Social niche shapes social behavior and cortisol concentrations during adolescence in female guinea pigs

Taylor L. Rystrom[1], S. Helene Richter[1], Norbert Sachser[1], Sylvia Kaiser[1]

[1]Department of Behavioural Biology, University of Münster, Münster, Germany

Corresponding author: Taylor L. Rystrom; rystrom@uni-muenster.de

ORCIDs:

Taylor L. Rystrom: 0000-0002-9939-6628

Sylvia Kaiser: 0000-0003-2770-7367

Acknowledgments

This research was funded by the German Research Foundation (DFG) as part of the SFB TRR 212 (NC^3) – Project numbers 316099922 and 396777165 (responsible PIs: S.K. and N.S.). We thank Sabine Kruse for conducting the endocrine analysis. We would also like to thank Hanna Sicking and Kimberley Kubski for animal caretaking and assistance with blood samples.

Abstract

Individualized social niches arise in social groups and are associated with behavior and hormone (e.g. cortisol) concentrations. During sensitive life phases, social interactions can have a profound impact on the development of social behavior. Focusing on adolescence, we aimed to investigate the relationship between the social niche, social behavior, and cortisol concentrations (baseline and response to challenge) in female guinea pigs. Females were pair-housed in early adolescence (initial social pair formation), and a social niche transition occurred after six weeks by replacing the partner with either a larger or smaller female, forcing the focal female to become dominant or subdominant. We show that social behavior and cortisol concentrations were influenced by dominance status. Dominant females were more aggressive in the initial social pairs, and this was immediately reshaped after the social niche transition. Submissive behavior toward the new social partner was also rapidly adjusted after the social niche transition. However, submissive behavior was not entirely reshaped because the previous dominance status from the initial social pair still affected the extent of submissive behavior three weeks after the social niche transition. Regarding cortisol concentrations, baseline levels were shaped by the social niche in early adolescence with higher concentrations measured in dominant females. After the social niche transition, cortisol responsiveness significantly increased for the females who were forced to become subdominant relative to those forced to become dominant. These results add substantially to our understanding of the effect of social interactions during adolescence on social behavior and hormone concentrations in females.

Significance statement

Adolescence is a sensitive life phase during which social behavior and hormone concentrations can be shaped to the social environment, but this is not well-understood, especially in females. We

found that manipulating the dominance rank of female guinea pigs during adolescence triggered a rapid shift in aggressive behavior and a subtle but significant shift in cortisol responsiveness. Furthermore, while subdominant females were consistently more submissive than dominant females, dominance status in early adolescence had a long-lasting effect on submissive behavior even after dominance status was manipulated. Therefore, social behavior and cortisol concentrations were flexible in response to changes in the social environment during adolescence, and social experiences in early adolescence had long-term impacts on how individuals interact with social partners.

Keywords

social status, individual variation, cortisol reactivity, phenotypic plasticity, social environment, individualized niche

Introduction

Despite inhabiting the same physical and social environment, individuals living together in a social group differ in how they interact with one another. This unique interaction with the social environment influences the individual's fitness and constitutes the individualized social niche (Bergmüller and Taborsky 2010; Montiglio et al. 2013; Saltz et al. 2016; Trappes et al. 2022). The individualized social niche concept is integral to understanding the social environment on the individual level (Trappes et al. 2022). Social interactions among groupmates can generate individualized social niches (social niche specialization; see Bergmüller and Taborsky 2010). Social niches are often frequency-dependent and can be influenced by factors such as body size and age (Montiglio et al. 2013). For example, if group mates differ in body size and larger individuals have an advantage to fill a certain social niche, a relatively smaller group mate would be forced to adjust its behavior to conform to an alternative social niche. Therefore, the social niche of an individual is not independent from the social niches of its groupmates.

Social groups are structured by dominance hierarchies in many species. According to the social niche concept, dominant and subdominant individuals occupy different social niches (Bergmüller and Taborsky 2010; Montiglio et al. 2013; Saltz et al. 2016; Trappes et al. 2022). Dominance hierarchies can develop in two different ways. Put simply, the social dynamics hypothesis postulates that dominance hierarchies can develop via social interactions among group members (Chase et al. 2002). Meanwhile, the prior attributes hypothesis states that dominance hierarchies can arise from individual differences in factors such as body size, age, or previous social experience (Chase et al. 2002). Therefore, dominance hierarchies are an emergent property of groups (Drews 1993), and phenotypes of group members are as important as the individual's own phenotype when determining the dominance status of an individual (Wilson et al. 2011; Schneider et al. 2017). These phenotypes can subsequently be shaped by the outcome of dominance interactions, forming feedback loops which affect future dominance interactions (Dehnen et al. 2022). Dominance status has consequences for behavior (Briffa et al. 2015; Niemelä and Santostefano 2015), hormone concentrations (Creel 2001; Creel et al. 2013), and fitness outcomes (Chelini et al., 2011; Côté & Festa-Bianchet, 2001; Pusey et al., 1997).

Dominance hierarchies reduce conflict, protecting group members from costly escalated aggression (Tibbetts et al. 2022). Nevertheless, establishing and navigating dominance hierarchies is stressful, and glucocorticoid (e.g. cortisol) concentrations can reflect the challenges associated with position in the dominance hierarchy (Creel 2001; Goymann and Wingfield 2004; Creel et al. 2013; Korzan and Summers 2021). Controlled by the hypothalamic-pituitary-adrenal (HPA) axis, glucocorticoids fluctuate throughout the day within their baseline range but rapidly rise in response to acute challenges, with wide-ranging effects on biological processes such as metabolism, reproduction, and behavior (Sapolsky et al. 2000). The relationship between glucocorticoid concentrations and dominance status has been studied across a wide range of taxa. The direction of this relationship varies; sometimes dominant individuals have higher glucocorticoid levels (Creel et al. 1996; Fichtel et al. 2007) while other studies find that dominant individuals have lower glucocorticoid levels (Alonso et al. 2012; Williamson et al. 2019) or no relationship is detected (Arce et al. 2010; Barrette et al. 2012; Zipser et al. 2013; Mutwill et al. 2021; Rystrom et al. 2022). Even within the same species, males and females can differ in the direction of this relationship (Cavigelli and Caruso 2015; Ode et al. 2015). Therefore, whether dominant or subdominant individuals have higher glucocorticoid concentrations depends on the types of social behaviors expressed and the impact of status on reproductive success (Creel et al., 2013). Furthermore, when an association is detected, it is often unclear whether glucocorticoid concentrations are the cause or the consequence of dominance status (Sapolsky 2004). It has both been shown that manipulation of dominance status causally affects glucocorticoid concentrations (Kohn et al. 2016) and that selection for individuals with high or low HPA axis reactivity predicts aggression and dominance acquisition (Pottinger and Carrick 2001; Touma et al. 2008).

The relationship between dominance status and glucocorticoid concentrations is particularly interesting to investigate in adolescence. During adolescence, individuals increasingly interact with unrelated conspecifics and experience the social environment directly rather than indirectly via maternal cues. The first reproductive opportunities arise; therefore, adjusting phenotypes based on information from the current social environment carries fitness benefits (Fawcett and Frankenhuis 2015). Additionally, the HPA axis and social behavior undergo extensive maturation during adolescence, and development is affected by experiences in this life phase (Romeo 2018; Sachser et al. 2018). Indeed, social behavior (Zimmermann et al. 2017a; Jäger et al. 2019; Sachser et al. 2020) as well as glucocorticoid concentrations (Zimmermann et al. 2017a; Emmerson and Spencer 2018; Mutwill et al. 2020) are shaped by social experiences during adolescence and even into adulthood.

Guinea pigs (*Cavia aperea* f. *porcellus*) are an excellent model species to investigate the effect of social interactions on phenotypes during adolescence. Guinea pigs are highly social mammals and have a long adolescence during which environmental cues can be used to shape phenotypes. After weaning at 21 days of age, sexual maturity occurs at approximately 1 month in females and 70 days in males, and full body size is reached at 8-12 months (Kaiser et al. 2010). Males become socially mature enough to become dominant in complex social environments at the age of seven months (Sachser 1986), and it is well-established that the social environment during adolescence and adulthood shapes social behavior and hormone concentrations in male guinea pigs (Lürzel et al. 2010, 2011; Zimmermann et al. 2017a, b; Sachser et al. 2018; Mutwill et al. 2020, 2021). However, it is not known whether there are comparable developmental trajectories in females, and few studies have addressed this in females (but see Thyen and Hendrichs 1990; Rystrom et al. 2022). This sex disparity in research does not only apply to guinea pigs. In general, physiological and behavioral correlates of social status are less studied in female rodents

(Fulenwider et al. 2022), in part due to the effect of estradiol on dominance behaviors (Wise 1974; Faruzzi et al. 2005; Hamilton et al. 2015; Pandolfi et al. 2021) and HPA axis activity (McCormick and Mathews 2007; Heck and Handa 2019). Regarding social interactions in adolescence and adulthood, it is known that female guinea pigs form linear dominance hierarchies that are temporally stable, with older and larger females generally becoming dominant over smaller and younger females (Thyen and Hendrichs 1990; Rystrom et al. 2022).

We aimed to investigate whether social behavior and hormone concentrations are shaped by the dominance status attained during adolescence in females. To do this, young female guinea pigs were pair-housed upon weaning for the first six weeks of adolescence, and dominance status was determined. There was then a social niche transition, and a new partner female was pair-housed with the focal female to manipulate her dominance status. We hypothesized that while dominant and subdominant females would have distinct social behavior profiles, the dominance status attained in the first social pair would have a long-lasting effect on social behavior observed throughout the experiment. Furthermore, we predicted that cortisol concentrations would be shaped to the dominance status attained in the initial social pairs and reshaped after the social niche transition.

Methods

Animals and housing conditions

Animals used in this experiment were bred at the Department of Behavioural Biology in Münster from a breeding program of multi-colored shorthaired guinea pigs (*Cavia aperea* f. *porcellus*). Breeding groups consisted of one male with two to three females and their pre-weaned offspring. Therefore, parentage was known for all individuals. In total, 48 focal females and 75 partner females were included in this experiment. Focal females were pair housed with a partner female throughout the duration of the experiment, and different partners were used for the two experimental phases. Some partner females were used multiple times with different focal females.

Throughout the experiment, the focal female was housed with her partner in enclosures measuring 1 m by 0.5 m in size, with a wall height of 0.5 m. Walls were constructed of wood with an opaque red plastic section at the bottom (approximately 7 cm in height). The floor was covered with wood shavings (Tierwohl Super, J. Rettenmaier & Söhne GmbH + Co KG, Rosenberg, Germany), and food (Höveler Meerschweinchenfutter 10700, Höveler Spezialfutterwerke GmbH & Co. KG) and water with ascorbic acid was available *ad libitum*. Hay was replenished daily. Two small shelters were placed on opposite ends of the enclosure; one was made from wood and one was made from red transparent plastic. These shelters were rectangular (9 cm by 20 cm) with a height of 13 cm. As a door, one of the smaller walls was missing and only one female comfortably fit within each shelter. Lights were on daily from 7:00-19:00 and room conditions were kept at 20-24°C and 40-70% humidity.

Partner females were housed in one large group when not pair-housed with a focal female. The size of this enclosure fluctuated based on the number of partner females within. Various shelters were provided, and all other enclosure and room conditions were comparable to the experimental enclosures.

Experimental procedure

For this experiment, adolescent females were pair-housed for nine weeks, with a social niche transition after six weeks (for overview see Fig. 1). When the focal female was weaned (21 ± 3 days), she was transferred from her natal group to pair-housing with another female of a similar age. The maximum age difference between the two females was five days, and the partner female was also directly transferred from her natal group to the pair-housing enclosure. The two females were weighed and simultaneously placed in their new shared enclosure at 9:00 ± 15 minutes. The social behavior was recorded for three hours immediately after transfer (d1), for one hour on the third day (d3), and for one hour weekly for the following five weeks (for more information see Methods subsection *Social behavior observations*). Additionally, cortisol concentrations were measured at 13:00 on d1 and d3. Baseline cortisol along with cortisol response one and two hours after the onset of a challenge was measured via a cortisol response test (CRT) on the day prior to beginning the experiment (CRT1) and day before the social niche transition (CRT2). Cortisol concentrations were determined from blood samples (for more information see Methods subsection *Cortisol response test (CRT)*). Body weight of the focal female was recorded after each blood sample. The partner female also experienced a modified CRT on d0 so that both females had a challenging experience prior to meeting one another since stress is known to affect the outcome of dominance interactions (Cordero and Sandi 2007). In the modified CRT, partner females were placed singly in a CRT arena for two hours, but no blood or saliva samples were taken. Body weight of the partner female was recorded at the beginning and end of the modified CRT1 on d0, on d1 directly before being housed with the focal female, in the afternoon of d1, and the day prior to the social niche transition when the focal female underwent CRT2. Guinea pigs become sexually mature at approximately one month of age (Kaiser et al. 2010), although first estrous has been observed as late as 50 d of age in female guinea pigs (Schöpper et al. 2012). Therefore, all focal females became sexually mature while in the initial social pairs because focal females were between 59 and 66 days old at the social niche transition.

