



## Evaluation of plasma cortisol levels and behavior in dogs wearing bark control collars

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### Abstract

On wk –1, 24 healthy mixed breed kennel dogs were screened by physical examination, complete blood count, serum biochemistry, and plasma cortisol measurement. Dogs were tested to ensure they barked at an unfamiliar dog. Dogs were randomly assigned to control, electronic bark collar or lemon spray bark collar groups ( $n = 8$  per group). On wk 0 (acclimation baseline), dogs wore inactivated collars 30 min/day for 3 consecutive days. On wks 1 and 2, dogs wore an activated collar 30 min/day for 3 consecutive days. Controls wore an inactivated collar. The bark stimulus was an unfamiliar dog walked in front of the run, three times, 30 s per presentation. Plasma cortisol was measured wk –1, wk 0 d 3, wk 1 d 1, wk 1 d 3 and wk 2 d 3. ACTH was measured wk 0 d 3 and wk 1 d 1. Barking and activity were measured each session.

**Results:** Dogs wearing electronic or lemon spray collars barked less than controls ( $P < 0.05$ ) by the 2nd day wearing an activated collar, with no significant difference in barking between collars. Mean numbers of collar corrections per dog on the 1st day wearing an activated collar were 4.0 (electronic) and 2.0 (lemon spray); the values decreased to 0 for both collars on the 3rd day. ACTH levels did not differ among groups ( $P > 0.05$ ). Mean plasma cortisol levels were within the reference range for all groups throughout the study. Overall, there was a significant time effect ( $P < 0.05$ ) but no significant difference in plasma cortisol between the control, lemon spray and electronic collar groups ( $P > 0.05$ ). Activity did not change significantly over time ( $P > 0.05$ ).

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**Conclusion:** Electronic and lemon spray bark collars significantly reduced barking, with no significant difference between collar types. Dogs with either type of bark collar had an increase in plasma cortisol the 1st day wearing the activated collar, but this was not statistically significant ( $P > 0.05$ ). There was no statistically significant difference in plasma cortisol levels between dogs wearing control, lemon spray or electronic collars.

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## 1. Introduction

Dog owners in the USA and United Kingdom who have been surveyed regarding canine behavior problems have typically listed excessive barking as one of the most common complaints (Beaver, 1994; Campbell, 1986; Wells and Hepper, 2000). When Patronek et al. (1996) conducted a case–control study of households that relinquished dogs for adoption, they found that if the work of caring for a dog was more than expected, then the dog was at increased risk of relinquishment. Unwanted barking was one of the factors which significantly contributed to care of the dog being more than expected. Recently, Villalobos (2005) pointed out that responsible pet ownership includes controlling excessive barking, and that veterinarians have a consultant role to guide pet owners who need help with this problem.

Anti-bark collars such as electronic (“shock”) collars have been considered effective bark deterrents (Hart and Hart, 1985), but some owners have concerns that these collars may inflict undue pain and stress (Rudolph and Myers, 2004). Citronella spray collars are an alternative. Studies comparing electronic and spray collars have indicated that citronella spray collars (Juarbe-Diaz and Houpt, 1996; Moffat et al., 2003) and unscented spray collars (Beaudet, 2001) also deter barking. Moffat et al. (2003) compared citronella and unscented spray collars. Both collars reduced barking, but there was a trend toward higher efficacy with the citronella spray collar.

Some authors have suggested that a citronella spray collar may be gentler and more effective than an electronic collar (Rudolph and Myers, 2004). The mechanism of action of the citronella spray collar is not known. Collar mounted training devices marketed to suppress barking are designed to deliver an aversive stimulus immediately, reliably, and consistently to a specific stimulus, such as vibrations of the throat and/or barking noise. The correction should startle the dog sufficiently to interrupt the unwanted behavior and deter repetition of that behavior (Coleman and Murray, 2000). The element of surprise of a disruptive stimulus may be responsible for deterring barking (Beaudet, 2001). Alternatively, dogs with citronella spray collars may be distracted by the odor and try to locate the source, or calmed by the odor, startled by the noise, or find the spray’s odor offensive (Juarbe-Diaz, 1997). But regardless of which collar is used, experts have stressed that no device should be recommended without concomitant behavior modification (Juarbe-Diaz and Houpt, 1996).

