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Behavioural and physiological reactions of goats confronted with an unfamiliar group either when alone or with two peers

Antonia Patt^{a,b,*}, Lorenz Gygax^a, Beat Wechsler^a, Edna Hillmann^b, Rupert Palme^c, Nina M. Keil^a

^a Centre for Proper Housing of Ruminants and Pigs, Federal Veterinary Office, Agroscope Reckenholz-Tänikon Research Station ART,

Tänikon, 8356 Ettenhausen, Switzerland

^b ETH Zurich, Institute of Agricultural Sciences, 8092 Zurich, Switzerland

^c Department of Biomedical Sciences/Biochemistry, University of Veterinary Medicine, 1210 Vienna, Austria

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ABSTRACT

When introduced into a new herd, goats are confronted with unfamiliar animals. Their behavioural and physiological reactions during this confrontation are likely to differ depending on the presence or absence of familiar conspecifics (peers). To assess these reactions, we confronted 12 goats both alone and with two peers (confrontees) with established groups (n=4 groups) consisting of goats unfamiliar to the confrontee (unfamiliar goats) $(12 \text{ goats} \times 2 \text{ confrontations} = 24 \text{ confrontations in total})$. Each confrontation lasted for one hour. Agonistic interactions, sniffing behaviour and level of activity were recorded throughout the confrontations. In addition, concentrations of cortisol metabolites were measured in faecal samples taken in the evening before the confrontation and three successive samples after the confrontation. Before the start of the experiment, we evaluated the dominance relationships of the involved goats within their respective housing groups by direct observations made during the main feeding times. Data were analysed using generalised linear mixed-effects models with the fixed effects presence of peers (yes, no), rank category (high, medium, low) and repeated confrontation (numeric variable). For the analysis of activity level and concentrations of faecal cortisol metabolites, period (minutes 0-15, 16-30, 31-45, 46-60) and sample (control, 13, 14, 15 h after the confrontation), respectively, were included as additional fixed effects. Unfamiliar goats directed fewer agonistic interactions towards confrontees when the latter were accompanied by peers compared to when they were alone (without peers: 57; with peers: 20 interactions per animal and confrontation). The same was true for the proportion of agonistic interactions involving physical contact (without peers: 69; with peers: 53%) and the number of sniffing behaviours (without peers: 16; with peers: 9 interactions per animal and confrontation). On the other hand, confrontees with peers were more likely to direct agonistic and sniffing behaviour towards unfamiliar goats than those on their own. Confrontees with peers had lower concentrations of faecal cortisol metabolites after confrontations (without peers: 273; with peers: 198 ng/g). For confrontees (with and without peers), activity level was highest during the first 15 min of the confrontation and decreased over its course. For the unfamiliar goats, the activity pattern was similar but was modulated by rank, with higher values for low-ranking goats than for medium- and high-ranking ones. In conclusion, our results indicate that the presence of peers is advantageous for goats being introduced into groups of unfamiliar goats.

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^{*} Corresponding author at: Centre for Proper Housing of Ruminants and Pigs, Federal Veterinary Office, Agroscope Reckenholz-Tänikon Research Station ART, Tänikon, 8356 Ettenhausen, Switzerland. Tel.: +41 52 368 33 43; fax: +41 52 365 11 90.

E-mail address: antonia.patt@agroscope.admin.ch (A. Patt).

1. Introduction

The introduction of individual goats into small established groups has considerable negative consequences for the introduced goat's welfare, including a substantial increase in lying duration (more than 20 h per day) and reduced feeding duration, as well as elevated levels of cortisol metabolites. Additionally, introduced goats received a considerable number of agonistic interactions on the first day of the introduction period (Patt et al., 2012). It is not known, however, whether these effects can be mitigated by introducing a goat together with familiar conspecifics (peers).

This question has been addressed for dairy cows in several studies (O'Connell et al., 2008; Gygax et al., 2009; Neisen et al., 2009) which aim to reduce the negative effects of introducing unfamiliar individuals, such as increased levels of aggression, impaired lying and feeding behaviour, and reduced performance (Brakel and Leis, 1976; von Keyserlingk et al., 2008). In our opinion, the presence of conspecifics could benefit the animals being introduced, either by providing social support, i.e. enhancing their coping ability (reviewed by Rault, 2012), and/or via a dilution effect, in which a more-or-less constant number of agonistic interactions is distributed equally among several animals, instead of being directed against just one (Neisen et al., 2009). Indeed, Neisen et al. (2009) reported fewer agonistic interactions after the introduction of a pair of heifers than after the introduction of just one. Further, heifers introduced as pairs were found to spend more time in the lying area (Gygax et al., 2009) and to spend more time lying when most animals of the herd were lying too (O'Connell et al., 2008). For dairy cows, therefore, the presence of conspecifics seems to be a promising approach to reducing the negative effects associated with introducing a new animal.