The focal female was transferred to a new enclosure with a new partner the day after the CRT2. In this social niche transition, focal females were forced to conform to a specific social niche. Focal females were forced to either become subdominant by being housed with an older, larger partner female or become dominant by being housed with a younger, smaller partner female. The older, larger partners were 52 ± 21 days older and 220 ± 137 g heavier than the focal female, and the younger, smaller partners were 35 ± 9 days younger and 193 ± 100 g lighter than the focal female. This method of manipulating the dominance status proved to be very effective, with 42 out of the 44 focal females acquiring the intended dominance status. The two females were simultaneously placed in their new shared enclosure at 9:00 ± 15 minutes. Half of the focal females were housed with a larger female (N = 22) and the other half were housed with a smaller female (N = 22). This created a full-factorial design whereby some females maintained their previous dominance status but with a new partner and other females were forced to transition to a dominance status that they had not yet experienced. Social behavior was observed for three hours immediately after transfer (d1), for one hour in the afternoon of the third day (d3), and for one hour weekly until the third week. Additionally, cortisol concentrations were measured at 13:00 on d1 and d3. Baseline cortisol along with cortisol responsiveness one and two hours after the onset of a challenge was measured via a CRT both one week (CRT3) and three weeks (CRT4) after the social niche transition. Focal females were weighed directly before being placed with the partner female on d1 and after each blood sample collection. All partner females were weighed directly before being placed with the focal female, in the afternoon of the first day, and on the final day of the experiment.

To control for a potential effect of estradiol on cortisol concentrations, estradiol concentration was measured. This was done by analyzing saliva samples that were collected directly after baseline cortisol (in CRTs) and cortisol measurements on d1 and d3 after initial pair formation and social niche transition (for more details see Methods subsections *Cortisol response test (CRT)* and *Statistical analysis*).

Of the 48 focal females, 44 completed the experiment because four females became ill in the initial social pairs and were excluded from the experiment. Of the 44 females who completed the experiment, 41 had full datasets. This is because one female did not meet our criterion for establishing a dominance hierarchy in the first pair (see *Dominance status determination*), one female achieved the status opposite of what was intended in the second pair, and one female both did not meet our criterion for establishing a dominance hierarchy in the first pair and achieved the status opposite of what was intended the second pair. In total, 25 females became dominant and 17 females became subdominant in the first pair. 19 females became dominant while 22 became subdominant in the second pair (for an overview of sample size for each treatment, see Fig. 1).

Social behavior observations

Social behavior was observed using video recordings of the home enclosure that were taken using a camera mounted approximately 1 meter above the enclosure. These videos were recorded for the first three hours after the females were introduced to one another on d1, for one hour in the afternoon of d3, and one hour each week thereafter (mornings, between 8:00 and 12:00). For all recordings aside from d1, fresh hay was added to the enclosure and a small piece of fruit or vegetable was placed in the food bowl to encourage activity and social interactions between the focal female and her partner. Videos were briefly checked afterwards for activity. If the animals were inactive, the video recording was repeated at an appropriate time.

Video analysis was done in successive blocks. First, all videos from week 4 through week 6 after initial social pair formation were analyzed. In case of a missing video, week 3 was also analyzed (N = 2). Then, all videos from week 3 after the social niche transition were analyzed. Finally, all videos from d1 (of initial social pair formation and social niche transition) were analyzed. Videos in each block were analyzed in a random order and the individual identity of the focal female was blinded, although the observer was aware which female in the pair was the focal female. After the social niche transition, it was not possible to blind the observer to dominance status due to the size difference of the focal and partner females. The behavioral coding software Interact (Interact, Lab Suite Version 2017, Mangold International GmbH) was used to count behaviors. All behaviors are defined in the ethogram in Table S1, and definitions are adapted from previous work with cavies (Rood 1972; Kaiser et al. 2015; Sangenstedt et al. 2018). Behaviors observed included approach, follow, sniffing behaviors (body sniff, nasal sniff, anal sniff), aggressive behaviors (brawl, chase, curved body posture, fixate, flip around, head thrust/bite, head up, kick/urine spray, rumba), play behavior (frisky hop, head twitch, run), and retreat. While rumba is generally seen as a courtship behavior in males, we observed it often in dominance interactions and have here included it as an aggressive behavior (for more information see Supplementary Note). Additionally, the amount of time that the two females spent together inside one shelter was counted.

Dominance status determination

A rank index was calculated to assist in the determination of the dominance status of the focal females. The rank index was calculated as the proportion of total retreats that were shown by the partner female. Since dominance behavior can be quite subtle in female guinea pigs, especially when the females are familiar to one another, a retreat was broadly defined as: A female moves away from another female so that she maintains a distance of more than one body length; this behavior is shown either after an interaction of the females or after an approach of one of the females involved. Therefore, retreats were counted even if the females did not explicitly interact. A rank index was calculated from each video observation for each female and had a range between 0 (focal female completely subdominant) and 1 (focal female completely dominant).

Dominance status after initial social pair formation was determined by compiling each focal female's rank indices from week 4, week 5, and week 6. Focal females were determined to be dominant if the average of her three indices was greater than 0.6 and subdominant if the average was less than 0.4 (N=33). For the 11 remaining individuals, behavior in week 3 was additionally observed. If rank index progressively increased or decreased over the four weeks, the individual was categorized by its final status (N=3). If at least three of the four rank indices were above 0.5, the individual was classified as dominant (N=5). If at least three of the four rank indices were below 0.5, the individual was classified as subdominant (N=1). The remaining individuals (N=2) were not assigned a dominance status for the initial social pairs as their rank index oscillated over the four observed weeks.

The rank index was also assessed three weeks after the social niche transition to ensure that the intended dominance status was acquired. All but one of the females who were housed with a smaller, younger female clearly became dominant, with a rank index of at least 0.6. All but three females who were housed with a larger, older female became subdominant (rank index < 0.45). For these four remaining females, behavior in week 2 was additionally analyzed. Two females clearly achieved the opposite status from what was intended and were excluded, and the rank index for the other two females clearly indicated subdominant status (rank indices of 0.17 and 0.33), in line with the intended status.

To assess the outcomes of dominance interactions, the rank index was inverted and used as a measure of submissive behavior. Therefore, submissive behavior was the proportion of total retreats in which the focal female retreated from her partner. A value closer to 1 indicated that a high proportion of the submissive behavior in a pair was expressed by the focal female. Submissive behavior was analyzed on d1 of initial social pair formation to determine whether the dominance hierarchies observed in weeks four, five, and six were already present on d1. Submissive behavior was also analyzed on d1 and wk3 after the social niche transition to determine whether the initial dominance status attained early in adolescence affected outcomes of future dominance interactions with a different social partner.

Cortisol response test (CRT)

The cortisol response test is described in detail elsewhere (Rystrom et al. 2022). Briefly, the focal female was collected from her home enclosure and a blood sample (approximately 150 µl) was taken from the ear within three minutes of entering her housing room. Afterward, a saliva sample was taken by inserting a cotton bud (Sterile applicators, Carl Roth GmbH + Co. KG, Karlsruhe, Volume 13 mL) into the mouth of the focal female and encouraging her to chew on it by slowly twirling the cotton bud. A complete saliva sample consisted of two saturated cotton buds. The female was then weighed and placed into the CRT arena located in a different housing room. The CRT arena was 1 m^2, built similarly to the housing enclosures, and contained food and water but no shelter. Exactly one hour after the onset of the test, the focal female was removed from the arena, another blood sample was taken within three minutes, and the female was weighed and returned to the enclosure. This procedure was again repeated exactly one hour later so that each female had three blood samples taken (baseline cortisol concentration, cortisol response after one hour, and cortisol response after two hours), one saliva sample at the first sampling timepoint (estradiol), and was weighed three times. The female was returned to her home enclosure after the final sample was taken.

Blood samples were immediately prepared (for more details see Rystrom et al. 2022). In brief, blood samples were centrifuged, and the plasma was isolated and frozen at -20°C. Cortisol concentrations of the samples were determined using enzyme-linked immunosorbent assays (Cortisol ELISA, RE52061, IBL International GmbH, Hamburg, Germany) once all samples were collected. The principle of the analysis is based on the following description (IBL International GmbH 2014): A certain amount of enzyme-labelled antigen and the antigen in the sample compete for the binding sites of the antibody-coated wells. After a certain incubation time, the enzyme-labelled antigens that had not bound were removed by washing. Cross-reactions with the antibody were as follows: prednisolone (30%), 11-desoxy-cortisol (7%), corticosterone (1.4%), cortisone (4.2%), prednisone (2.5%), 17α-oh-progesterone (0.4%), desoxy-corticosterone (0.9%), and 6α-methyl-17α-oh-progesterone. The intra-assay variances were on average CV=2.98% and the inter-assay variances were on average CV = 3.51%.

Saliva samples were also immediately prepared. The cotton bud tips were cut off and placed into a 1.5 ml Eppendorf tube which had a hole punctured in the bottom and was stacked within a 2 ml Eppendorf tube. These stacked tubes were then centrifuged at 13000 rpm for 10 minutes so that the saliva was extracted from the cotton bud and collected in the 2 ml Eppendorf tube. The saliva was then pipetted into a sterile 1.5 ml Eppendorf tube and centrifuged at 13000 rpm for 3 minutes. This was repeated until no visible pellet remained. The saliva samples were then frozen at -20°C. All saliva samples were analyzed upon completion of the experiment.

The estradiol concentration in the saliva was analysed with an in-vitro 17β-estradiol saliva luminescence immunoassay (Estradiol ELISA, RE62141/RE62149, IBL International GmbH, Hamburg, Germany) as described by IBL International GmbH (2016). This ELISA is usually used for the estradiol analysis of human saliva but has been standardized and used for guinea pig saliva at the Department of Behavioural Biology in Münster. The concentrations of estradiol were determined by using a standard curve. The compounds 17β-estradiol (100%), estrone (14%), deoxycortisol (0.58%), estriole (0.5%), fulvestrant (0.42%), estrone-3-sulfate (0.26%), ethinylestradiol (0.05%), estradiol-glucuronide (0.03%), epiestradiol (0.02%) and dihydrotestosterone (0.02%) were known to cross-react with the antibody. The intra-assay variances were CV=9.97% and the inter-assay variances were CV=11.30%.

Statistical analysis

Data analysis was carried out with R version 4.1.1 (R Core Team 2020). In general, two-tailed tests were used and significance was set at $P < 0.05$. Non-parametric Wilcoxon signed rank tests were used to determine whether there were differences in age, body weight, and age between females who became dominant and females who became subdominant during the initial pair formation. Paired Wilcoxon tests were used to analyze differences in behavior observed on d1 after initial pair formation and d1 after social niche transition. Linear models and linear mixed-effect models (package lme4 version 1.1-27.1; (Bates et al. 2015) were used to assess the relationship between cortisol concentrations and variables such as dominance status, age, and body weight as well as the relationship between behavior after the social niche transition and dominance status. Continuous explanatory variables were mean-centered and categorical explanatory variables were contrast coded. Therefore, in the model output, the intercept indicates the grand mean and the estimate for categorical variables indicates the difference between the two levels. ANOVA type III tables were generated with the lmerTest package (version 3.1-

3;(Kuznetsova et al. 2017) for mixed-effect models and the car package (version 3.0-11; Fox & Weisberg, 2019) for linear models. The performance package (version 0.7.3; (Lüdecke et al. 2021) was used to verify model assumptions.

To determine the repeatability of the rank indices calculated for weeks 4 through 6 after initial social pair formation, a mixed-effect model was fit with rank index as the response variable and individual identity as a random effect. Adjusted repeatability was estimated from this model using the rptR package (Stoffel et al. 2017) with number of parametric bootstraps set to 1000 and permutation set to 500. Behavior after the social niche transition was analyzed with linear models. These models were fit with each separate behavioral category (i.e. sniffing behavior, approach, follow, aggressive behavior, play, and submissive behavior) as the response variable. Explanatory variables included dominance status in the initial social pair in early adolescence, dominance status after social niche transition, and the interaction between the two. These models were fit separately for the behavior observed on d1 and week 3 after the social niche transition. “Follow” was not analyzed for week 3 because this behavior was observed in few females at this timepoint.

