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Looking into the eyes of a cow: Can eye whites be used as a measure of emotional state?

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ABSTRACT

A number of studies have looked at whether the percentage of visible eye whites could be a reliable and dynamic tool for measuring emotional state in cattle. In this study we have built upon previous research to further test this measure with different stimuli and different types of emotional states in order to assess its suitability as a welfare tool. We used positive and negative contrasts to elicit the emotional states of excitement and frustration in 22 Holstein dairy cows. We performed 10, 15 min focal observations with each cow. In the first four trials the cows were given standard feed, a substrate they have continuous access to. Then for the next five trials they were given concentrates, a high energy feed that is highly desired, and which they have limited access to. And for the final trial they were given inedible woodchip. The standard feed represented a neutral stimulus as it wasn't novel or highly desirable. The concentrates were a positive stimulus, and the inedible woodchip was a negative stimulus, especially as it followed the concentrates, and so the cow's expectations were thwarted. We measured both the cow's heart rate (beats per minute), and the percentage of visible eye whites throughout the focal observations. We found that the woodchip treatment elicited the highest heart: pre-feeding, $M = 83.01$ feeding, $M = 88.95$ and post-feeding $M = 84.51$, suggesting it was the most arousing of the three treatments, this was followed by the concentrates treatment. Results showed that the percentage of visible eye white significantly increased during the concentrates and woodchip treatments, compared with the standard feed treatment: pre-feeding ($p < 0.001$), feeding ($p < 0.001$) and post-feeding ($p < 0.001$. When we looked at the change in visible eye white within each treatment, during the concentrates treatment the eye white increased during the feeding segment compared with both the pre-feeding and post-feeding segments ($p < 0.001$). The visible eye white also increased significantly in the feeding segment of the woodchip treatment compared with during the post-feeding segment ($p < 0.001$), but not compared to the pre-feeding segment ($p = 0.25$). There is a need for more comparable research to be performed that explores both types of valence and arousal levels, before the effects can be fully understood. With this information and understanding, it would then be possible for visible eye whites to be used as a non-invasive measure of emotional state.

1. Introduction

Animal welfare is concerned with how well animals cope in their environment, and caregivers and owners are responsible for meeting their animal's needs (Broom, 2010, 1991). To do this, we need objective, animal based measures of how an animal is doing, both physically and emotionally (Boissy et al., 2007; Edgar et al., 2013).

In relation to using eyes whites as a measure of emotional state, we have previously shown that the percentage of visible eye whites significantly decreases when cows experience a low arousal, positive emotional state elicited through stroking (Proctor and Carder, 2015). In addition, Sandem and Braastad (2005), Sandem et al. (2002) found the opposite to occur when cows were exposed to a negative, high arousal stimulus such as being thwarted from accessing visible food, or a dam being separated from her calf (Sandem and Braastad, 2005; Sandem et al., 2002). Interestingly however, they found that the percentage of visible eye whites decreased below the original baseline levels once the negative stimulus ended and a positive stimulus was provided instead (access to the feed, or reunion with the calf) (Sandem and Braastad, 2005; Sandem et al., 2002). Both the stroking stimulus we provided in our previous study (Proctor and Carder, 2015), and the rewarding stimuli; being reunited with the calf, or given access to the feed, could all be considered to elicit low arousal states. The stroking for example, was considered to induce the affects 'relaxed' and 'calm', and the cows who were no longer frustrated by the visible feed, or searching for their calves were likely to be much less aroused than before (Proctor and Carder, 2015; Sandem and Braastad, 2005; Sandem

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et al., 2002). It is therefore unclear whether the effects on visible eye whites were indicative of a change in valence or arousal, or a combination of both. In the stroking study, we suggested that the cows did not experience a large drop in arousal, as they were already considered to be in a low state of arousal before the stroking began (Proctor and Carder, 2015). However, stroking is known to decrease cows' heart rates (Schmied et al., 2010), therefore a small drop in arousal levels may have occurred. It was therefore unclear whether the small change in arousal levels was the reason for the drop in visible eye whites, or whether it was the change in emotional valence (Proctor and Carder, 2015).

