Reviewer Report

Title: **Arabidopsis phenotyping through Geometric Morphometrics**

Version: **Original Submission** **Date:** 12/13/2017

Reviewer name: **Yoland Savriama**

Reviewer Comments to Author:

Reviewer's report
Title: Arabidopsis phenotyping through Geometric Morphometrics
Authors: Carlos A. Manacorda and Sebastian Asurmendi
Date: 13th December 2017

Review:
This work proposes a complete analytical approach for phenotyping leaf variation in Arabidopsis using geometric morphometrics. In general, the manuscript is well written, thorough and straightforward to read. The hypotheses are clearly presented and the methods seem appropriate for the study. The figures are also neat and very clear.

I much appreciated that the analysis of morphological variation via geometric morphometrics was done in great depth and carefully executed. This paper strongly differs from traditional studies that simply use linear measurements and/or discrete characters for phenotyping. Therefore, it has a particular merit and should set an example for future studies in this field.
I would include this reference (Berger et al. 2017) in the paper since this the very first study coupling geometric morphometrics and Virus Induced Gene Silencing (VIGS) to quantify the phenotypic effects of knocking down a single CYC2 paralog, FgCYC2A, as well as the reporter gene, FgANS in symmetry of flowers. Therefore, there is a common experimental and statistical background to both studies that could be briefly mentioned/discussed in this paper.

- Visualization grid obtained from thin plate splines (TPS) is potentially misleading for figure 9C. I am quoting a passage from Slice's book chapter to address this issue: "It is important that the initial grid cells be square so that deviations from "squareness" can be interpreted as oriented stretching within the cells of the resulting spline plot. This is not a mathematical requirement. It is just harder to assess how a cell has changed in a plot if you are unsure of its initial shape and distinguishing between initial rectangles and resultant quadrilaterals is more difficult than spotting deviations from squareness." (Slice 2005). It is quite straightforward to re-do this graph using Rohlf's TpsRelw software by simply ticking a box.

In my opinion, the manuscript is suitable for publication after these minor modifications have been taken into account.

References
Berger, B. A., Ricigliano, V. A., Savriama, Y., Lim, A., Thompson, V., & Howarth, D. G. (2017). Geometric morphometrics reveals shifts in flower shape symmetry and size following gene knockdown of CYCLOIDEA and ANTHOCYANIDIN SYNTHASE. BMC Plant Biology, 17.
Slice D.E., 2005. Modern morphometrics. In: Slice D.E. (Ed.). Modern morphometrics in physical anthropology. Klewer Press, New York, pp. 1-45

Methods

Are the methods appropriate to the aims of the study, are they well described, and are necessary controls included? Yes

Conclusions

Are the conclusions adequately supported by the data shown? Yes

Reporting Standards

Does the manuscript adhere to the journal's guidelines on minimum standards of reporting? Yes

Choose an item.

Statistics

Are you able to assess all statistics in the manuscript, including the appropriateness of statistical tests used? Yes, and I have assessed the statistics in my report.

Quality of Written English

Please indicate the quality of language in the manuscript: Acceptable

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