The purposes of this study were to (1) measure plasma cortisol and ACTH levels as indicators of physiological stress in healthy adult dogs wearing electronic and lemon spray bark control collars; (2) determine the effectiveness of the two collar types for control of barking.

## 2. Materials

### 2.1. Outline of study

On wk –1, 24 mixed breed kennel dogs were screened (physical examination, complete blood count, serum biochemistry, and plasma cortisol measurement) to ensure that they were healthy. Dogs also were

tested to ensure they barked at an unfamiliar dog. Dogs admitted to the study were randomly assigned to three groups: control, electronic bark collar or lemon spray bark collar ( $n = 8$  per group). On wk 0 (acclimation baseline), dogs wore inactivated collars for 30 min/day for 3 consecutive days. On wks 1 and 2, dogs wore an activated collar for 30 min/day for 3 consecutive days. Controls wore an inactivated collar. The bark stimulus was an unfamiliar dog walked in front of the run at specific time periods.

## 2.2. Subjects

The project was approved by the Animal Care and Use Committee and the Human Participant Review Committee, Tuskegee University. The original population was comprised of 24 dogs housed in a private no-kill shelter. All dogs had been relinquished by their owners. The study was approved by the shelter's manager and board of directors, who acknowledged that excessive barking was a major cause of relinquishment of dogs and an impediment to adoption. It was hoped that participating in the study would enhance the adoptability of the dogs. Three dogs were eliminated on wk 0 for failure to bark when presented with the bark stimulus (an unfamiliar dog on leash) while wearing an inactivated collar. Because these three dogs had barked at unfamiliar dogs during screening on wk -1, the investigators conjectured that the three dogs may have experienced bark collars previously (Moffat et al., 2003). The 21 remaining dogs included 11 males (9 intact, 2 neutered), and 10 intact females. All dogs were considered healthy based on physical examination, serum biochemistry, and complete blood count. Body weights ranged from 15.5 to 35.6 kg (mean, 22.5 kg). Mean age was 20 months (S.D. 14 months; range 10–64 months). Most dogs were mixed breed; 15 dogs appeared to be Labrador Retriever crosses. Mean duration of housing at the kennel prior to the study was 5 months (S.D. 3 months; range 1–15 months).

The initial screening prior to acceptance into the trial included a cursory behavior evaluation performed while observing each dog in its run (Schaffer and Schaffer, 1996). Dogs admitted to the trial were classified as playful ( $n = 5$ ), playful and attentive ( $n = 2$ ), attentive ( $n = 3$ ), submissive ( $n = 7$ ), submissive and attentive ( $n = 1$ ), passive submissive ( $n = 1$ ) and submissive and slightly fearful ( $n = 2$ ).

## 2.3. Housing

Dogs were housed individually in an animal shelter, in an indoor kennel with concrete floors, side walls with the lower half concrete and the upper half wire mesh, and wire mesh doors. Individual runs consisted of two sections (each approximately 1.2 m × 2.4 m) separated by a guillotine door. One section of each run faced a concrete wall toward the interior of the building and was air conditioned. The section on the other side of the guillotine door faced a concrete walkway and windows overlooking a yard, with fans but no air conditioning. Dogs were moved to this part of the run for recording sessions. Two dogs were studied at the same time, with approximately 10 empty runs and a hallway between each pair of dogs (total distance approximately 14 m). With this arrangement, the dogs were not in direct view of each other. Only personnel involved in the study were admitted to the area during testing in order to minimize distractions. All dogs had been housed in their run for a minimum of 7 days to avoid stress associated with exposure to a new environment.