In studies on the consequences of regrouping sheep and cows (Sevi et al., 2001; Schirmann et al., 2011) it was also found that adverse effects were stronger for animals having to deal with relocation to a new environment in addition to the grouping, whilst remaining in the home pen appeared to give residents an advantage over the relocated individuals. These two aspects potentially apply to both the residents' and the introduced animals' perspective and must be considered as confounding factors. Consequently, in order to specifically test the effects of the presence of familiar conspecifics during social confrontations, it is essential to use a place that is equally familiar to all of the involved animals.

In the present study, we assessed whether the presence of familiar conspecifics reduces negative effects during social confrontations with unfamiliar goats. The confrontations took place in a neutral arena, so as to exclude effects of residency and relocation to a new environment as potentially confounding factors. We were interested in the behavioural and physiological responses caused by confrontation both with and without peers. Moreover, we investigated whether these responses were modulated by the social rank of both confrontees and unfamiliar goats. We addressed these questions by consecutively confronting goats (= confrontees) which were either alone or with two peers, with an established group of six goats (= unfamiliar group). We expected confrontees with peers to be less affected by the confrontation than those introduced on their own. This might potentially be reflected in their being on the receiving end of fewer agonistic interactions, as well as in lower levels of activity and of faecal cortisol metabolites. Furthermore, we anticipated that unfamiliar goats would react differently when faced with an individual confrontee as opposed to a confrontee accompanied by two peers.

2. Methods

2.1. Animals and housing conditions

Four established groups of six (= unfamiliar groups) and four groups of three (= confrontees) horned, non-lactating female goats were used in the experiment. Confrontees were either confronted with an unfamiliar group individually or in the presence of the two peers with which they were housed. The goats had been grouped prior to the study in early March 2011 from individuals of various Swiss milking breeds (Saanen, Toggenburger, St Gallen Booted, Grisons Striped, Peacock, Nera Verzasca and Valais Blackneck) and their crossbreeds. Two goats were part Anglo Nubian. The animals were born between 2003 and 2009, and the experiment was conducted from May to July 2011. As far as possible, group composition was balanced in terms of breed, age and weight.

All groups were housed in the same building in identical pens, and had acoustic and visual contact as described in Patt et al. (2012). Two groups of unfamiliar goats were each housed adjacent to a group of confrontees, but separated by a wooden wall to prevent tactile contact. The total area of each pen was 15.3 m^2 (approx. $3 \text{ m} \times 5 \text{ m}$), consisting of a deep-bedded straw area of 11.7 m² and an elevated feeding place (3.6 m²) divided by a wooden wall into two equal-sized compartments. The deep-bedded area of each group of unfamiliar goats and each group of confrontees was further structured by a wooden platform and a freestanding partition providing climbing opportunities, and both elevated and protected lying areas. Hay was provided ad libitum in the feeding area from a 3 m hayrack refilled twice daily at around 8.45 am and 5 pm. One water trough, one licking stone and a brush were provided in each pen.

2.2. Dominance relationships

Shortly before the start of the experiment, the dominance relationships of the goats in each group ('unfamiliar' goats and 'confrontees') were evaluated by direct observation during morning and evening feeding times according to the method used by Aschwanden et al. (2008). With the help of the rank index (between 0 = omega and 1 = alpha), each goat was categorised as either low- (0.0–0.2), medium- (0.4–0.6) or high-ranking (0.8–1).

2.3. Confrontations

Based on the results of a previous study (Patt et al., 2012) confrontations of unfamiliar goats lasting longer than a few

hours had serious effects in terms of animal welfare. Additionally, confrontations of unfamiliar goats may bear a risk of being injured. However, in the study of Patt et al. (2012) the registered injuries were only mild. Thus, we considered confrontations lasting one hour to be acceptable for the present study and were prepared to terminate confrontations if a goat was attacked with risk of being injured. As the experimenter was present during all confrontations, interference could have been instantly but in effect was never required. The experiment was approved by the Cantonal Veterinary Office (Frauenfeld, Thurgau, Switzerland; Approval F4/09).

Twenty-four confrontations were initiated, each of which lasted for one hour. One-half of these confrontations (= 12) was initiated between a single confrontee and an unfamiliar group, whilst the other half (= 12) was between a confrontee accompanied by two peers, and an unfamiliar group. Each of the four unfamiliar groups was faced with three of the four groups of confrontees, as well as with each confrontee of the fourth group (six confrontations per unfamiliar group). This fourth group of confrontees was a different one for each unfamiliar group, so that each confrontee was confronted on her own once. Each confrontee therefore experienced a total of four confrontations (3× with peers, $1 \times$ without peers). The order of confrontations with or without peers was balanced across the 24 initiated confrontations from the viewpoint of both the confrontees and the unfamiliar group. Furthermore, each group/goat had a break of at least one day in between two confrontations.

