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Personality assortative female mating preferences in a songbird

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Abstract

Consistent individual behavioural differences ('animal personalities') are documented across a variety of animal taxa. Sexual selection, especially assortative mating has been suggested as a possible mechanism contributing to the maintenance of different personality types within populations but little is known about non-random pair-formation with respect to personality traits in unconstrained choice tests. We here tested whether female mating preferences were non-random with respect to male and female neophobia in zebra finches (*Taeniopygia guttata*), an important avian model of mate choice and animal personality research. Male and female neophobia was assessed by attaching novel objects to birds' feeders. Females' mating preferences were tested with randomly assigned, unfamiliar males in a four-way choice apparatus. Females associated most with males with neophobia scores similar to their own. These results provide evidence that mating preferences and personality traits can covary, supporting evolutionary scenarios of assortative mating contributing to the maintenance of personality traits.

Keywords

personality, neophobia, boldness, novel object, sexual selection, assortative mating, mate preference, zebra finch.

1. Introduction

Individuals within the same population often express consistent individual differences in their behaviour that persist through time and different contexts and are referred to as ‘animal personalities’ (Wilson et al., 1994; Gosling, 2001; Réale et al., 2007; Dingemanse et al., 2010). The increasing documentation of these phenomena raise the questions of their adaptive value and the selection pressures maintaining such pronounced between-individual variation (Réale et al., 2007; Wolf & Weissing, 2012). The potential importance of sexual selection in maintaining within-population variation in personality traits has only recently received attention (reviewed by Schuett et al., 2010). Observations of improved breeding performance of specific combinations of behavioural types in field and laboratory studies (Dingemanse et al., 2004; Both et al., 2005; Spoon et al., 2006; Schuett et al., 2011a; Gabriel & Black, 2012; Burtka & Grindstaff, 2015; Ihle et al., 2015; Rangassamy et al., 2015) suggest that animals would benefit from actively choosing compatible mates. However, such observations are inconclusive with respect to the cause of the associations as non-random mating patterns can arise from a variety of processes (Burley, 1983; Riebel et al., 2010). For instance, if different personality types differ in temporal and spatial grouping (Snijders et al., 2014; Johnson et al., 2017), territory choice (Both et al., 2005; Holtmann et al., 2017) or sampling activity (David et al., 2011) pair formation might be unduly influenced by non-random encounter rates between different personality types. Assortative pairs then might form simply because there are higher encounter rates among individuals of similar personalities and in this case assortative combinations could occur more often than expected by chance even in the absence of personality related mating preferences. Observations of apparently personality matched pairs can also result from behavioural convergence, where initially dissimilar partners become more and more similar after pair formation (Burley, 1983; Laubu et al., 2016). Hence, observations of non-random pairs are not sufficient to demonstrate whether mating preferences or other processes brought the pairs together (Burley, 1983). This distinction is non-trivial as different processes are likely

to show different responses to fluctuations in ecological factors affecting the temporal and spatial patterning of different personality types and hence show different evolutionary dynamics.

If assortative mating and sexual selection via mate choice contribute to the maintenance of interindividual variation in behavioural traits (for review see Schuett et al., 2010) then either mating signals have to be informative of personality (Godin & Dugatkin, 1996; Garamszegi et al., 2008; Amy et al., 2010) and/or choosing individuals must be able to directly assess the behavioural traits in question. These two mechanisms do not have to be mutually exclusive: in Trinidadian guppies (*Poecilia reticulata*), for example, male boldness (measured as predator inspection) and the intensity of orange colour ornamentation are correlated and both were found to be preferred by females after each was manipulated independently experimentally (Godin & Dugatkin, 1996) suggesting that both direct observation of behavioural and mating trait variation could provide females with information on personality traits in this species. This example notwithstanding, the links between variation in personality and mating traits for other species and taxa have been barely studied, but these phenomena seem phylogenetically wide-spread. Non-random mating preferences with respect to a personality trait have also been found in two spider species where both males and females show consistent interindividual variation in aggression levels and non-random mate choice with respect to these traits (Pruitt et al., 2011; Kralj-Fiser et al., 2013) but it is yet unclear which cues or signals mediate these choices. In birds, improved breeding performance for specific personality combinations has been reported in a number of species (e.g., Dingemanse et al., 2004; Both et al., 2005; Spoon et al., 2006; Schuett et al., 2011a; Gabriel & Black, 2012; Burtka & Grindstaff, 2015) but data were either observational or from experimental pairings, thus providing no answer as to whether birds actively choose their partners to form specific combinations of personality types. To manipulate exploration levels in zebra finches, Schuett et al. (2011b) let females observe two males: one was freely exploring his cage, whereas the other was confined to a small Perspex box. In subsequent mate choice trials, females preferred the previously exploring over the confined males. In control trials, however, where both males were unconstrained to explore the novel environment during the exploration phase, the naturally more exploring male was not necessarily preferred. This study, therefore, raises several follow-up questions: while the female subjects clearly discriminated against

the movement constrained birds (confined to a small box), they did not prefer the more explorative male in the conditions where two males could move freely. This suggests that either the experimental manipulation resulted in stronger contrast or the short confinement made the birds behave subtly different afterwards. Without knowing whether zebra finches would normally choose their mates non-randomly with respect to personality traits, it is difficult to interpret these results further.

However, it seems important to pursue this issue: for one, birds are well studied for personality differences both in the field and laboratory and there are well established and validated personality test paradigms that have revealed consistent individual differences in boldness and neophobia (Verbeek et al., 1994; Groothuis & Carere, 2005; Krause & Naguib, 2011; Schielzeth et al., 2011). Second, birds in general, and zebra finches in particular, are intensely studied models of mate choice (Riebel, 2009; Griffith & Buchanan, 2010). In zebra finches, preferences expressed in mate choice chambers predict actual pair formation (Clayton, 1990; reviewed in Forstmeier & Birkhead, 2004; Riebel, 2009). Moreover, neophobia can be reliably measured by placing a novel object next to a familiar feeder (Schielzeth et al., 2010; David et al., 2011). Here we made use of the validated methodologies for behavioural typing and measurements of mating preferences in unconstrained choice tests (ten Cate & Mug, 1984; Witte, 2006) in unmanipulated zebra finches to test whether mating preferences co-varied with male and female levels of neophobia (i.e., the exploration-avoidance axis, Réale et al., 2007). We focused on neophobia for multiple reasons. First, it may provide information on the overall strategy of an individual to cope with novel or challenging situations (including aggressiveness, speed of exploration, sensitivity to external stimuli and readiness to adjust behaviour to environmental changes; cf. Dingemanse et al., 2004). Second, within-pair similarity for such personality traits have been linked to fitness in avian species (Dingemanse et al., 2004; Burtka & Grindstaff, 2015). Third, neophobia is a personality trait that can be assessed easily. If sexual selection is indeed contributing to the maintenance of different behavioural types, a necessary pre-condition is that (independent of the actual choice mechanism) mating preferences should be non-random with respect to personality type of chosen males, choosing females or both.

