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Medium-term effects of repeated exposure to stray voltage on activity, stress physiology, and milk production and composition in dairy cows

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ABSTRACT

The medium-term effects of permanent or random exposure to stray voltage applied to the water trough were evaluated on milk production and stress physiology in lactating dairy cows. Seventy-four Holstein cows were assigned during two 8-wk experimental periods to 1 of 3 treatments. The treatments were permanent exposure to voltage (PERM, 1.8 V, n = 23) applied to the water trough, random exposure to voltage (RAND, 1.8 V, 36 h/wk, n = 25), and no exposure to voltage (control, n = 26). On the first day of voltage exposure, PERM cows had higher activity levels than control cows (9.8 ± 2.70 vs. −2.3 ± 2.74 14-s periods of movement/h). During the eighth week of exposure, RAND cows had higher activity levels than control cows (4.2 ± 3.64 vs. −7.7 ± 3.54 14-s periods of movement/h) and higher milk cortisol concentration than PERM cows (0.21 ± 0.024 vs. 0.14 ± 0.020 ng/mL). No differences were observed between treatments for cortisol response after an ACTH challenge during the seventh week of exposure. No effects of voltage exposure were observed on production traits and daily water intake. There was a transient decrease in milk yield on the second day of exposure in PERM cows (−1.4 ± 0.74 kg) and on the third day of exposure in RAND cows (−3.5 ± 1.03 kg) compared with control cows. In dairy cows, permanent or random exposure to stray voltage (1.8 V; 3.6 mA) could induce a transient stress response. Moreover, unpredictable voltage exposure could be considered a mild stressor, with slight modifications in stress physiology and activity but no impairment in production in the medium term.

Key words: stray voltage, dairy cow, unpredictability, chronic stress

INTRODUCTION

Electricity is essential to modern farming techniques and many electrically powered machines are used, such as milking machines, automatic feed dispensers, and electrically heated water bowls. Leakage of current from these types of equipment, electric and magnetic induction from high voltage lines, or faulty connections between the electrical circuit and the earth can lead to an undesirable electrical phenomenon called stray voltage (Hultgren, 1990). Stray voltage is defined as a small voltage (<10 V) measured between 2 points that can be simultaneously contacted by an animal (USDA, 1991). It can produce a low current that can flow through farm animals (Norell et al., 1983).

For several decades, stray voltage was considered a possible factor impairing production in dairy farms. Although producers and veterinarians reported reduced production as well as increased health problems and behavioral modifications in cows housed in buildings where stray voltage was detected (Hultgren, 1990), the direct implication of stray voltage among other possible factors was not clearly demonstrated. Therefore, experiments conducted in dairy cows evaluated the effects of stray voltage on behavior (Aneshansley et al., 1992; Aneshansley, 2003), milk production (Gorewit et al., 1992a; Reinemann et al., 2002), and health and stress physiology (Southwick et al., 1992; Aneshansley, 2003). Most studies were short-term experiments (except Gorewit et al., 1992a) and were performed on a limited number of animals.

Predictability of a stimulus or a stressor is important in relation to animal welfare (Désiré et al., 2002). When a stressor occurs in an unpredictable way, it is more stressful for the animal than if the stressor occurs in a predictable fashion (Quirce et al., 1981), which allows the animal to expect its occurrence and eventually to adapt. In most of the experiments on stray voltage, electric stressors were applied in a predictable fashion and cows habituated to voltage exposure (Gorewit et al., 1985; Henke Drenkard et al., 1985). After having

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observed strong behavioral responses (arching of the back or kicking) at the initial application of 4 mA to the spinal area, Gorewit et al. (1985) showed that after 24 h, dairy cows became habituated to the current and by the end of the experimental period (16 d), behavioral responses to the electric stressor were almost nonexistent. On farms, stray voltage can occur in a random manner and can be unpredictable for the animals (Hultgren, 1990). This may impair animal welfare more than if stray voltage was experienced in a permanent manner. Several studies (Gorewit et al., 1985; Reinemann et al., 2004) evaluated the effects of random current or voltage exposure on cow responses (behavior, stress physiology, and milk production). The conclusions were conflicting and the effects of random exposure to current need clarification.

The aim was to investigate with a multicriteria approach how random or permanent exposure to a 50-Hz stray voltage applied to the water trough affected behavior, stress physiology, and milk production of dairy cows during an 8-wk (medium-term) period. To identify when a possible habituation to voltage occurs, different time scale measurements were used. Thus, in addition to the short-term stress responses during the first week of exposure, measurements were performed in the medium term (8 wk) to detect chronic stress effects.

MATERIALS AND METHODS

The scientist in charge of the experiments was licensed to perform experiments on animals and the staff who applied the experimental procedures had attended a special course approved by the French Ministry of Agriculture (Paris, France).

Animals, Feeding and Management, and Housing

Animals. Eighty-six lactating Holstein cows were assigned to 1 of 3 treatments before the learning procedure according to their milk yield, parity, and stage of lactation (≤60 d, 61 to 180 d, and ≥181 d postcalving). The absence of differences between treatments for SCC was verified after the cows were assigned to their groups. Eleven cows were discarded during the learning procedure because of high stress responses in the stall or difficulties in learning to use the watering device correctly. The final partitioning before the beginning of treatment application is in Table 1. One cow had to be removed because of health problems in wk 2. One cow was removed in wk 6 because of a fall on the slatted floor that led to severe lameness (data from this cow were used until wk 6). Therefore, 74 cows were used during the 8-wk experimental period.

Table 1. Basal production characteristics (means ± SE) of Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 23) or randomly (36 h/wk; RAND; n = 25) or to no voltage exposure (control; n = 26)

	Treatment			
Item	Control	PERM	RAND	*P*-value
Milk yield[1] (kg/d)	35.4 ± 1.97	33.5 ± 1.53	33.6 ± 1.37	0.665
Parity	2.0 ± 0.17	1.7 ± 0.19	2.0 ± 0.25	0.762
DIM (d)	154 ± 21.9	160 ± 25.0	165 ± 20.8	0.741

[1]Average of the 2 wk before the start of exposure to voltage.