For the data from the CRTs, in order to select relevant variables for the linear models, linear mixed-effect models were fit with one of the cortisol measurements (baseline, responsiveness after one hour, and responsiveness after two hours) as the response variable and body weight, age, and estradiol concentration as explanatory variables. Data from all four CRTs were included, and animal identity was included as a random effect. Both age and body weight had a significant effect on at least one of the cortisol measurements while estradiol concentration did not significantly affect any of the cortisol measurements (see Table S2). Therefore, estradiol concentration was excluded as an explanatory variable for all subsequent models. Age was only included as an explanatory variable for the model using data from CRT1. At CRT1, the age range was 17 – 23 d while the age range was 58 – 65 d at CRT2. We did not expect that the cortisol concentrations of 58 d old females would differ from those of 65 d old females, however, age could feasibly have an effect when the females are between 17 and 23 d old.

Linear models were fit for the data from CRT1 and CRT2 separately. Within each CRT, three models were fit, each with one of the measured cortisol concentrations (baseline, response after one hour, and response after two hours) as the response variable. For CRT1, dominance status (dominant or subdominant), body weight, and age were included as explanatory variables. For CRT2, dominance status (dominant or subdominant), body weight, and the respective cortisol concentration measured at CRT1 for that individual were included as explanatory variables.

To analyze the cortisol concentrations after the social niche transition, linear mixed-effect models were fit with the data from CRT3 and CRT4 combined. Separate models were fit for the response variables baseline cortisol concentration, cortisol response after one hour, and cortisol response after two hours. Explanatory variables included body weight, CRT (CRT3 or CRT4), dominance status in the initial social pair, dominance status after the social niche transition, and all interactions between CRT and the two dominance statuses.

Cortisol concentrations on d1 and d3 were analyzed separately for initial social pair formation and social niche transition. Data from d1 and d3 was combined into one model with cortisol concentration as the response variable. For the initial social pair formation model, explanatory variables included body weight, day (1 or 3), dominance status, and the interaction effect between dominance status and day. For the social niche transition model, both dominance statuses were included as well as body weight, day, and all interactions between dominance statuses and day. Post hoc paired Wilcoxon tests were used to test whether cortisol concentrations on d1 and d3 were significantly different from baseline cortisol as measured on d0.

Results

Dominance status after initial pair formation

The rank index calculated four, five, and six weeks after the pair formation in early adolescence was significantly repeatable (R_{adj} = 0.643, SE = 0.076, 96% CI: [0.458, 0.758], P <0.001), suggesting that the dominance rank indices calculated for each female were stable over the three weeks in which the social interactions were observed. Focal females who became dominant or subdominant did not initially differ in their body weight (Wilcoxon test, N_{dom} = 25, N_{sub} = 17, P = 0.471). Furthermore, within each pair, the initial difference in body weight and age between the focal female and the partner female was not associated with the dominance status achieved by the focal female (Wilcoxon test, N_{dom} = 25, N_{sub} = 17, body weight: W = 254, P = 0.193, age: W = 200, P = 0.755). This suggests that age and weight did not play a substantial role in determining which female became dominant in the initial social pairs.

Effect of dominance status on behavior

Pair formation in early adolescence

On the first day of social pair formation in early adolescence, behavior did not significantly differ between the focal females who became dominant and the focal females who became subdominant. Behaviors observed included aggressive behavior, approaching, following, play behavior, and sniffing behavior (Wilcoxon test, N_{dom} = 25, N_{sub} = 17; aggressive: W = 200.5, P = 0.768; approaching: W = 181.5, P = 0.434; following: W = 253, P = 0.256; play: W = 200, P = 0.746; sniffing: W = 251.5, P = 0.318). However, when observed again four, five, and six weeks after pair formation, dominant females were significantly more aggressive than subdominant females (Wilcoxon test, N_{dom} = 25, N_{sub} = 17, W = 317, P = 0.008; Fig. 2a). Furthermore, the dominant focal females approached their partners significantly more often than the subdominant focal females did (Wilcoxon test, N_{dom} = 25, N_{sub} = 17, W = 316, P = 0.008; Fig. 2b).

Amount of play and sniffing behaviors did not significantly differ between dominant and subdominant focal females at this time point (Wilcoxon test, $N_{dom} = 25$, $N_{sub} = 17$, play: W = 244.5, P = 0.419, Fig. 2c; sniffing: W =213.5, P = 0.990, Fig. 2d).

After the social niche transition

Behavior in the three hours directly after social niche transition was affected by the new dominance status as well as the dominance status experienced previously in the first pair (Table 1). The current dominance status had a significant effect on the amount of aggressive behavior observed (Table 1a; Fig. 3a); focal females who had just become dominant were significantly more aggressive than females who had just become subdominant. Furthermore, the previous dominance status of the focal female from her first social pair significantly affected how often the focal female followed (Table 1c; Fig. 3c) and sniffed (Table 1e; Fig. 3e) her partner. Females who had previously been subdominant both followed and sniffed their new partner significantly more often than females who had previously been dominant (Fig. 3e). Amount of play behavior and approaches was not significantly affected by the dominance status in the original social pair, the dominance status attained after the social niche transition, or an interaction between the two (Table 1b,d; Fig. 3b,d).

Three weeks after the social niche transition, only aggressive behavior was significantly affected by the current dominance status (Table 2a), with aggressive behavior observed more often in dominant females than subdominant females (Fig. 4a). The interaction effect between the dominance status in each social housing pair on sniffing behavior was statistically a trend (Table 2d). Generally, the females that had undergone a transition in their dominance status sniffed their partner more often than females that had maintained the same dominance status in both pairs (Fig. 4d). Play behavior and amount of approaches were not significantly affected by dominance status in either pair (Table 2b,c; Fig. 4b,c).

Dominance status attained in the early adolescence pair significantly affected outcomes of dominance interactions even after the social niche transition. Females who had been dominant in the initial social pair and later became subdominant after social niche transition were more submissive than females were initially subdominant and later became dominant. However, these proportions of submissive interactions were relatively intermediate compared to the females who had always been dominant (lowest proportion of submissive outcomes) and the females who had always been subdominant (highest proportion of submissive outcomes). This was observed both one day (Table 1f, Fig 3f) and three weeks after the social niche transition (Table 2e, Fig. 4e).

Behavioral response to initial pair formation and social niche transition

Social behavior was observed for the three hours directly after social pair formation in early adolescence and social niche transition later in adolescence. Females were overall more active directly after social niche transition than when the first social pair was formed in early adolescence. Focal females approached (Paired Wilcoxon test, $N = 44$, $V = 12.5$, $P < 0.001$), followed (Paired Wilcoxon test, $N = 44$, $V = 1$, $P < 0.001$), and sniffed (Paired Wilcoxon test; $N = 44$, $V = 0$, $P < 0.001$) their partner significantly more often after social niche transition than after initial pair formation. Focal females were also significantly more aggressive toward their partner (Paired Wilcoxon test, $N = 44$, $V = 32$, $P < 0.001$) and played significantly more often (Paired Wilcoxon test; $N = 44$, $V = 33$, $P < 0.001$) after social niche transition. Meanwhile, focal females spent more time within one shelter with their partner upon pair formation in early adolescence (Paired Wilcoxon test; $N = 44$, $V = 754$, $P < 0.001$).

In the three hours after pair formation in early adolescence, females did not form a dominance hierarchy reflective of the stable hierarchy observed later in these pairs (i.e. proportion of submissive behavior did not significantly differ between focal females who were dominant and focal females who were subdominant in weeks 4-6; Wilcoxon test; $N_{dom} = 25$, $N_{sub} = 17$, $W = 158$, $P = 0.267$). However, already on the first day after social niche transition, focal females who were partnered with a larger, older female were significantly more submissive than focal females who were partnered with a smaller, younger female (Table 2e, Fig. 4e).

Cortisol concentrations

Baseline cortisol concentrations

Baseline cortisol was measured throughout the experiment: at each cortisol response test as well as on the first and third day after pair formation in early adolescence and social niche transition later in adolescence. The only time when dominance status was significantly associated with baseline cortisol was six weeks after the initial pairs were formed (CRT2; Table 3), when the baseline cortisol concentration of dominant females was significantly higher than that of subdominant females (Table 3).

There was a significant effect of body weight on the baseline cortisol concentration throughout the measurements taken when the focal females were in their initial social pairs (as measured at CRT1, Table S3; d1 and d3 after pair formation, Table 4; and CRT2, Table 3). At all of these time points, body weight was negatively correlated with baseline cortisol concentration; heavier animals had lower baseline cortisol concentrations. Age also had a significant effect on baseline cortisol, but only at the first CRT (Table S3). Again, this correlation was negative, meaning that older females had lower baseline cortisol concentrations. Body weight and age did not have a significant effect on baseline cortisol concentration at any point after the social niche transition (Table 4, Table 5).

Cortisol concentration significantly changed from d1 to d3 both after the initial pair formation and the social niche transition. Interestingly, this change was in opposite directions. Cortisol concentration significantly increased in the first three days after social pair formation (Table 4; Fig. 5). Meanwhile, in the three days after the social niche transition, cortisol concentration significantly decreased (Table 4; Fig. 5). Further statistical tests were carried out to determine how these cortisol concentrations compared to the baseline cortisol concentrations measured the day prior to pair formation and social niche transition. The cortisol concentration measured on d1 after social pair formation was higher than that measured the day prior (d0), although this was not statistically significant (Paired Wilcoxon test; N = 43, V = 320, P = 0.066). Furthermore, cortisol concentration at d3 was significantly higher than that measured on d0 prior to the transition from natal group to the female pair (Paired Wilcoxon test; N = 44, V = 254, P = 0.005). Meanwhile, after the social niche transition, cortisol concentrations at d1 were significantly higher than those measured at the same time the day before (Paired Wilcoxon test; N = 43, V = 96, P < 0.001), although cortisol levels had returned to the baseline level by d3 (Paired Wilcoxon test; N = 43, V = 442.5, P = 0.915).

Cortisol responsiveness in early adolescence

At the first cortisol response test, which took place the day prior to being housed with a same aged female partner, cortisol responsiveness after one or two hours did not differ between focal females who would later become dominant and focal females who would later become subdominant (Table S3). Age and body weight also had no significant effect on cortisol responsiveness at one or two hours (Table S3). When measured again six weeks after the initial social pair formation, cortisol responsiveness after one and two hours were again not significantly affected by dominance status, body weight, or cortisol response concentration measured at the first cortisol response test (Table 3).

Cortisol responsiveness after social niche transition

When measured one and three weeks after the social niche transition, cortisol responsiveness after one hour was not significantly affected by the dominance status in the previous pair, the current dominance status after social niche transition, or any interactions between dominance status and CRT (Table 5b). However, cortisol responsiveness after one hour was significantly correlated to body weight

(Table 5b) whereby heavier females had lower cortisol response concentrations. Additionally, the cortisol response concentration after one hour was significantly higher at three weeks after the social niche transition than at one week after the social niche transition (Table 5b).

The cortisol response after two hours was significantly affected by the interaction between the dominance status after social niche transition and time (Table 5c). The cortisol response of females who became subdominant increased from the first to third week after the social niche transition while the cortisol response of females who became dominant slightly decreased (Fig. 6). The interaction between the dominance status in the initial social pair and the dominance status after social niche transition tended to affect the cortisol response after two hours (Table 5c). Furthermore, cortisol responsiveness after two hours when measured three weeks after the social niche transition tended to be higher than when measured one week after the social niche transition (Table 5c). There was no significant main effect of body weight (Table 5c), previous dominance status (Table 5c), or current dominance status (Table 5c) on cortisol response after two hours.

Discussion

These results offer insight into how dominance hierarchies are formed among females during adolescence and characterize the behavioral and hormonal differences between dominant and subdominant individuals. When dominance status changed, aggressive behavior was rapidly reshaped so that the dominant female was consistently more aggressive. Dominance interactions reflected the new dominance status after the social niche transition, but the previous dominance status from the initial social pair had a long-lasting effect on how submissively females interacted with their new partners. Finally, when females were forced to become subdominant, the magnitude of their cortisol responsiveness to challenging situations increased.

