The valence of the long-lasting emotional experiences with various handlers modulates discrimination and generalization of individual humans in sheep

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ABSTRACT: Between-farm variation in animal reactions to humans can reflect different management styles and behavioral tendencies among farmers. Animals are well known to discriminate among humans, but less clear is the key issue of whether they more or less easily generalize their experience from specific humans to others depending on management style. Here, we chose 2 contrasted management styles by known handlers: “gentle” management, that is, long-lasting exposure to positive human interactions (with limited negative interactions), and “aversive” management including long-lasting exposure to various negative human interactions (with only food delivery considered a positive interaction) and aversive events. Over a period of 19 wk, 15 female lambs were exposed to the gentle management treatment (“gently treated” group) and another 15 lambs (“aversively treated” group) were exposed to the aversive management treatment. To facilitate discrimination by animals, experimenters wore white clothes for aversive events and green clothes for farming handling (positive handling and feeding for the gently treated group and only feeding for the aversively treated group). Sheep perception of the human was assessed after the management period by submitting lambs from each group to 2 standardized tests: 1) the presence of a stationary human (familiar human in white vs. familiar human in green vs. unknown human) and 2) the presence of a moving human (familiar human in white vs. familiar human in green vs. unknown human). As expected, during the stationary human test, aversively treated lambs spent less time in the human zone \((P < 0.0001)\), showed greater latency to approach the human \((P = 0.05)\), and had fewer contacts with the human \((P = 0.05)\) than gently treated lambs. During the moving human test, aversively treated lambs also showed a greater escape distance from humans than gently treated lambs \((P < 0.0001)\). Aversively treated lambs showed the same fear responses towards familiar and unknown humans and tended to generalize their aversive experiences with one handler to all humans. In contrast, gently treated lambs seemed to discriminate familiar humans from unfamiliar humans. Different management styles could modulate farm generalization to humans in farm animals.

Key words: human–animal relationships, lambs, welfare

INTRODUCTION

Animals’ perception of humans is strongly influenced by previous experiences with humans (Hemsworth and Coleman, 1998; Rushen et al., 1999a) based on stockperson attitudes and behaviors (Hemsworth and Boivin, 2011). Animals’ perception of humans can be positive or negative and is often the result of an associated learning process. Several studies have described the ability of farm animals to discriminate different humans [e.g., sheep (Davis et al., 1998), cattle (Rybarczyk et al., 2003), pigs (Tanida and Nagano, 1998)].

An important but as yet unresolved issue is whether farm animals generalize their experience from one human to others and whether they discriminate among humans. A number of experiments show that animals tend
to generalize positive or negative experiences with one handler to all other handlers (Rushen et al., 1999b). Between-farm variation in animal reactions to humans can reflect different management styles and behavioral tendencies among farmers (Hemsworth and Boivin, 2011). Animals do generalize responses to a familiar stockperson to unknown persons, but this ability to discriminate or generalize is not necessarily uniform and may vary with farm management styles, but this hypothesis has not yet been tested. This study set out to test the ability of sheep to discriminate and/or generalize among shepherds according to their individual experiences with humans.

In farm animals, a rough handler is often associated with negative valence (aversive cues such as hits and shouts) and the expression of sudden, unpredictable, and uncontrollable behaviors. According to the cognitive theory of emotion, these cognitive checks (i.e., suddenness, unpredictability, and uncontrollability) are implicated in the development of negative emotions such as fear (Désiré et al., 2006; Scherer, 1999).

We predicted that in response to an unknown person, aversively treated animals would quickly generalize their fear responses from the familiar aversive human to the unknown human as a safety strategy whereas gentled animals would show more discriminative behaviors and be more fearful of unknown humans than familiar humans. Therefore, we chose 2 contrasting management styles, that is, gentle and aversive, performed by the animals’ regular handlers.

**MATERIAL AND METHODS**

The experiment was carried out in accordance with the 1986 directive (86/609/EEC) on the protection of animals used for scientific purposes, and was approved by the local institutional review board (Comité d’éthique 1-11; Auvergne, France).

### Animals and Housing

The study used 30 female Romane (Romanov × Berri-chon-du-Cher) lambs (aged 5 mo) housed 5 lambs per pen in 6 indoor pens, 3 of which were in one room (room A) and 3 in another (room B) in the same livestock building to avoid carryover of management and handling treatments (visual, auditory, and olfactory stimuli).

