

Dear Recommender,
Dear Reviewer,

We would like to thank you for the opportunity to revise and resubmit our Stage 2 registered report (RR) entitled “Neophobia across social contexts in juvenile herring gulls”.

We are grateful for the comments provided by you and the reviewers. We have accepted all textual changes and added a detailed point-by-point response below. All adjustments have also been incorporated into the revised manuscript.

We would like to respond to two main comments made by reviewer 1, reiterated by the recommender here in detail:

(i) Species effect: There may be differences in neophobic responses between non-migratory species (e.g., herring gulls) and migratory species (e.g., lesser black-backed gulls), potentially resulting in species-specific distinctions. To account for this, we registered in the Stage 1 report that we would exclude lesser black-backed gulls from our primary analysis. To provide further transparency, we have included a supplementary table detailing the group compositions.

For completeness and to further address the comment of the reviewer, we repeated the analysis including lesser black-backed gulls. While the results remained broadly consistent, we observed that the fixed effect of context on latency to enter was no longer significant when lesser black-backed gulls were included. As discussed in the paper, latency to enter is not a direct measure of object neophobia, and this change does not affect the interpretation of our findings regarding neophobic responses. We included a table in the supplementary materials with the model output of the analysis including lesser black-backed gulls. Please note that given the small number of lesser black-backed gulls in our dataset ($n = 13$), the sample size was insufficient for meaningful direct species-level comparisons.

(ii) Sex effect: Thank you for highlighting this point. This was an oversight. We now account for sex in the analysis by including it both as a main effect and as an interaction term with object condition (to test for sex effects on neophobia) in our models. Interestingly, we found effects of sex for all three measures. Specifically, the time spent in the ZOI was significantly influenced by sex, but this interacted with object condition. Females spent more time in the ZOI in the control condition compared to the novel condition, whereas males exhibited a more stable response across both object conditions. For latency to enter and latency to eat, we found main effects of sex, but this did not interact with object condition (i.e. neophobia). However, given the constraints of our sample size and experimental design, we caution against concluding that sex does not influence these responses as well.

We have commented on these findings in the General Discussion. Furthermore, we have added a table to the supplementary material detailing the sex composition of each experimental group.

To ensure transparency, we have also provided details on the methods used for sex determination: “*Feather samples were collected for sex determination via PCR, following the*

protocol outlined by Fridolfsson and Ellegren (1999). This method targeted the CHD1W and CHD1Z introns using 2550F/2718R primers, with PCR conditions set to 30 cycles at an annealing temperature of 56°C.”

We declare that this revised Stage 2 RR remains original and unpublished. All authors approved the submission of the revised Stage 2 RR in its current form.

On behalf of all authors,
Reinoud Allaert

Review by anonymous reviewer 1, 08 Jan 2025 18:57

This is an interesting, well-written and concise manuscript exploring a very timely research question on differences in neophobia across social contexts in a highly social bird species. The Authors tested 54 juvenile herring gulls, in individual and social context (i.e., in groups of 4-5 individuals, containing no nestmates). The Authors tested three distinct research hypotheses: ‘risk dilution’, ‘negotiation’ and ‘social conformity’, and these hypotheses were accompanied with clear illustrations on the proposed effects. The Authors followed most of the recommendations from the Stage 1 report, and modified some aspects of Methods according to suggestions: in particular, by giving birds more time to habituate to the control objects and changing some of their novel objects. Data analyses followed the Stage 1 plan and were done using adequate statistical techniques. The Authors found that juvenile herring gull neophobia is modified by the social context: individuals tested in groups entered the test apparatus and ate food more quickly, as well as spent more time near the novel objects than when individuals were tested alone. These results give support to the risk-dilution hypothesis, whereby perception of risk is likely shared between the group members. I am looking forward to seeing this manuscript in print. Please see my specific comments as well as copy-edits below, and hope that the Authors will find them helpful.

One major comment regarding the study design is that two bird species (herring gulls and lesser black-backed gulls) were tested in this study, and the Authors only later in the analyses excluded lesser black-backed gulls from the analyses, which has led to a larger than expected dropout rate (out of the proposed 80 individuals, 54 individuals remained). The Authors do not adequately mention in text to which extent these two species are similar/different, how many of the lesser black-backed gulls were present in each testing group, they do not test for whether and to which extent the presence of lesser black-backed gulls during the tests influenced behaviour of the herring gulls, they also do not mention or argue how this mixed-species composition could have influenced the results. Could the Authors give some information on this? It would also be beneficial to provide some statistical comparisons between groups with different species composition.

Thank you for raising this point. We have addressed this comment in the rebuttal letter above.

How much food (stacked fish) was provided in the food plate (L208)?

We agree that this was unclear. We have specified the plate diameter and made the necessary adjustments to the manuscript accordingly: *“Next, a food plate (27 cm in diameter), completely filled with fish, and an object (novel or control, depending on the condition) were placed at the back of the enclosure, with the food plate placed in front of the object to rule out directional preference.”*

What was the length of the trial – was it ten minutes or until the bird first touched the food?