Social niche specialization and conformance

When social pairs were formed in early adolescence, dominance status attained was not predicted by cortisol concentrations, modest differences in body weight or age, or social behavior upon introduction. Aside from interactions with siblings and parents in the natal group, these females were initially naïve in social contexts and gained social experience by interacting with their new social partner. When social behavior was again observed four, five, and six weeks after pair formation, behavioral differences between dominant and subdominant individuals had developed. Therefore, since stable dominance hierarchies were observed later but were not established upon introduction, developing individual differences in social behavior (Stamps and Groothuis 2010; Sachser et al. 2020) and their effect on social interactions (Bergmüller and Taborsky 2010; David et al. 2011; Montiglio et al. 2013; Briffa et al. 2015) likely contributed to the dominance hierarchy formation, in line with the social dynamics hypothesis (Chase et al. 2002).

In contrast, female established a clear dominance hierarchy already on the first day after the social niche transition, with heavier and older females becoming dominant over their smaller and younger partners. This is in line with the prior attributes hypothesis, since females rapidly conformed to the appropriate social niche when there were pronounced differences in body size and age (Chase et al. 2002; Montiglio et al. 2013). Therefore, we conclude that different processes drove the formation of the dominance hierarchy depending on whether or not the females differed in the prior attributes important for dominance status acquisition. Studies assessing dominance status predictors across multiple social groups often observe that the dominance hierarchy of some groups are structured by age and body size while others are not (Holand et al. 2004; Kohn et al. 2016; Lu et al. 2016). Our study therefore contributes to the current understanding that the prior attributes and social dynamics hypotheses are not mutually exclusive but rather impact dominance hierarchy formation depending on demographic factors.

Shaping of the behavioral profile by dominance status

Regarding social behavior, there were two major findings. First, dominant females were consistently more aggressive than subdominant females, and there was no lingering effect of dominance status from the initial social pair on aggressive behavior after the social niche transition toward the new social partner. In a similar study with female rhesus macaques (*Macaca mulatta*), adult social groups were manipulated and changes in social behavior were correlated to the change in dominance status (Kohn et al. 2016). Dominant females were more aggressive, and when dominance status changed, aggression was rapidly reshaped. Indeed, females across a wide range of species flexibly adjust their aggressive behavior to match changing social conditions (Bowler et al. 2002; Pusey and Schroepfer-Walker 2013; Stockley and Campbell 2013; Taylor et al. 2014).

The second major finding is that the dominance status attained in early adolescence had a persistent effect on dominance interactions after the social niche transition, even when controlling for current dominance status. This could be due to winner-loser effects (Hsu et al. 2006), whereby females who retreat from their partner are more likely to retreat in future encounters. Winner-loser effects on dominance status acquisition have been demonstrated for example with male shore crabs (*Carcinus maenas*). Males who had won many encounters during a social disturbance event proceeded to acquire an even higher dominance status when original social groups were reformed (Tanner et al. 2011). However, since dominance status in the initial social pairs was not manipulated but rather emerged organically, it cannot be ruled out that the propensity to behave submissively was an intrinsic behavioral trait.

In addition, dominance status predicted the frequency of several other social behaviors. In the initial social pair, dominant females approached their partner more often. Contests are often won by the initiator (Jackson 1991; Kar et al. 2016), and dominance acquisition has been predicted by behaviors such as proactivity (David et al. 2011) and exploration (Favati et al. 2014; Fox et al. 2009; Kaiser et al. 2019; but see Devost et al. 2016). Therefore, the female who most often initiated contact might have become dominant. After the social niche transition, previously subdominant females and females that changed dominance status sniffed and followed their new social partner more often. A possible explanation is that subdominant females and females with an unstable dominance rank are more motivated to thoroughly assess new social partners. It has been previously shown that subordinate male mice assess unfamiliar odor cues longer than dominant male mice do (Garbe and Kemble 1994), and that male and female dogs of low status sniff urine of unfamiliar dogs longer than high status dogs do (Lisberg and Snowdon 2009).

Social factors shaped cortisol concentrations

There were three significant findings indicating that cortisol concentrations were shaped by social factors. First, cortisol concentrations responded differently to the initial pair formation and the social niche transition. Upon the formation of the initial social pairs, the main challenge was likely the removal from the natal group. Our results align with previous findings that cortisol levels of female guinea pigs increase upon separation from the mother and remain heightened for days (Hennessy et al. 2002). Meanwhile, the main challenge after the social niche transition was likely the social instability induced by the change in social partner. It is well-established that cortisol levels are higher during periods of social instability (Sapolsky 1982; Nuñez et al. 2014), and cortisol concentrations rise during social confrontations with unfamiliar female guinea pigs (Glenk et al. 2018). Since dominance hierarchies were quickly settled after the social niche transition, the first few highly socially-active hours post-introduction were likely the most challenging, with cortisol levels decreasing thereafter.

Second, baseline cortisol concentrations were shaped to the dominance status attained in the initial social pairs. Six weeks after the initial social pair formation, dominant females had a significantly higher baseline cortisol concentration than subdominant females. Higher concentrations of cortisol are often correlated to higher levels of aggression (Haller 2014), and in groups of size-matched individuals such as the female pairs early in adolescence, the individuals with higher circulating cortisol levels might be able to quickly mobilize the energy needed to express aggressive behavior. Furthermore, since the dominance hierarchy was not immediately settled after pair formation, rank instability and challenges by subdominant females may have resulted in a stressful social environment for dominant females (Bergman et al. 2005; Carvalho et al. 2018). Meanwhile, in hierarchies where dominance rank is clearly established by body size/age such as after the social niche transition, dominant individuals will not carry the stress of policing others (Creel et al. 2013).

Third, manipulating the dominance status triggered a shift in cortisol responsiveness. Subdominant females did not have a significantly higher cortisol responsiveness than dominant females, but the cortisol responsiveness of individuals who became subdominant increased over the three weeks after the social niche transition relative to that of the individuals forced to become dominant. Research on primate species indicates that differences in challenge-induced glucocorticoid concentrations emerge after dominance status is attained (Sapolsky 2004; Cavigelli and Chaudhry 2012; Michopoulos et al. 2012; Kohn et al. 2016). Notably, no correlation between dominance rank and cortisol response to a challenge has been detected in guinea pigs (Zipser et al. 2013; Mutwill et al. 2021; Rystrom et al. 2022). However, in these studies, dominance rank was observed in larger social groups rather than being manipulated in pairs as in the present study. Few other studies attempt to link challenge-induced glucocorticoid concentrations to dominance status in non-primate species (but see Pravosudov et al. 2003; Lindström et al. 2005; Poisbleau et al. 2005; Rubenstein 2007). These results support the hypothesis that shifts in dominance status can shape the magnitude of cortisol response to acute challenges.

Statements and Declarations

Contributions

SK, SHR, and NS conceived the experiment. SK and TLR planned the experiment. TLR collected the data and carried out the statistical analysis. SHR, NS, and SK assisted with interpretation of results. TLR wrote the manuscript with input from all authors.

Compliance with ethical standards

All procedures complied with the regulations covering animal experimentation within Germany (Animal Welfare Act) and the EU (European Communities Council DIRECTIVE 2010/63/EU) and were approved by the local and federal authorities (Landesamt für Natur, Umwelt und Verbraucherschutz Nordrhein-Westfalen “LANUV NRW”, reference number: 84-02.04.2015.A439).

Competing interests

The authors declare that they have no competing interests.

Data availability

All data analyzed during this study are included in the supplementary information files.

References

Alonso F, Honji RM, Moreira RG, Pandolfi M (2012) Dominance hierarchies and social status ascent opportunity: Anticipatory behavioral and physiological adjustments in a Neotropical cichlid fish. Physiol Behav 106:612–618. https://doi.org/10.1016/j.physbeh.2012.04.003

Arce M, Michopoulos V, Shepard KN, Ha QC, Wilson ME (2010) Diet choice, cortisol reactivity, and emotional feeding in socially housed rhesus monkeys. Physiol Behav 101:446–455. https://doi.org/10.1016/j.physbeh.2010.07.010

Barrette MF, Monfort SL, Festa-Bianchet M, Clutton-Brock TH, Russell AF (2012) Reproductive rate, not dominance status, affects fecal glucocorticoid levels in breeding female meerkats. Horm Behav 61:463–471. https://doi.org/10.1016/j.yhbeh.2011.12.005

Bates D, Mächler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using lme4. J Stat Softw 67:1–48. https://doi.org/10.18637/jss.v067.i01

Bergman TJ, Beehner JC, Cheney DL, Seyfarth RM, Whitten PL (2005) Correlates of stress in free-ranging male chacma baboons, *Papio hamadryas ursinus*. Anim Behav 70:703–713. https://doi.org/10.1016/j.anbehav.2004.12.017

Bergmüller R, Taborsky M (2010) Animal personality due to social niche specialisation. Trends Ecol Evol 25:504–511. https://doi.org/10.1016/j.tree.2010.06.012

Bowler CM, Cushing BS, Sue Carter C (2002) Social factors regulate female–female aggression and affiliation in prairie voles. Physiol Behav 76:559–566. https://doi.org/10.1016/S0031-9384(02)00755-2

Briffa M, Sneddon LU, Wilson AJ (2015) Animal personality as a cause and consequence of contest behaviour. Biol Lett 11:20141007. https://doi.org/10.1098/rsbl.2014.1007

Carvalho RR, Palme R, da Silva Vasconcellos A (2018) An integrated analysis of social stress in laying hens: The interaction between physiology, behaviour, and hierarchy. Behav Processes 149:43–51. https://doi.org/10.1016/j.beproc.2018.01.016

Cavigelli SA, Caruso MJ (2015) Sex, social status and physiological stress in primates: the importance of social and glucocorticoid dynamics. Philos Trans R Soc B Biol Sci 370:20140103. https://doi.org/10.1098/rstb.2014.0103

Cavigelli SA, Chaudhry HS (2012) Social status, glucocorticoids, immune function, and health: Can animal studies help us understand human socioeconomic-status-related health disparities? Horm Behav 62:295–313. https://doi.org/10.1016/j.yhbeh.2012.07.006

Chase ID, Tovey C, Spangler-Martin D, Manfredonia M (2002) Individual differences versus social dynamics in the formation of animal dominance hierarchies. Proc Natl Acad Sci 99:5744–5749. https://doi.org/10.1073/pnas.082104199

Chelini MOM, Palme R, Otta E (2011) Social stress and reproductive success in the female Syrian hamster: Endocrine and behavioral correlates. Physiol Behav 104:948–954. https://doi.org/10.1016/j.physbeh.2011.06.006

Cordero MI, Sandi C (2007) Stress amplifies memory for social hierarchy. Front Neurosci 1:175–184. https://doi.org/10.3389/neuro.01.1.1.013.2007

Côté SD, Festa-Bianchet M (2001) Reproductive success in female mountain goats: the influence of age and social rank. Anim Behav 62:173–181. https://doi.org/10.1006/anbe.2001.1719

Creel S (2001) Social dominance and stress hormones. Trends Ecol Evol 16:491–497. https://doi.org/10.1016/S0169-5347(01)02227-3

Creel S, Dantzer B, Goymann W, Rubenstein DR (2013) The ecology of stress: effects of the social environment. Funct Ecol 27:66–80. https://doi.org/10.1111/j.1365-2435.2012.02029.x

Creel S, Marusha Creel N, Monfort SL (1996) Social stress and dominance. Nature 379:212–212. https://doi.org/10.1038/379212a0

David M, Auclair Y, Cézilly F (2011) Personality predicts social dominance in female zebra finches, *Taeniopygia guttata*, in a feeding context. Anim Behav 81:219–224. https://doi.org/10.1016/j.anbehav.2010.10.008

Dehnen T, Arbon JJ, Farine DR, Boogert NJ (2022) How feedback and feed-forward mechanisms link determinants of social dominance. Biol Rev 97:1210–1230. https://doi.org/10.1111/brv.12838

Devost I, Jones TB, Cauchoix M, Montreuil-Spencer C, Morand-Ferron J (2016) Personality does not predict social dominance in wild groups of black-capped chickadees. Anim Behav 122:67–76. https://doi.org/10.1016/j.anbehav.2016.10.001

Drews C (1993) The concept and definition of dominance in animal behaviour. Behaviour 125:283–313. https://doi.org/10.1163/156853993X00290

Emmerson MG, Spencer KA (2018) Group housing during adolescence has long-term effects on the adult stress response in female, but not male, zebra finches (*Taeniopygia guttata*). Gen Comp Endocrinol 256:71–79. https://doi.org/10.1016/j.ygcen.2017.07.008

Faruzzi AN, Solomon MB, Demas GE, Huhman KL (2005) Gonadal hormones modulate the display of submissive behavior in socially defeated female Syrian hamsters. Horm Behav 47:569–575. https://doi.org/10.1016/j.yhbeh.2004.11.023

Favati A, Leimar O, Løvlie H (2014) Personality predicts social dominance in male domestic fowl. PLoS ONE 9:e103535. https://doi.org/10.1371/journal.pone.0103535