2. Materials and methods

2.1. Study population and housing conditions

The study took place at Göd Biological Station of Eötvös Loránd University. Subjects were 63 adult female and 110 adult male domesticated zebra finches without prior breeding experience originating from 12 different breeders in Hungary and Romania. Subjects wore one numbered aluminium leg ring (A C Hughes, Hampton, UK) for individual identification and were at least 240 days old at the beginning of the experiments (zebra finches are fully mature at around 100 days post-hatching, Zann, 1996).

All female subjects were first tested with random sets of 4 males for mating preferences, and all subjects were tested for neophobia within 1–2 months thereafter (in zebra finches, neophobia has been shown to be consistent for a period up to 2 years; Schielzeth et al., 2011). With the exception of the mate choice trials, subjects were housed individually in cages (42 × 30 × 44 cm) prior to and throughout the study, so that birds could hear but could not see other birds in the room. Males were housed in two male-only rooms (one male per cage, 40 cages per room, size of room: 3.4 × 3.4 × 2.4 m). Females were also housed in individual cages in three different rooms (20 female cages per room, size of room: 5.1 × 3.4 × 2.4 m) that also housed cages with non-experimental breeding pairs. In all rooms, there was an artificial 14:10 hour light:dark cycle (lights started at 06.00 a.m., local time, using full-spectrum tube lights (NASLI, Prague, Czech Republic) connected to a timer) and ambient temperature was kept between 18–21°C. Seed mixture (Foreign finches basic, Deli Nature, Belgium) and water were provided *ad libitum* (also during tests, except during the short food deprivation in neophobia tests, see below). In addition, every 2–3 days birds were provided with soft bird food (including green leaves, egg-protein and vitamins: Nekton E and Nekton S, Pforzheim, Germany).

2.2. Mate preference test

Female mating preference was tested using an established four-way choice apparatus paradigm (following Swaddle & Cuthill, 1994; Figure 1). The apparatus consisted of a central neutral arena from which a focal female could enter four different choice chambers with wire mesh walls. Four males in their home cages were placed adjacent to the distant end of each choice chamber behind this wire mesh partition ('stimulus compartments' in Fig-

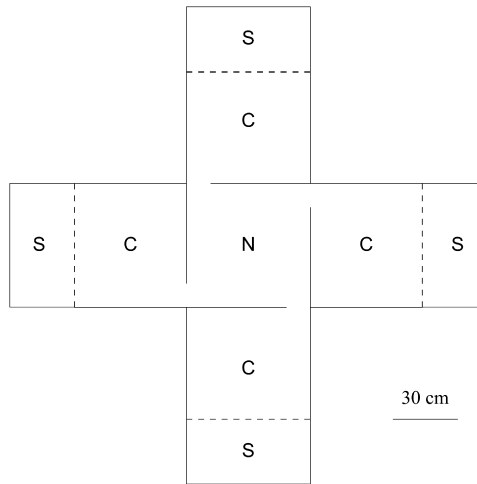


Figure 1. Plan view of the mate choice apparatus (Pogány et al., 2014b). The middle, neutral chamber (N) opens into four choice chambers (C). Stimulus compartments (S) housed one bird cage each, so males could be presented and easily exchanged between the different stimulus compartments while staying in their own cages. Infrared motion detectors were positioned on the top (centre) of each choice chamber and the neutral chamber (in the positions of letters 'C' and 'N'), facing downwards.

ure 1), such that they could not see each other and that the female could only see a stimulus male upon entering a given choice chamber. The testing phase was preceded by a familiarisation phase including females only; females were familiarized with the apparatus in groups of five for a total of 8 h, 1–8 days before their individual tests. The actual preference test of a female was run on two consecutive days with the same four males. Each testing day started with the female being moved into the apparatus and acclimatization between 08.00 a.m. and 08.45 a.m. The four stimulus males in her option set were then placed randomly adjacent to the four choice chambers and after 4 h of testing (between 9:00 a.m. and 1:00 p.m.) the opposing males' cages were exchanged and the second half day of testing started (between 1:15 p.m. and 5:15 p.m.). The next day started as the previous, but with each male's place rotated by 90 degrees. After 4 h males in opposite positions were exchanged once more, so that at the end of the second test day, each male had been in each position once for 4 h.

Allocation of stimulus males to focal females and female testing order was randomized, because we had no a priori prediction regarding the direction of

female preferences and whether it was uniform and directional (all females preferring the boldest of shyest males) or depending on females own personality type (in which case females at either end of the spectrum would never had males on offer that were shyer and bolder than themselves, while this would always be the case for females with intermediate scores) or how big a difference there had to be between stimulus males' (and subject females') personality traits. As we saw no a priori rules to construct comparable option sets for different females, we decided that four random males would yield the largest range of option sets without biasing the outcome towards a particular hypothesis (through the nature of the option set). Hence, males were assigned to stimulus sets randomly but if after randomization any of the envisaged stimulus males originated from the same breeder as the female, these males were individually replaced with another male randomly drawn from the pool until no stimulus male originated from the same breeder as the focal female. N.B. that each female was tested only once (for a two-day period) but most stimulus males were tested more than once (mean \pm SD = 2.3 ± 1.0 tests, range 1–6) but then in a different set of males.

During tests, a custom-built automatic infrared motion detector system and a ccd camera (Vidosec W-101, Euro Tech, Hungary) connected to a digital recorder (HDR 04 RP, Hunt Electronic, New Taipei City, Taiwan) recorded when, how often and for how long a female visited each chamber (a detailed description of the infrared detector system and its validation can be found in Pogány et al., 2014a).