Feeding and Management. Cows were milked twice daily at 0700 and 1700 h. A TMR (DM basis: 43% corn silage, 17% rapeseed meal, 13% sugar beet pulp, 11% brewery byproduct, 11% alfalfa hay, 3% high moisture corn, and 2% vitamins and minerals; NE_L: 6.76 MJ/kg of DM; true protein digested in the small intestine when fermentable N is limiting: 100 g/kg of DM; true protein digested in the small intestine when fermentable energy is limiting: 94 g/kg of DM) was given at 0930 h and refusals were pushed toward the cows at 1800 h. Hay, water, and salt licks were available ad libitum.

Housing. The animals were housed in the same enclosure (27 m × 14 m for the first period, n = 26; 35 m × 14 m for the second period, n = 48) consisting of a straw bedded area and a slatted floor area that gave access to the feeding table. Six individual watering stalls were placed on a concrete area of approximately 60 m^2 (Figure 1). The watering stalls consisted of a stall (1 m × 3 m) equipped with a gate providing access to water. The electromagnetic opening and closure of the gate was activated by an electronic key located around the neck of each cow and detected by an antenna located in the gate so that each cow had access to only 2 watering stalls, which were programmed according to treatment. The stalls were made from plain wood panels that were visually and electrically insulated from each other. The water troughs were electrically insulated from all the metallic parts of the pen and were equipped with a water meter. In addition, each trough was equipped with wooden dividers to visually isolate the animals and to prevent access to water by animals located in the neighboring stalls. An aluminum plate (1 m × 2.5 m), insulated from all the other metallic parts and insulated from the ground, was placed on the floor of each stall. For each repetition, water stalls were randomly allocated to each treatment. Electricité de France Research and Development (Moret sur Loing, France) provided the electricity exposure system (alternating 50-Hz voltage), the system for recording current intensity through the body of the cow, and the

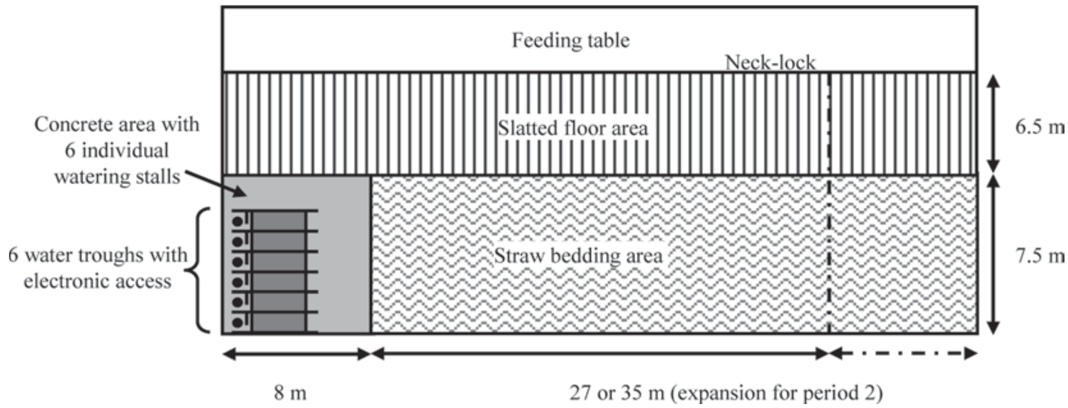

Figure 1. Experimental setting used to study the effects of an 8-wk exposure to voltage on behavior, stress physiology, and milk production in Holstein cows.

calculation of the resistance of the trough-cow-metallic plate set-up. The trough consisted of a constant-level polyethylene bowl with a capacity of 5 L. A metallic plate was immersed in the water to protect the float system (which regulated the level of water in the bowl). It was connected to the exposure system. While drinking, the muzzle of the cow was in contact with the water and with the metallic part of the trough. No pressure on a paddle was needed to obtain water.

Treatments

The experiment was conducted over 2 periods with different animals each time. The first period was from December to February and the second period was from March to May. Each period consisted of 3 identical phases: 1) 4 wk of learning and habituation to the watering device and to the experimental procedures such as blood sampling; 2) 2 wk of recording of the basal period of each animal (called basal period or wk 0 thereafter); and 3) 8 wk of exposure to voltage applied to a water trough (wk 1 to 8). Voltage was applied via the water trough to the aluminum plate placed on the floor of the stall to obtain a voltage pathway through the cow from the muzzle to the 4 hooves. A voltage of 1.8 V was chosen because it was the level of voltage that induced a permanent modification in behavior during a short-term choice experiment (K. Rigalma, unpublished data). During the 8-wk voltage exposure period, a voltage of 1.8 V was applied to the 2 water troughs assigned to each treatment, either permanently (**PERM**, n = 23; n = 8 for the first period and n = 15 for the second period) or randomly (36 h/wk with a duration of exposure varying from 4 to 16 h; **RAND**, n = 25; n = 10 for the first period and n = 15 for the second period). Twenty-six cows (control; n = 8 for the first period and n = 18 for the second period) were used as control animals and were never exposed to voltage.

Activity Measurement

Activity levels were recorded using an activity sensor (DeLaval Inc., Les Clayes sous Bois, France) connected to Alpro (DeLaval Inc.). This activity-sensing transponder, normally used for estrus detection, was securely attached around the necks of the cows with nylon bands. This system is based on detection of physical movement of the cow and records the number of 14-s periods per hour during which the cow is moving. Activity level (called activity hereafter) of 18 PERM cows, 16 RAND cows, and 15 control cows was measured during 24 h consecutively for each day of recording. The measurements were performed on wk 0 (2 d), the first and the third days of wk 1 (**W1D1** and **W1D3**, respectively), wk 2 (2 d), and wk 8 (2 d). When 2 d of measurements were performed per week, the data of the 2 d were averaged per hour. Differences between the measurement during voltage exposure and wk 0 were calculated: **ΔW1D1**, **ΔW1D3**, **ΔW2**, **ΔW4**, and **ΔW8** corresponded to the difference in activity between wk 0 and the first day of wk 1, the third day of wk 1, wk 2, wk 4, and wk 8, respectively. Cows in estrus were removed from the analysis.

Physiological Measurements

Plasma Glucose and Cortisol Concentrations. Blood samples were taken once during wk 0, 1, 2, 4, and 8. After restraining the animals with a neck lock, caudal venipunctures were performed at least 1 h after the return from morning milking, to avoid the peak in plasma cortisol associated with milking, and before the distribution of the TMR. Only blood samples that were taken in the first 2 min after the start of tail handling of the animal were analyzed to exclude a bias related to an acute stress response during blood collection (only 9/369 samples were discarded). This interval of 2 min was likely insufficient for plasma cortisol concentration to be affected by the handling associated with blood collection. Immediately after collection, samples were placed in ice-cooled water and were centrifuged within 1 h at 3,000 × *g* for 10 min at 4°C. Plasma was stored at −20°C until subsequent analysis.