Fawcett TW, Frankenhuis WE (2015) Adaptive explanations for sensitive windows in development. Front Zool 12:S3. https://doi.org/10.1186/1742-9994-12-S1-S3

Fichtel C, Kraus C, Ganswindt A, Heistermann M (2007) Influence of reproductive season and rank on fecal glucocorticoid levels in free-ranging male Verreaux's sifakas (*Propithecus verreauxi*). Horm Behav 51:640–648. https://doi.org/10.1016/j.yhbeh.2007.03.005

Fox RA, Ladage LD, Roth TC, Pravosudov VV (2009) Behavioural profile predicts dominance status in mountain chickadees, *Poecile gambeli*. Anim Behav 77:1441–1448. https://doi.org/10.1016/j.anbehav.2009.02.022

Fulenwider HD, Caruso MA, Ryabinin AE (2022) Manifestations of domination: Assessments of social dominance in rodents. Genes Brain Behav 21:e12731. https://doi.org/10.1111/gbb.12731

Garbe CM, Kemble ED (1994) Effects of prior agonistic experience on risk assessment and approach behavior evoked by familiar or unfamiliar conspecific odors. Aggress Behav 20:143–149. https://doi.org/10.1002/1098-2337(1994)20:2<143::AID-AB2480200207>3.0.CO;2-D

Glenk LM, Machatschke IH, Wallner B (2018) Fight or flight? Effects of vaginal oestrus on cortisol, testosterone, and behaviour in guinea pig female-female interaction. Behav Processes 157:625–631. https://doi.org/10.1016/j.beproc.2018.04.008

Goymann W, Wingfield JC (2004) Allostatic load, social status and stress hormones: the costs of social status matter. Anim Behav 67:591–602. https://doi.org/10.1016/j.anbehav.2003.08.007

Haller J (2014) The glucocorticoid/aggression relationship in animals and humans: An analysis sensitive to behavioral characteristics, glucocorticoid secretion patterns, and neural mechanisms. In: Miczek KA, Meyer-Lindenberg A (eds) Neuroscience of Aggression. Springer, Berlin, pp 73–109.

Hamilton LD, Carré JM, Mehta PH, Olmstead N, Whitaker JD (2015) Social neuroendocrinology of status: A review and future directions. Adapt Hum Behav Physiol 1:202–230. https://doi.org/10.1007/s40750-015-0025-5

Heck AL, Handa RJ (2019) Sex differences in the hypothalamic–pituitary–adrenal axis' response to stress: An important role for gonadal hormones. Neuropsychopharmacology 44:45–58. https://doi.org/10.1038/s41386-018-0167-9

Hennessy MB, Maken DS, Graves FC (2002) Presence of mother and unfamiliar female alters levels of testosterone, progesterone, cortisol, adrenocorticotropin, and behavior in maturing guinea pigs. Horm Behav 42:42–52. https://doi.org/10.1006/hbeh.2002.1794

Holand Ø, Gjøstein H, Losvar A, Kumpula J, Smith ME, Røed KH, Nieminen M, Weladji RB (2004) Social rank in female reindeer (*Rangifer tarandus*): Effects of body mass, antler size and age. J Zool 263:365–372. https://doi.org/10.1017/S0952836904005382

Hsu Y, Earley RL, Wolf LL (2006) Modulation of aggressive behaviour by fighting experience: Mechanisms and contest outcomes. Biol Rev 81:33–74. https://doi.org/10.1017/S146479310500686X

Jackson WM (1991) Why do winners keep winning? Behav Ecol Sociobiol 28:271–276. https://doi.org/10.1007/BF00175100

Jäger HY, Han CS, Dingemanse NJ (2019) Social experiences shape behavioral individuality and within-individual stability. Behav Ecol 30:1012–1019. https://doi.org/10.1093/beheco/arz042

Kaiser A, Merckx T, Van Dyck H (2019) Personality traits influence contest outcome, and vice versa, in a territorial butterfly. Sci Rep 9:2778. https://doi.org/10.1038/s41598-019-39155-9

Kaiser S, Krüger C, Sachser N (2010) The guinea pig. In: Hubrecht R, Kirkwood J (eds) The UFAW handbook on the care and management of laboratory and other research animals. Wiley-Blackwell, Chichester, UK, pp 380–398.

Kaiser S, Schwerdt B, Siegeler K, Sachser N (2015) Social instability during pregnancy and lactation alters female wild cavy offsprings' endocrine status and behaviour later in life. Behaviour 152:837–859. https://doi.org/10.1163/1568539X-00003256

Kar F, Whiting MJ, Noble DWA (2016) Influence of prior contest experience and level of escalation on contest outcome. Behav Ecol Sociobiol 70:1679–1687. https://doi.org/10.1007/s00265-016-2173-4

Kohn JN, Snyder-Mackler N, Barreiro LB, Johnson ZP, Tung J, Wilson ME (2016) Dominance rank causally affects personality and glucocorticoid regulation in female rhesus macaques. Psychoneuroendocrinology 74:179–188. https://doi.org/10.1016/j.psyneuen.2016.09.005

Korzan WJ, Summers CH (2021) Evolution of stress responses refine mechanisms of social rank. Neurobiol Stress 14:100328. https://doi.org/10.1016/j.ynstr.2021.100328

Kuznetsova A, Brockhoff PB, Christensen RHB (2017) lmerTest package: Tests in linear mixed effects models. J Stat Softw 82:1–26. https://doi.org/10.18637/jss.v082.i13

Lindström KM, Hasselquist D, Wikelski M (2005) House sparrows (*Passer domesticus*) adjust their social status position to their physiological costs. Horm Behav 48:311–320. https://doi.org/10.1016/j.yhbeh.2005.04.002

Lisberg AE, Snowdon CT (2009) The effects of sex, gonadectomy and status on investigation patterns of unfamiliar conspecific urine in domestic dogs, *Canis familiaris*. Anim Behav 77:1147–1154. https://doi.org/10.1016/j.anbehav.2008.12.033

Lu A, Borries C, Gustison ML, Larney E, Koenig A (2016) Age and reproductive status influence dominance in wild female Phayre's leaf monkeys. Anim Behav 117:145–153. https://doi.org/10.1016/j.anbehav.2016.04.020

Lüdecke D, Ben-Shachar M, Patil I, Waggoner P, Makowski D (2021) performance: An R package for assessment, comparison and testing of statistical models. J Open Source Softw 6:3139. https://doi.org/10.21105/joss.03139

Lürzel S, Kaiser S, Sachser N (2010) Social interaction, testosterone, and stress responsiveness during adolescence. Physiol Behav 99:40–46. https://doi.org/10.1016/j.physbeh.2009.10.005

Lürzel S, Kaiser S, Sachser N (2011) Social interaction decreases stress responsiveness during adolescence. Psychoneuroendocrinology 36:1370–1377. https://doi.org/10.1016/j.psyneuen.2011.03.010

McCormick CM, Mathews IZ (2007) HPA function in adolescence: Role of sex hormones in its regulation and the enduring consequences of exposure to stressors. Pharmacol Biochem Behav 86:220–233. https://doi.org/10.1016/j.pbb.2006.07.012

Michopoulos V, Reding KM, Wilson ME, Toufexis D (2012) Social subordination impairs hypothalamic–pituitary–adrenal function in female rhesus monkeys. Horm Behav 62:389–399. https://doi.org/10.1016/j.yhbeh.2012.07.014

Montiglio PO, Ferrari C, Réale D (2013) Social niche specialization under constraints: personality, social interactions and environmental heterogeneity. Philos Trans R Soc B Biol Sci 368:20120343. https://doi.org/10.1098/rstb.2012.0343

Mutwill AM, Schielzeth H, Zimmermann TD, Richter SH, Kaiser S, Sachser N (2021) Individuality meets plasticity: Endocrine phenotypes across male dominance rank acquisition in guinea pigs living in a complex social environment. Horm Behav 131:104967. https://doi.org/10.1016/j.yhbeh.2021.104967

Mutwill AM, Zimmermann TD, Hennicke A, Richter SH, Kaiser S, Sachser N (2020) Adaptive reshaping of the hormonal phenotype after social niche transition in adulthood. Proc R Soc B 287:20200667. https://doi.org/10.1098/rspb.2020.0667

Niemelä PT, Santostefano F (2015) Social carry-over effects on non-social behavioral variation: mechanisms and consequences. Front Ecol Evol 3:49. https://doi.org/10.3389/fevo.2015.00049

Nuñez CMV, Adelman JS, Smith J, Gesquiere LR, Rubenstein DI (2014) Linking social environment and stress physiology in feral mares (*Equus caballus*): Group transfers elevate fecal cortisol levels. Gen Comp Endocrinol 196:26–33. https://doi.org/10.1016/j.ygcen.2013.11.012

Ode M, Asaba A, Miyazawa E, Mogi K, Kikusui T, Izawa EI (2015) Sex-reversed correlation between stress levels and dominance rank in a captive non-breeder flock of crows. Horm Behav 73:131–134. https://doi.org/10.1016/j.yhbeh.2015.07.012

Pandolfi M, Scaia MF, Fernandez MP (2021) Sexual dimorphism in aggression: Sex-specific fighting strategies across species. Front Behav Neurosci 15:659615. https://doi.org/10.3389/fnbeh.2021.659615

Poisbleau M, Fritz H, Guillon N, Chastel O (2005) Linear social dominance hierarchy and corticosterone responses in male mallards and pintails. Horm Behav 47:485–492. https://doi.org/10.1016/j.yhbeh.2005.01.001

Pottinger TG, Carrick TR (2001) Stress responsiveness affects dominant-subordinate relationships in rainbow trout. Horm Behav 40:419–427. https://doi.org/10.1006/hbeh.2001.1707

Pravosudov VV, Mendoza SP, Clayton NS (2003) The relationship between dominance, corticosterone, memory, and food caching in mountain chickadees (*Poecile gambeli*). Horm Behav 44:93–102. https://doi.org/10.1016/S0018-506X(03)00119-3

Pusey A, Williams J, Goodall J (1997) The influence of dominance rank on the reproductive success of female chimpanzees. Science 277:828–831. https://doi.org/10.1126/science.277.5327.828

Pusey AE, Schroepfer-Walker K (2013) Female competition in chimpanzees. Philos Trans R Soc B Biol Sci 368:20130077. https://doi.org/10.1098/rstb.2013.0077

R Core Team (2020) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna.

Romeo RD (2018) The metamorphosis of adolescent hormonal stress reactivity: A focus on animal models. Front Neuroendocrinol 49:43–51. https://doi.org/10.1016/j.yfrne.2017.12.003

Rood JP (1972) Ecological and behavioural comparisons of three genera of Argentine cavies. Anim Behav Monogr 5:IN1-IN4. https://doi.org/10.1016/S0066-1856(72)80002-5

Rubenstein DR (2007) Stress hormones and sociality: integrating social and environmental stressors. Proc R Soc B 274:967–975. https://doi.org/10.1098/rspb.2006.0051

Rystrom TL, Prawitt RC, Richter SH, Sachser N, Kaiser S (2022) Repeatability of endocrine traits and dominance rank in female guinea pigs. Front Zool 19:4. https://doi.org/10.1186/s12983-021-00449-2

Sachser N (1986) Different forms of social organization at high and low population densities in guinea pigs. Behaviour 97:253–272. https://doi.org/10.1163/156853986X00630

Sachser N, Hennessy MB, Kaiser S (2018) The adaptive shaping of social behavioural phenotypes during adolescence. Biol Lett 14:20180536. https://doi.org/10.1098/rsbl.2018.0536

Sachser N, Zimmermann TD, Hennessy MB, Kaiser S (2020) Sensitive phases in the development of rodent social behavior. Curr Opin Behav Sci 36:63–70. https://doi.org/10.1016/j.cobeha.2020.07.014

Saltz JB, Geiger AP, Anderson R, Johnson B, Marren R (2016) What, if anything, is a social niche? Evol Ecol 30:349–364. https://doi.org/10.1007/s10682-015-9792-5

Sangenstedt S, Szardenings C, Sachser N, Kaiser S (2018) Does the early social environment prepare individuals for the future? A match-mismatch experiment in female wild cavies. Front Zool 15:13. https://doi.org/10.1186/s12983-018-0261-1

Sapolsky RM (1982) The endocrine stress-response and social status in the wild baboon. Horm Behav 16:279–292. https://doi.org/10.1016/0018-506X(82)90027-7

Sapolsky RM (2004) Social status and health in humans and other animals. Annu Rev Anthropol 33:393–418. https://doi.org/10.1146/annurev.anthro.33.070203.144000

