Cortisol Increases in Plasma of Holstein Heifer Calves from Handling and Method of Electrical Dehorning

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ABSTRACT
Changes in cortisol in plasma were used to assess stress when calves were restrained and then dehorned. Thirteen Holstein heifer calves between 3 and 4 wk of age were used over 4 d; each calf served as its own control. On d 1, 2, and 4, blood was sampled initially while calves were in a pen, 5 min after being placed in a restraint chute, and then at 5, 15, 30, and 45 min and 1, 2, 3, 4, 8, and 12 h after simulated or actual dehorning. On d 1, dehorning was simulated. On d 2 and 4, one horn bud of each calf was cauterized, respectively; sequence of horns (right, left) and dehorning instruments (conventional electrical, Buddex™) were alternated for all calves. Day or previous dehorning procedures had no effect on initial concentrations of cortisol in plasma. However, after calves were placed in a chute, cortisol in plasma increased with each entrance. Cortisol in plasma peaked at 5 min posthandling (d 1, 11.3 ng/ml) or 15 min postdehorning (electrical, 21.9 ng/ml; Buddex™, 20.7 ng/ml). These data suggest that both dehorning procedures resulted in similar rates of synthesis and secretion of cortisol.
(Key words: calves, electrical dehorning, plasma cortisol)

INTRODUCTION
Dehorning is a common management practice in most dairy rearing operations (5). Electrical dehorning, which has had wide application, normally is not performed on calves <3 wk of age and often is dependent on the brand of dehorner used and age of the calf when the horn buds erupt through the surface skin.

Previous studies (2, 8) indicated that dehorning with a conventional electrical dehorner had no effect on feed intake, growth, or health of 8-wk-old calves. However, cortisol concentrations in plasma were elevated up to 4 h after dehorning. Administration of a local anesthetic prior to electrical dehorning also did not prevent increased cortisol in plasma (8).
The Buddex™ dehorner (C.I.C. Pty. Ltd. Perth, Western Australia), a new type of electrical dehorner, was recently introduced to the US market (7). Calves can be dehorned with the Buddex™ dehorner as soon as a horn bud can be palpated beneath the skin, thus making dehorning of calves possible from d 1 to 3 wk of age. Growth of unerupted horn buds is arrested by cessation of blood flow. The Buddex™ dehorner is cordless, rechargeable, and heats to 816°C within milliseconds. Only 10 s of application are required to dehorn.
The purpose of this study was to determine the relative amount of stress caused by dehorning with the Buddex™ dehorner versus a conventional electric dehorner in calves of the same age using cortisol in plasma to indicate stress.

MATERIALS AND METHODS

Experimental Design
Thirteen Holstein heifer calves, 3 to 4 wk of age, were used in a 4-d study. Calves were housed in individual pens and fed whole milk at 10% of BW/d and offered a pelleted grower containing 18% CP (Agway Calf Grower, Agway Inc., Syracuse, NY) and high quality baled alfalfa grass hay for ad libitum intake.

Prior to d 1, a catheter (Abbocath T 16 gauge × 5.08 cm; Abbott Laboratories, North
Chicago, IL) was inserted into one jugular vein of each calf. The catheter was secured with duct tape wrapped in a butterfly fashion around the catheter and attached to the skin with tag cement (Nasco, Fort Atkinson, WI). The catheter was flushed with 0.5 to 1.0 ml of sodium heparin (100 IU/ml) after each use.

On d 1 (0600 h), jugular blood was sampled within 1 to 2 min after the pen of each calf was entered. All calves were then haltered and placed in a restraint chute located immediately outside of the pen. The restraint chute measured 106.7 x 50.8 x 113.0 cm and consisted of an adjustable headlock and width that allowed all calves to be immobilized. Jugular blood was again sampled 5 min post restraint. An unheated conventional electrical dehorner (Rhinehart 110 to 120 V of AC, Spencerville, IN) was then applied to one horn bud, simulating the pressure applied for 1 to 2 min during dehorning. Calves were then returned to their pen where jugular blood was sampled at 5, 15, 30, and 45 min and 1, 2, 3, 4, 8, and 12 h after the simulated dehorning.