2.4. Experimental room and experimental procedure

The confrontations took place between 9 and 10 am in a separate indoor arena about 150 m from the home pens. To habituate the goats to the experimental room, each group was taken to it four times over the two weeks preceding the start of the experiment, and was allowed to explore this indoor arena for an hour each time. By the end of the habituation, goats followed the experimenter readily into the arena and moved calmly within the arena. Further, vocalisation was reduced considerably over the habituation sessions. Afterwards and during confrontations, and so as to eliminate resource-based aggression, goats were not offered any food.

During confrontations, space per goat was kept at a constant 3 m². Consequently, the arena measured either 21 m² (5 m × 4.2 m) when the confrontee was confronted on her own, or 27 m² (5 m × 5.4 m) when she was confronted in the company of her two peers. The arena was further structured by two freestanding partitions identical to those in the goats' home pens (approx. 1 m in diameter and 0.8 m in height). These partitions allowed the goats to avoid each other more effectively during confrontations, similarly to such structural elements in loose housing that have been shown to effectively reduce agonistic interactions between goats (Aschwanden et al., 2009). On the confrontation days, the unfamiliar group was always led into the arena first. A few minutes later, the corresponding group of confrontees was led into the arena, and either all three goats were confronted (= with peers), or two goats were led back and the remaining confrontee was confronted alone (= without peers) with the unfamiliar group.

2.5. Data recording

To assess the effects associated with confronting a goat with or without peers, we measured the frequency of social interactions, activity values, and concentrations of faecal cortisol metabolites. For faecal cortisol metabolites, control values were collected on the day before each confrontation (day-1). Social interactions were recorded individually for all goats involved in the confrontations, whilst activity values and concentrations of cortisol metabolites were recorded in focal goats. In the 'unfamiliar' groups, the same three focal goats representing the three rank categories (high, medium and low) were chosen for recordings in all confrontations. In the 'confrontee' groups, each of the three goats served as a focal animal once during the three confrontations with peers.

2.5.1. Agonistic and affiliative interactions

During confrontations, the social behaviour of all goats was monitored continuously by direct observation. For agonistic interactions, we distinguished between whether physical contact was involved (head butt, fight and explicit displacement) or not (threat and implicit displacement). Sniffing, scratching, licking and mock fighting were recorded as affiliative behaviours. The definitions of the recorded behaviours are given in Patt et al. (2012). For each interaction, both initiator and recipient were noted. In the case of agonistic interactions, we also noted whether the initiator was successful, i.e. whether the recipient moved location. Thus, a goat lost an interaction if she was forced to leave her current location by another goat either explicitly (e.g. fight, butt, threat) or implicitly, e.g. after being approached by another goat which did not show obvious agonistic behaviour.

2.5.2. Activity values

To measure activity, we recorded acceleration values using a commercial 3D acceleration logger (MSR145WA, Modular Signal Recorder Electronics GmbH; $33 \text{ mm} \times 15 \text{ mm} \times 61 \text{ mm}$). The logger was attached to the goat's left-hind leg to minimise risk of injury when goats stepped onto freestanding partitions. Acceleration in the direction of the y-axis (= the axis parallel to the longitudinal axis of the goat's hind leg) was continuously recorded at a measurement range of twice the earth's gravity acceleration (= m/s^2) and a rate of 10 Hz. Due to the way the logger was attached to the goats' hind leg, acceleration values observed while standing quietly equalled -1 g. When the leg was moved, acceleration values reached both higher and lower values than the -1 g. We were only interested in deviations from the value at rest and we thus added +1 to the original values and then took their absolute values. These latter values were summed across the time period of interest (basically calculating an area under the curve).

Before the start of the experiment, we validated activity values with respect to different levels of activity. The increasing level of activity from 'standing' to 'walking regularly interrupted by halts' to 'continuous walking' was reliably reflected in the activity values. The lower (0.25) and upper (0.75) quartile of activity values reached 6.68–13.34 summed gravity acceleration/min, 20.73–34.21 summed gravity acceleration/min and 83.73–104.00 summed gravity acceleration/min for standing, interrupted walking, and continuous walking, respectively.

During confrontations, acceleration values were recorded with the acceleration logger, using the same settings (2 g, 10 Hz) and position on the goat's left-hind leg. To take into account changes of activity over time, the confrontation was divided into four successive 15-min periods (period 15, period 30, period 45, period 60) for analysis. The acceleration values were calculated for each 15-min period, providing an activity value per 15 min (summed gravity acceleration/15 min [m/s²]).