Milk Cortisol Concentration. Milk samples from evening milking were collected during wk 0, W1D1, W1D3, wk 2, and wk 8. Four milliliters of milk representative of the milk collected from each cow were frozen at −20°C until subsequent analysis.

ACTH Challenge. An ACTH challenge was performed on 42 cows (PERM, n = 14; RAND, n = 14; control, n = 14) at the end of wk 7 to assess the activity of the hypothalamic-pituitary-adrenal (**HPA**) axis. The animals were restrained in a neck lock and subsequently left undisturbed for 1 h. Blood samples were then taken for basal cortisol levels; this was followed by a single intravenous injection of ACTH (Synacthene, Novartis-Pharma, Rueil-Malmaison, France) at a dose of 1.98 IU/kg of $BW^{0.75}$ (Munksgaard and Simonsen, 1996) in the tail vein. Blood samples were collected 30, 120, and 180 min after the injection and treated as described previously to measure cortisol responses.

In addition to the maximal plasma cortisol concentration, the integrated response to exogenous ACTH was determined by calculating the area under the curve (**AUC**) using the following formula (Veissier et al., 2001): AUC = Σ (Ct + Ct + 1)/2 × dt, where Ct is the concentration at time t and dt is the time (min) between samples taken at t and t + 1.

Cortisol Analysis. Plasma and milk cortisol were measured by ELISA using an automated method (Elecsys, Roche Diagnostics, Meylan, France). The sensitivity of the cortisol assay was 0.362 ng/mL. The interassay coefficient of variation was 4.5% at 124.69 ng/mL. Milk samples were prepared as follows: 1 mL of skim milk was mixed with 4 mL of ethyl-acetate, shaken, and centrifuged. Part of the mixture (3.5 mL) was transferred into a tube and dried using an evaporator concentrator. The sample was reconstituted with assay buffer and assayed for cortisol.

Production Measurements

Water Intake. Three digital cameras connected to 3 time-lapse videocassette recorders were placed above each pair of troughs to identify the cows present in the watering stalls at all times. A digital camera, placed above the 6 water meters and connected to a hard disk drive recorder, allowed the recording of the 24-h water consumption in each trough (1 frame/s) during wk 0 (2 d), W1D1, W1D3, wk 2 (1 d), and wk 8 (1 d). By synchronizing the 3 video cameras and the water meter measurement, the total amount of water drunk by each cow (called daily water intake hereafter) was measured.

Milk Production and Composition. Each week, the average daily milk yield was calculated using the 7-d milk production (daily milk equaled milk collected in the evening and the milking of the following morning) measured per animal with milk meters (MM15, DeLaval Inc.) connected to Alpro (Alpro, DeLaval Inc.). The difference in milk yield between the second day of voltage exposure and the first day (**Δ2–1**) and between the third day of voltage exposure and the second day (**Δ3–2**) were calculated for each cow. Moreover, the number of cows treated for mastitis was recorded.

Milk samples, collected once weekly (corresponding to a mixture of milk from a successive evening and morning milking), were kept at room temperature with a preservative (Bronopol, Lanxess, Langenfeld, Germany). Samples were sent to the laboratory of the Milk Recording Organisation (Syndicat Interdépartemental de l'Elevage, Le Mée, France) to determine milk fat, protein, and lactose concentrations by infrared spectrophotometry (MilkoScan 6000, Foss Electric, Laurel, MD). Somatic cell counts were evaluated by flow cytometric measurement (Fossomatic 5000, Foss Electric).

Electrical Measurements

A multichannel transient recorder (Nicolet Data Acquisition System, Nicolet Technologies, Madison, WI) associated with an analyzer software (Nicolet Vision version 3.50, Nicolet Technologies) was configured to record current and voltage. Current measurements were performed using a current probe (A6302, Tektronix SA, Les Ulis, France) associated with a current probe amplifier (AM503, Tektronix SA) placed in the circuit at the exit of the power supply box. This current probe was connected to the Nicolet Data Acquisition System (Nicolet Technologies) through a 50-Ω coaxial link. The voltage was measured between the electrified trough and the aluminum plate. During the second period, voltage and current measurements (measured as root mean square) were performed on wk 3 over 4 d from 0900 to 1700 h. For each visit to the drinking station by PERM and RAND cows, 10 measurements evenly distributed during the drinking period were collected and aggregated to obtain the average resistance of the experimental set-up (trough-cow-metallic plate) and the average current crossing the cow (from muzzle to the 4 hooves).

Statistical Analysis

Statistical analyses were performed using the MIXED model procedure of SAS (version 9.1.3, SAS Institute,

Table 2. Activity of Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 18) or randomly (36 h/wk; RAND; n = 16) or to no voltage exposure (control; n = 15)[1]

	Treatment			*P*-value	
Item	Control	PERM	RAND	Treatment	Hour of day
ΔW1D1	$-2.3^{a} \pm 2.74$	$9.8^{b} \pm 2.70$	$2.1^{a} \pm 2.71$	0.023	0.026
ΔW1D3	3.4 ± 2.40	3.8 ± 2.54	0.2 ± 2.585	0.541	0.160
ΔW2	1.4 ± 2.96	0.1 ± 3.10	-4.0 ± 2.98	0.383	0.193
ΔW4	-0.3 ± 2.79	-0.2 ± 2.93	0.3 ± 2.84	0.985	0.086
ΔW8	$-7.7^{a} \pm 3.54$	$-1.0^{ab} \pm 3.70$	$4.2^{b} \pm 3.64$	0.069	<0.001

[a,b]Means within a row with different superscripts differ ($P < 0.05$).

[1]Differences in activity (measured in the experimental pen) between wk 0 and the first day of wk 1, the third day of wk 1, wk 2, wk 4, and wk 8 (ΔW1D1, ΔW1D3, ΔW2, ΔW4, and ΔW8, respectively) are presented.

