

Effects of human handling during early rearing on the behaviour of dairy calves

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Abstract

We examined the effects of daily positive or negative human handling on the behaviour of Holstein-Friesian dairy calves ($n = 20$ calves per treatment, five calves per group). The response to humans and indicators of positive emotions were examined at four weeks of age. Calves that received positive handling approached a familiar handler within 1 min in 50% of the handling sessions compared to 17% of the sessions for negatively handled calves but showed no difference when approaching an unfamiliar person. Calves that received positive handling showed less avoidance behaviour in their home pen to an approaching unfamiliar person (score, positive: 3.7, negative: 2.8) but there was no treatment effect on flight distance when tested outside the home pen. Both treatment groups responded similarly to a novel object and performed the same amount of play behaviour. Calves that received positive handling interacted more with cow brushes than calves that received negative handling (positive: 9.9%, negative: 7.9% of the total time). At three months of age, avoidance behaviour was re-tested, this time including 20 control animals of the same breed and age, reared routinely on-farm. Controls showed more avoidance behaviour (positive: 1.5, negative: 1.0, control: 0.3) and had a greater flight distance (positive: 3.3 m, negative: 3.7 m, control: 4.9 m). The results confirm existing literature demonstrating that the quantity and quality of handling influence the response towards humans. Little evidence was found that the type of early handling influences behaviours indicative of positive emotions.

Keywords: animal welfare, behaviour, dairy calves, flight distance, human handling, positive emotions

or hunger. More recently, research has focused on measures of positive emotions as an indication of good welfare and as a tool to assess an animal's welfare state. Behaviours that have been suggested as possible indicators of positive emotions include, for example, play, grooming, and exploration (reviewed by Boissy *et al* 2007). Calves frequently carry out play behaviour and the expression of play is reduced after a negative experience, such as weaning (Krachun *et al* 2010) or disbudding (Tucker *et al* 2008). Another possible indicator of positive emotions is exploratory behaviour, which most farm animals are highly motivated to carry out (Boissy *et al* 2007). Often, exploration is studied in situations where the animals are presented with a novel object that elicits both curiosity and fearfulness, thus exposing the animal to a trade-off between fear of novelty and motivation to explore (Boissy *et al* 2007; Bokkers *et al* 2009). The close link between fearfulness and motivation to explore suggests that exploration could be a possible indicator of good welfare.

The aim of this study was to examine how positive or negative human interactions during calf rearing influence the response towards humans and indicators of positive emotions. We predicted that calves receiving positive handling would show less avoidance behaviour and more approach behaviour towards people, less fearfulness and more exploration towards a novel object, and more play behaviour compared to calves that had received negative handling.

Materials and methods

All procedures involving animals were approved by the Ruakura Animal Ethics Committee and the University of Waikato Animal Ethics Committee under the New Zealand Animal Welfare Act 1999.

Study animals and housing

The study was undertaken at the Tokanui AgResearch Ltd farm near Te Awamutu, New Zealand, between 8 September and 16 November (southern hemisphere spring) 2008. Forty Holstein-Friesian heifer calves were used in the study and arrived at the farm when they were between two and five days of age. All calves were randomly allocated to either a positive or negative human-handling treatment and housed in groups of five (eight groups in total, four of each treatment). The two treatment groups were balanced for weight at arrival to the facility. At the start of the treatments, the positive treatment group weighed, on average, 41.0 (\pm 4.29) kg (\pm SD) and the negative treatment group 40.5 (\pm 4.04) kg. The groups were housed in indoor pens (4.8 \times 3.5 m; length \times width) on straw during the five-week trial period. All pens were in close vicinity and the calves had auditory but no visual contact with each other. Pens were cleaned daily and heavily soiled straw replaced. The calves were fed 2 L of milk replacement (100 g L⁻¹ Ancalf™, Fonterra Ltd, Auckland, New Zealand) twice daily (0830 and 1430h) using a five-teat calf feeder. Concentrates (Seales Ltd, Morrinsville, New Zealand), straw and water were provided *ad libitum* from the time the calves arrived on

the farm in accordance with normal farm practice. At three weeks of age, Fibrepro® (Fibre Fresh Feeds, Reporoa, New Zealand) was supplemented *ad libitum* into the diet to aid rumenal development. The feeding and cleaning routines were carried out by the handlers, however, during these times all interactions were maintained with minimal contact and use of voice.