2.5.3. Cortisol metabolites

To monitor an acute stressor by measuring faecal cortisol metabolites, samples should be collected within 12–15h after the event in guestion (Kleinsasser et al., 2010). As delay times vary between 12 and 15 h, samples should be taken several times in succession to allow measurement of the effect of a short-term stressor (i.e. one hour in our case). Samples were therefore collected 13, 14 and 15 h after the start of a confrontation beginning at 10 pm. To account for a possible circadian rhythm of cortisol levels, faecal samples were also taken at 10 pm on the control day (day -1). The samples were collected manually from the animal's rectum, with the goats being successively attached to the hayrack using a halter. Each sample was immediately put into a cooling box until the sampling was completed. Afterwards, all samples were frozen and stored at -20 °C until analysis. The concentrations of faecal cortisol metabolites were determined by a group-specific 11-oxoaetiocholanolone enzyme immunoassay (EIA; Möstl et al., 2002). This EIA has been successfully validated for monitoring adrenocortical activity in goats (Kleinsasser et al., 2010).

2.6. Statistical analysis

2.6.1. Model selection

In order to adequately reflect dependencies in the experimental design (nesting, repeated measurements), generalised linear mixed-effects models were used to evaluate the outcome variables. Statistical analysis was performed in R (version 2.14.1, R Development Core Team, 2011) using the lmer and glmer methods from the lme4 package (Bates et al., 2011), as well as the function dredge of the MuMIn package (Barton, 2012) to perform all subset analyses. The statistical approach taken here is based on the use of Akaike's information criterion (AIC) which provides probabilities for each of several concurrent models given the data (Burnham et al., 2011; Garamszegi, 2011; Symonds and Moussalli, 2011). In our case, AIC was further corrected for small sample sizes (AIC_c) . For each outcome variable, an all-subsets analysis was conducted, ranging from the minimal model including a constant (intercept) only (Burnham et al., 2011; Dochtermann and Jenkins, 2011) to the model including all fixed effects and their interactions. The former model corresponds to the null hypothesis that no explanatory variable has an influence, and that the responses vary randomly around a general mean. The choice among the different models was based on the Akaike weight (w_i), which can be interpreted as the probability of a given model to fit the data best within the set (all weights together add to one). For all outcome variables, the optimal model based on the Akaike weight (w_i) is shown in Table 1 and was at least 1.3 times more likely than the next best model. If models with a similar mode probability were nested and had similar AIC_c values, we followed the advice of choosing the simpler model (Richards et al., 2011). I.e. for Agonistic U-U and Cortisol U, we chose the second best model with model probabilities of 0.91 and 0.86 in relation to the best model.

Model selection is thus based on the models' relative fit within the set given the data. To visualise the relative strength of the best fitting model within the set, we also report the evidence ratio of the chosen model in comparison to the null model (E_0 in Table 1). Thus, ER_0 provides a measure of how much more likely the best fitting model is than the null model (Symonds and Moussalli, 2011). This statistical approach takes into account that any model is only an approximation of the hypothesis investigated. Thus single fixed effects are no longer 'significant', but the chosen model as a whole represents the approximation that is most likely to explain the obtained data and has to be presented. By considering effect sizes of fixed effects it can be decided whether or not the observed changes are biologically relevant (Symonds and Moussalli, 2011).

2.6.2. Outcome variables, random and fixed effects

Social behaviour was analysed from the recipient's point of view. Three types of social interactions were distinguished: a) both recipient and initiator were unfamiliar goats; b) the recipient was a confrontee or a peer, and the initiator was an unfamiliar goat; and c) the recipient was an unfamiliar goat, whilst the initiator was a confrontee or a peer. Since most types of agonistic interactions (head butts, fights, explicit displacements, threats and implicit displacement) occurred too rarely to be analysed separately, they were analysed under the comprehensive term 'agonistic interactions'. Most of the various sorts of affiliative interactions included in the ethogram were observed only sporadically. The exception was sniffing, which was therefore analysed.

The following were analysed as outcome variables (for information regarding transformation of the different variables, see Table 2):

- Number of agonistic interactions received by unfamiliar goats from other unfamiliar goats (Agonistic U-U, abbreviations as used in the tables) over the course of one confrontation (no./animal/confrontation);
- Number of agonistic interactions (Agonistic U-C) and number of sniffing interactions (Sniffing U-C) received by the confrontee or one of her peers from unfamiliar goats over the course of one confrontation (no./animal/confrontation);
- Proportion (%) of agonistic interactions lost by a confrontee or peer in relation to all classifiable agonistic



Criteria used to select the best-fitting model of each outcome variable analysed.