Cary, NC) with comparison of the estimates (*t*-test based). Physiological (plasma and milk cortisol concentration, cortisol response to ACTH challenge) and some production (daily water intake, Δ2–1, and Δ3–2) data were analyzed with the following model:

$$Y_{ijkl} = \mu + T_j + P_k + R_l + T_j \times P_k + BW_i + e_{ijkl},$$

where Y_{ijkl} is the dependent variable; μ is the overall mean; T_j is the fixed effect of the treatment j with 3 modalities (control, PERM, and RAND); P_k is the fixed effect of the period k with 2 modalities (periods 1 and 2); R_l is the fixed effect of the parity with 3 modalities (1, 2, and >2); $T_j \times P_k$ is the interaction between the treatment j and the period k; BW_i is the average BW of the animal i during the experiment as a covariate; and e_{ijkl} is the residual error. For daily water intake, the milk yield of the day was added as an additional covariate. For the activity level, the model was completed with H_z (the hour z; 1 to 24) as a repeated effect and $T_j \times H_z$ the interaction between the treatment j and the hour z.

Concerning milk yield and composition, the model was completed with W_m (the week m; 1 to 8) as a repeated effect; $T_j \times W_m$ as the interaction between the treatment j and the week m; the stage of lactation with 3 modalities (≤60 d, 61 to 180 d, and ≥181 d after calving); and a preexperimental covariate (mean value per cow for each variable obtained during the 2-wk of the basal level period).

The selection of the covariance structure was based on minimization of the Akaike criterion value. The unstructured, compound symmetry and first-order autoregressive covariance structures were tested. These covariance structures account for the repeated measurements within cow.

When assumptions of homogeneity of variance and normal distribution of the residuals were not verified, a log or inverse transformation was performed before conducting the analysis. Qualitative data such as number of cows treated for mastitis were analyzed with a chi-squared test.

Because no meaningful evidence of the effect of parity, stage of lactation, or interaction between treatment and period were found, these effects are not presented. Least squares means ± standard error are presented except when otherwise stated.

RESULTS

Activity

An effect of treatment ($P = 0.023$) and of hour ($P = 0.026$) was observed on ΔW1D1 (Table 2; Figure 2): activity of PERM cows was higher than activity of control cows ($P = 0.007$). An effect of hour ($P < 0.001$) was observed on ΔW8 (Table 2; Figure 2): activity of RAND cows was higher than activity of control cows ($P = 0.022$). No effect of treatment was observed on ΔW1D3, ΔW2, and ΔW4 (Table 2). An effect of period was found for ΔW1D3, ΔW2, ΔW4, and ΔW8 ($P < 0.01$).

Physiological Measurements

In wk 2, a trend for a treatment effect ($P = 0.056$) was found for plasma cortisol concentrations (Table 3). No treatment effects were observed on plasma cortisol concentrations in the other weeks. In wk 8, an effect of period was found on plasma cortisol concentrations ($P < 0.001$; Table 3).

In wk 8, an effect of treatment ($P = 0.049$) was found for milk cortisol concentrations: RAND cows had higher milk cortisol concentrations than PERM cows ($P = 0.022$; Table 3). No differences were observed between treatments for milk cortisol concentrations in the other weeks. In W1D1, an effect of period was found for milk cortisol concentrations ($P = 0.002$; Table 3).

After an ACTH challenge, no treatment effects were observed for the integrated response (AUC from 0 to 180 min: 13,558 ± 874.6, 13,206 ± 794.9, and 13,870 ± 760.4 ng/min per mL for control, PERM, and RAND cows, respectively; $P = 0.816$) and for the maximal plasma cortisol concentration (log back-transformed data: 95.3 ± 5.61, 94.8 ± 5.06, and 94.3 ± 4.81 ng/mL for control, PERM, and RAND cows, respectively; $P = 0.990$).

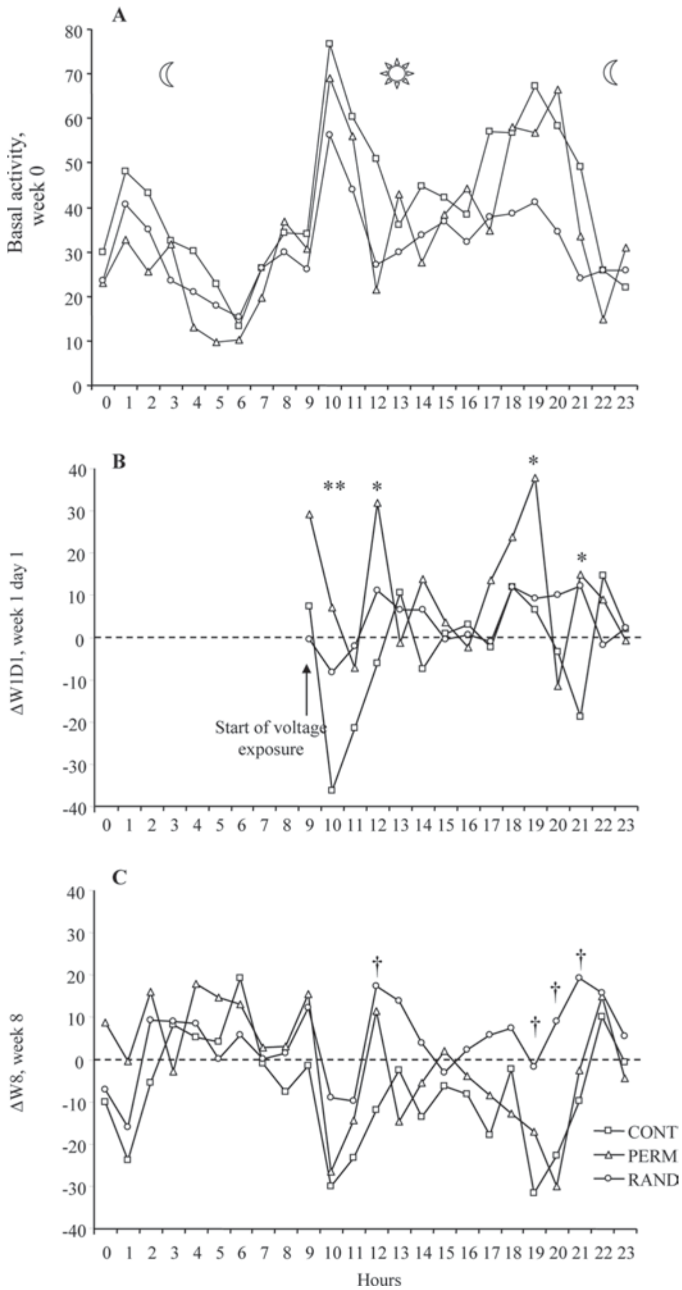

Figure 2. Activity level of Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 18) or randomly (36 h/wk; RAND; n = 16) or to no voltage exposure (CONT; n = 15). A) Basal activity in wk 0 (average of 2 d). B) Difference in activity between wk 1 d 1 and wk 0 (ΔW1D1). C) Difference in activity between wk 8 (average of 2 d) and wk 0 (ΔW8). †$0.05 < P < 0.10$; *$P < 0.05$; **$P < 0.01$.