On d 2 and 4, the sequence of events was the same except that only one horn bud was cauterized on each of the days; the sequence of horns (right, left) and electrical instruments used to dehorn (conventional, Buddex™) alternated between days and calves. The conventional electrical dehorner was applied for 1 to 2 min, and the Buddex™ dehorner was applied for 10 s. Cauterized areas were treated with an antiseptic spray (Furox Aerosol Powder; Smith Cline Animal Health Products, West Chester, PA). Jugular blood was sampled via the catheter into tubes containing sodium heparin (143 USP units/10 ml, Becton-Dickinson, Rutherford, NJ). No procedures were performed on calves on d 3.

Chemical Analysis

Blood was chilled immediately, and hematocrit was measured using a microcapillary reader (International Equipment Co., Needham, MA). Remaining blood was centrifuged at 1200 x g, and plasma was collected and stored at -20°C. Plasma cortisol, the predominant glucocorticoid in plasma of bovine species (1), was measured via a commercially available radioimmunoassay procedure (Coat-A-Count® Protocol and Kit Diagnostic Products Corp., Los Angeles, CA), utilizing iodinated cortisol and a highly specific antibody. To normalize unknown effects of interaction of calf plasma and human serum in the radioimmunoassay, charcoal-stripped calf plasma also served as the diluent.

Statistical Analysis

Data were analyzed using repeated measures of ANOVA (12). A model was developed to assess whether plasma cortisol changed significantly with time (pen, chute, 5, 15, 30, and 45 min and 1, 2, 3, 4, 8, and 12 h postdehorning) because of the effects associated with the instrument used to dehorn, day of the study (d 1, 2, or 4), horn bud cauterized (left vs. right), and their interactions with time. Data on plasma cortisol were logarithmically transformed prior to statistical analyses, but changes in plasma cortisol are presented in nanograms per milliliter (Figures 1, 2, and 3).

RESULTS AND DISCUSSION

Handling and dehorning significantly (P < .01) increased cortisol in plasma over time in calves (Figures 1, 2, and 3). The magnitude of the change in plasma cortisol was dependent (P < .09) on the day of the study and which horn bud (left or right) was cauterized. Interactions

Figure 1. Changes in cortisol concentrations in plasma when calves (n = 13; pooled SE = 1.64) were handled on d 1 (O) and when one horn bud was cauterized on d 2 (■) and d 4 (□), respectively. Time increments reflect the following: -10 min, initial concentrations of calves in individual pens; -5 min, concentrations after calves were placed in a restraint chute; and 5 min through 12 h, concentrations postdehorning of calves in individual pens.

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3726 WOHLT ET AL.
Figure 2. Changes in cortisol concentrations in plasma when calves (n = 13; pooled SE = 1.64) were handled (C) or one horn bud dehorned using a conventional electrical dehorner (M) or Buddex™ dehorner (O). Time increments reflect the following: -10 min, initial concentrations of calves in individual pens; -5 min, concentrations after calves were placed in a restraint chute; and 5 min through 12 h, concentrations postdehorning of calves in individual pens.

of day × time and horn bud × time were significant (P < .05) when d 2 and 4 were included in the model.

At 0600 h, plasma cortisol averaged 4.4 ng/ml in calves 3 to 4 wk of age that had been fed one-half of their daily allotment of whole milk. Day or previous dehorning procedures conducted during the 4-d study had no effect on these initial concentrations of cortisol in plasma measured while calves were in pens (Figures 1 and 2). These baseline cortisol concentrations in plasma were similar to those reported by Boandl et al. (2) and Ladden et al. (8).

Each day, within 5 min after calves had been placed in a restraint chute, plasma cortisol was increased above the previous day's concentration (Figure 1; 3.5, 4.6, and 8.5 ng/ml on d 1, 2 and 4, respectively). Five minutes were allotted before sampling to determine whether plasma cortisol increased because of a calf's anticipation by association from the previous day's activity over the 4 d. No procedures were performed on d 3 to prevent such a carry-over effect between instruments used to dehorn on d 2 and 4, respectively.

