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Re: Thesis Oldřich Tomášek

Report of Prof. Dr Simon Verhulst on the PhD-thesis of Oldřich Tomášek: Condition-dependence of sexually selected ornaments in birds

This thesis consists of five papers, of which three have been published and one is in revision, all in good quality journals. All papers have a fair number of authors (5-10), and Tomášek is first author of three of the five papers and second author on two papers. Papers 1-3 are on sexual selection in barn swallows, while papers 4 and 5 report results of one experiment with zebra finches on signalling and sperm respectively. The papers are preceded by an 'Introduction and synthesis' chapter, which is eminently readable and provides a useful overview of the field as well as the contents of the five papers.

Please note that I have concentrated my efforts on the unpublished papers, where there is the potential opportunity to further improve the paper.

Paper 1: Extra-pair paternity patterns in European barn swallows *Hirundo rustica rustica* are best explained by male and female age and not male ornamentation

Tail streamers of European barn swallow males are a textbook example of a sexually selected trait, with previous publications reporting increased mating success of males with longer tail streamers. The present publication challenges the generality of this result, based on the absence of an association between streamer length and success in extra-pair fertilisation success. Instead, it is argued, male age is a more important factor determining extra-pair fertilisation success. This is an interesting suggestion, but it is not fully resolved. One question is how different the coefficient of the association between streamer length and EPF success really was from the coefficients in previous studies, and whether the analyses are otherwise comparable. In the present study, this coefficient is not significantly different from zero, but it may not be significantly different from previous estimates either. Another point to consider is whether the variance in ornamentation is comparable between study populations – a smaller variance in streamer length could in itself generate a difference in selection.
Another question is whether females can assess male age in other ways than through sexual ornaments. Male age can be measured exactly, while ornamentation is complex, i.e. may well be multi-modal and involve behaviour, and hence the estimates of ornamentation in the study are unavoidably incomplete when compared to the information that a choosing female has available. Or is it so that older females are able to select for age more strongly, because they have prior experience with the male? Do yearling and older females differ in the extent to which their extra-pair young are sired by older males?

Male tarsus length came out as a predictor of cuckoldry in using one statistical approach, but not when using another statistical approach, and the difference in result appears unresolved. A potential explanation to explore is a difference in statistical power – perhaps the effect is very similar in the two approaches, but is significant in only one approach due to higher statistical power. This explanation is testable using e.g. simulated data.

Older females had more EPP. Do they arrive earlier? Perhaps they have more information on males than the younger females? Can swallows individually recognize each other; this is a prerequisite for such a mechanism. Or perhaps new individuals can be recognized by their behaviour in the barn?

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Paper 2: Age-dependence and viability signalling function of tail streamer length in the European barn swallow

In this paper, the association between age and streamer length is investigated in a way that separates between- and within–individual variation and hence also informs on the extent to which mortality is associated with streamer length. In addition, the effect on survival and next year streamer length is reported of an experiment in which streamer length was experimentally manipulated.

Streamer length increased with age from the 1st to 2nd year, but not thereafter, and there was no evidence for 'streamer senescence'. Individuals with longer streamers as yearling enjoyed a longer lifespan, in particular in the Czech population. These results are broadly in agreement with what one expect, except that the absence of streamer senescence deviates from what was found earlier. It is argued that methodological differences may explain this difference, but, not mutually exclusively, a different signalling role in the study populations may also play a role.

Streamer length manipulation was carried out between the 1st and 2nd breeding attempt, which contrasts with earlier studies (when I remember correctly). Signalling may well continue to be important after pair-formation, and hence I am curious whether the manipulation had an effect on occurrence and timing of 2nd broods, and, if so, whether this may have confounded possible effects on survival. In either case, that the manipulation was carried out later in the season compared with previous studies may in itself explain that the survival effect is weaker, simply because birds did not suffer the manipulation during a shorter period and not during what may well be a critical period (i.e. pair formation and raising of the first brood).
A further methodological point worth noting was that pre-manipulation streamer length differed between the experimental groups. How this could arise was not clear, and I am not sure to what extent controlling for initial streamer length in the statistical analyses resolves this bias.