Dogs were kept on the same commercial dog food and feeding schedule over the entire study. Recordings were performed between 11:00 and 16:00 h. Dogs were fed once per day at 16:45 h. Ambient temperature and humidity were measured at the start and end of each session. Ambient temperature ranged from 23 to 33 °C. Humidity ranged from 57 to 97%.

## 2.4. Study schedule and data acquisition

*Wk -1 (screening).* Dogs were screened by physical examination, blood collection for complete blood count, serum biochemistry, and plasma cortisol measurement, and presentation of the bark stimulus (an unfamiliar dog on leash) to ensure that they barked. Then, dogs were randomly assigned to one of three groups (control, electronic bark collar or lemon spray bark collar,  $n = 8$  per group), by picking a card out of a hat.

*Wk 0 (acclimation baseline)*. Dogs wore inactivated collars 30 min/day for 3 consecutive days. Data were recorded on wk 0 d 3.

*Wks 1 and 2*. Dogs wore an activated collar 30 min/day for 3 consecutive days. Controls wore an inactivated collar. Data were recorded on 3 consecutive days for both weeks.

Plasma cortisol was measured five times (wk -1, wk 0 d 3, wk 1 d 1, wk 1 d 3 and wk 2 d 3). ACTH was measured wk 0 d 3 and wk 1 d 1. Barking duration and activity were measured each session.

Two observers evaluated two dogs simultaneously in individual runs spaced 10 empty runs apart (14 m). Dogs were paired based on the location of their kennel. Approximately equal distances were maintained between all pairs of test dogs. A third person presented the bark stimulus. The observer sat quietly, approximately 1.2 m in front of the run, in full view of the test dog. A digital camcorder mounted on a tripod was positioned a fixed distance behind the observer to videotape each session. The observer timed barking with a stopwatch, and recorded the number of collar corrections. Each observer held a pocket voice recorder and clipboard to record observations. At the end of each 30 min session, personnel entered the run, turned off activated collars, and took blood samples on designated days.

### 2.5. *Bark stimulus*

The bark stimulus was an unfamiliar dog walked on leash in front of the run of the test dog. A different dog was presented at each of the three presentations during the 30 min session (at 5, 15 and 25 min), for 30 s per presentation. The unfamiliar dogs were selected to be dogs which themselves did not bark during testing. These dogs were housed in another part of the kennel.

### 2.6. *Test collars*

Test collars were a lemon spray bark control collar (Model SBC100 Spray bark control collar containing lemon spray [Lemon Spray RFA-164, Petsafe Corp., Knoxville TN]) and an electronic bark control collar (Deluxe Bark Collar Model DBC100 electrical stimulation collar; Radio Systems Corp., Knoxville, TN). The spray collar emitted a lemon-scented spray but did not contain citronella. Vibration detection triggered the spray. The electronic collar used a combination of vibration and sound detection techniques such that the device activated when both the sound and vibration of a bark were detected simultaneously. The approximate time from detection of the bark to correction is 152 ms for the electronic collar, and 67 ms for the spray collar.

Activated electronic collars were set at the “off” position for wk 0 acclimation, and “low” intensity setting on wks 1 and 2. The electronic collar was positioned high on the neck, immediately below the jaw. The lemon spray collar was positioned 2–4 cm lower, so that the lower jaw did not block the spray outlet of the canister. Collars were checked each week to ensure proper functioning. During most of the study, each dog wore its own test collar.

Half the controls wore an electronic collar that was turned off, and the other half wore a lemon spray collar that had previously been filled with spray but was empty and turned off.

### 2.7. *Barking and activity measurements*

The observers measured barking duration with a stop watch for the entire 30 min, at each recording session. Barking data were verified by review of the videotapes at a later time. Measurements included whining as well as barking.