Sapolsky RM, Romero LM, Munck AU (2000) How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. Endocr Rev 21:55–89. https://doi.org/10.1210/edrv.21.1.0389

Schneider J, Atallah J, Levine JD (2017) Social structure and indirect genetic effects: genetics of social behaviour. Biol Rev 92:1027–1038. https://doi.org/10.1111/brv.12267

Schöpper H, Palme R, Ruf T, Huber S (2012) Effects of prenatal stress on hypothalamic-pituitary-adrenal (HPA) axis function over two generations of guinea pigs (*Cavia aperea* f. *porcellus*). Gen Comp Endocrinol 176:18–27. https://doi.org/10.1016/j.ygcen.2011.12.010

Stamps JA, Groothuis TGG (2010) Developmental perspectives on personality: Implications for ecological and evolutionary studies of individual differences. Philos Trans R Soc B Biol Sci 365:4029–4041. https://doi.org/10.1098/rstb.2010.0218

Stockley P, Campbell A (2013) Female competition and aggression: Interdisciplinary perspectives. Philos Trans R Soc B Biol Sci 368:20130073. https://doi.org/10.1098/rstb.2013.0073

Stoffel MA, Nakagawa S, Schielzeth H (2017) rptR: repeatability estimation and variance decomposition by generalized linear mixed-effects models. Methods Ecol Evol 8:1639–1644. https://doi.org/10.1111/2041-210X.12797

Tanner CJ, Salali GD, Jackson AL (2011) The ghost of social environments past: dominance relationships include current interactions and experience carried over from previous groups. Biol Lett 7:818–821. https://doi.org/10.1098/rsbl.2011.0325

Taylor RW, Boutin S, Humphries MM, McAdam AG (2014) Selection on female behaviour fluctuates with offspring environment. J Evol Biol 27:2308–2321. https://doi.org/10.1111/jeb.12495

Thyen Y, Hendrichs H (1990) Differences in behavior and social organization of female guinea pigs as a function of the presence of a male. Ethology 85:25–34. https://doi.org/10.1111/j.1439-0310.1990.tb00382.x

Tibbetts EA, Pardo-Sanchez J, Weise C (2022) The establishment and maintenance of dominance hierarchies. Philos Trans R Soc B Biol Sci 377:20200450. https://doi.org/10.1098/rstb.2020.0450

Touma C, Bunck M, Glasl L, Nussbaumer M, Palme R, Stein H, Wolferstätter M, Zeh R, Zimbelmann M, Holsboer F, Landgraf R (2008) Mice selected for high versus low stress reactivity: A new animal model for affective disorders. Psychoneuroendocrinology 33:839–862. https://doi.org/10.1016/j.psyneuen.2008.03.013

Trappes R, Nematipour B, Kaiser MI, Krohs U, van Benthem KJ, Ernst UR, Gadau J, Korsten P, Kurtz J, Schielzeth H, Schmoll T, Takola E (2022) How individualized niches arise: Defining mechanisms of niche construction, niche choice, and niche conformance. BioScience 72:538–548. https://doi.org/10.1093/biosci/biac023

Williamson CM, Lee W, DeCasien AR, Lanham A, Romeo RD, Curley JP (2019) Social hierarchy position in female mice is associated with plasma corticosterone levels and hypothalamic gene expression. Sci Rep 9:7324. https://doi.org/10.1038/s41598-019-43747-w

Wilson AJ, Morrissey MB, Adams MJ, Walling CA, Guinness FE, Pemperton JM, Clutton-Brock TH, Kruuk LEB (2011) Indirect genetics effects and evolutionary constraint: An analysis of social dominance in red deer, *Cervus elaphus*. J Evol Biol 24:772–783. https://doi.org/10.1111/j.1420-9101.2010.02212.x

Wise DA (1974) Aggression in the female golden hamster: Effects of reproductive state and social isolation. Horm Behav 5:235–250. https://doi.org/10.1016/0018-506X(74)90032-4

Zimmermann TD, Kaiser S, Hennessy MB, Sachser N (2017a) Adaptive shaping of the behavioural and neuroendocrine phenotype during adolescence. Proc R Soc B 284:20162784. https://doi.org/10.1098/rspb.2016.2784

Zimmermann TD, Kaiser S, Sachser N (2017b) The adaptiveness of a queuing strategy shaped by social experiences during adolescence. Physiol Behav 181:29–37. https://doi.org/10.1016/j.physbeh.2017.08.025

Zipser B, Kaiser S, Sachser N (2013) Dimensions of animal personalities in guinea pigs. Ethology 119:970–982. https://doi.org/10.1111/eth.12140

Table 1 Focal female behavior on day one after the social niche transition. Dominance status in initial social pair and after social niche transition were contrast coded, and reference levels are “subdominant”. Estimates were obtained using the summary() function and ANOVA type III tables were generated using the Anova() function from the car package. Differences were considered significant at p < 0.05 (bold); 0.05 ≤ p < 0.1 was considered a trend (bold and italic)

Fixed effects	Estimate ± SE	95% CI	Sum Sq	df	F value	P
a) Aggressive behavior (square root transformed)						
intercept	6.244 ± 0.401					
Initial dominance status	1.199 ± 0.802	-0.426, 2.824	13.89	1	2.237	0.143
Dominance status after transition	3.681 ± 0.802	2.056, 5.306	130.85	1	21.074	**<0.001**
Initial * transition dominance status	-0.134 ± 1.604	-3.383, 3.116	0.04	1	0.007	0.934
residuals			229.73	37		
b) Approach (untransformed)						
intercept	54.900 ± 5.195					
Initial dominance status	-2.312 ± 10.389	-23.363, 18.739	52	1	0.050	0.825
Dominance status after transition	11.534 ± 10.389	-9.516, 32.585	1285	1	1.233	0.274
Initial * transition dominance status	23.291 ± 20.779	-18.811, 65.392	1310	1	1.256	0.270
residuals			38565	37		
c) Follow (untransformed)						
intercept	8.758 ± 0.780					
Initial dominance status	-3.484 ± 1.560	-6.646, -0.322	117.21	1	4.985	**0.032**
Dominance status after transition	1.901 ± 1.560	-1.261, 5.062	34.88	1	1.484	0.231
Initial * transition dominance status	-6.199 ± 3.121	-12.522, 0.125	92.76	1	3.945	***0.055***
residuals			869.99	37		
d) Play behavior (square root transformed)						
intercept	6.427 ± 0.577					
Initial dominance status	-1.257 ± 1.154	-3.596, 1.081	15.27	1	1.187	0.283
Dominance status after transition	-0.033 ± 1.154	-2.371, 2.305	0.01	1	0.001	0.977
Initial * transition dominance status	0.473 ± 2.308	-4.204, 5.150	0.54	1	0.042	0.839
residuals			475.90	37		
e) Sniffing behavior (log transformed)						
intercept	3.453 ± 0.103					
Initial dominance status	-0.568 ± 0.206	-0.986, -0.151	3.12	1	7.605	**0.009**
Dominance status after transition	-0.093 ± 0.206	-0.510, 0.325	0.08	1	0.203	0.655
Initial * transition dominance status	-0.613 ± 0.412	-1.449, 0.222	0.91	1	2.214	0.145
residuals			15.18	37		
f) Submissive behavior (untransformed)						
intercept	0.534 ± 0.025					
Initial dominance status	-0.123 ± 0.050	-0.225, -0.022	0.147	1	6.056	**0.019**
Dominance status after transition	-0.384 ± 0.050	-0.486, -0.283	1.427	1	58.739	**<0.001**
Initial * transition dominance status	-0.174 ± 0.100	-0.377, 0.029	0.073	1	3.009	***0.091***
residuals			0.899	37		

Table 2 Focal female behavior three weeks after the social niche transition. Dominance status in initial social pair and after social niche transition were contrast coded, and reference levels are "subdominant". Estimates were obtained using the summary() function and ANOVA type III tables were generated using the Anova() function from the car package. Differences were considered significant at $p < 0.05$ (bold)

Fixed effects	Estimate ± SE	95% CI	Sum Sq	df	F value	P
a) Aggressive behavior (square root transformed)						
intercept	2.234 ± 0.176					
Initial dominance status	-0.097 ± 0.351	-0.808, 0.615	0.090	1	0.076	0.785
Dominance status after transition	1.705 ± 0.351	0.993, 2.416	28.056	1	23.555	**<0.001**
Initial * transition dominance status	-0.591 ± 0.702	-2.014, 0.832	0.844	1	0.709	0.405
residuals			44.069	37		
b) Approach (log transformed)						
intercept	2.309 ± 0.124					
Initial dominance status	-0.183 ± 0.247	-0.684, 0.319	0.323	1	0.546	0.465
Dominance status after transition	0.071 ± 0.247	-0.431, 0.572	0.049	1	0.082	0.776
Initial * transition dominance status	0.009 ± 0.495	-0.994, 1.012	0.000	1	0.0004	0.985
residuals			21.885	37		
c) Play behavior (square root transformed)						
intercept	2.738 ± 0.336					
Initial dominance status	-0.794 ± 0.671	-2.154, 0.567	5.761	1	1.324	0.245
Dominance status after transition	-0.169 ± 0.671	-1.530, 1.191	0.538	1	0.124	0.802
Initial * transition dominance status	-0.536 ± 1.343	-3.257, 2.185	0.693	1	0.159	0.692
residuals			161.055	37		
d) Sniffing behavior (square root transformed)						
intercept	1.343 ± 0.141					
Initial dominance status	-0.017 ± 0.282	-0.588, 0.555	0.009	1	0.011	0.917
Dominance status after transition	0.016 ± 0.282	-0.555, 0.587	0.097	1	0.127	0.724
Initial * transition dominance status	-1.005 ± 0.564	-2.148, 0.137	2.440	1	3.178	***0.083***
residuals			28.406	37		
e) Submissive behavior (untransformed)						
intercept	0.515 ± 0.022					
Initial dominance status	-0.107 ± 0.043	-0.195, -0.019	0.111	1	6.115	**0.018**
Dominance status after transition	-0.555 ± 0.043	-0.642, -0.467	2.972	1	164.453	**<0.001**
Initial * transition dominance status	-0.090 ± 0.087	-0.265, 0.086	0.020	1	1.076	0.306
residuals			0.669	37		

Table 3 Statistical analysis of cortisol concentrations six weeks after the formation of the initial social pairs (CRT2). Dominance status (subdominant or dominant) was contrast coded, and subdominant is the reference category. Continuous fixed effects (body weight and cortisol concentration at CRT1) were mean centered for better interpretability of main effect estimates. Estimates were obtained using the summary() function from the lmerTest package and ANOVA type III tables were generated using the Anova() function from the car package. Differences were considered significant at $p < 0.05$ (bold)

	Estimate ± SE	95% CI	Sum Sq	Df	F value	P
Baseline cortisol (untransformed)						
intercept	474.742 ± 30.529					
dominance status	150.611 ± 63.974	20.988, 280.234	199847	1	5.543	**0.024**
body weight	-1.880 ± 0.664	-3.225, -0.534	288820	1	8.010	**0.007**
cortisol concentration at CRT1	-0.121 ± 0.080	-0.284, 0.041	82231	1	2.281	0.139
residuals			1334096	37		
Cortisol responsiveness after one hour (Transformation: x^2)						
intercept	2436279 ± 118957					
dominance status	-99741.2 ± 247446	-600671, 401188	9.2713×10^{10}	1	0.163	0.689
body weight	-189.9 ± 2383.6	-5015, 4635	3.6201×10^{9}	1	0.006	0.937
cortisol concentration at CRT1	194.2 ± 326.9	-468, 856	2.0132×10^{11}	1	0.353	0.556
residuals			2.1684×10^{13}	38		
Cortisol responsiveness after two hours (untransformed)						
intercept	1951.585 ± 37.999					
dominance status	-99.654 ± 78.937	-259.453, 60.146	92648	1	1.594	0.214
body weight	-0.565 ± 0.763	-2.110, 0.980	31895	1	0.549	0.463
cortisol concentration at CRT1	0.014 ± 0.114	-0.216, 0.243	824	1	0.014	0.906
residuals			2208991	38		

Table 4 Cortisol concentrations on the first and third day after initial pair formation and social niche transition. Body weight was mean-centered for better interpretability of main effect estimates. Day and dominance status were contrast coded, and reference levels are “day 1” and “subdominant”. Estimates were obtained using the summary() function and ANOVA type III tables were generated using the anova() function from the lmerTest package. Differences were considered significant at $p < 0.05$ (bold)