Paper 3: Signal traits and oxidative stress: a comparative study across populations with divergent signals

In this study, the association between signal traits and oxidative stress proxies is compared between populations that apparently differ in the role of those signal traits in sexual selection. The study makes good use of the well described difference between Europe and North America in the average level and role in sexual selection of two sexual ornaments: tail streamer length and breast colouration. As predicted, there were indeed significant interactions between trait values and population with respect to different oxidative stress related variables. This is in itself an interesting finding, but, as recognized in the paper, it does also raise some questions.

An important assumption in the study is that the oxidative stress proxies reflect health, which I take to mean that they are a predictor of fitness prospects. I was wondering what evidence there is for this assumption in swallows.

In your comparison, you assume that European females prefer long tail streamers – is this assumption justified for your study population?

Table 1 – some interactions are not significant, but then again, the models without the interaction are probably not substantially better models.

Why would there be an association between colouration and redox state when the feathers have already been formed previously? With the streamers, this question does not arise, because the idea is that the costs are primarily incurred the effect of long streamers on flight, and not so much by their development. Such an effect is not so obvious with respect to colouration, which makes me wonder why would there be an association between colouration and redox state.

A general question about the study is whether one year of data sufficient to compare populations. When sampling in only one year, the implicit assumption is made that between-year differences in a population are negligible relative to the between-population differences. I am wondering if this is so, and suggest that knowledge of between-year variation in the studied relationships at either site could shed some preliminary light on this assumption.

Paper 4: Opposing effects of oxidative challenge and carotenoids on antioxidant status and condition-dependent sexual signalling

In this paper, the results are presented of an experiment in which in a 2x2 design zebra finches were administered a pro-oxidant (diquat) and carotenoids. The aim of the experiment was to test the effect of carotenoids on oxidative stress. This is of interest, because carotenoids are important in sexual selection, because it colours body parts and feathers that are at least in some cases under sexual selection. However, the nature of the role of carotenoids is under debate, with different scenario's assuming a different effect of carotenoids on oxidative stress. Because of its experimental methodological approach and the results this chapter was for me one of the highlights of the thesis. In particular the suggestion, and confirmation, that antioxidant activity of carotenoids should be measured in lipophilic medium has consequences for the field. A remaining question is what we would have gained from having multiple rather than two levels of treatment? This is particularly relevant because it is difficult to ascertain to what extent the treatment levels were within the natural range, and multiple treatment levels would shed some light on the importance of finding out what the natural range is. A second question is whether the treatments resulted in differences in consumption of water and food, because this would complicate interpretation of the results.

Paper 5: Trade-off between carotenoid-based sexual ornamentation and sperm resistance to oxidative challenge

This paper is based on the same experiment as paper 4, but focuses on the experimental effects on sperm-traits. It appears that the treatments had some effects in interaction with other traits, but no overall effects (in particular when a correction of the p-values for multiple testing is applied).

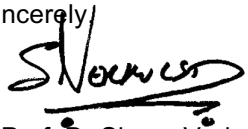
There is no mention of the oxidative stress measurements reported in paper 4, based on the same experiment. What were their associations with sperm traits? And why were they not reported? Do they not provide information on the question posed in this paper?

The models in Table 1 in most cases include the initial value of a trait while the dependent variable is the difference between the initial value and the final value. This implies that the initial value is corrected for twice, which does not seem optimal and the effect on the results is not obvious; the resulting correlations are a clear example of a phenomenon known as 'regression to the mean'. However, little is made of these correlations, and hence I expect little effect of this approach on the results.

Conclusion:

Based on my reading of the thesis I am happy to report that I consider the thesis suitable for the defense and that its quality fulfills the criteria necessary for obtaining the Ph.D. degree.

Yours sincerely,



Prof. Dr Simon Verhulst