**Editorial comments:**  
Changes to be made by the Author(s):  
1. Please take this opportunity to thoroughly proofread the manuscript to ensure that there are no spelling or grammar issues.

We proofread and revised the manuscript as instructed.

2. Please upload each Figure individually to your Editorial Manager account as a .png, .tiff, .svg, .eps, .psd, or .ai file.

We uploaded all figures as 300-dpi .png files as instructed.

3. Please upload each Table individually to your Editorial Manager account as an .xls or .xlsx file.

We converted Tables into separated .xlsx files and uploaded as instructed.

4. Please remove the titles and Figure Legends from the uploaded figures. The information provided in the Figure Legends after the Representative Results is sufficient.

We removed figure title and legends from uploaded figures.   
  
5. Please shorten the figure legends. The Discussion of the Figures should be placed in the Representative Results. Details of the methodology should not be in the Figure Legends, but rather the Protocol.

We revised figure legends to be more concise, as instructed. If the figure is self-explanatory (e.g., Figure 1), we removed the legend entirely and instead added references in the appropriate places in the Protocol.   
  
6. The current Abstract is over the 150-300 word limit. Please shorten it.

The Abstract in the original submission contained 292 words. We have further shortened it to 266 words in this revision.

7. Please define all abbreviations before use.

We spelled out all abbreviations at their first appearance in the revision.

8. Please remove all commercial language from your manuscript and use generic terms instead. All commercial products should be sufficiently referenced in the Table of Materials and Reagents. For example: Osmocote, Scotts Miracle-Gro Co., MEGAquick-spin, iNtRON biotechnology, Gateway cloning technology, Invitrogen Co., Zymo Research, Gateway LR ClonaseII, USA Scientific, MicroPulser, BIO-RAD, Silwet L-77, etc.

We replaced all commercial product names with generic terms as instructed. We added to the information on generic terms and their respective commercial products used in to the experiment, to the “List of Materials”. Where the generic term of a commercial product was used in the revised text, we added references to the list of materials as [see Materials].

9. Please simplify the Protocol so that individual steps contain only 2-3 actions per step and a maximum of 4 sentences per step. Please revise 2.6, 4.1, etc. accordingly.

We revised the text as instructed.

10. 2.7: Please write the text in the imperative tense. Any text that cannot be written in the imperative tense may be added as a “Note.”

We revised the text as instructed.

11. Please add more details to your protocol steps. Please ensure you answer the “how” question, i.e., how is the step performed? Alternatively, add references to published material specifying how to perform the protocol action. Please revise 2.1, 6.5.3-6.5.5 accordingly.

We revised the text as instructed, by adding references or providing more details.

12. 2.1: Please provide more details or add a reference for gel extraction of PCR product.

We added the information on the gel extraction kit, as well as a couple references.

13. 6.5.3: Please list PCR conditions and primers.

We added PCR conditions and primer sequences for the amplification of the *bar* gene.

14. Please reference Figure 1 in the Protocol section.   
We added references to Figure 1, as notes to the step 1.3.2 and 5.1.

15. Please reference Table 1 in the Protocol section for composition of growth media and infiltration solution.

We added references to Table 1 at steps 2.3 and 4.3.

16. There is a 2.75 page limit for filmable content. Please highlight 2.75 pages or less of the Protocol (including headings and spacing) that identifies the essential steps of the protocol for the video, i.e., the steps that should be visualized to tell the most cohesive story of the Protocol. Remember that non-highlighted Protocol steps will remain in the manuscript, and therefore will still be available to the reader.

We highlighted from the step 4 to step 6.4.3 by adding a gray shade to the text. We believe these steps are crucial because they contain the necessary modifications from conventional flower-dipping methods used for *Arabidopsis thaliana*.

17. Please ensure that the highlighted steps form a cohesive narrative with a logical flow from one highlighted step to the next. Please highlight complete sentences (not parts of sentences). Please ensure that the highlighted part of the step includes at least one action that is written in imperative tense.

Please see our response to the comment #16.

18. Discussion: Please also discuss any limitations of the technique.

The transformation efficiency lower than the most popular model plant *Arabidopsis thaliana* (ecotype Col-0) can be considered as a limitation. We added a more detailed discussion on this point.

19. References: Please do not abbreviate journal titles.

We revised the references as instructed.