Outcome variable ^a	Selected models ^b	AIC _c ^c	w_i^d	ER0 ^e
Agonistic U-U (no./animal/confrontation)	Presence of peers \times rank + repetition	237.30	0.31	>62.00
Agonistic U-C (no./animal/confrontation)	Presence of peers + repetition	110.00	0.64	>128.00
Agonistic lost U-C (proportion, %)	Presence of peers	177.30	0.34	26.15
Agonistic physical U-C (proportion, %)	Presence of peers	153.60	0.51	>102.00
Sniffing U-C (no./animal/confrontation)	Presence of peers	109.80	0.63	3.94
Agonistic C-U (yes/no)	Presence of peers + repetition	163.50	0.31	>62.00
Sniffing C-U (yes/no)	Presence of peers	195.70	0.40	2.50
Activity U (m/s ²)	Presence of peers + repetition + rank × period	289.20	0.12	>24.00
Activity C (m/s^2)	Period + repetition	173.70	0.48	>96.00
Cortisol U (ng/g)	Presence of peers × repetition	276.80	0.12	6.00
Cortisol C (ng/g)	Presence of peers + rank	110.10	0.29	>58.00

^a U-U: initiator = unfamiliar goat, recipient = unfamiliar goat; U-C: initiator = unfamiliar goat, recipient = confrontee/peers; C-U: initiator = confrontee/peers, recipient = unfamiliar goat; U = unfamiliar goat, C = confrontee.

^b Fixed effects included in the best-fitting model.

^c Akaike's Information Criterion value corrected for small sample sizes.

 d w_i: Akaike weight which can be interpreted as the probability of a given model to fit the data best within the presented set.

^e *ER*₀: Evidence ratio between the chosen model and the null model (including the intercept only).

interactions received by a confrontee or one of her peers from an unfamiliar goat (Agonistic lost U-C);

- Proportion (%) of agonistic interactions involving physical contact in relation to all agonistic interactions received by a confrontee or one of her peers from an unfamiliar goat (Agonistic physical U-C);
- Whether agonistic interactions (Agonistic C-U) and sniffing (Sniffing C-U) were received by an unfamiliar goat from a confrontee or one of her peers;
- The activity values (summed gravity acceleration/15 min) of focal unfamiliar goats (Activity U) and the confrontee (Activity C) for each period;
- The concentrations of faecal cortisol metabolites (ng/g faeces) of focal unfamiliar goats (Cortisol U) and the confrontee (Cortisol C) using each sample taken at 13, 14 and 15 h after confrontations individually.

In all models, random effects consisted of the recipients being either confrontees or unfamiliar goats nested in their housing group and crossed with the group identity of the unfamiliar group or confrontees' group, respectively. For activity values and concentrations of cortisol metabolites, the sequence number of the confrontation was also nested within recipient.

Model assumptions were verified using graphical analysis of residuals focusing on normality of errors and random effects as well as homoscedasticity of the errors in the case of normally distributed errors, and on normality of random effects and absence of bias in the mean errors for the generalised models.

The presence of peers (factor with two levels: yes, no), rank category (factor with three levels: high, medium and low) and the fact that goats were confronted repeatedly (repetition as a numeric variable) were explanatory variables in all models. A fourth explanatory variable was added in some models: period (factor with four levels: period 15, 30, 45 and 60) in the case of activity values, and sample (factor with four levels: control, 13, 14 and 15 h after confrontation) in the case of faecal cortisol metabolites.

Table 2

Estimates and 95% confidence intervals^a for the fixed-effects presence of peers of all outcome variables analysed with main-effects models.

Outcome variable	Transformation	Presence of peers	
		Without peers	With peers
Agonistic U-U ^b (no./animal/confrontation)	log	-	-
Agonistic U-C (no./animal/confrontation)	log	58 [54; 62]	20 [18; 23]
Agonistic lost U-C (proportion, %)	logit	77 [43; 95]	92 [75; 98]
Agonistic physical U-C (proportion, %)	logit	69 [40; 90]	53 [27; 79]
Sniffing U-C (no./animal/confrontation)	log	16 [9; 26]	9 [6; 13]
Agonistic C-U ^c (yes/no)	logit link function	17 [8; 34]	54 [33; 73]
Sniffing C-U ^c (yes/no)	logit link function	43 [31; 56]	65 [49; 78]
Activity U ^d (m/s ²)	log	40 [27; 61]	46 [31; 70]
Activity C (m/s ²)	log	-	-
Cortisol U (ng/g)	log	-	-
Cortisol C (ng/g)	log	273 [186; 387]	198 [138; 282]

^a All values rounded to zero decimal places.

^b See Table 3 for estimates and 95% confidence intervals of the interaction between the fixed-effects peers and rank.

^c Using generalised models with binomial error distribution.

^d See Table 4 for estimates and 95% confidence intervals of the interaction between the fixed-effects period and rank.