Very little research has been performed to explore the suitability of eye whites as an indicator of emotional state, however the majority of research to date has been performed on cows. Sandem et al. (2006) tested the effects of positive anticipation on the visible eye whites of cows. The cows were conditioned to associate the arrival of a stockperson with the delivery of feed. They found that the cow's eye whites significantly increased when the stockperson entered, and then decreased considerably once they received the feed, compared with when the stockperson first entered. The eye whites did not significantly drop below the baseline levels until between 40 s and 2 min after the food was provided. The authors concluded that these findings show that an increase in visible eye whites is associated with a strong emotional response, both positive and negative. Furthermore, because the eye whites took time to decrease to the baseline levels, they suggest that the very low eye white levels they consider to be associated with rewarding and consummatory behaviours, develop slowly (Sandem et al., 2006). Reefmann et al. (2009a) found that treatment did not have an effect on the percentage of visible eye white in sheep. However, in a different study Reefman found that the relative eye aperture of sheep was highest during separation from group members (negative valence). The eyes were open less wide during an intermediate valence (standing in a feed area), and even less when they were being groomed by a human, which was considered to be a positive valence (Reefmann et al., 2009b).

In our current study, the aim was to determine whether similar effects on visible eye whites in dairy cows could be found with different positive and negative stimuli to those used before. Furthermore, we aimed to investigate the effects of valence and arousal on visible eye whites to determine whether they can be used to detect different aspects of a cow's emotional state.

2. Materials and methods

2.1. Ethics

The study was performed in line with both the journal and the Royal Veterinary College's ethical procedure, and it did not require a Home Office License.

2.2. Subjects and housing

In this study we used 22 Holstein lactating dairy cows, who ranged in age from three to seven years, and were in good physical health. Eleven of the cows were randomly selected from a high-yielding group, and the other 11 were randomly selected from a low-yielding group. The cows came from a commercial dairy herd of 92 cows, and were housed at Boltons Park Farm, Hertfordshire, UK, part of the Royal Veterinary College's farm animal practical teaching facility. Data collection took place over 6 weeks from May to July 2015.

We worked with a new group of four cows each week from Monday to Friday. For two of the weeks, only three cows were tested due to time restrictions. Each day, the focal cows were separated from the main herd following the first milking session, and placed into an adjacent pen (home pen) by the farm staff for the duration of the experiment (9am to 3pm). While in the home pen the cows were not restricted in any way, and had continuous access to the standard feed. At 3pm the cows re-joined the main herd for milking and remained with the herd until after the next morning's milking at 7:30 am The focal cows were kept indoors in their usual housing system; a deep litter, free housing system, for the five days they were used. For the experiment, the cows were moved from their home pen, into a handling stall, measurements of the handling stall were 170 cm × 71 cm × 206CM (H × W × L). We only moved one cow at a time, and each cow was only used twice a day (in the morning and in the afternoon) every day for five days, ensuring that there was a minimum of 1.5 h between trials.

The cows were very familiar with being held in the stalls as they were part of a teaching herd, and were regularly held in the stalls both singly and socially for varying periods of time. We only held each cow in the stall for a maximum of 25 min at one time. The period when the cows were held in the stall consisted of a 10-min period of acclimatisation and equipment fitting, whilst we let their heart rate return to normal after the brief activity of walking, then 15 min for the focal observation. To ensure that the process of being brought into the stall, with the presence of certain equipment as visual and olfactory cues, was not confused with any other experience or any anticipatory effects were diluted, the focal cows were not brought into the stall for any other purpose during the study week, and the same stall was used throughout the week. The layout and presence of the equipment in the stall remained the same throughout the study. This consisted of three identical sealed buckets, containing woodchip, concentrates feed, and standard feed; a small table with a laptop and saline spray on it; a feed trough, and a monopod with a video camera attached to it.

Prior to the study all of the study cows were habituated to a physiological monitoring telemetry device (BioHarness 3.0, Telemetray Syste, Zephyr Technology Corporation), by gradually exposing them to wearing it over a number of days leading up to the start of the data collection. They were also habituated to the presence of the experimental equipment. The cows were also habituated to the presence of unfamiliar people during the regular teaching sessions they were previously exposed to, but the cows had no prior experience of the researchers or the experimental procedure.

2.3. Experimental procedure

Five researchers were responsible for data collection and so to ensure consistency, inter-observer tests were performed at the start of each week during the 6-week data collection period. Each researcher observed the same focal observations and compassions were made between the data for each observation. Each test achieved >95% agreement in the Kappa coefficient test analysis.

2.3.1. Treatment 1: standard feed

All of the focal cows underwent the same procedure throughout a 5-day period. First the focal cow was secured in the closed stall. Then the BioHarness, which was attached to an elasticated strap, was placed and tightened around the cow's middle, just behind the cow's front legs. The contact area for the electrodes was shaved, and the BioHarness was sprayed with saline to promote conductivity. One of the researchers observed the focal cow for any adverse reactions such as kicking or stamping, whilst the equipment was fitted. None of the 22 cows were considered to react adversely to the equipment. The cow was then left until a total of 10 min had passed since she had entered the stall, in order to allow her heart rate to revert to the standing rate. Prior to the start of the study we tested the same cows to determine how long it took for their heart