The calves were identified individually using ear tags and coloured collars. In addition, each animal was marked using tail paint (Tell-tail paint, FIL NZ Ltd, Mount Maunganui, New Zealand) on the head, tail and across the rump and shoulders, to facilitate observations. All testing in Experiment one (see below) was carried out when the calves were four weeks old (with the exception of play behaviour which was recorded weekly). The testing was in the following order: exploration test (three consecutive days); avoidance towards an unfamiliar person (one day after the exploration test); novel object test (two days after the avoidance test); and individual flight distance (one day after the novel object test). All testing was carried out after the morning feeding (between 1000 and 1200h).

Handling treatment

Handling was carried out twice daily (5 min per group on each occasion) five days per week directly after feeding in the morning and before feeding in the afternoon, and once per day during weekends (5 min per group after feeding in the morning), for five weeks. Multiple people were used as handlers imposing both the positive and the negative handling treatments. All handlers were females, wearing blue or green overalls. The treatments were always carried out in the home pens, all other interactions with the animals were maintained with minimal contact and use of voice. The positive treatment consisted of the handlers moving slowly (< 1 step s⁻¹) and calmly around in the pen, speaking in a quiet (low, conversational), calm voice and encouraging interactions including pats and scratches. The handler aimed to interact with all animals in the group, but interactions were never forced (for example, if an animal was lying down, it was not approached). Sucking on fingers and clothes was not encouraged. The handler in the negative treatment alternated between standing still in the pen for 45 s (inactive, looking directly at the animals) and moving around for 45 s (active) during the handling period (ie four inactive bouts and three active bouts per 5 min). The active negative phase consisted of the handler performing fast movements and speaking with a harsh voice. Different tools (white plastic bag, plastic bottle with rattling stones in it, and a 53-cm long polyurethane plastic stick) were introduced every week and used only during the negative treatment every second day for one week. The tools were used in order to prevent habituation and create noise, however, the tools were never used directly on the animals. The handler would sometimes push the calves away, but no animals were ever physically harmed during the handling treatments.

Experiment one (1–5 weeks of age)

Voluntary approach to the familiar handler

Voluntary approach to the familiar handler was recorded every weekday throughout the five weeks of handling before the afternoon handling session. The handler stood motionless for 1 min in the middle of the pen, and recorded how many of the five calves in the pen approached; defined as a calf touching the person with its nose, and the latency to approach (s).

Voluntary approach and avoidance behaviour to an unfamiliar person

Voluntary approach and avoidance behaviour to an unfamiliar person, who had not interacted with the calves prior to the test, were recorded once at four weeks of age in the home pen. The voluntary approach was measured using the same definition as above.

The response to an approaching unfamiliar human was recorded in the home pen immediately after measuring the voluntary approach using the avoidance test described by Bokkers *et al* (2009). Each calf was given a score between zero and four depending on whether the observer could make eye contact (score 1), take one (score 2), or two steps towards the calf (score 3), or touch the calf (score 4) before it moved away, defined as moving both forelegs. One female observer wearing blue overalls carried out all testing.

Individual flight distance

The individual flight distance to an approaching human (the same person as in the avoidance test) was recorded in a raceway (15.0 × 1.5 m; length × width) outside the home pen. The test order was randomised. The raceway was not novel to the calves as they could see it from their home pens, and were brought down the race once weekly to be weighed and for preparation for the play recording (see below). The raceway was wide enough to allow the calf to turn around and move away from the approaching person. The person approached the calf in a standardised way (approximately 1 step s⁻¹, no eye contact) from a starting point 10 m away from the calf. The distance to the calf was measured at the point that the calf moved away from the human (defined as moving both front legs). The individual flight distance (m) was recorded twice and the average flight distance used for analysis. The time between the two test occasions for each calf was approximately 20 min. The distance was measured to the nearest 0.5 m using a scale painted on one of the walls. It was also recorded if the calf could be touched or not. The focal calf always had visual contact with its pen-mates.