Production Measurements

No differences were observed between treatments for daily water intake during wk 0, W1D1, W1D3, wk 2, and wk 8 (Table 4). No differences were observed between treatments for the number of cows treated for mastitis: 4 control cows out of 25, 2 PERM cows out of 23, and 5 RAND cows out of 26 during the 8 wk of exposure (chi-squared = 0.427; $P = 0.808$). No differences between treatments were observed for weekly milk composition (Table 5).

No treatment differences were observed for average daily milk yield calculated weekly (Figure 3), whereas an effect of week ($P < 0.001$) was observed. However, a treatment effect was observed on Δ2–1 ($P = 0.026$): a decrease in milk yield was observed in PERM cows compared with control and RAND cows (Table 6). In addition, a treatment effect was observed for Δ3–2 ($P = 0.004$): a decrease in milk yield was observed in RAND cows compared with control and PERM cows (Table 6). No differences between treatments were observed the following days.

Electrical Measurements

The average resistance of the set-up trough-cow-metallic plate was 516 ± 13.4 Ω (mean ± SE). The resistance varied between 411 and 680 Ω. When a cow was drinking, the average current crossing its body was 3.6 ± 0.08 mA.

DISCUSSION

The aim was to study the medium-term effects of a 50-Hz stray voltage on activity, stress physiology, milk yield, and composition. Measurements were performed to measure acute as well as chronic stress responses. The voltage was applied to a water trough either permanently or randomly. The reason for applying voltage in an unpredictable way was to mimic farm conditions (Deschamps, 2002). No drastic reductions in water intake or unsafe behavior (i.e., kicking, agonistic behaviors, struggling or violent movements in the watering stall) during voltage exposure were observed in the present experiment.

Activity

An effect of the period was observed on cow activity. This could be explained by differences in the environ-

Table 3. Plasma and milk cortisol concentrations in Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 23) or randomly (36 h/wk; RAND; n = 25) or to no voltage exposure (control; n = 26) during a basal period (wk 0, 2 d), d 1 and 3 of wk 1 (wk 1 d 1 and wk 1 d 3, respectively), wk 2 (1 d), and wk 8 (1 d) of exposure to voltage

	Treatment			*P*-value	
Item	Control	PERM	RAND	Treatment	Period
Plasma cortisol concentration (ng/mL)					
wk 0	3.2 ± 0.48	3.0 ± 0.44	3.2 ± 0.47	0.947	0.845
wk 1	4.5 ± 0.65	6.0 ± 0.59	4.8 ± 0.59	0.191	0.459
wk 2	3.8 ± 0.59	4.4 ± 0.59	5.8 ± 0.59	0.056	0.531
wk 8	4.8 ± 0.96	5.6 ± 0.90	4.9 ± 0.83	0.757	0.001
Milk cortisol concentration (ng/mL)					
wk 0	0.17 ± 0.036	0.20 ± 0.036	0.25 ± 0.056	0.536	0.534
wk 1 d 1	0.22 ± 0.032	0.24 ± 0.034	0.21 ± 0.030	0.863	0.002
wk 1 d 3	0.24 ± 0.036	0.18 ± 0.029	0.19 ± 0.028	0.403	0.758
wk 2	0.12 ± 0.060	0.16 ± 0.063	0.25 ± 0.056	0.263	0.440
wk 8	0.15^{ab} ± 0.020	0.14^{a} ± 0.020	0.21^{b} ± 0.024	0.049	0.623

[a,b]Means within a row with different superscripts differ ($P < 0.05$).

ment during the 2 experimental periods (temperature, humidity, day length) that may influence cow activity.

An effect of voltage exposure was observed on cow activity. Indeed, on the first day of exposure, cows permanently exposed to voltage exhibited higher activity than control cows and, at the end of the experiment, cows randomly exposed to voltage showed higher activity than control cows. Activity is a criterion that can be used to evaluate comfort and welfare in farm animals. For example, activity of cattle can be influenced by lack of rest (Munksgaard and Simonsen, 1996) or social instability (repeated changes of conspecifics and pens; Veissier et al., 2001).

In our experiment, during wk 0, 2 diurnal peaks of activity were observed around 1000 and 1800 h (Figure 2), supporting Linnane et al. (2001). These peaks corresponded to feeding (first diurnal peak) and to the return from the afternoon milking accompanied by the push-up of feed refusals (second diurnal peak). In addition, feed intake was usually followed by drinking activity. On the first day of exposure, cows permanently exposed to voltage had higher peaks of activity across the day, which could be explained because they were confronted by voltage for the first time. This extra activity could be partly attributed to the numerous visits to the water trough where the cows were confronted by the novelty and the discomfort of the voltage. This extra activity was more pronounced at the period of the day when drinking activity was usually observed; approximately 66% of the visits to the watering stall were performed between 1000 and 1900 h. In wk 8, cows experiencing random voltage exposure had higher activity than control cows, especially at the end of the day. This increase in activity at the end of the day was reported in calves subjected to repeated regrouping and relocation that were feeding and stepping (taking at least 1 step) more from 1800 to 2000 h compared with control calves (Veissier et al., 2001).

Stress Physiology

During the first 2 wk of exposure to voltage, no differences between treatments were observed for basal plasma cortisol concentrations. Higher milk cortisol concentrations were recorded during wk 8 in cows randomly exposed to voltage compared with permanently exposed cows.