Fixed effects	Estimate ± SE	95% CI	SumSq	DenDF	F value	P
Initial pair formation, d1 and d3 combined (log transformed)						
Intercept	7.022 ± 0.033					
body weight	-0.004 ± 0.001	-0.005, -0.002	1.941	78	22.345	**<0.001**
day	0.163 ± 0.066	0.037, 0.290	0.535	78	6.154	**0.015**
dominance status	0.086 ± 0.067	-0.043, 0.215	0.142	78	1.635	0.205
dominance status * day	-0.030 ± 0.132	-0.283, 0.223	0.005	78	0.053	0.819
Social niche transition, d1 and d3 combined (log transformed)						
Intercept	6.266 ± 0.050					
body weight	-0.001 ± 0.001	-0.003, 0.001	0.100	39.321	1.582	0.216
day	-0.295 ± 0.058	-0.407, -0.185	1.611	39.512	25.453	**<0.001**
initial dominance status	0.065 ± 0.102	-0.127, 0.259	0.026	35.622	0.405	0.529
transition dominance status	-0.122 ± 0.107	-0.322, 0.081	0.083	35.906	1.309	0.260
initial * transition dominance status	0.148 ± 0.199	-0.226, 0.523	0.035	35.417	0.553	0.462
day * initial dominance status	-0.074 ± 0.114	-0.293, 0.144	0.027	36.314	0.422	0.520
day * transition dominance status	-0.064 ± 0.115	-0.283, 0.154	0.020	36.339	0.317	0.577
day * initial * transition dominance status	0.188 ± 0.229	-0.249, 0.625	0.042	36.415	0.670	0.418

Table 5 Statistical analysis of cortisol concentrations after the social niche transition (data from CRT3 and CRT4). Body weight was mean-centered for better interpretability of main effect estimates. CRT, dominance status in initial social pair, and dominance status after social niche transition were contrast coded, and reference levels are "CRT3" and "subdominant". Estimates were obtained using the summary() function and type three anova tables were generated using the anova() function from the lmerTest package. Differences were considered significant at $p < 0.05$ (bold); $0.05 \le p < 0.1$ was considered a trend (bold and italic)

Fixed effects	Estimate ± SE	95% CI	Sum Sq	DenDF	F value	P
a) Baseline cortisol (Transformation: Log(x))						
intercept	6.117 ± 0.053	6.018, 6.216				
body weight	0.0001 ± 0.0010	-0.002, 0.002	0.002	41.361	0.021	0.886
CRT	0.106 ± 0.085	-0.055, 0.266	0.147	63.761	1.555	0.217
initial dominance status	-0.012 ± 0.108	-0.215, 0.191	0.001	36.272	0.012	0.912
transition dominance status	-0.016 ± 0.110	-0.224, 0.192	0.002	36.496	0.022	0.882
initial * transition dominance status	0.050 ± 0.210	-0.345, 0.446	0.005	36.018	0.057	0.813
CRT * initial dominance status	0.236 ± 0.140	-0.030, 0.503	0.269	36.889	2.846	0.100
CRT * transition dominance status	-0.206 ± 0.140	-0.472, 0.061	0.204	36.913	2.159	0.150
CRT * initial status * transition status	0.192 ± 0.280	-0.342, 0.725	0.044	37.038	0.468	0.498
b) Cortisol response after one hour (Transformation: √x)						
intercept	7.318 ± 0.030	7.261, 7.375				
body weight	-0.0013 ± 0.0006	-0.0024, -0.0002	0.080	46.417	5.212	**0.027**
CRT	0.100 ± 0.039	0.026, 0.174	0.101	71.196	6.586	**0.012**
initial dominance status	-0.013 ± 0.062	-0.129, 0.105	0.001	36.128	0.041	0.840
transition dominance status	-0.083 ± 0.064	-0.201, 0.038	0.026	36.532	1.700	0.200
initial * transition dominance status	0.139 ± 0.121	-0.089, 0.367	0.020	35.675	1.323	0.258
CRT * initial dominance status	0.009 ± 0.056	-0.099, 0.116	0.0003	36.428	0.025	0.876
CRT * transition dominance status	-0.061 ± 0.056	-0.168, 0.047	0.018	36.470	1.154	0.290
CRT * initial status * transition status	0.083 ± 0.113	-0.132, 0.299	0.008	36.692	0.536	0.469
c) Cortisol response after two hours (Transformation: Log(x))						
intercept	7.567 ± 0.023	7.524, 7.610				
body weight	-0.0006 ± 0.0004	-0.0014, 0.0002	0.018	45.363	1.788	0.188
CRT	0.054 ± 0.031	-0.004, 0.113	0.032	70.233	3.063	***0.084***
initial dominance status	0.034 ± 0.047	-0.055, 0.123	0.005	36.478	0.514	0.478
transition dominance status	-0.045 ± 0.048	-0.136, 0.046	0.009	36.838	0.888	0.352
initial * transition dominance status	0.161 ± 0.092	-0.013, 0.334	0.031	36.073	3.045	***0.090***
CRT * initial dominance status	-0.040 ± 0.046	-0.128, 0.048	0.008	36.856	0.745	0.394
CRT * transition dominance status	-0.094 ± 0.046	-0.182, -0.006	0.042	36.894	4.117	**0.049**
CRT * initial status * transition status	-0.137 ± 0.093	-0.313, 0.040	0.022	37.092	2.172	0.149

Num DF for all predictors of all models = 1.

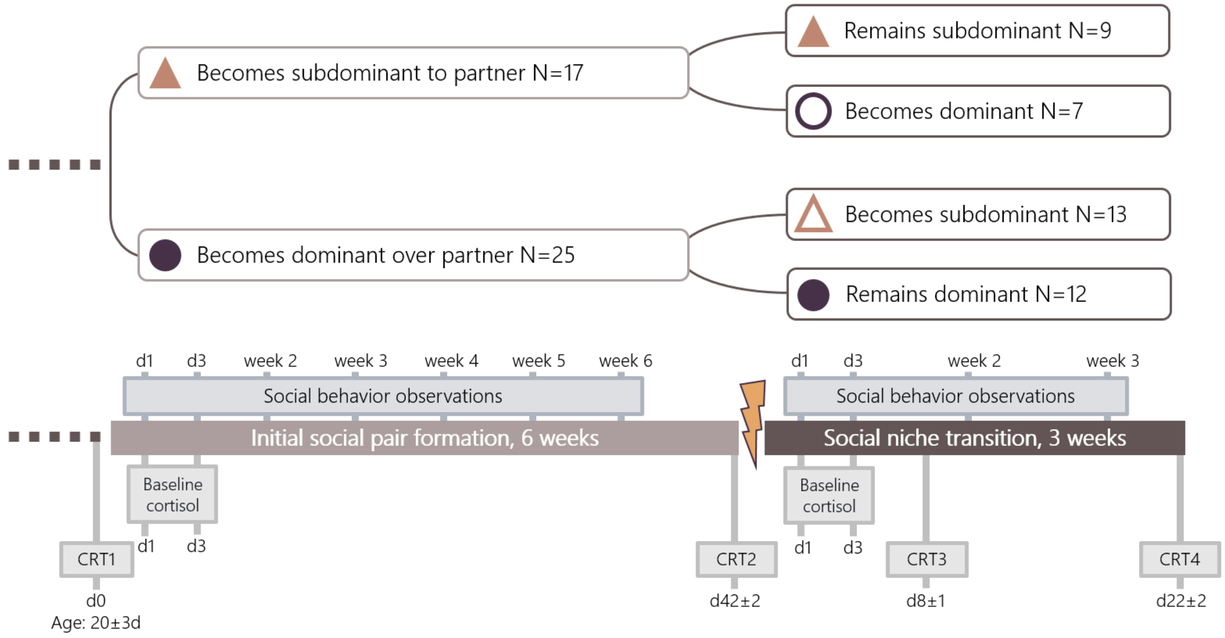

Figure 1 Experimental design. For the initial social pair formation, focal females were pair-housed with a same-aged female partner for six weeks. Then, a social niche transition occurred in which focal females were housed with either an older, larger or a younger, smaller female partner for three weeks. Videos of social behavior in the home enclosures were recorded on d1 (3h), d3 (1h) and weekly (1h). Cortisol response tests (CRT1-4) occurred the day before the start of the initial social pair formation (CRT1), on the day before the social niche transition (CRT2), approximately one week after the social niche transition (CRT3), and again three weeks after the social niche transition (CRT4). Baseline cortisol concentration was additionally measured on d1 and d3 after both the initial social pair formation and social niche transition

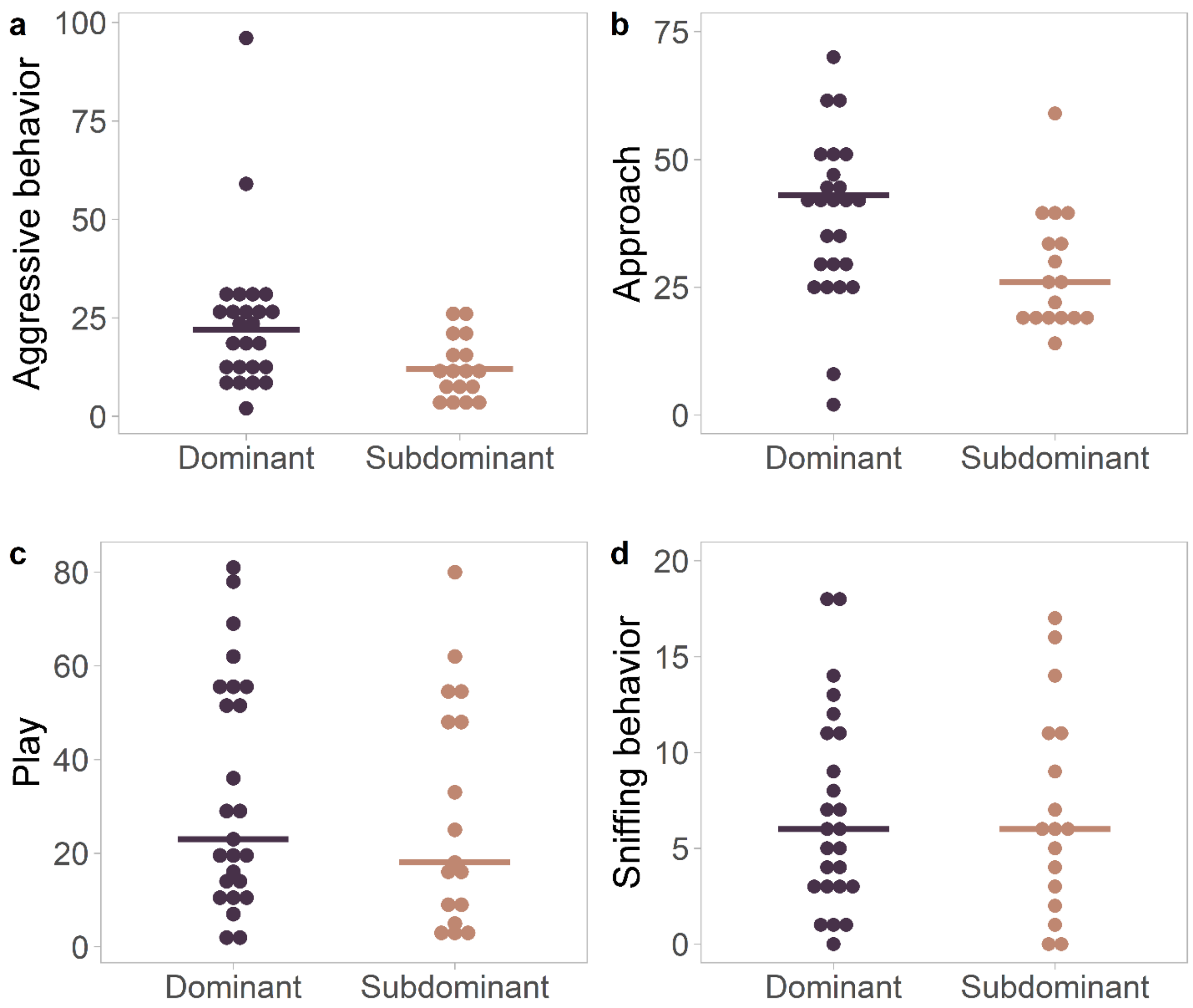

Figure 2 Behavior from weeks four, five, and six combined after initial social pair formation for dominant (N = 25; purple) and subdominant (N = 17; orange) individuals. Plotted are individual data points (dots) and medians (bars) of number of observations per three hours. Dominant and subdominant females significantly differed in the amount of aggressive behavior and approaches; dominant females were more often aggressive (Wilcoxon test, W = 317, P = 0.008) and approached their partners more often (Wilcoxon test, W = 316, P = 0.008). Dominant and subdominant females did not differ in the amount of play behavior (Wilcoxon test, W = 244.5, P = 0.419) or sniffing behavior (Wilcoxon test, W = 213.5, P = 0.989) observed