2.3. Neophobia test

Several studies in zebra finches have shown repeatable interindividual differences in neophobia tests, involving novel objects placed in the vicinity of feeders (Schielzeth et al., 2010; David et al., 2011; Krause et al., 2018). As in previous studies, the neophobia tests were carried out in each subject's home cage. These tests took place after the mate choice tests (mean \pm SD days females: 24.5 ± 34 , range 2–118; males: 64.4 ± 47 , range 2–197 (with the exception of one male tested 45 days before its first mate choice test)). The novel object was one of two types of small flags (2×3 cm) of either plain unpainted metal ('plain') or painted with black and yellow stripes ('striped'). All tests took place between 8:00 a.m. and 4:00 p.m. and always started by removing all food from the cage. After 2 h of food deprivation,

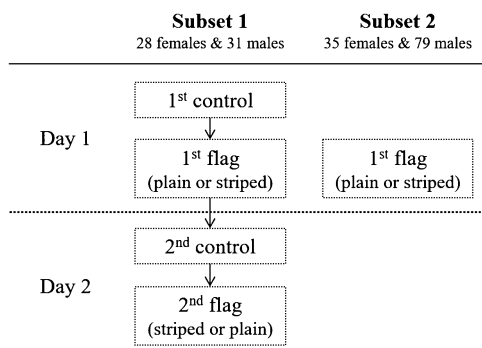


Figure 2. Overview and time line of neophobia tests. Subjects (63 females and 110 males) were randomly allocated to two subsets. Subset 1 individuals were repeatedly tested in no-flag (1st and 2nd control) and flag trials (1st and 2nd flag, with both flag types: plain and striped, in random order). Subset 2 individuals were tested only once with either a plain or striped flag.

the feeder was either returned unmanipulated (control tests) or with a flag attached (flag tests).

Birds were randomly assigned to either of two subsets (see schematic overview in Figure 2). Subset 1 ($N = 28$ females and 31 males) was used to validate the procedure: subjects were first tested in a control then in a flag test. To this end, the feeder and all food were removed for 2 h and then returned without flag (1st control test). Birds were now allowed to feed during the next 2 h. The feeder was then removed again to be returned after another 2 h, but now it had one of two types of small flags (plain or striped) attached (1st flag test). This was to establish whether behavioural changes in response to placing back the feeder with the novel object were indeed due to the novel object rather than the disturbance caused by the experimenter when replacing the feeder (these trials will be referred to as ‘validation’ trials). Whenever the feeder was replaced (with or without flag attached), latency to feeding was measured (see below). The next day, the procedure was repeated (2nd control and 2nd flag tests), now using the previously unused type of flag (i.e., if the 1st flag was plain, the 2nd flag was striped or vice versa; these trials will be referred to as ‘consistency’ tests). The remainder of the birds, henceforth called subset 2 ($N = 35$ females and 79 males) were tested only once: their feeder was removed and returned after two hours with a randomly chosen plain or striped flag attached (1st flag test; Figure 2).

In all trials, latency to feeding was observed in real time and also recorded by video camera (installed ca. 2 m from the cage at the time of food removal,

video equipment as described above). In all test trials, birds were observed until they started to eat from the feeder, or if they did not start feeding for 30 min, the trial was terminated. In either case, the time stamp of the video system at the start and end of the test was used to calculate the latency to feeding.

2.4. Statistical analyses

Statistical analyses were carried out using R 3.2.3 (R CoreTeam, 2015). Latency to first feeding (response variable) was analysed using Cox Mixed Models (R package “coxme”; Therneau, 2015a) and in Cox Models (R package “survival”; Therneau, 2015b) with occurrence of feeding as terminal event. Individuals that did not feed within the 30 min of a trial were treated as censored observations (1st flag test: $N = 28$ (of 89) and $N = 40$ (of 88) censored individuals with plain and striped flags, respectively). Latency to feeding was analysed in three separate models for validation, consistency and to test potential carry-over effects of the neophobia tests (Table 1). The Cox Mixed Model of validation included data from subset 1 only ($N = 28$ females and 31 males, Figure 2) and allowed us to compare the reaction to removing and returning the feeder with and without flag (control). The full model included experimental treatment (1st control or 1st flag) and sex as fixed factors and ring ID as random factor. To test for individual consistency in responses to novel objects the repeated flag tests of subset 1 individuals (Figure 2) were also analysed in a Cox Mixed Models. The full model included experimental treatment order (1st flag test or 2nd flag test), sex and flag type (plain or striped) as fixed factors and ring ID as random factor. Influential factors on latencies in 1st flag tests of all individuals (i.e., subset 1 + subset 2) were analysed in a Cox Model. The full model included time of the day, date of the test, condition (residuals of body mass regressed on tarsus length), flag type, sex and experimental group (subset 1 vs. subset 2) as fixed factors. Model selection was based on AIC values, and the effects of explanatory variables were analysed by likelihood ratio tests in all three survival analyses.

This analysis revealed that sex and flag type induced differences in latencies, and that responses in the two experimental groups tended to differ: subset 1 individuals, that were tested in a control (no-flag) test first, started to feed faster in their 1st flag trial than subset 2 individuals that had not been tested previously (see results and Table 1). To control for the absolute

Table 1.

Novel object tests using latency to feeding as response variable. Parameter estimates of three final models (following AIC-based model selection) are given for validation, consistency in novel object tests, and the influential factors on latency, separately. Hazard ratio $\text{Exp}(\beta)$ between levels of a given fixed effect with 95% confidence interval, and χ^2 and p values of likelihood ratio tests (LRT) of models with and without the explanatory variable are given.

Model	$\text{Exp}(\beta)$ [95% CI]	LRT χ^2	LRT p
Validation			
Trial (1st control \rightarrow 1st flag)	0.39 [0.24; 0.61]	16.91	<0.001
Consistency			
Flag type (plain \rightarrow striped)	0.22 [0.13; 0.37]	28.8	<0.001
Influential factors in 1st flag test			
Flag type (plain \rightarrow striped)	0.56 [0.38; 0.84]	8.34	0.004
Sex (male \rightarrow female)	1.62 [1.09; 2.39]	5.56	0.018
Experimental group (subset 1 \rightarrow subset 2)	0.68 [0.46; 1.03]	3.18	0.075

differences arising by sex, flag type and experimental groups in our subsequent analyses, we ranked feeding latency (in 1st flag test of all individuals) within each of the six combinations of sex, flag type and experimental group separately (with shy individuals receiving low and bold individuals receiving high ranks). Since sample sizes varied between these groups (mean $N \pm \text{SD} = 21.8 \pm 12.1$; range 11–44), the quantiles of these ranks ('neophobia quantiles' henceforth) were calculated by dividing ranks by the maximum rank within each group. Therefore, the range of neophobia quantiles was 0–1 in each group and high neophobia quantile values represented the less neophobic (bolder) individuals. These quantiles were used in all analyses of mating preferences.