The lambs were hand fed a daily ration of concentrate in the morning (0830 h) and hay and straw in the afternoon (1200 h). The floor of the pen was bedded with straw and the room received artificial lighting from 0730 to 1930 h. For nonexperimental reasons, all animals in the experiment were exposed twice to blood sample collection by an experimenter wearing white clothes and who imposed the aversive management and handling treatments.

### Table 1. Aversive events used during the 19-wk aversive management treatment

<table>
<thead>
<tr>
<th>Predator signals</th>
<th>Conspecific signals</th>
<th>Human signals</th>
</tr>
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<tbody>
<tr>
<td>Sheep dog handling</td>
<td>• Odor of a dog’s feces</td>
<td></td>
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<tr>
<td>Lambs were moved in groups of 5 using a sheepdog, from their pens and along the corridors of the building</td>
<td>• Odor of blood taken in a slaughterhouse</td>
<td></td>
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<tr>
<td>• Aversive contact with a dog</td>
<td></td>
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<tr>
<td>A very big barking Beauceron dog (resembling a wolf) entered in or walked around the lambs’ home pen</td>
<td>Boxes of sheep blood taken in a slaughterhouse were placed in the home pen</td>
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<tr>
<td>• Howling dogs and wolves</td>
<td></td>
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<tr>
<td>At fixed times the sound of howls was played during the night in the home pen</td>
<td>Boxes of sheep urine taken in a slaughterhouse were placed in the home pen</td>
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<tr>
<td>• Odor of a dog’s feces</td>
<td></td>
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<tr>
<td>Boxes of dog feces were placed in the home pen</td>
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### Experimental Design

Over 19 wk, sheep in room A were exposed to aversive management involving both human and nonhuman aversive stimuli. Aversive management included long-lasting exposure to various unpredictable and uncontrollable aversive events regularly encountered in farming systems (i.e., aversively treated animals), at a rate of 5 aversive events/wk during the first 7 wk and 3 aversive events/wk during the next 12 wk. Aversive events were classified into 3 classes known to be biologically significant for sheep: predators, conspecifics, and human cues (Table 1). Each exposure to an aversive event was uncontrollable and unpredictable for the animals because it occurred at different hours of the day and/or night.

During the same period, sheep in room B (i.e., gently treated animals) were exposed to gentle management involving both human and nonhuman positive stimuli. Gentle management included long-lasting exposure to positive events:
- Random presentation of 4 objects given by a familiar human: balls or plastic tubes or tires used for exploration behavior twice weekly for a period of 10 min and a wool brush used for scratching behavior once per week for 1 h.

- Predictable food distribution for positive anticipation: a light coming on daily to announce the morning food distribution by a familiar human (5 min before feeding).

- Positive farming handling: exposure to a familiar gentle human carried out twice weekly for 10 min. A familiar human wearing green clothes entered in the pen, remained passive, and only gave positive tactile contacts to animals that actively initiated contacts. Durations of exposure to gentle human contact were approximately similar to the durations of exposure to negative handling events in the aversively treated group.

Experimenters (n = 3) wore white clothes for husbandry, blood sample collection, and for all aversive events imposed on aversively treated animals, grey striped clothes when leading animals to the tests, and green clothes for farming handling (positive handling and feeding for the gently treated group and feeding only for the aversively treated group) to discriminate humans via the color of their clothing (Rushen et al., 1999b). The shape of the stripes was used to distinguish the different types of people. Sheep can discriminate between different color and brightness cues (Hargreaves and Hutson, 1997). The unknown human wore horizontal black and white stripes and the other humans wore full green, full white, or vertical grey-and-red stripes.

Tests of Reactivity to Humans

**Test 1: Stationary Human Test.** The test was a 3 × 2-min phase (isolation–human presence–isolation) test adapted from Boivin et al. (1997). The indoor pen of each group was used as test pen (3 by 4.5 m; Fig. 1). The lambs were tested in their own group. One at a time, the 6 groups of 5 lambs were moved to the corridors of the livestock building in another room. The test pen was divided into three 4.5 m² zones by white lines painted on the fences. The tested group was carried over the 10 m between the corridor and the test pen by a human wearing a grey coverall, and introduced into the test pen via the door. The test comprised three 2-min phases. In phase 1, the animals were left alone. In phase 2, a human entered the pen via the door and crouched in the middle of the opposite side of the pen (Fig. 1). In phase 3, the human left the pen and the animals were again left alone for 2 min. This test was repeated 3 times on the following days (1 test/d) in order to test the lambs’ responses to different humans: a familiar human wearing white clothes (human in white), another familiar human wearing green clothes (human in green), and an unfamiliar human wearing a black-and-white-striped lab coat (unknown human). During the 3 d of test, each human (i.e., human in white, human in green, and unknown human) was tested in a crossover design to balance the order of human presentation (i.e., the humans were presented in a different order for each group for the 3 d of testing).