3. Results

3.1. Qualitative observations

Confrontations were characterised by many short intervals of high levels of activity and agonistic interactions. During these intervals, which had no recognisable trigger, unfamiliar goats directed many agonistic interactions towards confrontees and/or their peers, and chased them around. Unfamiliar goats often ran towards confrontees and/or their peers at full speed and butted them. Although many agonistic interactions that unfamiliar goats initiated against the confrontee or one of her peers involved physical contact, the mean number of fights was low with only 3.5 initiated fights per confrontation. In between these intervals, confrontees and peers tended to remain mostly along the outer wall of the arena, in corners, or on the wooden partitions. Where peers were present, they and the confrontees tended to stay close together, often maintaining bodily contact. During confrontations, goats were almost never observed to lie down. The 24 confrontations resulted in a total of eight injuries (seven abrasions and one haematoma) on five different goats (four confrontees and one unfamiliar goat).

3.2. Effects of the presence of peers (yes vs. no)

Whether or not a peer was present appeared to be important for all outcome variables, except for the activity of confrontees (Table 1, Activity C). Confrontees were on the receiving end of the greatest number of agonistic interactions/confrontations initiated by unfamiliar goats – around 54 – in the absence of peers. Accordingly, the number of instances of both agonistic interactions and sniffing behaviour was substantially lower for confrontees with peers as opposed to those on their own (Table 2: Agonistic U-C, Fig. 1a and Sniffing U-C, Fig. 1b). When looking at proportions rather than the absolute number of agonistic interactions, the proportion of agonistic interactions involving physical contact in relation to all agonistic interactions that were directed by unfamiliar goats towards confrontees and/or their peers was slightly lower where confrontees were with peers rather than alone (Table 2: Agonistic physical U-C, Fig. 2a). Although the proportion of agonistic interactions from which the confrontees and/or their peers emerged as losers in relation to all agonistic interactions was generally high, it was even higher when confrontees were confronted together with peers as opposed to without (Table 2: Agonistic lost U-C, Fig. 2b). Moreover, although the activity values of the confrontees did not vary depending on whether or not they were accompanied by peers (Activity C), concentrations of faecal cortisol metabolites were lower when peers were present (Table 2: Cortisol C).

Looking at the agonistic interactions initiated by confrontees and/or peers, it is apparent that very few unfamiliar goats were on the receiving end of agonistic interactions initiated by an unaccompanied confrontee (Fig. 3a). The proportion of unfamiliar goats receiving agonistic interactions were much higher when confrontees were accompanied by peers as opposed to being confronted on their own (Table 2: Agonistic C-U). Confrontees and/or peers directed sniffing behaviour towards a higher proportion of unfamiliar goats than agonistic interactions. Moreover, there was a greater probability of confrontees sniffing at unfamiliar goats when the former were accompanied by peers rather than alone (Table 2: Sniffing C-U; Fig. 3b). Fewer agonistic interactions were directed against low-ranking unfamiliar goats by other unfamiliar goats when confrontees were accompanied by peers than when they were alone (Table 3: Agonistic U-U). Furthermore, the activity values of unfamiliar goats were somewhat higher when the confrontee was accompanied by peers (Table 2: Activity U). Whereas concentrations of faecal cortisol metabolites in the unfamiliar goats (Cortisol U) decreased between confrontations when the confrontee was alone, they remained constant when the confrontee was with peers. Thus, when the confrontee was alone concentrations were reduced from 379 ng/g [274; 509] during the 1st confrontation to 202 ng/g [148; 271] during 6th





Fig. 1. a) Total number of agonistic interactions and b) number of sniffing behaviours directed by unfamiliar goats towards confrontees confronted either individually (without peers) or accompanied by peers (with peers). Box-and-whiskers plot: boxes = 1st and 3rd quartile, thick line = median, whiskers = range from minimum to maximum value. Solid lines = model estimates, dotted lines = 95% confidence intervals. Model estimate of agonistic interactions takes repeated testing into account.



Fig. 2. a) Proportion (%) of agonistic interactions with physical contact and b) proportion (%) of agonistic interactions lost by the confrontee or one of her peers out of all agonistic interactions directed by unfamiliar goats towards either individually confronted goats (without peers) or the confrontee or one of her peers (with peers). Box-and-whiskers plot: boxes = 1st and 3rd quartile, thick line = median, whiskers = range from minimum to maximum value. Solid lines = model estimates, dotted lines = 95% confidence intervals.



Fig. 3. Proportion of unfamiliar goats towards whom a) agonistic interactions and b) sniffing behaviour were directed either by individually confronted goats (without peers) or by confrontees accompanied by peers (with peers). Filled circles = proportion in the four experimental groups. Solid lines = model estimates taking repeated testing into account, dotted lines = 95% confidence intervals.

confrontation. When the confrontee was with peers, concentrations during the 1st confrontation were 274 ng/g [201; 368] compared to 279 ng/g [206; 377] during the 6th confrontation.