For the mate choice trials, we calculated relative time for each male separately. First, we summed the total of times with each male for the 4×4 hour blocks (mean $\pm \text{SD} = 3.1 \pm 2.3$ h, range 0.1–12 h). This gave us time spent with each male and the total time spent choosing (the sum of the values for the four males). The time spent in the neutral chamber was omitted from the calculations of total time (% time in neutral chamber of the 16 h mate choice test, mean $\pm \text{SD} = 22.1 \pm 13.2\%$, $N = 63$). Relative time spent with and number of visits to a particular male were moderately correlated (Pearson's correlation coefficients of four data point pairs (relative time vs. relative number of visits) per female: mean $r \pm \text{SE} = 0.64 \pm 0.06$, $N = 63$ females). In line with previous work we used time spent with a focal male as measure

of female preference as this measure in choice apparatus predicts pair formation in aviaries (Clayton, 1990; Witte, 2006). Using the R package “rptR” (Stoffel et al., 2017), we confirmed our mate choice protocol by investigating repeatability of total time females spent with the four offered males during the four 4 hour blocks (i.e., whether females ‘followed’ their most preferred male, whenever males were moved). Likewise, we calculated repeatability estimates between the two test days of females.

Directional preference for males with low neophobia values was tested in Linear Mixed Models (LMM, R package “nlme”; Pinheiro et al., 2016) with relative time as response variable (arcsine transformed), male neophobia quantile as a covariate and female ring ID as a random factor.

To test assortative mate choice for neophobia, the neophobia quantile (response) of the preferred male (i.e., the male with the highest relative time value for a given female) was analysed in Linear Models (LM) with female neophobia quantile as explanatory variable. By selecting the offered four males randomly for a given female to choose from, some females might have had more chance to choose assortatively than others for two reasons. First, if there was high variation among the four males’ neophobia levels, it might have been easier for females to discriminate between males. Second, choosing assortatively is possible only if at least one of the offered males has similar neophobia to that of the female, i.e., the lower the minimum distance between neophobia quantile of the female and that of the four males, the more potential there is for personality-assortative choice (Figure 3). Therefore, two separate weights were calculated for each female’s test based on the standard deviation of the neophobia quantile of the four males (W_{SD}) and minimum distance of the neophobia quantile of the female and the four males in each set (W_{MND}). We ran four separate LMs including the same terms but either without weights or with W_{SD} , W_{MND} or the mean of W_{SD} and W_{MD} entered as weights; we report results of the model that provided the best fit.

In addition to the main question of our study, we used data collected on female movements in the choice arena during preference tests to analyse female activity levels and sampling strategies which has been implied to covary with personality (David & Cezilly, 2011). First, we analysed the repeatability of the number of switches between all compartments of the offered males between test day 1 and test day 2 (using R package “rptR”; Stoffel et al., 2017). Second, we correlated total number of switches over the two test days with the female’s neophobia rank.

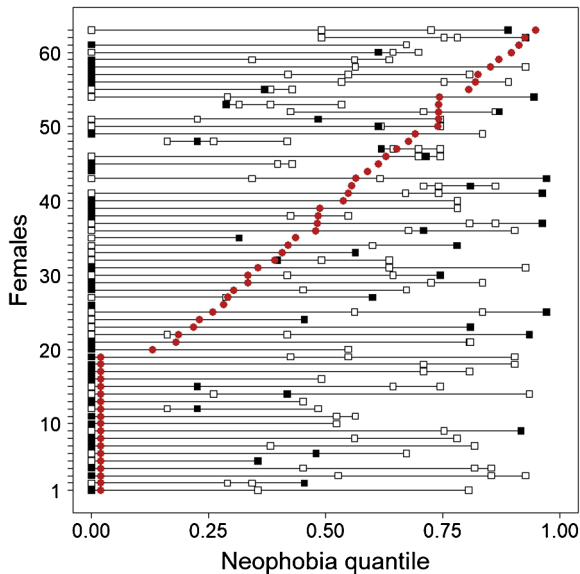


Figure 3. Assortative mating preference for neophobia and opportunity for assortment in our experiment. Squares indicate the neophobia quantile scores of the offered four males: open squares are the three non-preferred, whereas the filled square is the preferred male. Females (indicated by red filled circles) are ordered along y-axis based on their neophobia quantiles (from the most neophobic to the least neophobic). Symbols are staggered for the most neophobic females (i.e., at 0 on the neophobia axis) to avoid overlapping with the most neophobic males.

2.5. Ethical note

The study was conducted in line with national laws and the ethical guidelines of ASAB and of our respective home universities. Our study was revised and permitted by the Ethical Board of Eötvös Loránd University (permit no.: ELTE MÁB 02/2014). The 2 h food deprivations during the neophobia tests were implemented to increase feeding motivation. A food deprivation of 2 h is unlikely to compromise zebra finch welfare or health, as domesticated zebra finches on ad libitum feeding conditions only spent about 8.5% of their daily time budget on feeding even when housed outdoors at low winter temperatures (Koetsier & Verhulst, 2011). In addition, an experimental study where food was removed daily for 2 h during a two-week period found no effect on overall mass or daily mass fluctuations (Dall & Witter, 1998).

3. Results

3.1. Novel object tests

The novel object clearly induced neophobia: the validation revealed that after temporary removal of the feeders for two hours, birds returned to the feeders with a flag later than to feeders without flag (mean \pm SE latency with flag vs. without flag: 12.4 ± 1.7 min vs. 4.1 ± 0.9 min; Table 1, Validation).

Individual responses in repeated flag trials were consistent: 1st flag and 2nd flag tests of the same individuals revealed consistent latency to feeding (LR test of the final model with sex and flag type as explanatory variables with and without random term: $\chi_1^2 = 20.11$, $p < 0.001$, Table 1, Consistency; Spearman rank correlation between latency with 1st and 2nd flag: $r_s = 0.34$, $N = 59$, $p = 0.008$).

Flag type affected latency; the comparison of the latencies in 1st flag tests of all individuals revealed that latency to feeding was longer with a striped flag than with a plain one, and longer for males than for females (Table 1, Influential factors in 1st flag test). In addition, individuals that had received a control (no-flag) treatment previously tended to start feeding faster in their 1st flag test than individuals without previous control treatment.

3.2. Female mating preferences

Females spent $49.5 \pm 2\%$ (mean \pm SE) of their choice time (calculated as the sum of time in front of all four males) in front of their most-preferred male. Female preference (more precisely, time spent with each offered male) was repeatable both between the four 4-h test blocks ($r = 0.45$, 95% CI [0.38; 0.51], $p < 0.001$) and between the two test days ($r = 0.61$, 95% CI [0.52; 0.67], $p < 0.001$).

