our study. These gaps are further elucidated in the manuscript.

**Reviewer comment:**  
133. Expand this general scope with more text.

**Author response:**

We appreciate the suggestion for expansion, but we would like to request clarification regarding the specific section or content the reviewer is referring to. Please provide additional details so that we can address this comment effectively.

**Reviewer comment:**  
Not clear how many replicates and subreplicates were used, clarify.

**Author response:**

It is not clear what section the reviewer refers to. Yet, in general, we used ten plants per treatment and control to assess the effect of raw-frass-exposed plants on the performance of *Plutella xylostella* larvae. This is mentioned in the Methods section. Number of replicates are indicated in the figures as well.

**Reviewer comment:**

524. This is the standard procedure of the authorization of frass, regardless of the overall effects of the thermal treatments. This can be further clarified.

**Author response:**

Our samples underwent extended oven drying, potentially impacting the population of beneficial microbes, which differs from the standard procedure for authorizing frass.

**Reviewer comment:**  
531-3. Why- perhaps due to their increased conductance?

**Author response:**

We appreciate the reviewer's question. We have expanded the discussion to address the potential reasons behind the negative effects observed in *B. rapa* when exposed to raw BSFF or MWF. These effects are likely due to a combination of factors, including the salinity of the frass, which we have discussed in the revised manuscript.

**Reviewer comment:**

552. Merge with the next paragraph.

**Author response:**

We have merged lines 552-554 with the following paragraph as suggested.

**Reviewer comment:**

607. No need to have a separate conclusion section, use this text as the closing paragraph in the discussion.

**Author response:**

As recommended, we have eliminated the heading 'Conclusion' and integrated the text as the closing paragraph in the discussion section.

**RESPONSE TO ADDITIONAL COMMENTS BY THE EDITOR**

**Reviewer comment:**

I will add a few comments to those of the reviewers:

**Author response:**

Thank you for your additional comments on our manuscript. We appreciate your feedback and have addressed these new comments along with the ones provided by the two reviewers in our revised manuscript.

**Reviewer comment:**

Line 44, use of the word “protective” here creates ambiguity Protective might imply to some readers that plants were protected from herbivores under this treatment, which was not the case.

**Author response:**

We appreciate your observation. We have now revised the sentence to ensure that readers do not misinterpret the intended meaning of our statement. Specifically, we have eliminated the word "protective" to accurately convey that the herbivores exhibited enhanced performance under this treatment.

**Reviewer comment:**

Line 65, move the definition of PGPR to line 61, after first mention?  I think this would make the paragraph flow better.

**Author response:**

Thank you for your suggestion. We have moved the definition of PGPR to enhance the flow of the paragraph.

**Reviewer comment:**

Line 122, delete comma.

**Author response:**

Thank you for your suggestion. We have deleted the comma as suggested.

**Reviewer comment:**

Lines 142-143, it would be extremely helpful to know something about the soil (e.g., pH, organic content, N, P, K levels, soil type) used in the experiments.

**Author response:**

We appreciate your observation and suggestion. We have cited a previous study for the soil's physical and chemical properties in our revised manuscript.

**Reviewer comment:**

Line 202, :”after” instead of “since”; insert “and” after the comma.

**Author response:**

We have replaced the word “since” with “after”, as suggested.

**Reviewer comment:**

Line 211, delete comma.

**Author response:**

We have deleted the comma as suggested.

**Reviewer comment:**

Figure 2, in the third and fourth panels, the letters designating the Tukey results appear to be in error – shouldn’t BSFF be accompanied by a “b” and both “NoFrass” and “MWF” have “a”’s?.

**Author response:**

You are right. We have corrected this error, ensuring that BSFF is now accompanied by a "b," while both "NoFrass" and "MWF" have "a"s. Thank you for pointing this out. The conclusion remains unchanged.

**Reviewer comment:**

Line 382, were patterns similar in second experiment.

**Author response:**

Yes, the patterns were similar. In the revised sentence, we clarified that there were no significant differences among treatments.

**Reviewer comment:**

Line 416, were patterns similar in the two experiments?

**Author response:**

Yes, the patterns were similar. In the revised sentence, we have now clarified that there were no significant differences among treatments.

**Reviewer comment:**

Lines 514-515 – again, any information about the base soil used in the experiments would be helpful – also, 2 g frass per kg of soil does not seem to be an inordinate amount to me – how feasible is it that the frass led to soil compaction or waterlogging?

**Author response:**

We acknowledge that 2 g frass per kg of soil may not seem excessive. While we agree that it is unlikely to cause soil compaction or waterlogging, we cannot rule out these possibilities, as they are mentioned in the literature, although we did not measure them specifically in our study.

**Reviewer comment:**  
Line 525, when what was “introduced to the frass”? Unclear.

**Author response:**

We apologize for the confusion. In the revised manuscript, we have clarified the paragraph to specify that heat treatment eliminated detectable amounts of pathogens in frass.

**Reviewer comment:**

Line 533, italicize species name.

**Author response:**

We have italicised the species name as suggested.