Exploration/self-grooming behaviour

Exploration and self-grooming behaviour was examined in the home pen at four weeks of age. Interactions with a cow brush (DeLaval stationary cow brush B2, Hamilton, New Zealand) that was divided and mounted as two separate brushes at calf height on two walls in the home pens were recorded. The location of the brushes was the same in all pens. Interactions with the brushes were recorded using 1-min scan observations (Martin & Bateson 1993) for 1 h

during three consecutive days per group. The brushes were only available during the observation periods, thus the calves had access to the brushes for 3 h in total. Two groups (one of each treatment group) were observed simultaneously by one observer. Behaviours recorded were licking, sniffing, biting the brush, and rubbing the head or body against the brush. Lying behaviour was also recorded and defined as flank in contact with the ground, no bodyweight supported by any of the legs. One observer carried out all testing.

Novel object

The reactions to a novel object were recorded in the home pen at four weeks of age following the procedure previously described by Bokkers *et al* (2009). A red rubber ball (diameter: 45 cm) was tied to a stand that was located outside the pen. Observers remained outside the pen. The ball was dangling just above the calves' heads (approximately 1.5 m above the floor). The latency to approach the ball and the total duration of the interaction were recorded for 5 min using handycams (Sony Digital 8 DCR-TRV355E, Sony, Japan). For each calf, we used two measures of latency to approach the object: i) latency to touch and ii) latency to stand within a calf length of the ball. The latency to touch was recorded by direct observations whereas the latency to stand within a calf length was determined from video recordings. Total duration of interaction was recorded as the time spent within a calf length of the ball. Time within a calf length from the ball was defined as when the calf's head was directed towards the ball within a calf length of the object. One observer carried out all testing and video analyses.

Play behaviour

Play behaviour was recorded continuously for all calves once per week for five weeks, after the morning feeding and handling session. Two pens (one of each treatment group) were recorded simultaneously for 30 min using the same handycams as for the novel object test, two per pen. Play behaviour was encouraged by adding fresh straw to the home pens when pens were cleaned. During cleaning, the calves were kept in their treatment groups in an enclosed area of the raceway (3.5 × 1.5 m; length × width), directly outside the home pen for approximately 10 min. Play behaviour was never observed during this period of time. After cleaning, the animals were returned to their home pen and video recording began, without the presence of any humans. Behaviours observed from the video recordings were: duration of running (any gait faster than walking), and number of bucks (one or both hind legs lifted off the ground in a tucked position that is < 90° angle from the hock without a leg extension and with the head at the same level or lower than the top of the shoulders); buck kick (same as buck except at least one hind leg is lifted off the ground and extended outwards from the body); moving kick (with all hooves moving and one hind leg extended outwards in one movement with head above the shoulders); stationary kick (one or both hind legs extended in one movement while the calf is standing still); other kick (same as for buck kick and moving kick, except that the head

cannot be seen); head shake (head is moved from side-to-side, up and down, or rotated); and head to object (with at least two hooves moving, the calf touches its forehead against straw or other animal). Furthermore, it was recorded if the calves were lying (flank in contact with ground, no weight supported by any legs).

As part of a separate study (Stewart *et al* unpublished data), the behavioural and physiological responses to common husbandry procedures (restraint, ear tagging and disbudding) were examined at five weeks of age, after completion of the behavioural testing reported in the present study.

