

Interactive comment on “Impact of climate and hydrochemistry on shape variation – a case study on Neotropical cytheroidean Ostracoda” by Claudia Wrozyzna et al.

Anonymous Referee #1

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General comments

This paper reports the morphological difference of ostracod valves among three geographic regions and its relation to climatic and hydrochemical factors. The research theme is interesting and appealing, but there are some major concerns in analysis and discussion. Further analyses and discussion may improve the paper; therefore, major revision is recommended.

The authors analyzed data by relative warp analysis, PLS analysis, and regression analysis. One of my major concerns is why the authors did not conduct MANOVA/MANCOVA or Procrustes ANOVA, but conducted regression analysis on

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each relative warps. The former is appropriate and usually applied for understanding effects of covariates on shape deformation, and is implemented for example in tpsRegr (software by Rohlf FJ, available at <http://life.bio.sunysb.edu/morph>) and geomorph package in R (Adams & Otárola-Castillo 2013). Moreover, if geographic distance or genetic distance among three geographic regions can be incorporated as phylogenetic information (geomorph implements this analysis), the effects of climatic and hydrochemical factors will become clear. Each RWs as response variables might not reflect shape deformation that covariates with climatic and hydrochemical factors.

In addition, further inspection on the result of PLS analysis is recommended. The authors emphasize the similarity between Florida and Brazil based on the plot of RW1 vs RW2, but Fig. 5 indicated the similarity between Florida and Mexico. Graphical presentation of shape deformation indicated by PLS singular axis of shape variables should be added for further discussion. Loadings of environmental variables in PLS singular axis will be helpful for understanding the effects of climatic and hydrochemical factors. The authors reported first PLS singular axes, but second PLS singular axes might also indicate relationship between shape and environmental factors.

Discussion in the paper descriptively indicates that there are morphological differences that possibly relates to environmental variables, but there are little discussions on why shape of ostracod valves differ depending on environments (e.g. are there any functional meanings? or merely due to physicochemical consequences?) and how ostracods respond to environmental changes. It is impossible to achieve definitive conclusions in the paper, but at least proposing some hypotheses is required.

Literature cited Adams DC, Otárola-Castillo E (2013) geomorph: an R package for the collection and analysis of geometric morphometric shape data. *Methods Ecol Evol* 4: 393–399.

Specific comments

Page 3 Line 1: The authors focus on “the causes for the regional differences”, so how

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the factors cause the morphological differences should be more discussed in Discussion.

Line 8: For identifying “the morphological characteristics and environmental variables that contribute most to the relationship”, my recommendation is to inspect further the result of PLS analysis and to apply MANOVA/MANCOVA or Procrustes ANOVA, rather than regression analyses conducted in the study.

Line 20: Add explanation more about “water sampling”.

Page 5 Lines 9–10: Even if selecting variables that contain high loading values in the PLS analysis, separately applying regression analyses might diffuse the environmental effects on shape.

Lines 11–12: The authors concern multicollinearity. One idea is to conduct principle component analysis of environmental variables, and use PC scores as explanatory variables.

Page 6 Line 18: Show the result of statistical testing of the “clear relationships” in the PLS analyses. Statistical testing is possible by using resampling technique, such as permutation test.

Lines 27–28: Table 1 does not show that higher warps (the authors mean RW4, RW5, etc.?) have minor influence on shape variation. In this paragraph, do the authors show the results of relative warp analysis?

Page 8 Lines 5 & 23: The authors only noted that the reasons why shape differences relate to environmental factors are not or poorly understood. However, at least proposing some hypotheses is required.

Page 9 Lines 22–29: Incorporating phylogenetic information in the analysis may be helpful.

Page 10 Lines 6 & 19: The authors emphasize the similarity between Florida and

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Brazil, but the results of PLS analyses seem to indicate the similarity between Florida and Mexico.

Technical corrections

Page 2 Line 1: “individuals” instead of “species” may be better.

Line 9: “extant and extinct” instead of “recent and fossil” may be better.

Page 3 Line 2: “is the knowledge of ... restricted” should be “the knowledge of ... is restricted”.

Page 4 Lines 17–18: The types mean Bookstein’s (1991) types?

Page 5 Line 28: I could not understand which specimens are from Punta Laguna in northern Yucatan. Please show them in Figures 3 and 4.

Line 30: What are “both clusters”?

Page 6 Lines 10–11: The direction of the valve (i.e. antero-posterior axis and dorso-ventral axis) is suggested to be clarified in Fig. 2. Clarify what “consistently” means here.

Page 7 Line 1: “significant” is confusing because it seems to mean “statistically significant”. Here, for example, “valid” is better.

Lines 27–28: The sentence is a little complicated. Suggestion: Annual minimum and maximum temperatures in Florida are 16°C and 3°C, and those in southern Brazil are 30°C and 10°C, respectively.

Page 9 Line 5: “comparatively” or “relatively” instead of “comparably”? And low values of what?

Table 1 The table is complicated and difficult to understand. Captions and/or footnotes should be added for making it stand-alone.

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