Editor’s letter BEAS

Dear Mr Recio,  
  
Based on the evaluations of two referees and an Associate Editor's recommendation, I have to reject the present version of your manuscript for publication in Behavioral Ecology and Sociobiology. However, although reviewers identified significant deficiencies in your study, they also recognized that your work has merit and the potential to develop into a substantial contribution.  If after careful consideration of reviewer comments you are able to fully address all concerns, you may resubmit a thoroughly revised manuscript for reconsideration.   
  
When submitting a revision, please reply point-by-point to the referees' comments.  Provide a clear rationale if you disagree with any comment. Refer to the new line numbers to identify your revisions. Submit your work as a new submission while referring to the original manuscript number together with a marked manuscript in which the changes have been highlighted.  
  
For resubmissions, the marked manuscript should be uploaded as "supplementary material" and response to reviewers as "authors' response to reviewers' comments".  
  
The reviewers' comments can be found at the end of this email.  
  
Thank you for giving us the opportunity to consider your work.  
  
Sincerely,  
Theo C. M. Bakker  
Editor-in-Chief for Behavioral Ecology and Sociobiology, vertebrates  
  
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COMMENTS FOR THE AUTHOR:  
  
Associate Editor:  
I have finally, after inviting numerous reviewers, been able to find two expert reviewers to provide comment on this manuscript. One of them states “reject” the other “reject but resubmission possible”. I will therefore provide the authors with the possibility to submit a revised version of the manuscript where all the comments provided by the reviewers are adequately addressed. If I find the revised version of the manuscript has accomplished this, it MIGHT become suitable for publication in BEAS.  
Thomas Madsen  
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Reviewer #2:   
After carefully reading this manuscript, I regret to say that the experimental results of this study, although quite interesting, are still far from providing publishable data. Although the researchers have carefully designed the experiment and clearly clarify the study's structure, it appears that none of the treatment conditions allowed this lizard species to exhibit numerical discrimination ability under the current experimental design.  
  
I acknowledge the significance of the researchers' experimental design, particularly the investigation of glucocorticoid concentrations and incubation temperature on juvenile lizards, which are academically important and worthy of study. However, the entire paper is structured around comparing their numerical discrimination ability across different experimental treatments. However, since none of the four experimental groups successfully demonstrated the expected behavior, it is difficult to compare an effect that does not exist. Therefore, I sincerely recommend that the authors first refine their experimental design until the animals can reliably exhibit the target behavior before drawing conclusions. Otherwise, despite various controls over experimental conditions, it would be challenging to determine the actual effects of these variables.

We sincerely appreciate the reviewer’s constructive feedback. We fully acknowledge that without the animals exhibiting the target behaviour, comparisons between experimental conditions could be misleading. However, we believe it is important to: a) transparently acknowledge that our primary research question focused on the effects of prenatal treatments, even though our experimental design did not allow us to fully investigate this question as initially intended; and b) highlight that, despite our inability to conclusively assess the impact of early life conditions on numerical discrimination with this design, our data still offer valuable insights into how early environmental factors influence cognitive processes and decision-making.

In our revised manuscript, we will adjust the introduction to reflect the aspects of the research that can be effectively addressed. Additionally, we will provide a more detailed explanation of the experimental design decisions and demonstrate how the current design still allows us to explore potential effects of early conditions on cognitive outcomes.

Additionally, I have a few specific concerns:  
  
1. The researchers attempted to alter the placement of cricket food items to standardize object area or length. However, under the current experimental conditions, I believe such control is premature. It remains unknown what cues lizards use to perceive numerical information—whether it is occupied space, area, length, or other visual cues. Before confirming that they indeed possess numerical discrimination ability, introducing a spatial illusion to confuse the animals may not achieve the intended effect.

2. Furthermore, how exactly was this placement manipulation expected to achieve the described effect? The authors did not clearly explain this in the manuscript (Lines 210-218).  
  
3. Line 318: "the relevant stimuli involved either vegetable or large quantities." This sentence seems not precise. Based on my understanding of these species, being herbivorous or preferring larger food items is unlikely to be the key determinant of numerical discrimination ability. Carnivorous animals are also known to possess numerical abilities. Improving the experimental design may be more critical than focusing on whether the animals are herbivorous or carnivorous.  
  
  
  
  
  
Reviewer #3:   
This paper is an important contribution because it is the first study that I am aware of that tackles the combined effect of both cort and temperature on cognitive ability, in this case quantity discrimination. the authors use the garden skink (Lampropholis guichenoti) as a model system, which is entirely appropriate. Overall, the paper is well-written and certainly well analysed. Having said that, the introduction fell a little short when it came to describing the two core systems (see below) used for quantity discrimination. This is an issue that is easily addressed. However, it is perhaps a prelude to an unorthodox method for measuring quantity discrimination. I going to this in more detail below (see comments about Fig. 1). The authors themselves raise this as a potential issue in the discussion. I guess the key question here is to what degree their design addresses the role of court and temp on a measure of cognitive ability - quantity discrimination. Added to this is  
the issue that the authors don't find any quantity discrimination ability, which makes testing for the role of cort and temperature slightly problematic. I can't help thinking they likely have some QD ability that just wasn't uncovered using their design but I could certainly be wrong.   
  