Experiment two (3 months of age)

Avoidance behaviour and individual flight distance

The avoidance and individual flight distance tests were repeated when the animals were three months old. For this experiment, we included a control group consisting of 20 calves of the same age and breed that had received routine on-farm handling in the same facility as the treatment groups from Experiment one. The control calves had been reared in groups with similar stocking density as the calves in Experiment one and with the same feeding and cleaning routines, except that all feeding and cleaning was carried out by female farm staff. From six weeks of age all calves had been reared on pasture without any particular handling treatment following normal farm practice. All 60 animals were kept in two groups while on pasture (10 from each treatment group). The animals were fed once daily with 5 L of milk replacement (100 g L⁻¹) until weaning at approximately three months of age and otherwise were fed *ad libitum* pasture with access to straw, commercial calf meal and water at all times, following normal farm practice. The avoidance test to an unfamiliar person (described above) was carried out in the same groups of five as in Experiment one in an outdoor enclosure measuring 9 × 10 m (length × width) with concrete flooring. The test was carried out twice per animal at the same testing occasion and scores were averaged (approximately 5 min elapsed between each test occasion). The test order was randomised. Immediately after the avoidance testing, individual flight distance was measured to the nearest 0.5 m in a nearby grass enclosure (15 × 3 m; length × width) using a painted scale on the fence. The individual flight distance was recorded twice per calf (approximately 15 min between each test occasion) and distances averaged. The calf that was tested always had visual contact with its group mates that were held in an adjacent pen (the same pen as in the avoidance test) behind the focal calf. One unfamiliar female observer in blue overalls carried out the avoidance testing and flight distance.

Statistical analysis

All data were recorded on an individual level; repeated observations on the same animal were averaged and then averaged across calves for each pen. All analyses were carried out on these pen means. The treatment effect in Experiment one was assessed using a one-way analysis of variance with one degree of freedom for the treatment effect and six degrees of freedom for the residual (eight

groups in total). The effects of handling treatment were tested against the latency to approach the handler (s), the proportion of voluntary approaches of the total number of observations per calf (20 test occasions per calf in total) to the handler, latency to approach the unfamiliar person (s), avoidance behaviour towards the unfamiliar person (score between 0 and 4), individual flight distance (m), lying behaviour (s) and total interactions with the cow brush (numbers of lick, sniff, bite and rub events pooled into total number of interactions), responses to a novel object (latency to move within a calf length of the object, latency to touch, and total duration of interaction [s]), and play behaviour (number of play events and duration of play running [s] and lying behaviour [s] during play recording). All data were normally distributed except for number of play events, where a natural log-transformation was used to normalise the data. Back-transformed means for play events are presented in the *Results*. The treatment effect in Experiment two was tested using a one-way analysis of variance with two degrees of freedom for the treatment effect and nine degrees of freedom for the residual (12 groups in total). The effects of treatment were tested against avoidance behaviour (score between 0 and 4) and individual flight distance (m). Pair-wise comparisons between the treatment groups were performed using Tukey's test. Mean differences in response to treatment are provided with the standard error of the difference (SED). All statistical analyses were conducted using the statistical package GenStat, version 12.2 (VSN International, Hemel Hempstead, UK).

Results

Experiment one (1–5 weeks of age)

Voluntary approach to the familiar handler

All calves in the positive handling treatment approached the familiar handler within 1 min at least once during the five weeks of recording, whereas three calves in the negative treatment never approached the handler. Calves that received positive handling were more likely to approach the handler voluntarily, compared to negatively handled calves; calves that received positive handling approached the handler within 1 min in 50% of the handling sessions compared to 17% of the handling sessions for negatively handled calves (SED: 9.8%, $P = 0.015$). Calves that received positive handling also approached the handler faster than negatively handled calves (positive: 37 s, negative: 53 s, SED: 1.1 s, $P = 0.026$).

Voluntary approach and avoidance behaviour to an unfamiliar person

Forty-five percent of the calves that received positive handling approached the unfamiliar person within 1 min, compared to 25% of the calves that received the negative handling (SED = 21%, $P = 0.382$). Both treatments had the same latency to approach the unfamiliar person (positive: 42 s, negative: 48 s, SED: 1.9 s, $P = 0.538$).

Calves that received positive handling showed less avoidance behaviour compared to calves that received negative handling ($P = 0.039$; Figure 1).