In our experiment, the choice was made not to measure plasma cortisol concentrations immediately after exposure to the electrified trough (i.e., acute response

Table 4. Daily water intake (L/d) by Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 23) or randomly (36 h/wk; RAND; n = 25) or to no voltage exposure (control; n = 26) during a basal period (wk 0, 2 d), d 1 and 3 of wk 1 (wk 1 d 1 and wk 1 d 3, respectively), wk 2 (1 d), and wk 8 (1 d) of exposure to voltage

	Treatment			
Item	Control	PERM	RAND	*P*-value, treatment
wk 0	72.9 ± 2.59	76.8 ± 2.64	70.8 ± 2.36	0.241
wk 1 d 1	74.3 ± 3.37	75.9 ± 3.25	74.8 ± 3.02	0.933
wk 1 d 3	71.0 ± 4.06	77.1 ± 3.58	76.4 ± 3.37	0.485
wk 2	78.9 ± 2.75	79.5 ± 2.71	76.6 ± 2.54	0.697
wk 8	79.5 ± 3.95	72.1 ± 3.93	77.1 ± 3.68	0.382

Table 5. Milk production and milk composition of Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 23) or randomly (36 h/wk; RAND; n = 25) or to no voltage exposure (control; n = 26)

	Treatment			*P*-value			
Item	Control	PERM	RAND	Treatment	Week	Treatment × week	Period
Milk yield (kg/d)	34.7 ± 0.59	34.5 ± 0.63	33.8 ± 0.59	0.550	<0.001	0.643	0.645
Fat (g/L)	38.0 ± 0.60	37.7 ± 0.64	38.7 ± 0.61	0.400	<0.001	0.448	0.012
Protein (g/L)	30.9 ± 0.50	30.5 ± 0.53	31.0 ± 0.49	0.773	<0.001	0.283	0.459
Lactose (g/L)	50.8 ± 0.28	50.8 ± 0.30	50.5 ± 0.28	0.746	<0.001	0.308	0.076
SCC (10^3 cells/mL)	262 ± 95.9	276 ± 101.8	256 ± 96.9	0.260	0.808	0.204	0.432

to the stressor). This was in order not to disturb the activity of the animals by restraining them to obtain blood samples. In other experiments, exposing cows to a similar electric stressor (4 mA during milking) induced an acute stress response with an increase in plasma cortisol (Henke Drenkard et al., 1985) 10 min after exposure to the stressor. Yet, in other experiments (Lefcourt et al., 1986; Reinemann et al., 2003) no effects of electric current (from 2.5 to 12.5 mA) were reported on plasma cortisol concentrations 15 min after exposure to the stressor.

In the literature, it was reported that chronic stress could induce either a decrease or an increase in basal cortisol concentrations. Indeed, van Reenen et al. (2000) observed a slight decrease in basal cortisol concentrations in calves chronically stressed by isolation, whereas Barnett et al. (1988) observed an increase in basal cortisol concentrations in pigs chronically stressed by housing conditions (tether stalls). However, Jensen et al. (1996) observed no differences in basal cortisol concentrations in pigs receiving unpredictable and inescapable electric shocks (2.7 V, 0.25 mA, 60 to 65 pulses/s) applied to the neck region for a month compared with control pigs. Nevertheless, it is difficult to demonstrate the presence of a chronic stress without vascular catheterization and frequent blood sampling. Indeed, in cattle, basal plasma cortisol concentrations were only slightly increased in chronic stress and vary greatly because of ultradian and diurnal rhythms (Mormède et al., 2007). Thus, additional cortisol measurements could be useful for evaluating chronic stress.

Cortisol in the milk can be used as a noninvasive way of measuring cortisol. It has been shown that measurements of cortisol concentrations in milk and blood were closely correlated (Verkerk et al., 1998). If the interval between 2 milkings is short, milk cortisol instantaneously reflects changes in blood cortisol (Termeulen et al., 1981). It is usually accepted that milk cortisol concentrations are a good indicator of the response to an acute stressor in lactating cows when it occurs within a period up to 2 h before milk sampling (Verkerk et al., 1998). Thus, the higher milk cortisol recorded in cows randomly exposed to voltage could indicate that, even after 8 wk, the cows were not completely habituated to random voltage. The opposite was observed in cows exposed permanently to stray voltage. This result could indicate that the cows exposed to unpredictable voltage experienced a mild chronic stress.

Chronic stress can be studied by assessing the reactivity of the HPA axis. The stimulation of the HPA axis is usually performed by injecting synthetic ACTH intravenously and by measuring the area under the cortisol response curve (Veissier et al., 2001). During our experiment, after 7 wk of exposure to electricity, no modifications in the area under the cortisol response curve or the maximal cortisol response after ACTH injection were observed. Induction of chronic stress in dairy cows does not automatically result in a change in the reactivity of the adrenal gland (Munksgaard and Simonsen, 1996).

Permanent exposure to stray voltage did not modify any of the stress cortisol indicators, probably indicating a habituation of the animals to the stressor when it was experienced in a permanent and predictable way, whereas random voltage exposure seemed to have at least some effects on the stress physiology of the cows,

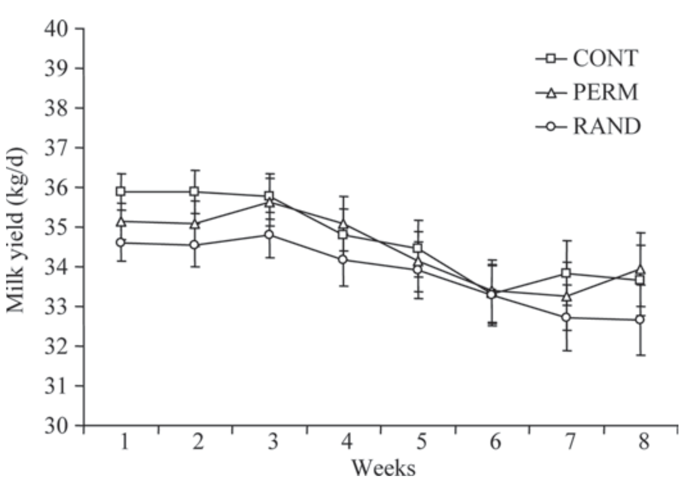

Figure 3. Evolution in milk yield (kg/d) of Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 23) or randomly (36 h/wk; RAND; n = 25) or to no voltage exposure (CONT; n = 26).