There was no directional preference for males with low neophobia: females did not spend overall more time with males with low than high neophobia (LMM, male neophobia quantile: $b = -0.03 \pm 0.04$, $t_{188} = -0.67$, $p = 0.505$). Instead, females showed assortative preferences: females' own neophobia scores predicted best the neophobia score of the male she spent most time with (LM weighted with the minimum neophobia quantile distance W_{MND} between the female and the four males, male boldness quantile: $b = 0.36 \pm 0.13$, $t_{61} = 2.66$, $p = 0.010$; Figure 4). Since qualitative evaluation of our results (see Figure 4) suggests that assortment may mostly be driven by the individuals showing the most pronounced neophobia (i.e., the

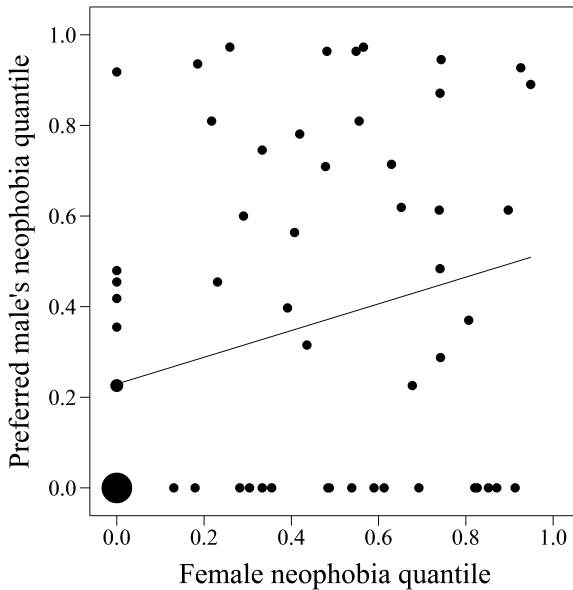


Figure 4. Assortative mating preference for neophobia in the zebra finch. Female neophobia predicts that of her preferred male. Neophobia values are quantiles of latency to feeding in the novel object test (for calculation see methods). Low values represent more neophobic individuals. Preferred male was established based on relative time in front of males. Symbol size corresponds to overlapping data points.

individuals that did not feed within the 30-min observation time and thus were censored), we tested for independence between occurrence of feeding in the neophobia tests between females and their preferred males. This analysis confirmed our previous result by showing a weak trend for non-independence ($\chi^2_1 = 3.21$, $p = 0.073$; Figure 5), and revealed ca. two-times more cases when both the female and her preferred male fed than cases in which both the male and the female were censored i.e., in which none of the sexes fed (43 vs. 19% of $N = 63$ cases, respectively; Figure 5).

3.3. Female activity levels and strength of preference based on their neophobia

Females' activity levels were assessed by counting the total number of switches between all compartments per day. Females showed substantial inter-individual variation in how often they visited the different compartments in total during the two test days (mean \pm SD = 1168 ± 984 visits, range 29–5112) and this behaviour was highly repeatable between test days

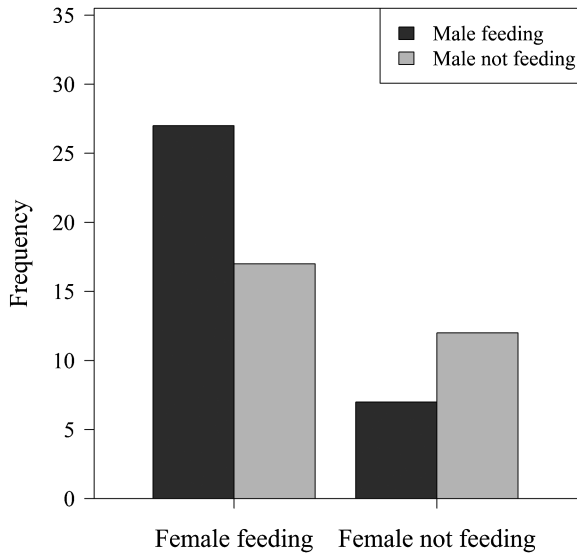


Figure 5. The number of females that did and did not feed within 30 min in the neophobia test and the number of their preferred males that would also feed/not feed. Categorisation refers to the 30 min test period from feeder replacement with a novel object attached. Preferred males were established based on time the female spent in front of each of four offered males in separate mate choice trials.

($r = 0.70$, 95% CI [0.55; 0.80], $p < 0.001$). Females' activity levels were independent of their neophobia: females' ranks based on the total number of visits to different compartments were not correlated with their neophobia quantiles (Spearman $r_s = 0.02$, $N = 63$, $p = 0.856$). Also, the strength of female preference was independent of their own neophobia score (Spearman $r_s = -0.03$, $N = 63$, $p = 0.816$).

4. Discussion

We here tested whether female zebra finches expressed non-random mating preferences with respect to their own and their chosen mates' neophobia levels. When allowed to choose between four different unfamiliar males, females chose males with neophobia levels similar to themselves more often than expected by chance. Our experimental test of neophobia, consisting of unfamiliar flags attached to otherwise familiar feeders, delayed approach to these feeders, and individual differences in this behaviour were consistent. Next to individual differences, birds' sex and types of stimulus flags

also affected feeding latencies. The yellow-black striped flag caused longer latencies than the plain flag possibly because its similarity to natural aposematic warning coloration (Schuler & Roper, 1992; Ruxton et al., 2005; Ham et al., 2006). With either flag type attached, females approached the feeder faster than males. This fits other observations in this species, for instance, the higher vulnerability to food stress in female nestlings (Martins, 2004; Wolf & Weissing, 2012) and sex-specific social foraging strategies in adulthood (Katz & Lachlan, 2003; Schuett & Dall, 2009; Riebel et al., 2012) and is in line with predictions from life history theory (Wolf & Weissing, 2010).

In our study, female zebra finches did not prefer less neophobic males in general but males that were similar to themselves. In line with this, Schuett et al. (2009) reported improved breeding performance in personality matched zebra finch pairs while McCowan et al. (2014) observed higher breeding success in pairs with more explorative males. The latter, however, tested exploration and breeding success in large aviary housed groups, while exploration in our study and Schuett et al. (2011) was tested in cages and single individuals and/or pairs. Schuett & Dall (2009) report that exploration scores were different for individually versus pair-tested birds, but repeatable in either context, suggesting that solo versus group exploration tests different personality aspects or is confounded by co-occurring social learning (Katz & Lachlan, 2003; Riebel et al., 2012) which could also explain the lack of correlation between exploration of a novel environment in the laboratory versus subsequent discoveries of novel feeders in the field in a population of wild zebra finches (McCowan et al., 2015). In the present study, focal birds were tested individually for neophobia. The observed interindividual differences in these tests covaried with the outcomes of the 4-way mating preference tests where females with relative high (low) neophobia preferentially chose males of likewise relatively higher (lower) neophobia. Such assortative preferences with respect to personality traits have been suggested to be one possible mechanism underlying non-random pairings of different personality types observed in the wild (Dingemanse et al., 2004; Both et al., 2005; Gabriel & Black, 2012) and thus contribute to maintaining within-population variation of such traits (Dingemanse & Réale, 2005; Schuett et al., 2011a) but so far have only been reported in two spider species (Pruitt & Riechert, 2009a,b; Kralj-Fiser et al., 2013) and were absent in wild zebra finches personality typed for exploration (McCowan et al., 2015, $N = 33$ pairs).