An activity indicator was obtained by counting the number of times the left front foot of the dog crossed one of the grid lines painted on the floor. The left front foot was chosen arbitrarily for ease of counting, and as a consistent standard of measure. Grid lines, approximately 2.5 cm wide, had been painted on the floor of each run. Grid lines were oriented from left–right and front–back, resulting in 0.6 m × 0.6 m squares. The criterion for crossing a line was that the entire foot print crossed the line. The activity data were determined by the evaluators by reviewing the videotapes at a later time.

## 2.8. Cortisol and ACTH measurements

Blood samples were taken within 4 min of opening the door to the run at the end of the 30 min recording sessions. Blood was drawn from the cephalic vein into a 6 ml syringe. Each dog was returned through the guillotine door to the other side of its run immediately after blood sampling. The blood was divided into two EDTA vacutainer tubes (3 ml volume) and stored on ice for transport. Several hours later, the tubes were centrifuged (4000 rpm  $\times$  10 min) and aliquots of plasma were frozen at  $-28^{\circ}\text{C}$ . Previous studies indicated that samples could be kept on ice for at least 6 h with no change in cortisol (Steiss, unpublished data). The frozen samples were assayed within 4 days.

The exception to the protocol of drawing blood within 4 min of approaching the dog was during the initial screening (wk  $-1$ ), when some dogs were walked on leash from their run to the treatment room and were subjected to the stress of manual restraint for physical examination before drawing blood. Therefore, wk 0 values are considered more accurate indicators of baseline than wk  $-1$ .

Assays were performed by the Endocrinology Laboratory, College of Veterinary Medicine, Auburn University. Cortisol and ACTH concentrations were assayed in duplicate using commercial radio-immunoassay kits (Coat-A Count Cortisol, Diagnostic Products Corp., Los Angeles, CA; ACTH, Nichols Institute Diagnostics, San Clemente, CA). The methodology for the cortisol assay (Kemppainen et al., 1983) and ACTH assay (Gould et al., 2001) have been reported previously. The cortisol assay sensitivity is 5 nmol/l. The only steroids which have been shown to have significant ( $>3\%$ ) cross reaction are prednisolone and 11-deoxy-17-hydrocorticosterone (Kemppainen et al., 1983). The limit of detection of the ACTH assay is 1 pg/ml (Gould et al., 2001). The manufacturer states that related peptides such as alpha MSH and beta endorphin show no cross reaction.

## 2.9. Statistics

The experiment was evaluated as a repeated measures over time for the two treatments (electronic and lemon spray collars) and the control group. Data were analyzed by the general linear model (GLM) procedure of SAS (SAS Institute, 1990) with treatments and time as the main effects. Barking, plasma cortisol, ACTH and activity were measured repeatedly over time. Significantly different main effects (treatments and time) for various response measurements were further classified using the all pairs Tukey–Kramer honestly significant difference (HSD) test (Tukey, 1953). Within the main effect of time, the data were also analyzed for treatment effect by the GLM procedure of SAS (SAS Institute, 1990) and Scheffe's test for multiple comparisons.

## 3. Results

### 3.1. Barking

Starting the 2nd day wearing either an activated electronic or lemon spray collar (wk 1 d 2), barking time was reduced to a mean of 1.8 s or less during the 30 min recording period,  $P < 0.05$ . The values on wk 1 d 3 were not significantly different compared to controls probably due to the large standard deviation in the control group on that day. There was no significant difference between collar types. We did not notice any barking that could be attributed to interaction between the dogs (Table 1).

### 3.2. Number of collar corrections

The mean number of corrections during the 30 min recording sessions decreased to 0.0 for both collar types on the 3rd day wearing an activated collar (wk 1 d 3) (Table 2).

Table 1  
Summary of barking (measured in s, mean  $\pm$  S.E.M.)

