Response to Reviews: Manuscript ID JIS-2019-0327 entitled "Investigating behavior of the potato psyllid *Bactericera cockerelli* (Šulc) (Hemiptera: Triozidae) on three potato genotypes with putative resistance to “*Candidatus* Liberibacter solanacearum”"

**Reviewer: 1**

Comments to the Author

Thank you for submitting this manuscript, which contributes to an area of study that is both complex and globally important. Some minor suggestions have been included in the attached file.

It would be interesting to know if the genotypes studied here differed in physical morphology, both at a whole plant and at a leaf surface level.

***Response from the authors to Reviewer 1:***

**Thank you for your help reviewing the article. I have addressed inconsistencies in my usage of resistance, tolerance and susceptibility in the paper. The A07781 family exhibits tolerance: they exhibit high tolerance and low susceptibility to *Candidatus* Liberibacter solanacearum. I made the other minor edits as well. Regarding your question about physical morphology: There were no apparent differences in morphology between these plants, therefore morphological differences were not compared. I added a section in the discussion to address the need for additional study in this area.**

**Reviewer: 2**

Comments to the Author

Review of ‘Investigating behavior of the potato psyllid *Bactericera cockerelli* (Sulc) (Hemiptera:Triozidae) on three potato genotypes with putative resistance to “*Candidatus* Liberibacter solanacearum”

By Fife et al.

Varietal resistance to arthropod pests is an area that involves time and more so in case involving insect vectors of plant diseases. The authors have studied mechanisms of host plant resistance of potato psyllid on putatively Lso-resistant breeding clones derived from *Solanum chacoense* Bitter. It is rather interesting to note that resistance is primarily to the pathogen and not observed on psyllid settling behavior.

Manuscript is well written and deserves merit. Few clarifications are sought and some typos are also listed.

Objectives and scope are not clearly defined. Authors should mention the purpose of this study and how this study will be helpful in furthering the research on potato psyllid and Lso?

Some essential details of the research methods are found to be missing as in line 230. In ovipositional assays, how many pots were maintained within the PVC-framed cages and it is not clear how many such cages were maintained? ‘Plants were arranged in a randomized complete block in rows of four and placed inside mesh-covered PVC-framed cages’. How was this blocked? Did cage serve as block? I do not see a blocking variable involved in which case it should be designed as CRD.

61 – remove ‘to’

119 – ‘exhibits high tolerance and low susceptibility to Lso’ – high tolerance and low susceptibility, both mean the same.

152 – 154 When were psyllids tested for Lso – how long before start of the experiment?

157 – Forty adult psyllids

159- 185 There is no need to repeat the DNA extraction procedure. These lines may be removed.

266 – remove ‘the’

270 – this line appears to contradict the previous line. Please modify.

327 – ‘psyllid probing and feeding behavior’ Psyllid probing cannot lead to Lso transmission unless they hit phloem, it is when feeding begins. Probing cannot be eliminated, whereas only feeding can be inhibited by introducing resistant/tolerant genotypes.

338 – In the present study,

386 – ‘potato psyllids may require multiple mates’ It is not clear if the authors meant mating multiple times or mating with several males? There are evidence that with a single male and female each female psyllid can lay >1000 eggs in multiple matings. Based on discussion, it appears that experiments could be better planned.

***Response from the authors to Reviewer 2:***

**I appreciate your help with reviewing my article. I reworded some of introduction to clarify the purpose of these experiments: to test if the tolerance of the A07781 family was due to antibiotic/antixenotic effects of these plants on psyllid feeding. Video recordings were used due to a lack of available resources and as a point of comparison of techniques. The next set of experiments would be to compare video recordings to EPG analysis in order to see how well the different behavioral recording methods correlate with host acceptance. I corrected the methods section to clarify that each cage contained 4 rows of the different genotypes and clarified locations of the pots within each PVC framed cage. I mention that each PVC framed cage was considered a blocking unit for statistical analysis. I attempted to correct and clarify the other points mentioned as well. Thank you for your time and assistance.**

**Reviewer: 3**

Comments to the Author

This manuscript reports the results of experiments to quantify the initial host acceptance/rejection behaviors of potato psyllids in response to susceptible and putatively resistant potato germplasm. Several lines of new germplasm are compared to a common susceptible cultivar (Russet Burbank). The goal of the experiments is to determine whether probing/feeding behavior by the psyllids is altered, thereby reducing the exposure risk for inoculation of the putative causal agent of Zebra chip disease. The authors also explore longer-term psyllid-plant interactions using oviposition assays during which each female only receives nutrition from one of the experimental genotypes or the control. The manuscript is well-written and contains appropriate literature. Some issues arise in the discussion, namely that much of this section is spent justifying use of certain methods that do not allow conclusions to be drawn conclusively.