Zebra finches are an important model of avian mate choice (Riebel, 2009; Griffith & Buchanan, 2010). Previous studies addressing putative links between personality and mate choice have as yet not involved preference tests by and for unmanipulated individuals in involving personality typed individuals of both sexes. However, a number of studies tested aspects of mate choice or sampling behaviour in personality typed females. Schuett et al. (2011b) tested whether females would prefer males that they had seen move around a cage freely ('explorers') over those that had been restrained in a small box, as well as in control conditions involving both males being allowed to move freely. Females showed random preferences with respect to males' actual exploration scores (i.e., in the control condition), but high exploring females' choices avoided males that had been restrained prior to the test. While it is unclear by which mechanisms this manipulation affected female choice, the non-independence of females' mating preferences from their own exploration levels was also found in another population (David & Cezilly, 2011) in which females' search tactics (selectivity, preference strength and consistency) and female explorative tendencies covaried. Unfortunately, this study did not report males' personality and provide no information on whether females preferred males with similar personalities. In an aviary study, Schielzeth et al. (2011) found no assortative mate choice for neophilia (measured as approach to a new object in the home cage), or fitness benefits for pairs with similar neophilia scores in zebra finches when allowed to breed freely in small communal aviaries. However, inter- and intrasexual competition in small aviary groups quickly remove potentially preferred partners from the available pools of mates and could have masked mating preferences in the aviary but not in the unconstrained choice context of the 4-way experimental apparatus used in our tests. Without additional studies replicating our results for this and/or other personality traits, we cannot yet explain the differences between the assortative preferences for neophobia in this study and an absence thereof for neophilia (Schielzeth et al., 2011) as measured traits and preference testing methods differed. In addition, although neophilia and neophobia are generally assumed to be both part of the exploration-avoidance axis (Réale et al., 2007) this is not the case for all species studied to date and should be validated per species (Carter et al., 2013). In zebra finches, neophobia and exploration of novel environments were not correlated within individuals in two different studies in two different populations (Martins et al., 2007; David et al., 2011). In yet another study

and in a different study population (David & Cezilly, 2011) exploration in a novel environment and activity in a mate choice apparatus were correlated (although exploration and neophobia were not, David et al., 2011). We also observed consistent inter-individual differences in activity and total number of visits (which might indicate different mate sampling behaviour), but no correlation between neophobia levels and females' activity rates which is in line with earlier observations (David et al., 2011).

A number of studies have reported enhanced breeding performances of matched pairs (ca. 37% higher reproductive success) in captive zebra finch pairs resulting from free mate choice (i.e., mate choice for compatibility, see Ihle et al., 2015). Similar results were reported from experimentally matched personality- and quality-assortative pairs in other captive zebra finches (Holveck & Riebel, 2010; Schuett et al., 2011a) as well as in natural pairs of other bird species (Dingemanse et al., 2004; Both et al., 2005; Gabriel & Black, 2012). Higher reproductive success could result from increased behavioural coordination and cooperation (resulting in e.g., similar nest defence strategies as in eastern bluebirds *Sialia sialis*; Burtka & Grindstaff, 2015), and reduced sexual conflict over care provisioning (Royle et al., 2002, 2010; Schuett et al., 2010) which would confer adaptive advantages to mating preferences that are non-random with respect to personality traits (Schuett et al., 2010). This however, raises the question as to how individuals might recognise different personalities. In our 16-h-long mating preference tests, both males and females were well acclimated to the choice chamber, making it unlikely that birds' could directly observe neophobic behaviour in their prospective mates. Moreover, screening potential mates' general behaviour may be more time consuming (as opportunities have to occur for males to show and for females to watch explorative behaviour) than basing mate choice on much more prominent displayed secondary sexual signals (cf., Laubu et al., 2017). In the type of choice chambers we used in this study, substantial variation in female preference can be explained by variation in male secondary sexual traits like song or beak colour (Riebel, 2009; Simons & Verhulst, 2011), raising the question as to whether variation in mating signals provides information about personality (Godin & Dugatkin, 1996; Naguib et al., 2010; Schuett et al., 2010). Clearly, such hypothesized associations between male mating and personality traits require more study in the future, only then can we answer the question of whether female preference was driven directly by male personality, or whether other behavioural

or morphological traits that covaried with neophobia guided female' choices. Moreover, zebra finches show mutual mate choice (for references and discussion see Holveck *et al.*, 2011), so follow-up studies should test whether males' like females' mating preferences covary with personality traits.

For now, our study showed that female zebra finches, choosing from unmanipulated, randomly assigned option sets of 4 males showed mating preferences that are non-random with respect to their and their preferred male's neophobia scores. Females associated more often than expected by chance with males that had similar neophobia levels to themselves. If such personality-assortative mating preferences are also leading to successful pair formation and improved breeding, then one way personality traits could be maintained is by sexual selection through mate choice. Future research needs to address the relatively unexplored link between sexual selection and personality, in particular, the questions of how frequent assortative mate choice for personality traits is in various species, and to what extent it affects fitness. Furthermore, theoretical and empirical work are needed to estimate the relative significance of sexual versus natural selection in maintaining within-population variation in personality traits.

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References

- Amy, M., Sprau, P., de Goede, P. & Naguib, M. (2010). Effects of personality on territory defence in communication networks: a playback experiment with radio-tagged great tits. — *Proc. Roy. Soc. Lond. B: Biol. Sci.* 277: 3685-3692.
- Both, C., Dingemans, N.J., Drent, P.J. & Tinbergen, J.M. (2005). Pairs of extreme avian personalities have highest reproductive success. — *J. Anim. Ecol.* 74: 667-674.