Group <sup>a</sup>	Wk 0 d 3 <sup>b</sup> (D) acclimation to inactivated collar	Wk 1 d 1 (EJ) 1st day wearing active collar	Wk 1 d 2 (FJ) 2nd day wearing active collar	Wk1 d 3 (GJ) 3rd day wearing active collar	Wk 2 d 1 (HJ) 4th day wearing active collar	Wk 2 d 2 (IJ) 5th day wearing active collar	Wk 2 d 3 (J) 6th day wearing active collar
Control collar (A) ( <i>n</i> = 7)	48.1 $\pm$ 11.0 (100%) <sup>c</sup>	32.7 $\pm$ 9.8 (69%)	26.0 $\pm$ 7.9 (54%)	40.9 $\pm$ 23.3 (85%)	55.4 $\pm$ 18.4 (115%)	38.4 $\pm$ 16.1 (79%)	71.6 $\pm$ 27.4 (150%)
Electronic collar (BC) ( <i>n</i> = 6)	102.4 $\pm$ 18.1 (100%)	16.0 $\pm$ 5.6 (16%)	1.8 <sup>d</sup> $\pm$ 1.3 (2%)	1.0 $\pm$ 1.0 (1%)	1.3 <sup>d</sup> $\pm$ 1.0 (1%)	0.7 $\pm$ 0.7 (1%)	0.7 <sup>d</sup> $\pm$ 0.7 (0%)
Lemon spray collar (C) ( <i>n</i> = 8)	65.4 $\pm$ 20.4 (100%)	12.8 $\pm$ 4.3 (20%)	0.9 <sup>d</sup> $\pm$ 0.4 (1%)	0.2 $\pm$ 0.2 (0%)	0.0 <sup>d</sup> $\pm$ 0.0 (0%)	0.0 <sup>d</sup> $\pm$ 0.0 (0%)	0.4 <sup>d</sup> $\pm$ 0.2 (0%)

Barking was measured over 30 min. The bark stimulus was a strange dog walked in front of the run for 30 s on three separate occasions during the 30 min test period. Overall comparisons within columns are designated by letters A–C. Treatment groups bearing the same letter are not significantly different,  $P > 0.05$ . Overall comparisons within rows are designated by letters D–J. Times bearing the same letter are not significantly different,  $P > 0.05$ .

<sup>a</sup> Treatments were significantly different,  $P < 0.05$  (d.f. = 2;  $F = 16.71$ ).

<sup>b</sup> Time was significantly different,  $P < 0.05$  (d.f. = 6;  $F = 8.28$ ).

<sup>c</sup> The percentage in parentheses represents the mean on that day divided by the mean of the acclimation period (wk 0 d 3).

<sup>d</sup> Interaction between treatment and time was significantly different,  $P < 0.05$  (d.f. = 12;  $F = 2.81$ ). Significantly different from control at this time point,  $P < 0.05$ .

Table 2

Summary of number of corrections per dog from bark control collars during the first 3 days of wearing activated collars (mean  $\pm$  S.E.M.)

Group	Wk 1 d 1 1st day wearing active collar	Wk 1 d 2 2nd day wearing active collar	Wk 1 d 3 3rd day wearing active collar
Control collar ( $n = 7$ )	Not applicable	Not applicable	Not applicable
Electronic collar ( $n = 6$ )	4.0 $\pm$ 0.55	0.0 $\pm$ 0.05	0.0 $\pm$ 0.05
Lemon spray collar ( $n = 8$ )	2.0 $\pm$ 0.53	1.0 $\pm$ 0.21	0.0 $\pm$ 0.04

Observations were made over 30 min.

### 3.3. Activity

There was no significant difference between the lemon spray and electronic collar groups or electronic and control groups. The lemon spray group was significantly less active than the control group throughout the study,  $P < 0.05$ . The time periods were not significantly different,  $P > 0.05$  (Table 3).