3.3. Effects of repetition

Apart from the unfamiliar goats' cortisol metabolites concentrations (Cortisol U), where the effect of repetition

Table 3Estimates and 95% confidence intervals for agonistic interactions betweenunfamiliar goats (Agonistic U-U; no./animal/confrontation).

Presence of peers × Rank				
High	n Medium			
	Without peers			
1.1 [0.8; 1.5]	1.3 [0.9; 1.8] With peers	7.7 [5.7; 10.5]		
0.9 [0.7; 1.3]	1.2 [0.9: 1.6]	3.4 [2.5; 4.6]		

interacted with presence of peers, repetition was included as a main effect in the best-fitting models of some other variables. Being confronted repeatedly reduced the number of agonistic interactions initiated by unfamiliar goats and experienced by confrontees and/or peers (Agonistic U-C, a reduction from 44 interactions [23; 84] during the 1st confrontation to 18 interactions [9; 34] during the 4th), as well as reducing the proportion of unfamiliar goats being on the receiving end of agonistic interactions initiated by confrontees and/or peers (Agonistic C-U, i.e. a reduction from a proportion of 50% [30; 70] unfamiliar goats during the 1st confrontation to 22 [11; 41] during the 6th). Similarly, activity values of both unfamiliar goats and confrontees decreased with repeated confrontations (Activity U, i.e. from 50 m/s^2 [33; 70] during the 1st confrontation to 36 m/s² [24; 52] during the 6th; Activity C, i.e. from 70 m/s^2 [42; 109] during the 1st confrontation to 20 m/s^2 [9; 50] during the 4th). Despite this, the number of agonistic interactions between unfamiliar goats increased slightly (Agonistic U-U, i.e. from 1.6 interactions [1.2; 2.2] during



Fig. 4. Activity values for confrontees with respect to the four 15-min periods during a confrontation. Box-and-whiskers plot: boxes = 1st and 3rd quartile, thick line = median, whiskers = range from minimum to maximum value. Solid lines = model estimates taking repetition/repeated testing into account, dotted lines = 95% confidence intervals.

the 1st confrontation to 2.1 interactions [1.5; 2.9] during the 6th).

3.4. Effects of period (period 15, 30, 45, 60)

The confrontees' activity values were higher during the first period of a confrontation than during the following three periods (Activity C, period 15: 112 m/s^2 [72; 181], period 30: 57 m/s², [36; 90]; period 45: 50 m/s² [31; 80]; period 60: 33 m/s² [20; 53]; Fig. 4). In focal unfamiliar goats, the decrease in activity values from period 15 to period 60 interacted with social status, since the activity levels of low-ranking goats were higher than those of medium-and high-ranking goats, especially in period 15 (Table 4: Activity U).

3.5. Effects of rank (high, medium, low)

Rank as a main effect was included in the bestfitting model of the confrontees' cortisol metabolites concentrations (Cortisol C). Further, rank was included in two other models: agonistic interactions between unfamiliar goats (Table 3: Agnostic U-U), where rank interacted

Table 4

Estimated effects and 95% confidence intervals a for activity values of unfamiliar goats (Activity U; m/s^2).

$Rank \times Period$			
Period 15	Period 30	Period 45	Period 60
High 50 [33; 79] Medium	33 [22; 52]	27 [18; 42]	24 [16; 38]
75 [48; 120] Low	41 [26; 64]	38 [25; 61]	39 [25; 60]
99 [62; 162]	58 [37; 93]	45 [29; 75]	36 [23; 58]

^a All values rounded to zero decimal places.

with the presence of peers, and in the case of unfamiliar goats' activity values (Table 4: Activity U), where it interacted with period. Concentrations of faecal cortisol metabolites decreased along with the goat's rank, from high- to medium- and low-ranking goats (high-ranking: 316 ng/g [223; 451], medium-ranking: 275 ng/g [195; 381]; low-ranking: 143 ng/g [102; 202]).

4. Discussion

In the present study, we tested whether the presence of peers reduced the negative effects of social confrontation experienced by confrontees when confronted without peers. In addition, we aimed to test whether these two confrontation paradigms had different effects on the reactions of the unfamiliar goats with which the confrontees were faced.

Our results show that the presence of peers reduced the total number of agonistic interactions, as well as the proportion of agonistic interactions involving physical contact directed against the confrontees and/or peers by unfamiliar goats. Moreover, the confrontees' cortisol metabolites concentrations were lower in the presence of peers than in their absence. Consequently, the presence of familiar conspecifics was shown to be advantageous for goats during confrontations.