Figure 1

Mean (\pm SED) avoidance score of calves that received either positive, negative or control (routine on-farm) handling treatments from 1 to 5 weeks of age ($n = 4$ groups per handling treatment, 5 calves per group). Avoidance behaviour was recorded at 4 weeks of age and repeated at 3 months of age. Low scores indicate greater avoidance behaviour.

* $P < 0.05$; ** $P < 0.01$.

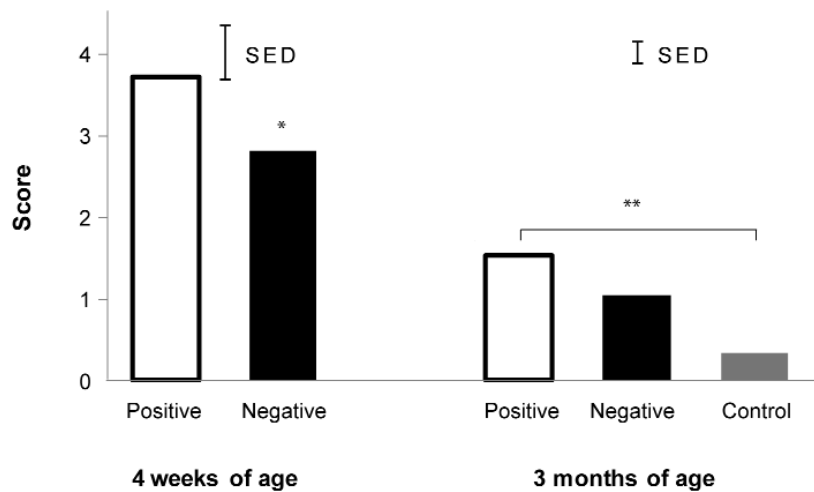
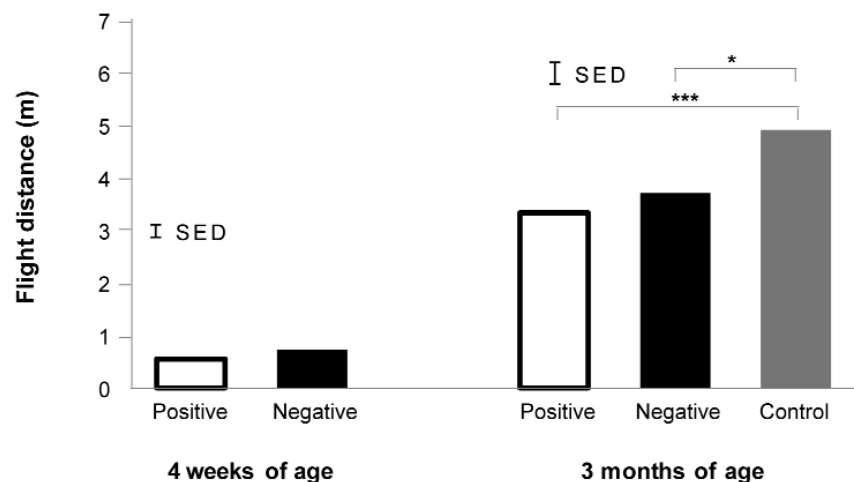


Figure 2

Mean (\pm SED) individual flight distance of calves that received either positive, negative or control (routine on-farm) handling treatments from 1 to 5 weeks of age ($n = 4$ groups per handling treatment, 5 calves per group). Flight distance was measured at 4 weeks of age and repeated at 3 months of age.

* $P < 0.05$; *** $P < 0.001$.



Individual flight distance

The two treatment groups had the same flight distance when tested individually in the raceway ($P = 0.526$; Figure 2). The greatest flight distance recorded was 2 m (for three animals only, one negative, two positive). Of the 20 animals in each group, four of the negative calves could be touched compared to nine of the positive calves.