For each test, locomotion activity (number of zones crossed) and time spent in the human zone were measured for each phase and for each animal. In phase 2, latency to human contact within 2 min and number of contacts with the human also were recorded.

**Test 2: Moving Human Test.** The same design was repeated on d 4 but each human (aversive, gentle, and unknown) was mobile in the phase 2. This test was repeated 3 times on the same day, with each human tested in crossover. The human entered the pen via the door and stopped in the middle of the opposite side of the pen. The person then walked towards an animal until this animal moved to escape and noted the distance between him or her and the animal. The human started over for each animal of the group from the same starting point. The order in which sheep were approached was randomized. Locomotion activity (number of zones crossed) and time spent in the human zone were measured for each individual for phases 1 and 3.

During tests 1 and 2, animal behavior was recorded on cameras (Sony, Tokyo, Japan) overlooking the test pens and connected to a video recorder (Sony SVT-96LP; Sony Corp., Tokyo, Japan). The video tapes were converted to digital media files and later analyzed on
The Observer software (Noldus, Wageningen, The Netherlands) using several behavioral patterns.

**Statistical Analysis**

Data were analyzed using SAS software (version sas9x; SAS Inst. Inc., Cary, NC). Data on in-test behavior met the requirements for parametric tests. For data from test 1 (locomotion and time spent in the human zone), the effects of management treatment (aversive vs. gentle management), phases (1, 2, and 3), type of human (human in white vs. human in green vs. unknown human), and the interactions human × management × phase, human × management, phase × management, and phase × human were tested using the MIXED procedure of SAS (generalization of the standard linear model with REML and ML estimation methods implemented with a Newton–Raphson algorithm; SAS Inst. Inc., 1999 and with the individual in their group as a random effect. Post hoc comparisons after the MIXED procedure were run using least square differences. For data collected in phase 2 (latency to approach the human, and number of contacts with the human), the same model was used but without the effect of the phase.

Data from test 2 (locomotion and time spent in the human zone) was tested in exactly the same way as for data from test 1, with data on distance of escape in phase 2 tested using the same model but again without the effect of the phase.

Values are expressed as means ± SEM. Limit of significance was set at $P = 0.05$.

**RESULTS**

**Test 1**

**Locomotion Activity (Number of Zones Crossed; Figure 2).** In phase 1, there was no significant difference between treatments whatever the type of human. In phase 2, aversively treated animals crossed fewer zones than gently treated animals in presence of the known human (i.e., humans in white and in green). There was no significant difference between treatments in presence of the unknown human. In phase 2, gently treated animals crossed fewer zones with the unknown human than with the known human in green. In phase 3, after the presence of the human in white, aversively treated sheep showed lower levels of movement than gently treated sheep.
Gently treated animals increased their locomotion activity between phase 1 (without human) and phase 2 (with human) whereas aversively treated animals showed no change in locomotion activity in presence of the human in green.

**Time Spent in the Human Zone (Figure 3).** In phases 1 and 3 (without human), there was no significant difference between treatments in terms of time spent in the human zone, whatever the type of human. In phase 2 (with stationary human), gently treated animals spent more time in the human zone than aversively treated animals in presence of humans in white and in green. There was no significant difference in presence of the unknown human.

Gently treated animals spent less time in the human zone with the unknown human than with the human in green whereas aversively treated animals stayed the same time in the human zone whatever the type of human.

**Latency to Approach the Human (Figure 4).** Whatever the type of human, aversively treated animals took more time to approach the human than gently treated animals. Gently treated animals took more time to approach the unknown human than familiar humans in white and in green whereas aversively treated animals showed no difference in time taken to approach all types of human (humans in white and in green and unknown human).

**Number of Contacts with the Human (Figure 5).** Gently treated animals had more contacts with the familiar human (in white or in green) than aversively treated animals. There was no significant difference between aversively treated and gently treated animals in terms of number of contacts with the unknown human.

Gently treated animals had more contacts with the human in green than with the human in white or the unknown human. Aversively treated animals had the same number of contacts humans whatever the type of human.