This advantage might be the result of both social support (Rault, 2012) and a dilution effect (Neisen et al., 2009), with the data providing indications for both. On the one hand, the number of agonistic interactions directed by unfamiliar goats towards confrontees accompanied by two peers is roughly one-third of the number directed at non-accompanied confrontees, and suggests a dilution effect. On the other hand, the observation that confrontees lost more agonistic interactions when accompanied by peers than when unaccompanied, but still had lower levels of cortisol metabolites, might suggest an element of social support. In a study of pigs, it was shown that losing most agonistic interactions is associated with higher cortisol concentrations than winning most interactions (Mendl et al., 1992). In addition, the increased probability of agonistic interactions being directed against unfamiliar goats by confrontees accompanied by peers could be interpreted as a result of social support. As the positive effect of peers was linked with familiarity in our study, it would be interesting to know if the effect of the presence of peers could also be confirmed by using unfamiliar goats. However, when introducing sows that were familiar to one another into a dynamic group the amount of aggression between residents and introduced sows was lower than was the case when introducing sows that were unfamiliar to one another (Durrell et al., 2003).

Both in the present study and an earlier study in which goats were introduced individually into established groups (Patt et al., 2012), goats were housed in the same building and had acoustic and visual contact. Thus, they were not completely unfamiliar to each other. Between mothers and their offspring and within established groups, acoustic and visual contact seems to be important to recognise each other and to keep in contact (Siebert et al., 2011; Briefer et al., 2012; Keil et al., 2012; Patt et al., 2013). However, in both studies familiarisation between goats was only based on acoustic and visual contact and seemed to be minimal as it did not allow goats to establish dominance relationships quickly after introductions or confrontations. The fact that acoustic and visual contact did not ease confrontations in the earlier and the present study additionally highlights the difficulty of introducing goats into established herds.

During the confrontations, we observed a high proportion of agonistic interactions involving physical contact. Far more of these were initiated by unfamiliar goats than by confrontees and/or peers, irrespective of the rank of the unfamiliar goats. This finding is unusual for horned goats in stable groups, where agonistic interactions without physical contact normally predominate (Aschwanden et al., 2008). Rather than being associated with the establishment of dominance relationships, the agonistic interactions during confrontations might have been motivated by unfamiliarity, intended to drive away unfamiliar animals, as has been suggested by Puppe (1998) in the context of mixing unfamiliar pigs. This hypothesis is also supported by the qualitative observation that confrontees and peers stayed mainly along the outer wall of the arena, in corners, or on the wooden partitions. We had also found a similar picture in an earlier study, where goats introduced individually into groups minimised social interactions by hiding in lying niches (Patt et al., 2012).

The higher concentrations of cortisol metabolites in unaccompanied confrontees than in goats accompanied by two peers might be explained by the higher number and larger proportion of agonistic interactions involving physical contact experienced by confrontees without peers. In pigs, physical agonistic interactions during social confrontations were shown to increase cortisol concentrations (Otten et al., 1999), and agonistic interactions with physical contact are characterised by a higher heart rate than those without physical contact (Marchant et al., 1995). However, the mere fact of being confronted on their own could also have caused the higher concentration of faecal cortisol metabolites in confrontees, as separation from the group has itself been shown to activate the hypothalamic-pituitary-adrenal axis (Guesdon et al., 2012; Patt et al., 2013). Nevertheless, since the factor sample was not included in the best-fitting model for cortisol metabolites, it must be assumed that the differences between the reference values and the samples 13, 14, and 15 h after the stressor were not statistically relevant.

A number of the unfamiliar goats' reactions varied depending on whether or not confrontees were accompanied by peers. Increasing the ratio of confronted to unfamiliar goats from 1:6 to 1:2 led to a reduced level of agonistic interactions directed against low-ranking unfamiliar goats, a higher proportion of agonistic interactions lost by confrontees and/or peers, and a higher level of activity values in unfamiliar goats. Furthermore, whereas unfamiliar goats' concentrations of faecal cortisol metabolites decreased when repeatedly confronted with just one goat, they remained more or less constant when confronted with three goats. All in all, this indicates that unfamiliar goats paid more attention to three goats than to one, as well as putting more effort into agonistic interactions and being more restless when faced with three goats. Consequently, it would be interesting to investigate whether increasing the number of peers would further increase the impact on unfamiliar goats while decreasing the impact on confrontees, and whether such effects depend on group size. Further, it would be of practical relevance to investigate independently the potential positive and negative effects of remaining in the home environment and being relocated to a novel environment.

In conclusion, the results of our study show that the presence of familiar conspecifics mitigates the adverse effects associated with social confrontations taking place in a neutral environment. Given our experimental design, however, it is necessary to verify whether the results are also valid for goats introduced into larger groups. Nevertheless, we recommend putting the husbandry procedure tested in our experiment into practice when unfamiliar goats are introduced into established herds. This would involve introducing groups of goats familiar with one another instead of individuals, as well as mixing animals in a neutral environment (e.g. on pasture) rather than in their home pen, provided that all animals are equally accustomed to the location.

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