Exploration/self-grooming behaviour

During the 60-min period when the calves had access to the two mounted cow brushes, the positively handled calves spent 9.9% of the time interacting with the brush compared to 7.9% of the time for negatively handled calves (SED: 0.77%, $P = 0.039$). Similar amounts of time were spent on licking, biting and sniffing (4.8 and 4.0% of the time for positive and negative treatments, respectively) and rubbing the head or other body parts (5.1 and 3.9% for positive and negative

treatment groups, respectively). The treatment groups spent similar amounts of time lying (56.5 and 62.7% for positive and negative, respectively, SED: 7.44%, $P = 0.463$).

Novel object

Both treatment groups showed the same latency to touch the red ball (positive: 2.5 min, negative: 1.9 min, SED: 2.52 min, $P = 0.517$) and to move within a calf length of the ball (positive: 1.3 min, negative: 1.0 min, SED: 0.32 min, $P = 0.491$). Both groups also spent the same total amount of time interacting with the ball (positive: 2.4 min 5 min⁻¹, negative: 3.2 min 5 min⁻¹, SED: 0.39 min, $P = 0.395$).

Play behaviour

During the play recordings, both groups spent a large proportion of the time lying down (positive: 12 min 30 min⁻¹, negative: 13 min 30 min⁻¹, SED: 1.17 min, $P = 0.735$). Both groups spent on average 0.7% of the total

time running (12.6 s 30 min⁻¹ per treatment group). Head shaking (on average, 2.0 times 30 min⁻¹ per treatment group) and kicks (on average, 1.1 kicks 30 min⁻¹ per treatment) were the most common play events. However, there was no difference between the treatments in the time spent running and performing play events ($P \geq 0.122$).

Experiment two (3 months of age)

Avoidance behaviour and individual flight distance

Animals that had received routine on-farm rearing (controls) showed more avoidance behaviour ($P = 0.004$; Figure 1). A *post hoc* analysis revealed that the control group showed greater avoidance behaviour compared to the group that had received positive handling ($P < 0.01$). The control animals also had greater individual flight distances when tested in a grass enclosure ($P < 0.001$; Figure 2). A *post hoc* analysis revealed that the control group had a greater flight distance than both the positive group ($P < 0.001$) and the negative group ($P < 0.05$).

Discussion

Aversive human handling can result in more reactive or fearful animals (Rushen *et al* 1999) whereas non-aversive handling at an early age can have the opposite effect and may reduce fear of people (Boissy & Bouissou 1988; Boivin *et al* 1994). Calves that received positive handling were more likely to approach the familiar handler within 1 min in their home pen, compared to negatively handled calves, whereas there was no difference between the treatment groups in how they approached an unfamiliar person. Our results agree with findings by de Passillé *et al* (1996) where two-week-old dairy calves were more likely to approach a handler who treated them positively, whereas they avoided an aversive handler after 12 handling treatments (de Passillé *et al* 1996). The calves in the present study were fed by the handlers and this may have influenced the results. Positive reinforcement on-farm typically involves feeding, and several studies have demonstrated that food is perceived as more rewarding than handling alone (Boivin *et al* 1992; de Passillé *et al* 1996; Jago *et al* 1999). For example, calves that are fed by humans are faster to interact with a human compared to animals that have been handled only (Jago *et al* 1999). However, temporary handling during the first nine months of life without the positive reinforcement of feeding has been shown to reduce the avoidance behaviour and improve handling ability in Friesian calves (Boissy & Bouissou 1988). Both treatment groups showed the same flight distance when tested individually outside the home pen. The greatest flight distance recorded was 2 m (for three animals only), which agrees with Jago *et al* (1999) where dairy calves never showed flight distances greater than 2 m at 17 and 32 days of age. Indeed, most calves did not show a flight response at all, which also agrees with Jago *et al* (1999). One possible explanation for this could be that calves at this age do not have fear of humans, especially if they depend on humans to feed them. However, because the negatively treated calves responded differently to an approaching unfamiliar

person in their home pen, a second more likely explanation is that the calves were more motivated to explore the relatively novel environment rather than focusing on the approaching human. This is consistent with de Passillé *et al* (1996) who found that some calves responded differently to people depending on whether they were tested in their home pen or in an unfamiliar location. From general observations, we found that it was difficult to attract the attention of the calves when testing outside the home pen. These results suggest that flight distance outside the home pen is not a suitable measure of avoidance behaviour in young calves; however, the results from the present study confirm existing literature that calves are able to discriminate between people and that positive handling increases approach behaviour and reduces avoidance behaviour.