- Burley, N. (1983). The meaning of assortative mating. — *Ethol. Sociobiol.* 4: 191-203.
- Burtka, J.L. & Grindstaff, J. (2015). Similar nest defence strategies within pairs increase reproductive success in the eastern bluebird, *Sialia sialis*. — *Anim. Behav.* 100: 174-182. DOI:10.1016/j.anbehav.2014.12.004.
- Carter, A.J., Feeney, W.E., Marshall, H.H., Cowlshaw, G. & Heinsohn, R. (2013). Animal personality: what are behavioural ecologists measuring? — *Biol. Rev.* 88: 465-475.
- Clayton, N.S. (1990). Mate choice and pair formation in Timor and Australian mainland zebra finches. — *Anim. Behav.* 39: 474-480.
- Dall, S.R.X. & Witter, M.S. (1998). Feeding interruptions, diurnal mass changes and daily routines of behaviour in the zebra finch. — *Anim. Behav.* 55: 715-725.
- David, M., Auclair, Y. & Cezilly, F. (2011). Personality predicts social dominance in female zebra finches, *Taeniopygia guttata*, in a feeding context. — *Anim. Behav.* 81: 219-224.
- David, M. & Cezilly, F. (2011). Personality may confound common measures of mate-choice. — *PLoS One* 6: e24778.
- Dingemanse, N.J., Both, C., Drent, P.J. & Tinbergen, J.M. (2004). Fitness consequences of avian personalities in a fluctuating environment. — *Proc. Roy. Soc. Lond. B: Biol. Sci.* 271: 847-852.
- Dingemanse, N.J., Kazem, A.J.N., Réale, D. & Wright, J. (2010). Behavioural reaction norms: animal personality meets individual plasticity. — *Trends Ecol. Evol.* 25: 81-89.
- Dingemanse, N.J. & Réale, D. (2005). Natural selection and animal personality. — *Behaviour* 142: 1159-1184.
- Forstmeier, W. & Birkhead, T.R. (2004). Repeatability of mate choice in the zebra finch: consistency within and between females. — *Anim. Behav.* 68: 1017-1028.
- Gabriel, P.O. & Black, J.M. (2012). Behavioural syndromes, partner compatibility and reproductive performance in Steller's jays. — *Ethology* 118: 76-86.
- Garamszegi, L.Z., Eens, M. & Török, J. (2008). Birds reveal their personality when singing. — *PLoS One* 3: e2647.
- Godin, J.G.J. & Dugatkin, L.A. (1996). Female mating preference for bold males in the guppy, *Poecilia reticulata*. — *Proc. Natl. Acad. Sci. USA* 93: 10262-10267.
- Gosling, S.D. (2001). From mice to men: what can we learn about personality from animal research? — *Psychol. Bull.* 127: 45-86.
- Griffith, S.C. & Buchanan, K.L. (2010). Maternal effects in the zebra finch: a model mother reviewed. — *Emu* 110: 251-267.
- Groothuis, T.G. & Carere, C. (2005). Avian personalities: characterization and epigenesis. — *Neurosci. Biobehav. Rev.* 29: 137-150.
- Ham, A.D., Ihalainen, E., Lindstrom, L. & Mappes, J. (2006). Does colour matter? The importance of colour in avoidance learning, memorability and generalisation. — *Behav. Ecol. Sociobiol.* 60: 482-491.
- Holtmann, B., Lagisz, M. & Nakagawa, S. (2017). Metabolic rates, and not hormone levels, are a likely mediator of between-individual differences in behaviour: a meta-analysis. — *Funct. Ecol.* 31: 685-696. DOI:10.1111/1365-2435.12779.
- Holveck, M.J., Geberzahn, N. & Riebel, K. (2011). An experimental test of condition-dependent male and female mate choice in zebra finches. — *PLoS One* 6: e23974.

- Holveck, M.J. & Riebel, K. (2010). Low-quality females prefer low-quality males when choosing a mate. — *Proc. Roy. Soc. Lond. B: Biol. Sci.* 277: 153-160.
- Ihle, M., Kempnaers, B. & Forstmeier, W. (2015). Fitness benefits of mate choice for compatibility in a socially monogamous species. — *PLoS Biol.* 13: 21.
- Johnson, K.V.A., Aplin, L.M., Cole, E.F., Farine, D.R., Firth, J.A., Patrick, S.C. & Sheldon, B.C. (2017). Male great tits assort by personality during the breeding season. — *Anim. Behav.* 128: 21-32.
- Katz, M. & Lachlan, R.F. (2003). Social learning of food types in zebra finches (*Taeniopygia guttata*) is directed by demonstrator sex and feeding activity. — *Anim. Cogn.* 6: 11-16.
- Koetsier, E. & Verhulst, S. (2011). A simple technique to manipulate foraging costs in seed-eating birds. — *J. Exp. Biol.* 214: 1225-1229.
- Kralj-Fiser, S., Mostajo, G.A.S., Preik, O., Pekar, S. & Schneider, J.M. (2013). Assortative mating by aggressiveness type in orb weaving spiders. — *Behav. Ecol.* 24: 824-831.
- Krause, E.T. & Naguib, M. (2011). Compensatory growth affects exploratory behaviour in zebra finches, *Taeniopygia guttata*. — *Anim. Behav.* 81: 1295-1300.
- Krause, E.T., Bischof, H.-J., Engel, K., Golüke, S., Maraci, Ö., Mayer, U., Sauer, J. & Caspers, B. (2018). Olfaction in the zebra finch (*Taeniopygia guttata*): what is known and further perspectives. — *Adv. Stud. Behav.* 50: 37-85.
- Laubu, C., Dechaume-Moncharmont, F.X., Motreuil, S. & Schweitzer, C. (2016). Mismatched partners that achieve postpairing behavioral similarity improve their reproductive success. — *Sci. Adv.* 2: 7.
- Laubu, C., Schweitzer, C., Motreuil, S., Louapre, P. & Dechaume-Moncharmont, F.X. (2017). Mate choice based on behavioural type: do convict cichlids prefer similar partners? — *Anim. Behav.* 126: 281-291.
- Martins, T.L.F. (2004). Sex-specific growth rates in zebra finch nestlings: a possible mechanism for sex ratio adjustment. — *Behav. Ecol.* 15: 174-180.
- Martins, T.L.F., Roberts, M.L., Giblin, I., Huxham, R. & Evans, M.R. (2007). Speed of exploration and risk-taking behavior are linked to corticosterone titres in zebra finches. — *Horm. Behav.* 52: 445-453.
- McCowan, L.S., Rollins, L.A. & Griffith, S.C. (2014). Personality in captivity: more exploratory males reproduce better in an aviary population. — *Behav. Proc.* 107: 150-157.
- McCowan, L.S.C., Mainwaring, M.C., Prior, N.H. & Griffith, S.C. (2015). Personality in the wild zebra finch: exploration, sociality, and reproduction. — *Behav. Ecol.* 26: 735-746, <https://doi.org/10.1093/beheco/aru239>.
- Naguib, M., Kazek, A., Schaper, S.V., van Oers, K. & Visser, M.E. (2010). Singing activity reveals personality traits in great tits. — *Ethology* 116: 763-769.
- Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D. & R Core Team (2016). nlme: Linear and non-linear mixed effects models. — R package version 3.1-125. R Foundation for Statistical Computing, Vienna, available online at <https://CRAN.R-project.org/package=nlme>.
- Pogány, Á., Heszberger, J., Szurovecz, Z., Vincze, E. & Székely, T. (2014a). An infrared motion detector system for lossless real-time monitoring of animal preference tests. — *Acta Biol. Hung.* 65: 385-395.