Title: I would probably have gone with "Does early environmental experience impact quantity discrimination ability in a lizard?"  
  
22.     Again, I think "quantity discrimination" is more appropriate than quantitative abilities. For example, on a very elementary level an animal might simply be able to tell whether one quantity is larger than another. In this case, they are not really doing any form of mathematics or quantification. Bear this in mind for the rest of the manuscript, I will not highlight any other instances.  
  
Abstract. 33. Yes, and were you examining numerical vs size discrimination or both?  
  
83-85 I wasn't aware of any studies? You should cite them.  
  
The intro is very well written and nicely tackles the role of the early thermal developmental environment and glucocorticoids, in affecting cognition. It's missing some useful background information on quantity discrimination (QD). For example, the brain uses two core systems, the object file system (OFS) and the approximate number system (ANS). The OFS considers items as discrete units. So, this system employs absolute numbers albeit typically only up to 4. The ANS is ratio dependent and deals more with magnitude. The ANS does seem to follow Weber's law, which the authors mention, and allows an animal to discriminate when one quantity is larger than the other. The authors appear to be focusing on the ANS and not the OFS. This is fine given the context, but they just need to make this clear.  
  
159.     snout-vent length, tail length (lower case).  
  
170.     I'm quite surprised that the control was 100% ethanol. I see that the court was dissolved in 100% ethanol, but wouldn't ethanol itself potentially have an adverse effect on development? Or is it too small a volume to worry about and perhaps just short-term?  
  
202.    Maybe add that lizards were happy to eat dead crickets. Okay, I see you do this in 203. That being said, why the new pg? I'm surprised these were dusted with calcium and multivitamins in an experiment with a food reward, but totally fine.  
  
213-215. This is the first mention of discriminating between using the ANS vs OFS although these terms are not used. This needs to be addressed in the introduction and mentioned early in the methods.  
  
Fig. 1. I'm slightly confused by this experimental design. Typically, you would separately test discrete (OFS) versus overall quantity/size (ANS). When testing OFS the different quantities would still have the same surface area. When testing ANS it would be the same number of objects but they would differ in overall size. I get this is a little difficult with crickets. But the current design seems to make a few assumptions. I think the authors need to explain this a little bit more in light of what people typically do in these studies and do a better job explaining what we can infer from the results using this design. I think the key result is that cort and temperature have no significant effect on QD. I'm just not too sure about the specific metric of QD that is being measured. (After writing this, I saw in the discussion [line 340-352] they mention potential issues with their design.) But perhaps more importantly, the authors don't actually find any QD ability and as such,  
a reader may wonder why you are measuring an effect of cort and temp on a phenomenon that has not been demonstrated. This perhaps needs to be fleshed out a bit more.

We appreciate the reviewer’s thoughtful concerns and would like to clarify our experimental design decisions. We acknowledge that the cues lizards use to perceive numerical information are not fully understood, and we intentionally included conflicting cues—both numerical (number of items) and size (area/volume)—to explore whether these cues are processed independently or in relation to each other. Given that size or length has been shown to influence quantity discrimination in lizards, we chose to test the potential for numerical discrimination under these conflicting conditions to better understand how early environmental factors might affect decision-making. We realize that this approach is unconventional but felt it was necessary to test the boundary between these conflicting cues in a single design.

We agree that our approach could have been clarified further in the manuscript, particularly regarding the distinction between object file system (OFS) and approximate number system (ANS) strategies. We will revise the methods section to better explain our reasoning for combining both size and number in a single trial and how this design attempts to capture any potential interaction between these cues. We will also provide a more detailed explanation of what we can infer from the current results, despite not having demonstrated clear evidence of numerical discrimination.

Regarding the lack of observed quantity discrimination (QD) ability, we will further elaborate in the discussion about how this result—while not supporting the hypothesized QD—still provides important insights into how early conditions (cortisol levels and temperature) may influence cognitive abilities and decision-making. This will clarify why we pursued this design despite the absence of significant QD ability.  
  
236-237. Was PR also blind to the actual 'amount' in addition to the treatment? Maybe that was not possible?  
  
The data were rigorously and appropriately analysed, and well presented.  
  
325.    Spelling. Maybe do a spell check on entire document.  
  
The discussion is thorough and I appreciate the authors detailing their literature search queries in the ESM.

 **ou were NOT adding noise or confusion—your goal was to dissociate size from number.**

* Instead of the expected pattern (more crickets = more total size), your setup deliberately **broke** that relationship to see if lizards relied on number alone.
* This is a common method in numerical cognition studies! But you need to emphasize that this was an **intentional design choice**, not a flaw.

 **Your experiment still allows testing of numerical discrimination.**

* If lizards rely on number, they should **still choose based on numerosity** even when size is held constant or reversed.
* If they do not, then they likely rely more on continuous variables like area/length rather than discrete numbers.

 **You controlled for size as much as possible and tested whether it mattered.**

* The **Prey Orientation Test** suggests that lizards did not simply default to using orientation/size cues, supporting the idea that size was not the driving factor.
* If you have data showing that total size across conditions was truly equal, highlight this more clearly.