### 3.4. Cortisol

During the initial screening prior to admission to the study, the mean cortisol level for all 21 dogs was 83.6 nmol/l (S.E.M., 13.4 nmol/l). Mean plasma cortisol values remained within the reference range for all groups throughout the study. On the 1st day wearing an activated bark collar, mean plasma cortisol increased to 169% of acclimation baseline levels for both collar types, but this was not statistically significant ( $P > 0.05$ ). There was no significant difference between the control, electronic collar and lemon spray collar groups,  $P > 0.05$ . There was a significant overall time effect ( $P < 0.05$ ) (Table 4).

Table 3

Summary of activity (crossing grid lines on floor, mean  $\pm$  S.E.M.) for dogs in bark collar study

Group <sup>a</sup>	Wk 0 d 3 acclimation to inactivated collar	Wk 1 d 1 1st day wearing active collar	Wk 1 d 3 3rd day wearing active collar	Wk 2 d 3 6th day wearing active collar
Control collar (AB) ( $n = 7$ )	399.7 $\pm$ 193.1 (100%) <sup>b</sup>	378.9 $\pm$ 108.4 (95%)	374.0 $\pm$ 110.0 (94%)	337.3 $\pm$ 119.8 (84%)
Electronic collar (BC) ( $n = 6$ )	283.3 $\pm$ 58.2 (100%)	217.0 $\pm$ 54.4 (77%)	236.7 $\pm$ 68.6 (84%)	285.5 $\pm$ 95.8 (101%)
Lemon spray collar (C) ( $n = 8$ )	262.2 $\pm$ 86.5 (100%)	193.6 $\pm$ 49.9 (74%)	166.1 $\pm$ 48.7 (63%)	155.5 $\pm$ 29.3 (60%)

Activity was measured over 30 min. Measurements were calculated by an observer counting the number of times the dog's left front foot crossed over a grid line during the 30 min test period. Grid lines (2 cm wide) were painted on the floor of each run resulting in a 2 ft.  $\times$  2 ft. square pattern. Overall comparisons within columns are designated by letters A–C. Treatments groups bearing the same letter are not significantly different,  $P > 0.05$ .

<sup>a</sup> Treatments were significantly different,  $P < 0.05$  (d.f. = 2;  $F = 3.84$ ). Time, and treatment  $\times$  time interactions were not significant,  $P > 0.05$ .

<sup>b</sup> The percentage in parentheses represents the mean on that day divided by the mean of the acclimation period (wk 0 d 3).

Table 4  
Summary of canine plasma cortisol concentrations (nmol/l) in bark collar study (mean  $\pm$  S.E.M.)

Group	Wk 0 d 3 <sup>a</sup> (AB) acclimation to inactivated collar	Wk 1 d 1 (A) 1st day wearing activated collar	Wk 1 d 3 (B) 3rd day wearing activated collar	Wk 2 d 3 (B) 6th day wearing activated collar
Control collar ( $n = 7$ )	33.4 $\pm$ 5.0 (100%) <sup>b</sup>	31.6 $\pm$ 7.4 (97%)	22.0 $\pm$ 2.3 (67%)	27.3 $\pm$ 4.5 (82%)
Electronic collar ( $n = 6$ )	61.3 $\pm$ 19.1 (100%)	103.3 $\pm$ 31.8 (169%)	35.0 $\pm$ 7.1 (57%)	39.3 $\pm$ 9.1 (64%)
Lemon spray collar ( $n = 8$ )	36.0 $\pm$ 4.7 (100%)	61.1 $\pm$ 14.9 (169%)	37.2 $\pm$ 9.0 (103%)	33.9 $\pm$ 5.0 (94%)

Reference range for canine baseline cortisol, Endocrine Diagnostic Service Laboratory, Auburn University: 10–160 nmol/l. Mean plasma cortisol concentration for all 21 dogs at screening prior to admission to the study was 83.6 nmol/l (S.E.M., 13.4 nmol/l). Overall comparisons within rows are designated by letters A and B. Times bearing the same letter are not significantly different,  $P > 0.05$ .

