

#PreprintReviewChallenge - ‘Consistent variations in personality traits and their potential for genetic improvement in the biocontrol agent *Trichogramma evanescens*’

Reviewers:

Ana Pereira, Broad Institute of MIT and Harvard

Joshua Byrne, National Coalition of Independent Scholars

1. Overall Statement

One-paragraph summary of what the main findings reported are and how the work fits in the context of knowledge in the field.

In this paper the authors quantify several behavioral traits of the biocontrol agent *Trichogramma evanescens*, that are used to define phenotypes based on animal personality. The authors correlate these features with traits classically measured in the lab with the aim of creating a platform that may improve the selection of biological control agents (BCAs). The results show that individuals of different genetic lines exist in a continuum of exploratory behavior and activity (personality scores), and that there is a correlation between fitness-related traits such as number of offspring and one of the personality scores, activity. The authors also found that there are significant differences in personality traits between lines, and that a genetic correlation between a personality and a fitness-related trait could be established. This potential link between desirable life history traits of BCAs and individual differences in behaviour illustrates the ecological relevance of personality when assessing the suitability of BCAs for their use in crop protection.

2. Highlight the things you liked about the work

A paragraph outlining what you liked about the study, what results did you find interesting and why? Was there a new angle to the research approach or results that bring a new perspective to the field? What avenues for further research does the preprint open?

I find the topic of this research really interesting and an unusual way to approach the relevance of the study of animal behavior. I think the text is very clear and easy to follow. The documented variability amongst isogenic lines is a quite interesting finding.

The results are presented in a logical and concise way to be able to identify both statistical and meaningful results of the analysis. I also liked how the figures clearly display important features of the experimental set-up so the behaviour observed can be visualised by the reader. A particularly elegant feature of this study was how isogenic lines allowed the authors to tease apart the potential genetic and environmental influences on the expression of animal personality.

The aim is very clearly specified in the introduction and answered by the subsequent discussion of the findings presented in the paper.

3. Outline any questions for the authors or areas of concern

Major points - relating to the methodological rigor of the study or to whether the conclusion can stand), consider the following:

- *Do the title and abstract accurately reflect the work carried out?*

The title is very descriptive and the abstract sufficiently describes the work carried out

- *Study design and methodology: is the design adequate to address the research question? Were the techniques employed appropriate and in line with the state of the art in the field? Were suitable controls included? Were the statistical methods used appropriate?*

The methodology used is adequate to the purpose of the study and the animal model being tested. Regarding the statistical methods and data analysis, the author's mostly use a Spearman's rank correlation coefficient that assumes that there is a monotonic relationship between the variables. It would be beneficial to have information about this assumption, as well as to the distribution of the data, in order to evaluate if the data has been analysed in the proper way.

- *Do the results support the authors' conclusions?*

The shortcomings and implications of the findings are discussed equally. The conclusions obtained in this paper are well supported and are set into the findings of other studies, such as the discussion of the R (repeatability) values found by the authors compared against those from other studies.

- *Are there any gaps in terms of how the study is contextualized within its field, or concerns in the interpretation of the results?*

I don't have enough knowledge of the field to be able to answer this point.

Although I cannot comment on the assessment of BCA's, the citations used to support the material related to animal personality is relevant and sets the paper within the field. Also, there are couple of points that could be addressed:

- The relationship between the light and the dark used in the observations of behaviour could be better explained; not obvious in the paper presently. Also, the paper could discuss the significance of darkness for the focal species. Depending on the context, shade can mean safe cover or the risk of predator presence. Highlighting this distinction for the wasps will clearly define the behaviour being captured in these experiments, such as boldness in response to risk.
 - Could add a paragraph to the introduction that explains the wasps behaviour, so that the ecological context of the experimental design for this species is clear. For example, that this species disperses mostly by walking rather than flight so their activity and exploration within a two-dimensional space is more relevant. Another point could be highlighting how much/little social behaviour is expressed in this species so the impact of females being isolated can be addressed.
- *What additional experiments or changes to the manuscript would you recommend to address any concerns relating to the items above?*
None in particular.

Minor points - additional clarifications or small corrections that improve the paper but do not impact the standing of the work or the conclusions, consider the following:

- *Is the reporting of the methodology sufficient to allow the work to be replicated?*
Yes - as mentioned above the figures clearly illustrate the experimental set-up used. Also, the housing and animal care of the focal species is thoroughly explained.
- *Do the authors adequately discuss the limitations of the study?*
Yes - highlights how the low heritability of the traits observed could affect long-term evolutionary projects and discusses the potential issue for other species where large numbers, in this case hundreds, of subjects may not be feasible.
- *Does the manuscript adequately place the work in the context of knowledge in the field?*
Yes - Again, cannot comment on the literature surrounding BCA's but the animal personality material is relevant and significant to what is known in the field.
- *Are there any aspects of the language or presentation that need to be polished?*

There are no syntactical errors or recurring issues with presentation. Language is clear and concise.

- *Have the authors made the underlying data/code available?*

Not yet, such as the sample sizes from each isogenic line, but the authors will make them available

- *Are there any competing interests that should be considered?*

No

4. Any additional comments or questions for the authors?

- *The potential for differing effects of personality on male and female behaviour?*

The authors focused on females because they have larger significance in this species role as a BCA by parasitising the eggs of pests. However, opposing effects of personality traits between genders is still a possibility that can produce a trade-off in the context of BCAs. For example, this study finds low exploration scores in females may increase egg number, and thus the species impact as a BCA, but this behavioural score could affect male dispersal. If fewer females are fertilised this could reduce the benefits of selecting lines that have lower exploration scores.

- *Do you keep the female isolated? Do you release them as a group? How the personality traits are affected by the fact that they are isolated?*

Technical limits of the experiment. When they release them in the field, they are easily dispersed. In lab rearing, there is an interaction between individuals. This species is not particularly social so it is assumed by the authors that isolation would not affect behaviour.

- *Is the behaviour while walking different from the flying behaviour ?*

These wasps mostly disperse by walking. They are inefficient fliers so their behaviour in flight may not be as significant for this species impact as a BCA

- *Tablet screen vs paper? Effects on the wasps?*

There was a glass barrier between the wasps and the screen so they were not directly in contact. But did not check if radiation, such as potential vibration or harsh light from the equipment, may affect the wasps. However, temperature was maintained at a constant and replicable level.

- *Measurement of the activity: does activity means moving?*

Cleaning the antenna is not considered as an activity. Not possible to take into account any other parameter than movement.

- *Value of repeatability? Could you explain?*

Repeatable behaviour: in relation to the other individuals of the group. The higher the value, the less the individual shows variability of this trait. In other words, it is a measure of consistency in behaviour over time. This helps ascertain if a key feature of animal personality, that individuals are restricted to a particular response over time, is present.