The trial duration was 10 minutes for each bird after it left the start area. The trial did not end when the bird first touched the food; it continued for the full 10-minute period. We’ve implemented this in the manuscript:

“In both group and individual trials, individuals were given a maximum of 10 minutes to leave the start area and enter the test arena. Once an individual entered, the trial duration was a fixed 10 minutes. During this period, individuals had the opportunity to feed, but the trial continued for the full 10-minute duration regardless of whether the bird first touched the food. This approach aligns with previous novel object studies.”

Table 1. Did the Authors measure all times that the animals spend within the zone of interest overall (also after the food was touched), or just how long the animal spent in ZOI before it first started eating?

We measured all the time the animals spent in the zone of interest throughout the entire 10-minute trial, not just the time before they started eating. We have revised the information in the table:

“The bird is considered to enter or leave the zone of interest when the front half of its body crosses the (notional) boundary of the zone.”

Details on sex ratios and sex analyses that are mentioned in the supplementary material are missing from the manuscript. In the Stage 1 report it was mentioned that sex will be considered in the statistical analyses, but I could not find any mention of these analyses. Could you provide more detail on this? Was there any sex effect?

Please see our reply above. In short, we have updated our methods to account for sex as a fixed effect in the analysis. Interestingly, we found a significant main effect of sex on two of our dependent variables, and an interaction effect on the third. These findings have been briefly discussed in the manuscript.

Abstract should include more methodological details like the number of tested individuals.

We have included the number of tested individuals in the abstract and revised it as follows: *"To this end, we exposed juvenile herring gulls (N = 54) to novel objects in both individual and group settings (4-5 individuals), replicating each condition twice."*

The authors use some abbreviations that are not explained on their first mention in the text. For instance, what is "RM" and "cor." in line 127-129? Please check for other such abbreviations in text too.

We removed the abbreviations in the text and have adjusted the main manuscript accordingly: *"Power = 0.80 ; correlation among repeated measures = 0.5), as well as an interaction between context and object with small effect size (0.11; Power = 0.80 ; correlation among repeated measures = 0.5)."*

.

L9. "to establish reproducibility" -> "to establish repeatability"

We appreciate the suggestion. However, rather than aiming to establish reproducibility or repeatability, the primary goal of repeating the experiment was to obtain replicates. For clarity, we have revised the sentence as follows: *"To this end, we exposed juvenile herring gulls (n = 54) to novel objects in both individual and group settings, replicating each condition twice."*

L11. It is somewhat unclear what is meant by "by distributing it among group members". Please rephrase.

To improve clarity, we have revised the sentence to: *"The results of our study suggest that the presence of group members reduces perceived individual risk, allowing individuals to behave less cautiously."*

L43-44. It would be good to support this sentence with some references.

Thank you for the suggestion. We intentionally did not include citations in this sentence because the subsequent paragraphs provide detailed examples and references supporting this statement.

L55. "causing them to behave more similarly" does not follow logically from the sentence, please clarify further as was done in L67-68.

We have clarified the sentence to better explain the link between shared risk perception and uniform behaviour. The revised sentence now reads: *"These theories predict that neophobia, or fear responses in general, are reduced in the presence of others, as individuals in a group collectively share the potential risks associated with novel situations or threats. This shared*

risk perception will also lead to more uniform behaviour within the group, as individuals adapt their actions in response to the behaviour of conspecifics.”

L176. Following my previous comment, did any measures differ between the groups of different sizes?

Due to model constraints, we did not include group size in the models, as we already had to simplify the preregistered version of the models to ensure convergence and interpretability. Additionally, we do not expect meaningful differences between groups of four and five birds.

L277-279. Could you provide more details on how the contrasts were calculated?

To fit the multivariate model, we transformed the dataset into a long format and created the “eat_vs_leave_contrast” variable (with eating = 0.5 and leaving = -0.5) to contrast the two behaviours. As the specific values of the contrast are arbitrary and do not affect the analysis, they were not explicitly mentioned in the manuscript. We did not modify the latency values themselves but rather added a column to the dataset allowing us to distinguish between the two behaviours in the multivariate model.

L280-281. Why is it mentioned that these results are in the main manuscript?

Thank you for pointing this out, we’ve adapted it in the manuscript as follows: *“However, for ease of interpretation, the results of the univariate models are presented here, as they allow a more straightforward interpretation of the individual effects of each predictor on the dependent variables.”*

Review by Claudia Mettke-Hofmann, 05 Dec 2024 08:01

This stage 2 manuscript follows the introduction and methodological protocol outlined and approved in stage 1 and any deviations (e.g. sample size) are justified and documented.

Quality checks were performed and when necessary, transformations conducted.

Results are well presented with effects of both factors (context and object) and their interactions showing indicating that the lower sample size did not hamper detection of significant differences.

The discussion centres around the questions proposed in stage 1 and the respective outcomes.

The conclusion is based on the evidence provided.

Overall, this is a very nice and interesting study that is well presented.

I have only one comment (see below).

Comments:

Methods:

Lines 143-145: shouldn’t it be within subject factors rather than within species factors for the repeated measure?

Thank you for highlighting this point. We have made the changes accordingly.

“Although this is a significant reduction from our planned sample size ($N = 72$ after exclusion of LBBG), it is important to note that our sensitivity analyses were based on repeated measures MANOVAs (within subjects factors).”