We were interested in investigating whether the type of handling influences measures that may be indicative of positive emotions. Exploration behaviour has been suggested as a possible indicator of positive emotions since most animals are motivated to explore their environment and the behaviour is closely related to and affected by fearfulness (Boissy *et al* 2007). The calves spent, on average, 55% of the test time interacting with the novel object (a ball) with no detectable treatment differences, and from general observations very few calves showed any signs of being fearful of the object. The latency to touch the object was similar to that found in veal calves by Bokkers *et al* (2009) but we are not convinced that the delay in touching the object was due to fearfulness as the animals often were eating or lying. Our findings agree with others where handling decreased fear of humans in beef cattle and veal calves, but did not affect the animals' responses to novel and surprise stimuli (Hemsworth *et al* 1996; Lensink *et al* 2000b). Calves that received positive handling explored and used cow brushes mounted in their home pen more compared to the negatively handled calves. Self-grooming has been suggested as an indicator of positive emotions, but more in the context of excessive grooming occurring in sub-optimal conditions (Boissy *et al* 2007). We speculate that the positive handling treatment, which included patting and scratching the animals, caused a change in self-grooming behaviour in the presence of the brushes or a change in how these animals explore their environment.

Despite our predictions, calves that had received positive handling did not show more play behaviour than negatively handled calves. Play has been suggested as a potential indicator of good welfare due to young animals being highly motivated to play when their primary needs, such as sufficient food, thermal comfort and health, are met (Jensen *et al* 1998). Furthermore, the performance of play seems to be reinforcing, and may indicate the presence of good welfare and positive feelings (Jensen *et al* 1998). Play behaviour was stimulated by adding new straw to the pen, as novel environments have been shown to encourage play behaviour (de Passillé *et al* 1995). We chose to stimulate play rather than record naturally occurring play behaviour

as play is a relatively rare activity and short in duration. Dairy calves spend 0.5% of their active time playing at 4 to 6 weeks of age (Jensen *et al* 1998), or 54 s 24 h⁻¹ at five weeks of age (Jensen & Kyhn 2000) compared with 12.6 s of the 30-min period in the current study. It is possible that the motivation to play was strong in both treatment groups due to play behaviour being stimulated. Furthermore, there were no humans present during the video recording of the play behaviour, and a suggestion for further research is to investigate how play behaviour is influenced by the presence of gentle or aversive handlers.

There is evidence that minimal contact with humans elicits more fearful behaviour compared to more handling, be it positive or negative (Petherick *et al* 2009a,b), possibly because of the lack of habituation to human contact. Animals that had received routine on-farm rearing showed more avoidance behaviour in a group and had greater individual flight distances, thus supporting existing literature which report that the amount of human interactions influences how animals react to humans. For example, beef cattle that had more contact with humans for three months were easier to handle and less aggressive than range animals that had been reared with less human contact (Boivin *et al* 1994). However, reactions towards humans will depend on not only the type of handling, but on other factors such as the familiarity with the human, previous experience and the test situation itself (Boivin *et al* 1998).

Animal welfare implications

Dairy calves that received daily positive handling were more likely to approach the familiar handler and showed less avoidance behaviour towards an unfamiliar person at four weeks of age compared to calves that received negative handling. Little evidence was found that the type of handling influences measures indicative of positive emotions, but further research in this area is required. At three months of age, calves that had received routine on-farm rearing showed more avoidance behaviour compared to calves that had received positive and negative handling. The results confirm existing literature that the type of human interaction during early rearing influences the response towards humans. The amount of human contact had greater impact at three months of age than the quality of handling.

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