<sup>a</sup> Time was significantly different,  $P < 0.05$  (d.f. = 3;  $F = 7.36$ ). Treatments, and treatment  $\times$  time interactions were not significant,  $P > 0.05$ .

<sup>b</sup> The percentage in parentheses represents the mean on that day divided by the mean of the acclimation period (wk 0 d 3).

### 3.5. ACTH

Groups showed no significant difference in plasma ACTH levels,  $P > 0.05$ . The plasma ACTH values (pg/ml, mean  $\pm$  S.E.M.) for the control group were 14.3  $\pm$  1.8 (wk 0 d 3) and 15.0  $\pm$  1.7 (wk 1 d 1). The corresponding values for the electronic collar group were 20.2  $\pm$  5.6 (wk 0 d 3) and 28.2  $\pm$  9.8 (wk 1 d 1); values for the lemon spray collar group were 13.5  $\pm$  1.3 (wk 0 d 3) and 16.7  $\pm$  1.7 (wk 1 d 1). The reference range for resting ACTH (Endocrine Diagnostic Service Laboratory, Auburn University) is 10–80 pg/ml.

## 4. Discussion

In this study, barking was significantly decreased starting the 2nd day of wearing either an electronic or lemon spray collar, with no significant difference between the collar types. Evaluations of bark control collars have been reported previously. Investigators in Australia randomly surveyed people who had used a collar mounted electronic training device (Coleman and Murray, 2000). They reported that 97% of respondents were satisfied with the product, and 30% considered their dogs to be calmer after using the electronic collar. Juarbe-Diaz and Houpt (1996) conducted an in-home study comparing citronella spray collars and electronic shock collars for efficacy and user satisfaction after a 2 week trial. They found that citronella spray collars decreased barking 89% whereas electronic collars decreased barking 44%. There was no explanation for this difference between collar types. Most owners in that study expressed a preference for the citronella spray collar and perceived it as more humane. Moffat et al. (2003) used citronella collars on dogs in a veterinary hospital and a kennel; out of 62 dogs, barking ceased in 40 and decreased in 17 dogs with the collar. In a colony of Beagles, citronella collars reduced barking noise from a mean of 106 to 70 dB (Moffat et al., 2003). The results of our study cannot be compared to a more recent report on electronic (“shock”) collars because that study examined electronic collars during guard dog training, not as bark deterrents (Schilder and van der Borg, 2004).

Elevation in blood cortisol concentration has long been used as an indicator of stress in mammals (Beerda et al., 1998). Rushen (1991) has cautioned about attempts to make claims concerning animal welfare based on physiological data, given the complexity of the pituitary–adrenocortical axis. Corticosterone levels have been reported to fluctuate systematically in



response to changes in emotional state (Hennessy et al., 1979). In the present study, on the 1st day of wearing an activated bark collar, mean plasma cortisol increased to 169% of acclimation baseline levels for both collar types, but this was not statistically significant ( $P > 0.05$ ). Throughout this study, the average cortisol level in dogs wearing either bark collar remained within normal range. In a study which examined dogs in an animal shelter, Hennessy et al. (2002) reported plasma cortisol concentrations ranging from approximately 10 to 20 ng/ml (equivalent to 27.5–55 nmol/L), comparable to the values in this study.

Concern has been expressed that the stress of blood sampling limits the usefulness of cortisol measurement (Wielebnowski, 2003). However, there is an interval after a stressful stimulus before cortisol rises. Hennessy et al. (1979) documented increases in corticosterone and ACTH in rats after 10 min, but not 2.5 min, of exposure to a stressful stimulus. In later studies, Hennessy collected samples from dogs within 4 min after removing the dog from its cage or the test area (Hennessy et al., 1997, 1998, 2002). In this study, in order to avoid a rise in plasma cortisol which could be associated with the stress of restraint and venipuncture, blood samples on wks 0–2 were collected within 4 min of the operator opening the door to the dog's run.