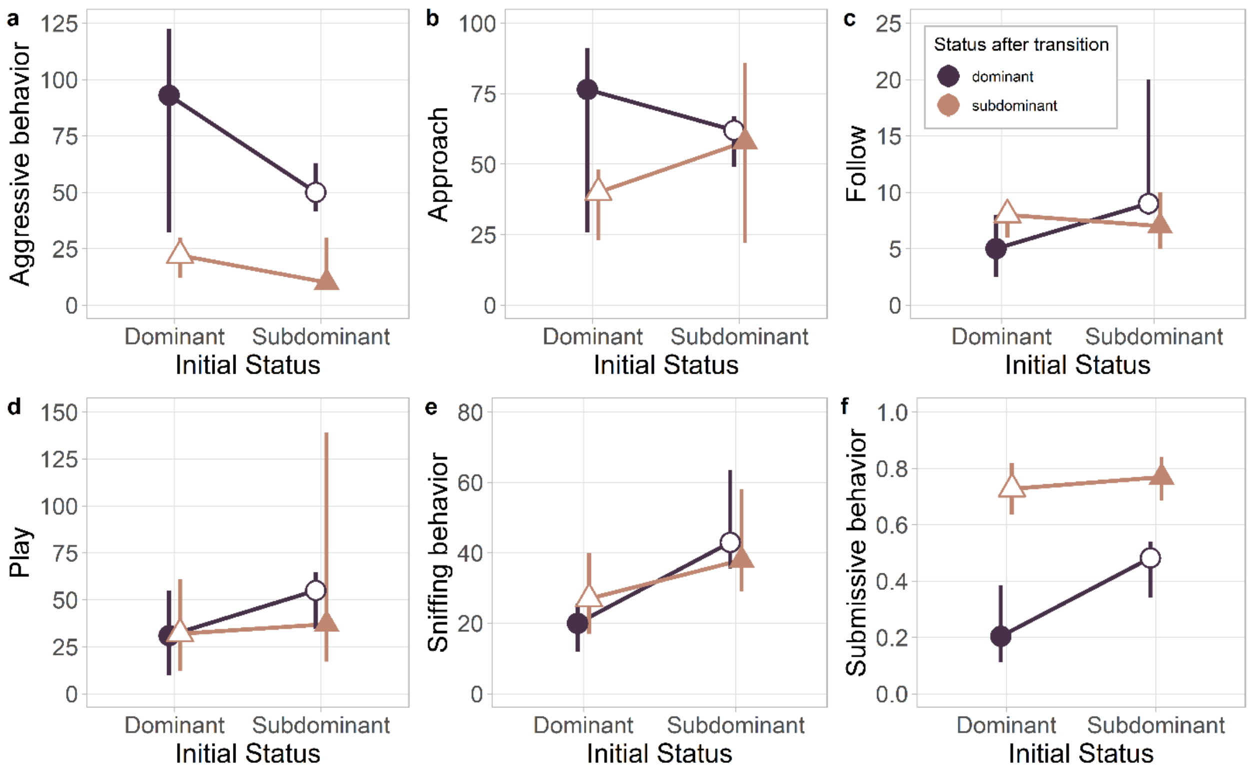

Figure 3 Behavior on d1 after the social niche transition was affected by the previous dominance status in the initial pair (follow, sniffing behavior, submissive behavior), the current dominance status after social niche transition (aggressive behavior, submissive behavior), and an interaction between the dominance status in both pairs (follow, submissive behavior). See Table 1 for statistics. Plotted are medians; error bars represent first and third quartiles. The dominance status from the initial social pair is grouped on the X axis and dominance status after the social niche transition is indicated by the connected points (purple circles: dominant; orange triangles: subdominant). Treatments that underwent a shift in dominance status are represented by hollow shapes; treatments that maintained the same dominance status are represented by filled shapes

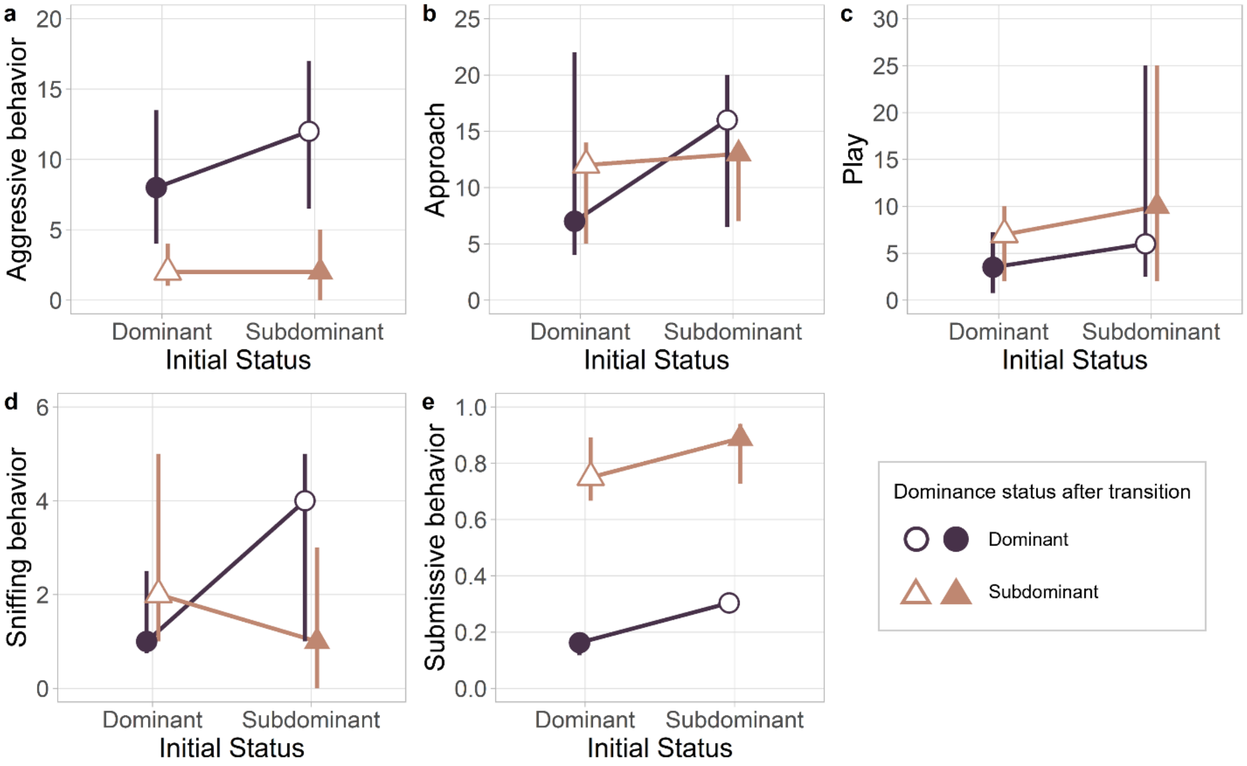

Figure 4 Behavior three weeks after the social niche transition was affected by the current dominance status (aggressive behavior, submissive behavior), previous dominance status in the initial social pair (submissive behavior), and an interaction of dominance status in both social pairs (sniffing behavior). See Table 2 for statistics. Plotted are the medians grouped by previous dominance status in the initial social pair (x axis) and current dominance status after the social niche transition (connected points; purple dots: dominant, orange triangles: subdominant). Error bars reflect first and third quartiles. Treatments that underwent a shift in dominance status are represented by hollow shapes; treatments that maintained the same dominance status are represented by filled shapes. See Table 2 for statistics

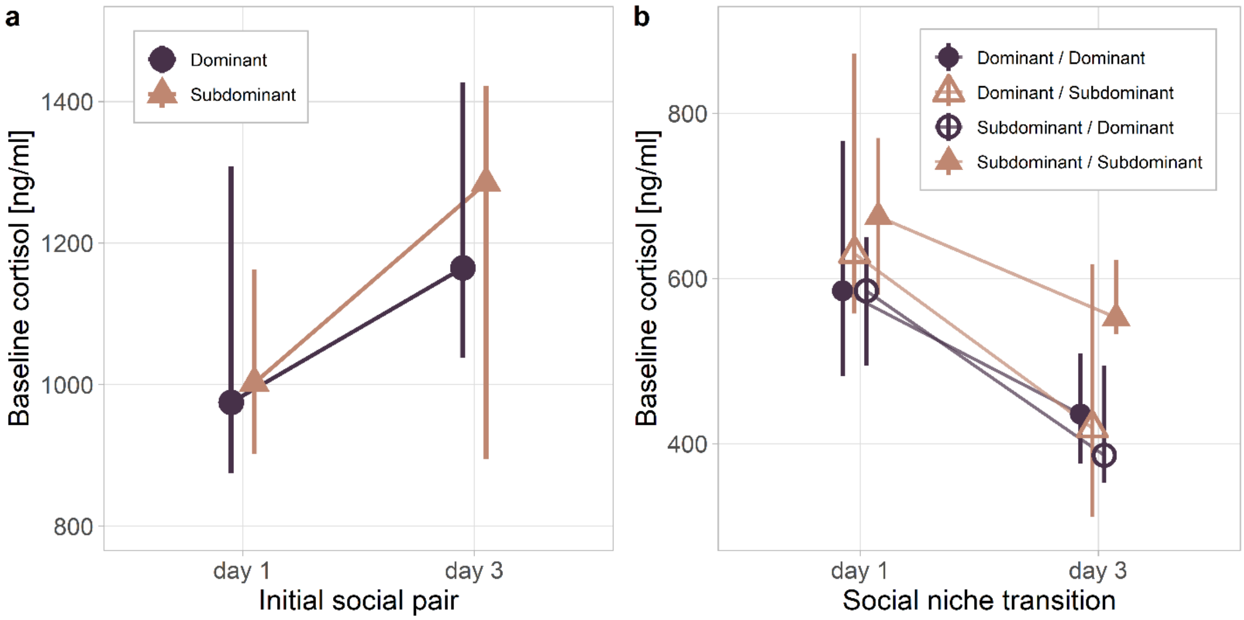

Figure 5 Baseline cortisol concentration on the first (d1) and third (d3) day after initial social pair formation (a) and after social niche transition (b). Baseline cortisol was significantly correlated to day (see Table 4); baseline cortisol increased from d1 to d3 after the initial social pair formation and decreased from d1 to d3 after social niche transition. Plotted are medians with first and third quartiles, grouped by dominance status

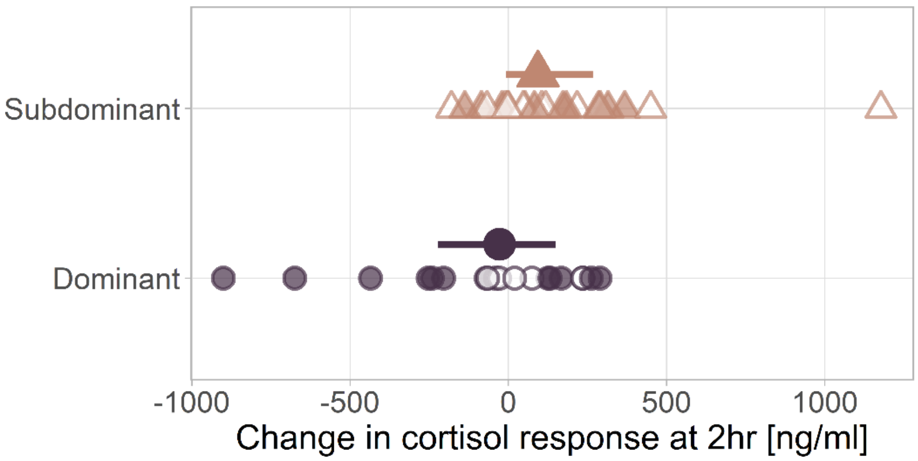

Figure 6 The change in cortisol responsiveness at two hours [ng/ml] from one (CRT3) to three (CRT4) weeks after the social niche transition. There was a significant interaction effect between CRT and dominance status after social niche transition (see Table 5 for statistics); the cortisol response of individuals who became subdominant after the social niche transition increased over time while the cortisol response of individuals who became dominant after the social niche transition decreased over time. Individual data points are plotted, grouped by dominance status after the social niche transition. Filled points indicate individuals who maintained the same dominance status in both social pairs; empty points indicate individuals who had changed dominance status from the first to the second social pair. Medians and first and third quartiles are plotted above individual data points for dominant (purple circle) and subdominant (orange triangle) status after social niche transition