Plasma cortisol peaked 5 min poststimulation of dehorning on the handling day and 15 min postdehorning on d 2 and 4 (Figure 1). Handling increased plasma cortisol twofold to 11 ng/ml, which has been reported previously by Boandl et al. (2). Dehorning on d 2 and 4 increased cortisol in plasma four- to fivefold; the increase tended to be greater on d 4 versus d 2 (23.3 vs. 19.2 ng/ml). Cortisol in plasma was higher on d 4, regardless of whether the left or right horn bud was dehorned (Figure 3).

Dehorning with a conventional electrical dehorner or a Buddex™ dehorner resulted in similar plasma cortisol peaks (Figure 2; 21.9 and 20.7 ng/ml, respectively). Although the Buddex™ dehorner was applied for a shorter time than the conventional electrical dehorner (10 s vs. 1 to 2 min), the temperature of the ceramic tip of the Buddex™ dehorner was higher than the heated conventional electrical dehorner (816 vs. 538°C). The data suggest that both dehorning procedures created conditions...
that resulted in similar rates of synthesis and secretion of cortisol from the adrenal cortex. Also, both dehorning procedures were equally effective in preventing horn growth.

Peak cortisol concentrations in plasma were always less when the right horn bud versus the left horn bud was dehorned (Figure 3), regardless of day or instrument used to dehorn. Asymmetry in human pain perception has been described for several sensory modalities. This asymmetry frequently produces greater sensitivity to pain on the left side of the body (4, 9), although not all studies are in agreement (10). Furthermore, neurons transmitting signals for pain and temperature cross over in the central nervous system, so that pain from one side of the body is processed by the contralateral cerebral hemisphere (11). In humans, the right cerebral hemisphere is involved in some kinds of emotional behavior (3). Whether cerebral hemispheric functions in cattle correspond to those in humans is not known. However, the consistently higher cortisol response to left horn bud removal is suggestive of a pattern of cerebral organization of emotional function in cattle analogous to that in humans.

Handling and dehorning elevated cortisol in plasma for 1 h (Figures 1 and 2), which is not in agreement with Ladden et al. (8), who observed that cortisol in plasma returned to baseline values at 4 h postdehorning. However, when Ladden et al. (8) performed conventional electrical dehorning, both horn buds were dehorned and calves were 8 wk of age, but, in the present study, a single horn bud was dehorned, and calves were at 3 to 4 wk of age. Horn buds are larger at 8 wk than at 4 wk of age, contributing to more stress from the application time of the conventional electrical dehorner and surface skin area affected. Another factor that may have extended the return of cortisol in plasma to baseline in the study of Ladden et al. (8) was that blood was sampled by jugular venipuncture, but an indwelling catheter was used to sample blood in the present study.

At 8 h postdehorning, cortisol in plasma significantly (P < .05) increased in all treatments. Cortisol in plasma responds to a diurnal rhythm. Controversy exists over whether diurnal rhythms of plasma cortisol occur in cattle because of absence of deep sleep in ruminants (13). Changes in plasma cortisol are thought to be associated with daytime activity and regularly occurring events in the daily management schedule. In rats and calves, diurnal plasma cortisol concentrations peak before meals. In feed-deprived calves, plasma cortisol peaked at the scheduled feeding (6). Cortisol in plasma at 8 h postdehorning increased prior to a regularly scheduled feeding of whole milk.

CONCLUSIONS

Use of a Buddex™ dehorner versus a conventional electrical dehorner did not lower cortisol in plasma postdehorning. The Buddex™ dehorner had the advantage of a shorter application time and capability of dehorning calves <3 wk of age, when calves are smaller and easier to handle. Additional studies need to be conducted in which behavior is also measured to establish whether use of a Buddex™ dehorner is less stressful than a conventional electric dehorner for calves.

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