Mean plasma cortisol levels in this study were higher on wk –1 (initial physical examination) than wk 0 (acclimation). On wk –1, some dogs were moved from their run to a treatment room and were restrained for physical examination before drawing blood. This change in handling the dogs was not due to any difference in the dogs. Transfer to a different room and/or handling and restraint of the dogs likely account for the higher mean cortisol levels. Therefore, wk 0 values were considered more accurate indicators of resting baseline than wk –1. Hennessy et al. (1998) found that handling a dog and taking a venous blood sample raised plasma cortisol. Some investigators have expressed concern that dogs housed long term in shelters or laboratories are unable to increase their cortisol levels due to chronic stress (Hennessy et al., 2002). The findings on wk –1 show that the dogs were able to elevate their blood cortisol.

Several factors in the experimental design were controlled in order to avoid extraneous causes of elevated plasma cortisol concentrations. Dogs have been shown to have higher plasma cortisol concentrations during their first 3 days of confinement in an animal shelter compared to dogs housed in the shelter for longer periods (Hennessy et al., 1997). In our study, dogs were housed a minimum of 7 days in the kennel in which the study was conducted. Also, 10 empty runs separated each pair of dogs, thereby avoiding the possibility of dogs in adjacent runs being exposed to lemon spray, or being influenced by the behavior of the dog in a neighboring run. To minimize diurnal variations in cortisol, dogs were studied within a time frame of 11:00–16:00 h. Blood sampling was scheduled more than 2 days apart to allow for recovery of the adrenal axis between sampling. The dogs were screened by age (approximately 1–4 years of age), breed and behavior. Breeds that might be considered to have a higher threshold for pain, such as Pit Bulls and Jack Russell terriers, were not studied. Dogs that demonstrated aggressive tendencies on the behavioral screening were not used.

Measurements of plasma cortisol and behavior may be preferable to measurement of either one alone. Our study reports one behavioral measure of activity, namely, the number of times dogs moved across sectors (grid lines) marked on the floor of each run (Beerda et al., 1998). The citronella group was less active than the other groups at all time points in this study. That result is likely due to the small number of animals and individual variation.

Some studies have reported barking as “episodes per minute” (Moffat et al., 2003). However, distinguishing a single bark from episodes of multiple short barks can be difficult and could yield unacceptable inter-rater reliability. Therefore, in this study, vocalization time, measured with a stop watch, has been reported instead.

Some studies indicate that dogs may habituate to the citronella spray collar (Moffat et al., 2003; Wells, 2001) although habituation has not always been found (Beaudet, 2001). No evidence of habituation was seen in this study with dogs wearing bark collars intermittently over a 2 week period. Juarbe-Diaz and Houpt (1996) commented that dogs quickly learned not to bark when they wore citronella spray collars and to bark when the collar was not worn. The possibility of habituation to the collars over a longer time period than the present study would be a relevant issue to investigate in future studies.

## 5. Conclusions

Dogs wore a lemon spray bark collar, electronic bark collar or an inactivated bark collar ( $n = 8$  per group) for 30 min/day, 3 days per week for 2 weeks, after an acclimation period the previous week. Both electronic and lemon spray bark collars significantly reduced barking, with no significant difference between the two types of collars. The amount of barking was significantly reduced starting the 2nd day that the dogs wore a bark collar. The mean number of corrections decreased to 0 by the 3rd day for both collar types. Mean plasma cortisol levels were within the reference range for all groups throughout the study. Plasma cortisol, as well as ACTH levels, did not differ among groups ( $P > 0.05$ ). Activity did not change significantly over time ( $P > 0.05$ ). The findings of this study may contribute additional information in the animal welfare debate regarding whether the use of bark control collars is humane. In the present study, with dogs wearing bark control collars intermittently over a 2 week period, the collars effectively deterred barking without statistically significant elevations in plasma cortisol, compared to controls, at any of the time points measured.

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