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**Reviewers' comments:**  
**Reviewer #1:**  
Manuscript Summary:  
This submission describes a plant transformation protocol that is suitable for use with Schrenkiella parvula, a highly stress tolerant extremophile relative of the far less stress tolerant genetic model plant, Arabidopsis thaliana. To fully exploit a species as a genetic model, it is essential to have a way of manipulating genes to validate their role(s) in generating a particular phenotypic response. An impediment to exploiting S. parvula has been the virtual absence of a transformation protocol. One commonly used approach that works well for Arabidopsis involved dipping flowers into a solution containing Agrobacterium carrying a cloned gene of interest. As the authors point out, not only has the conventional approach for transforming S. parvula been problematic, the plant continues to flower after the dipping is done and the screening for successfully transformed T0 plants is complicated by a screening strategy that leaves many false positives among very few true transformants. These are all deterrents to using S. parvula for finding genes associated with stress tolerance despite the extraordinary physiological stress tolerance features that this plant displays and the excellent genomic tools available for its study. The solutions to these concerns that this protocol provides make sense and are laid out clearly: multiple dipping treatments works well with a plant that is difficult to transform and has indeterminate flowering (and more chances to generate transgenic seeds) and considering the narrow leaves and wax as impediments to the penetration of a selection agent are factors that should be obvious…but no doubt also easily overlooked. The protocol will offer suggestions to those working with other extremophytes that suffer from poor transformation and/or selection efficiencies and this should increase the research promise of other plants highly adapted to environmental stress.  
The protocol is well written, the authors highlight steps that require extra precautions and they provide reasons why following the precautions yields better results. The title describes the contents of the manuscript well. It might be advantageous for the authors to include a former name in the keywords, for example, as changes in the taxonomy for several extremophytes has made for confusion in the literature. The illustrations are also clear and support the text well.  
Thank you for the comments. Per reviewer’s suggestion, we included two former names ofthe extremophyte, *Eutrema parvulum* and *Thellungiella parvula*, among the keywords.

Major Concerns:  
No major concerns. The protocol is detailed and the results are described well. Although a recovery of 3 or 4 transformants from 10,000 seeds may seem problematic, this low recovery approximates the rates obtained for some Arabidopsis ecotypes that are also considered recalcitrant to transformation (eg. C24). It may well be that some of the floral features that make S. parvula resistant to transformation could be some of the same features that allows the plant to resist extreme stresses. However, that consideration cannot be addressed without having at least some means of generating transgenics in the first place. Finally, this submission also provides an important benchmark against which researchers can compare differences related to the use of other strains/plasmids/treatment conditions to devise an improved recovery of transformants in the future.  
  
Minor Concerns:  
There is a lot of good information and detail on how to propagate healthy plants. In that regard, it would be useful if the authors named the soil they used in the text as it only seems to be named in the list of supplies. Can any soil be used equally well or is the one given the only suitable substrate?  
We appreciate the reviewer’s comment. However, we were advised not to use commercial names in the main text, hence the reference to the exact soil mix was only in the list of supplies. In our experience, any soil mix where *Arabidopsis thaliana* can grow will also work for *S. parvula*. We added this information as a note to the step 1.3.1  
  
**…**

**Reviewer #2:**  
Manuscript Summary:  
The manuscript by Dassanayake et al. focuses on a transformation protocol for Schrenkiella parvula, a halophytic relative of Arabidopsis thaliana. Along with the related Eutrema salsugineum, S. parvula has been developed as a powerful tool for comparative physiological, biochemical and molecular analysis of adaptive mechanisms that allow plants to cope highly saline conditions. Such are the importance of these two halophytes, that they have been declared as new model organisms (Zhu et al., 2015, The next top models. Cell 163: 18-20). The development of functional genomics tools and protocols is essential to further the development of these model halophytes, and therefore this manuscript is not just another transformation protocol for an extra plant species but a critical platform to further our understanding of salt tolerance mechanisms in halophytes.  
Overall the protocol described in the manuscript can be clearly followed and the figures nicely augment the written protocol to provide an excellent visual aid as well as confirming transformation.  
  
Major Concerns:  
None.  
  
Minor Concerns:  
1. My only question is whether PCR of the Bar gene alone is sufficient to prevent false positives, as plants can sometime be transformed with the selective marker but not with the target gene (or the target gene can suffer deletions/insertion etc). Would it not be wise to add that PCR with target gene-specific primers should be performed on the plants exhibiting the presence of the Bar gene?

We appreciate the reviewer’s comment. We have also confirmed our transformants using one primer from the Bar gene and another from the target sequences. We added this as a recommendation in the note to the step 6.5.3 in the revised manuscript.

2. There are numerous English grammatical mistakes and erroneous use of terminology. While I understand that the manuscript will be copy-edited, I want to point out the terminological errors:  
(a) Lines 140, 181 and 190: Bacteria are not "poured" onto plates or "dissolved" in liquid media. The plates or liquid are "inoculated" with bacteria.  
(b) Lines 197-198: Bacterial pellets are not "dissolved" in a solution. They are "re-suspended".

We thank the reviewer for pointing out these. We have revised those two points as well as the entire procedures to replace casual languages to more appropriate scientific terms.