**Test 2**

**Locomotion Activity (Number of Zones Crossed; Figure 6).** There was no significant difference between aversively treated and gently treated animals in terms of number of zones crossed in any phase of the test. Gently treated animals crossed a greater number of zones between phases 1 and 3 whereas aversively treated animals showed no change in number of zones crossed between phases 1 and 3.
There were no significant management × type of human interactions or management × type of human × phase interactions.

**Time Spent in the Human Zone.** There was no significant between-treatment differences in time spent in the human zone in phases 1 and 3 (df = 2, \( F = 0.2, P = 0.7 \)). All animals spent more time in the human zone in phase 1 vs. phase 3 (92.1 s in phase 1 vs. 31.2 s in phase 3; \( df = 2, F = 288.2, P < 0.0001 \)).

There was no interaction between management and types of human.

**Distance of Escape (Figures 7 and 8).** Distance of escape from the moving human was greater for aversively treated animals than gently treated animals (Fig. 7). For all animals, there was an effect of type of human on distance of escape, which was highest in response to the unknown human and lowest in response to the human in green (Fig. 8). There was no interaction between management and types of human.

**DISCUSSION**

This study set out to test the ability of sheep to discriminate and/or generalize among shepherds according to their individual experience with humans. Here, the management style adopted for 19 consecutive wk modulated the human–animal relationship: gentle management improved the human–animal relationship whereas aversive management damaged it. During the stationary human test, aversively treated lambs spent less time in the human zone with familiar humans in green and in white, showed greater latency to approach different humans (familiar human in white, familiar human in green, and unknown human) and had fewer contacts with them than gently treated lambs. Moreover, aversively treated lambs moved farther from the human during test 2 compared to gently treated lambs, suggesting that aversively treated lambs had a greater flight zone (Grandin, 1997). Aversively treated lambs were more fearful of all types of humans than gently treated lambs. Previous studies also have found that management style modulates human–animal relationship. For example, Hemsworth et al. (1987) showed that pigs submitted to an unpredictable treatment (randomly applied unpleasant and pleasant handlings) were more fearful of humans than pigs submitted to a pleasant handling treatment, as indicated by the fact that pigs submitted to an unpredictable treatment were warier of approaching a stationary human. Likewise, sheep submitted to aversive handling did not approach a stationary human whereas sheep submitted to gentle handling did (Hild et al., 2011). In the stationary human test performed here, we found that aversively treated lambs crossed fewer zones in presence of familiar humans (in white and in green) and after the presence of the familiar human in white compared to gently treated lambs. Dantzer and Mormede (1983) hypothesized that experiences of unavoidable aversive events are known to lead to apathy. Aversively treated lambs may perceive the presence of a human as an unavoidable aversive event and therefore demonstrate less activity than gently treated lambs. Moreover, aversively treated lambs showed a greater distance of escape than gently treated lambs. Sheep show 2 different types of fear-related reactions to a human: a passive defensive reaction where they stand still and glance or gaze at the human and an active defensive reaction where they move around and vocalize (Vandenheede et al., 1998). In our study, when the human was immobile, aversively treated animals decreased their locomotion activity. In contrast, when the human was moving, aversively treated animals showed an active defensive reaction associated with a higher distance of escape. This points to the hypothesis that the fear-related reaction reflected by the locomotion of
the lambs was dependent on the human’s behavior. In contrast, in gently treated lambs, the gentle interactions seemed to have increased the animals’ motivation to interact with humans. This result is in accordance with previous studies showing that the quality of interactions with the familiar stockperson influences lambs’ responses to his or her presence in the rearing pen (Tallet et al., 2005). Human contact, especially stroking and feeding, seems very important to induce a lasting improvement in the human–animal relationship (Boivin et al., 2000).

Aversively treated lambs showed the same number of contacts, same latency of approach, same number of zones crossed, and same time spent in the human zone in presence of a familiar human in white vs. a familiar human in green vs. an unknown human. Aversively treated animals thus had the same fear-related reactions towards a “gentle” human, that is, a familiar human in green who fed them, an “aversive” human, that is, a familiar human in white who negatively handled them, and an unknown human. We can posit that aversively treated lambs generalized negative events with the familiar aversive human (wearing white clothes) to the presence of the familiar gentle human (wearing green clothes) or the unknown human (wearing white and black striped clothes) in their indoor pen. According to Estep and Hetts (1992), human–animal relationships are based on the history of interactions between the 2 individuals. Throughout our experiment, the human wore white clothes for aversive events and green clothes for positive events. Hence, despite feeding with a human in green, aversive events with a human in white seemed to have damaged the human–animal relationship in the aversively treated group.