Table 6. Difference in milk yield between the second day of experiment and the first day (Δ2–1) and between the third day of experiment and the second day (Δ3–2) in Holstein cows submitted to an 8-wk exposure to voltage (1.8 V applied to a water trough) permanently (PERM; n = 23) or randomly (36 h/wk; RAND; n = 25) or to no voltage exposure (control; n = 26)

	Treatment			
Item	Control	PERM	RAND	*P*-value, treatment
Δ2–1	$0.1^{ab} \pm 0.58$	$-1.4^{b} \pm 0.74$	$1.4^{a} \pm 0.71$	0.026
Δ3–2	$0.2^{a} \pm 0.89$	$1.1^{a} \pm 0.85$	$-3.5^{b} \pm 1.03$	0.004

[a,b]Means within a row with different superscripts differ ($P < 0.05$).

even if only transient modifications could be detected. In cows, habituation to an electric stressor supports observations by Gorewit et al. (1985) and Henke Drenkard et al. (1985). But, in the present experiment, additional criteria of chronic stress such as an ACTH challenge did not confirm modifications in stress physiology in cows exposed to electricity. More research is needed to fully understand the stress physiology of cows in the presence of a long-lasting stressor.

Water Intake and Milk Production and Composition

No modifications in daily water intake were observed after treatment application, which supports others (Gorewit et al., 1992a; Reinemann et al., 2005). In addition, daily water intake was similar to that of Cardot et al. (2008).

An effect of week was observed on milk yield and reflected the physiological decrease in milk yield with increasing stage of lactation after peak production. No differences were observed between treatments for milk production and milk composition. The lack of effect of voltage exposure on milk yield and milk composition supports others (Henke Drenkard et al., 1985; Gorewit et al., 1992a; Reinemann et al., 2002). During the first 3 d of voltage exposure, a transient decrease in milk production was observed in cows permanently or randomly exposed to voltage compared with control cows. No other differences were observed the following days. Our results support Reinemann et al. (2005), who observed a decrease in milk yield in the first 2 d of current exposure (with a peak current from 8.5 to 20 mA). This result indicates that cows were transiently disturbed by exposure to stray voltage during the first 2 d when it was applied in a permanent manner. This disturbance occurred with a longer delay when voltage exposure was unpredictable.

Field observations indicated that stray voltage impairs production in dairy cows (a decrease in milk yield and an increase in SCC; Wilson et al., 1996). No studies in controlled environments confirmed these results (Lefcourt et al., 1985; Gorewit et al., 1992b; Southwick et al., 1992). In dairy cows, the application of a voltage (1, 2, or 4 V) to a trough over the entire lactation had no effect on milk yield (Gorewit et al., 1992a) or on the incidence of mastitis (Gorewit et al., 1992b). During our experiment, cows were maintained in good housing conditions: adequate space, no competition for feed, and no mixing with foreign animals. These conditions allowed the animals to express their full production potential. These comfortable rearing conditions may in part explain the lack of effects of stray voltage on the production measures.

Electrical Measurement and Individual Variability

In the present experiment, the alternating (50 Hz) voltage of 1.8 V corresponded to an average current (crossing the circuit trough-cow-metallic plate) of 3.6 mA. The calculated resistance of the trough-cow-metallic floor was 516 ± 13.4 Ω, slightly higher than the average value cited in the literature for the mouth-all hooves pathway of 359 Ω (from 244 to 525 Ω; Norell et al., 1983). This difference could be explained because Norell et al. (1983) experimented with a specific piece of equipment (e.g., a metallic grid in the mouth of the cows) used to reduce the contact resistance. The variability of the resistance of the trough-cow-metallic plate (from 411 to 680 Ω) could be partly explained by the resistance of the muzzle-trough set-up. Indeed, cows positioned their muzzle differently in the water trough and this could have led to a variation in the resistance of the set-up muzzle-trough: some cows put their muzzle deep in the bowl, others maintained their muzzle on the surface of the water, and some drank while leaning the head on the side of the trough. Drinking behavior was reported as a possible variation factor in the electrical resistance of cows. Indeed, Reinemann et al. (2005) observed a change in drinking behavior after voltage exposure: some cows exerted a pressure on the trough and increased the contact surface muzzle-trough and reduced the current density at the level of their muzzle. Further investigation is necessary to relate the position of the muzzle in the bowl to the resistance of the contact point.

Unpredictability

On the farm, stray voltage is an intermittent or random phenomenon, which makes it unpredictable for animals (Deschamps, 2002). In our experiment, random exposure to voltage consisted of several periods of different durations from 4 to 16 h. Permanent exposure to stray voltage is classically used in the literature: animals are routinely subjected to the electric stressor. Unpredictability of electricity can be more disruptive to the animal. Indeed, studies on stress in livestock indicate that the response of an animal to its environment did not depend on the situation itself but rather on the interpretation that the animal makes of this situation (Boissy et al., 2007). Predictability is one criterion that would allow the animal to assess a situation (Désiré et al., 2002) and may modify behavior. In our experiment, unpredictable stray voltage seemed to generate a higher and more persistent response than permanent (predictable) stray voltage. Because the exposure of the cows to 1.8 V (3.6 mA) applied to a water trough did not impair production, it suggests that such a low stray voltage is a mild stressor, inducing only behavioral and physiological changes.

CONCLUSIONS

Repeated exposure to stray voltage induced a response of acute stress at the beginning of the experiment, with a transient decrease in milk production and an increase in activity in cows permanently exposed to voltage. These effects were short lived. The unpredictable exposure to stray voltage may generate a chronic stress, with a slight increase in basal milk cortisol and an increase in activity. The response to an ACTH challenge, used to test the reactivity of the adrenal gland, was not modified at the end of the experiment by stray voltage. As opposed to field observations, our experiment, using 1.8 V applied to a water trough, did not affect milk yield and composition and SCC. In the context of this experiment, stray voltage could be considered a mild chronic stressor in dairy cows, especially when it was unpredictable, with only slight modifications in stress physiology accompanied by changes in activity. Moreover, no impairment of milk production was observed.

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REFERENCES

Aneshansley, D. J. 2003. Stray voltage research at Cornell University. Pages 63–71 in Proc. Stray Voltage and Dairy Farms: A Conference for Farm Advisors, Educators, Utilities, and Public Policy Advisors. NRAES, Camp Hill, PA.

Aneshansley, D. J., R. C. Gorewit, and L. R. Price. 1992. Cow sensitivity to electricity during milking. J. Dairy Sci. 75:2733–2741.

Barnett, J. L., P. H. Hemsworth, G. M. Cronin, C. G. Winfield, T. H. McCallum, and E. A. Newman. 1988. The effects of genotype on physiological and behavioural responses related to the welfare of pregnant pigs. Appl. Anim. Behav. Sci. 20:287–296.