- Pogány, Á., Szurovecz, Z., Vincze, E., Barta, Z. & Székely, T. (2014b). Mate preference does not influence reproductive motivation and parental cooperation in female zebra finches. — *Behaviour* 151: 1885-1901.
- Pruitt, J.N. & Riechert, S.E. (2009a). Male mating preference is associated with risk of pre-copulatory cannibalism in a socially polymorphic spider. — *Behav. Ecol. Sociobiol.* 63: 1573-1580.
- Pruitt, J.N. & Riechert, S.E. (2009b). Sex matters: sexually dimorphic fitness consequences of a behavioural syndrome. — *Anim. Behav.* 78: 175-181.
- Pruitt, J.N., Riechert, S.E. & Harris, D.J. (2011). Reproductive consequences of male body mass and aggressiveness depend on females' behavioral types. — *Behav. Ecol. Sociobiol.* 65: 1957-1966.
- R CoreTeam (2015). R: a language and environment for statistical computing. — R Foundation for Statistical Computing, Vienna, available online at <https://cran.r-project.org/>.
- Rangassamy, M., Dalmas, M., Féron, C., Gouat, P. & Rödel, H.G. (2015). Similarity of personalities speeds up reproduction in pairs of a monogamous rodent. — *Anim. Behav.* 103: 7-15.
- Réale, D., Reader, S.M., Sol, D., McDougall, P.T. & Dingemanse, N.J. (2007). Integrating animal temperament within ecology and evolution. — *Biol. Rev.* 82: 291-318.
- Riebel, K. (2009). Song and female mate choice in zebra finches: a review. — *Adv. Study Behav.* 40: 197-238.
- Riebel, K., Holveck, M.J., Verhulst, S. & Fawcett, T.W. (2010). Are high-quality mates always attractive? State-dependent mate preferences in birds and humans. — *Commun. Integr. Biol.* 3: 271-273.
- Riebel, K., Spierings, M.J., Holveck, M.J. & Verhulst, S. (2012). Phenotypic plasticity of avian social-learning strategies. — *Anim. Behav.* 84: 1533-1539.
- Royle, N.J., Hartley, I.R. & Parker, G.A. (2002). Sexual conflict reduces offspring fitness in zebra finches. — *Nature* 416: 733-736.
- Royle, N.J., Schuett, W. & Dall, S.R.X. (2010). Behavioral consistency and the resolution of sexual conflict over parental investment. — *Behav. Ecol.* 21: 1125-1130.
- Ruxton, G.D., Sherratt, T.N. & Speed, M.P. (2005). *Avoiding attack: the evolutionary ecology of crypsis, warning signals and mimicry.* — Oxford University Press, New York, NY.
- Schielzeth, H., Bolund, E., Kempnaers, B. & Forstmeier, W. (2011). Quantitative genetics and fitness consequences of neophilia in zebra finches. — *Behav. Ecol.* 22: 126-134.
- Schuett, W. & Dall, S.R.X. (2009). Sex differences, social context and personality in zebra finches, *Taeniopygia guttata*. — *Anim. Behav.* 77: 1041-1050.
- Schuett, W., Dall, S.R.X. & Royle, N.J. (2011a). Pairs of zebra finches with similar "personalities" make better parents. — *Anim. Behav.* 81: 609-618.
- Schuett, W., Godin, J.G.J. & Dall, S.R.X. (2011b). Do female zebra finches, *Taeniopygia guttata*, choose their mates based on their "personality"? — *Ethology* 117: 908-917.
- Schuett, W., Tregenza, T. & Dall, S.R.X. (2010). Sexual selection and animal personality. — *Biol. Rev.* 85: 217-246.
- Schuler, W. & Roper, T.J. (1992). Responses to warning coloration in avian predators. — *Adv. Study Behav.* 21: 111-146.

- Simons, M.J.P. & Verhulst, S. (2011). Zebra finch females prefer males with redder bills independent of song rate — a meta-analysis. — *Behav. Ecol.* 22: 755-762.
- Snijders, L., van Rooij, E.P., Burt, J.M., Hinde, C.A., van Oers, K. & Naguib, M. (2014). Social networking in territorial great tits: slow explorers have the least central social network positions. — *Anim. Behav.* 98: 95-102.
- Spoon, T.R., Millam, J.R. & Owings, D.H. (2006). The importance of mate behavioural compatibility in parenting and reproductive success by cockatiels, *Nymphicus hollandicus*. — *Anim. Behav.* 71: 315-326. DOI:10.1016/j.anbehav.2005.03.034.
- Stoffel, M.A., Nakagawa, S. & Schielzeth, H. (2017). rptR: repeatability estimation and variance decomposition by generalized linear mixed-effects models. — *Methods Ecol. Evol.* 8: 1639-1644.
- Swaddle, J.P. & Cuthill, I.C. (1994). Preference for symmetrical males by female zebra finches. — *Nature* 367: 165-166.
- ten Cate, C. & Mug, G. (1984). The development of mate choice in zebra finch females. — *Behaviour* 90: 125-150.
- Therneau, T.M. (2015a). coxme: Mixed Effects Cox Models. R package v. 2.2-5. — R Foundation for Statistical Computing, Vienna, available online at <https://cran.r-project.org/package=coxme>.
- Therneau, T.M. (2015b). A package for survival analysis in S. R package v. 2.38. — R Foundation for Statistical Computing, Vienna, available online at <https://cran.r-project.org/package=survival>.
- Verbeek, M.E.M., Drent, P.J. & Wiepkema, P.R. (1994). Consistent individual differences in early exploratory behavior of male great tits. — *Anim. Behav.* 48: 1113-1121.
- Wilson, D.S., Clark, A.B., Coleman, K. & Dearstyne, T. (1994). Shyness and boldness in humans and other animals. — *Trends Ecol. Evol.* 9: 442-446.
- Witte, K. (2006). Time spent with a male is a good indicator of mate preference in female zebra finches. — *Ethol. Ecol. Evol.* 18: 195-204.
- Wolf, M. & Weissing, F.J. (2010). An explanatory framework for adaptive personality differences. — *Philos. Trans. Roy. Soc. Lond. B: Biol. Sci.* 365: 3959-3968.
- Wolf, M. & Weissing, F.J. (2012). Animal personalities: consequences for ecology and evolution. — *Trends Ecol. Evol.* 27: 452-461.
- Zann, R.A. (1996). The zebra finch: a synthesis of field and laboratory studies. — Oxford University Press, Oxford.