Further confirming a number of experiments (Rushen et al., 1999b), aversively treated animals may tend to generalize aversive experiences with one handler to all other handlers. This generalization process may be related to a pessimistic-like judgment bias of human presence. Doyle et al. (2011) showed a pessimistic-like judgment in chronically stressed sheep that negatively perceived ambiguous situations (e.g., much less frequently approached an ambiguously located reward than control lambs). Here, the presence of an unknown human may have been perceived as similarly ambiguous, prompting the aversively managed sheep to quickly generalize their fear responses from the familiar aversive person to the unknown one as a safety strategy. In addition, several negative situations in the aversive management treatment were not directly related to negative human associations or interactions (conspecific and predatory cues) and several of the aversive events labeled as “human signals” may be in fact “environmental.” For example, it is highly unlikely that sheep can generalize “wet straw bedding,” “lights at night,” or “disturbances with food distribution” as directly related to human behavior. This makes it difficult to firmly conclude that the responses of aversively managed sheep are exclusively the result of human–animal interaction rather than a general increased fearfulness of all situations. The human interaction tests were novel situations and may therefore...
have been perceived as ambiguous and fearful. Hence, in sheep, increased fearfulness towards humans may be a symptom of general fear. To support this hypothesis, further research is needed using general fearfulness tests such as novelty or suddenness tests (Doyle et al., 2011).

Independently of management style, all lambs showed a greater distance of escape from an unknown human than a familiar human wearing green clothes. Our results confirmed that sheep are able to discriminate individual humans, as originally shown by Davis et al. (1998). The regular positive handling by the familiar human wearing green clothes could overcome a generalized fear of humans resulting from aversive handling, as already observed in calves by De Passillé et al. (1996) and in pigs by Hemsworth et al. (1996). Hemsworth et al. (1994) found that pigs did not make such discriminations between humans but instead tended to generalize fear caused by aversive handling to all other humans. This may reflect species differences or methodological differences such as different degrees of aversive practice between studies. For example, all the handlers in Hemsworth’s study wore same-color overalls. As Hemsworth et al. (1994) pointed out, discrimination between humans would be easier if animals had a larger number of “discriminable” cues. Here, the handlers were dressed differently to maximize the lambs’ ability to tell them apart. However, the color of the clothes worn may not have been the only cue used by the sheep to distinguish between humans [see Kendrick et al. (2001) for discussion on facial cues]. Here, gently treated lambs had higher latency to approach humans, spent less time in the human zone, and crossed fewer zones in presence of the unknown human than the familiar humans wearing green or white clothes. They also made fewer contacts with the unknown human than the familiar human in green. Gently treated lambs appeared to discriminate between humans and the results clearly showed the persistent impact of gentle interactions between lambs and the human wearing green clothes on the subsequent lamb responses to their familiar stockperson. Boivin et al. (1997) also demonstrated that lambs can distinguish among different humans and that familiarity with their shepherd has a strong impact on lambs’ reactions to a human: bottle-fed lambs vocalized less and were quicker to approach and show more interaction with a known than an unknown shepherd. This increased approach behavior to the human wearing green clothes may be a consequence of learning processes at work during treatment (conditioning). The presence of humans was associated with elements that may be rewarding to the animals, such as petting by the experimenter and close-up exploration of the experimenter. However, Boivin et al. (1997) found that at 14 wk, lambs no longer behave differently to known vs. unknown handlers. This may reflect methodological differences. Here, gently treated lambs continued to receive gentle interactions whereas in Boivin’s study contacts were reduced after weaning.

To conclude, sheep are able to discriminate or generalize among shepherds according to their individual experiences with humans. Long-lasting exposure to unpredictable and uncontrollable aversive events with humans appeared to damage the human–lamb relationship whereas long-lasting exposure to positive events appeared to improve it. Moreover, whereas aversively treated lambs appeared to generalize their fear of a known stockman to unknown humans, gently treated lambs still showed different reactions to humans according to their degree of familiarity with them. The observed difference in lamb behavior towards the human dressed in a specific color compared to humans in other colors and the greater number of contacts demonstrated suggests a specific relationship with the familiar human.

**LITERATURE CITED**