Examples:

Authors immediately point out downsides of using the video recording method. What are the up-sides beyond expense? What other aspects of this method are user friendly? How did this method help you to focus on the question at hand and avoid tedious EPG analysis? How can these methods be useful for other researchers?

Authors mention “limited observations” of longer-term recordings. Monitoring video recordings long-term is just as tedious as EPG analysis. Is there a way to more quantitatively demonstrate that what you observe during the first five minutes of the assay is highly correlated with later behaviors – i.e., it would seem prudent to provide good evidence that your five minute recording is an adequate proxy for being able to settle and feed in the phloem on the host. Otherwise, it is difficult to draw conclusions about exposure risk for Lso inoculation for the different genotypes.

Authors discuss volatiles, but assays did not discriminate selection based on volatile vs. contact cues. Discussion of volatiles does not seem warranted as this was not tested explicitly. At best, volatiles could be discussed as a component of the spectrum of contact/taste cues being experienced. There is not very good evidence that volatiles strongly drive hemipteran probing during the first five minutes, at least with aphids, which probe any surface by reflex upon tarsal contact. Do psyllids do similarly?

Authors mention that “plants were all uninfected” in line 341. They were clearly uninfected when used in the assays, and that status is not likely to change. However, the oviposition plants could have been exposed to sufficient Lso to become infected during the period of time each female was on the plant. Were these plants tested for Lso infection? This seems like additional data that should have been collected to help differentiate between resistance to Lso via reduced exposure (psyllid feeding difficulty) vs. resistance to Lso pathogen via direct pathogen suppression. It is naturally a good set up for this because the age and duration of feeding by a single psyllid is tightly controlled.

Authors should clearly state if the cited papers provide evidence that multiple matings are needed for full fertility. Says now “it is possible” and this forces the reader to go consult those references. This is a yes or no question, so please state whether the cited references found that multiple matings are or are not required (line 387).

Was survival measured for the psyllid females during the fertility assays? These data are potentially important if antibiosis is functioning and has an effect on ability of psyllids to survive on putatively resistant germplasm.

The discussion should be revised to focus on what the authors can reliably extrapolate about the resistant germplasm, with less time spent on “what ifs.” The experiments are sounds and well-executed. Tell the reader why your approach is adequate and consider whether you are reporting all the relevant data collected during these experiments (e.g., plant infection rates, insect survival). The video recording approach may also be useful for other researchers. Other literature is cited regarding this technique (“similar studies” on line 309) but details are not given. Have other researchers tested how this technique relates to later behaviors? How long did they run their experiments? The discussion should convey confidence in the approach and conclusions. Please also provide more detail on the next experiments to be pursued – i.e., is one genotype more promising? What are the next experiments?

***Response from the authors to Reviewer 3:***

**I addressed the benefits of video recording more in depth and added detail to explain the nature of the overnight recordings. The overnight recording were brief preliminary tests to justify the duration of recording. During these recordings, we saw that the psyllids did not move very far from their initial placement on the plant. The psyllids were allowed free access to the potato for a 12-hour period, yet they did not even leave the leaf of the A07781 genotype. There was greater initial activity when the psyllid was first placed on the plant, which is why we chose the 5-minute time period, a length where we expected to observe most of the expected behaviors. We were expecting more obvious rejection of the A07781 lines, which did not occur. I eliminated the volatiles discussion. We did not test volatile attraction in these psyllids. We did not test the plants used for the oviposition assays for Lso, which now appears to be a regrettable oversight. I added the conclusions of the references about multiple matings in the discussion. Survival was the same for females across all genotypes, which has been noted in the results and discussion. The ‘similar studies’ referenced were similar in that they observed the same targeted behaviors, but this is likely the first time video recordings have been used to observe the feeding behaviors of this psyllid. I edited the concluding remarks to reflect more confidence in our methods and to suggest future directions for research. Thank you for your helpful suggestions.**