Boissy, A., C. Arnould, E. Chaillou, V. Colson, L. Désiré, C. Duvaux-Ponter, L. Greiveldinger, C. Leterrier, S. Richard, S. Roussel, H. Saint-Dizier, M. C. Meunier-Salaün, and D. Valance. 2007. Emotion and cognition: A new strategy to achieve animal welfare. INRA Prod. Anim. 20:17–21.

Cardot, V., Y. Le Roux, and S. Jurjanz. 2008. Drinking behavior of lactating dairy cows and prediction of their water intake. J. Dairy Sci. 91:2257–2264.

Deschamps, F. 2002. L'électricité dans l'environnement et les exploitations agricoles. Bull. Soc. Vet. Prat. France 86:174–181.

Désiré, L., A. Boissy, and I. Veissier. 2002. Emotions in farm animals: A new approach to animal welfare in applied ethology. Behav. Processes 60:165–180.

Gorewit, R. C., D. J. Aneshansley, and L. R. Price. 1992a. Effects of voltages on cows over a complete lactation. 1. Milk yield and composition. J. Dairy Sci. 75:2719–2725.

Gorewit, R. C., D. J. Aneshansley, and L. R. Price. 1992b. Effects of voltages on cows over a complete lactation. 2. Health and reproduction. J. Dairy Sci. 75:2726–2732.

Gorewit, R. C., N. R. Scott, and C. S. Czarniecki. 1985. Responses of dairy cows to alternating electrical current administered semi-randomly in a non-avoidance environment. J. Dairy Sci. 68:718–725.

Henke Drenkard, D. V., R. C. Gorewit, N. R. Scott, and R. Sagi. 1985. Milk production, health, behavior, and endocrine responses of cows exposed to electrical current during milking. J. Dairy Sci. 68:2694–2702.

Hultgren, J. 1990. Small electric currents affecting farm animals and man: A review with special reference to stray voltage. I. Electric properties of the body and the problem of stray voltage. Vet. Res. Commun. 14:287–298.

Jensen, K. H., L. J. Pedersen, E. K. Nielsen, K. E. Heller, J. Ladewig, and E. Jørgensen. 1996. Intermittent stress in pigs: Effects on behavior, pituitary-adrenocortical axis, growth, and gastric ulceration. Physiol. Behav. 59:741–748.

Lefcourt, A. M., R. M. Akers, R. H. Miller, and B. Weinland. 1985. Effects of intermittent electrical shock on responses related to milk ejection. J. Dairy Sci. 68:391–401.

Lefcourt, A. M., S. Kahl, and R. M. Akers. 1986. Correlation of indices of stress with intensity of electrical shock for cows. J. Dairy Sci. 69:833–842.

Linnane, M. I., A. J. Brereton, and P. S. Giller. 2001. Seasonal changes in circadian grazing patterns of Kerry cows (*Bos taurus*) in semi-feral conditions in Killarney National Park, Co. Kerry, Ireland. Appl. Anim. Behav. Sci. 71:277–292.

Mormède, P., S. Andanson, B. Auperin, B. Beerda, D. Guemene, J. Malmkvist, X. Manteca, G. Manteuffel, P. Prunet, C. G. van Reenen, S. Richard, and I. Veissier. 2007. Exploration of the hypothalamic-pituitary-adrenal function as a tool to evaluate animal welfare. Physiol. Behav. 92:317–339.

Munksgaard, L., and H. B. Simonsen. 1996. Behavioral and pituitary adrenal-axis responses of dairy cows to social isolation and deprivation of lying down. J. Anim. Sci. 74:769–778.

Norell, R. J., R. J. Gustafson, R. D. Appleman, and J. B. Overmier. 1983. Behavioural studies of dairy cattle sensitivity to electrical currents. Trans. ASAE 26:1506–1511.
Quirce, C. M., M. Odio, and J. M. Solano. 1981. The effects of predictable and unpredictable schedules of physical restraint upon rats. Life Sci. 28:1897–1902.
Reinemann, D. J., M. D. Rasmusssen, and S. D. LeMire. 2002. Milking performance of dairy cows subjected to electrical current and induced milking machine problems. Trans. ASAE 45:833–838.
Reinemann, D. J., L. E. Stetson, N. E. Laughlin, and S. D. LeMire. 2005. Water, feed, and milk production response of dairy cattle exposed to transient currents. Trans. ASAE 48:385–392.
Reinemann, D. J., L. E. Stetson, and S. D. LeMire. 2004. Comparison of dairy cow aversion to continuous and intermittent current. Trans. ASAE 47:1257–1260.
Reinemann, D. J., M. C. Wiltbank, M. D. Rasmussen, L. G. Sheffield, and S. D. LeMire. 2003. Comparison of behavioral to physiological response of cows exposed to electric shock. Trans. ASAE 46:507–512.
Southwick, L. H., D. J. Wilson, and P. M. Sears. 1992. Milk production, water consumption, and somatic cell count responses of cows subject to one or two volts of alternating current. J. Am. Vet. Med. Assoc. 201:441–444.
Termeulen, S. B., W. R. Butler, and R. P. Natzke. 1981. Rapidity of cortisol transfer between blood and milk following adrenocorticotropin injection. J. Dairy Sci. 64:2197–2200.
USDA. 1991. Effects of electrical voltage/current on farm animals: How to detect and remedy problems. Agriculture handbook no. 696. A. M. Lefcourt, ed. USDA, Washington, DC.
van Reenen, C. G., M. H. Mars, I. E. Leushuis, F. A. M. Rijsewijk, J. T. van Oirschot, and H. J. Blokhuis. 2000. Social isolation may influence responsiveness to infection with bovine herpesvirus 1 in veal calves. Vet. Microbiol. 75:135–143.
Veissier, I., A. Boissy, A. M. de Passille, J. Rushen, C. G. van Reenen, S. Roussel, S. Andanson, and P. Pradel. 2001. Calves' responses to repeated social regrouping and relocation. J. Anim. Sci. 79:2580–2593.
Verkerk, G. A., A. M. Phipps, J. F. Carragher, L. R. Matthews, and K. Stelwagen. 1998. Characterization of milk cortisol concentrations as a measure of short-term stress responses in lactating dairy cows. Anim. Welf. 7:77–86.
Wilson, D. J., L. H. Southwick, and D. R. Kaeser. 1996. Improvement in milk production and udder health following correction of stray voltage on computer feeders. Agric. Practice 17:24–29.