

A longitudinal evaluation of urinary cortisol in kennelled dogs, *Canis familiaris*

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Abstract

Urinary cortisol levels (based on cortisol : creatinine ratios) were evaluated in a randomly selected sample of shelter dogs kennelled over a 31-day period. Urine was collected on days 2, 5, 10, 17, 24 and 31 (with day 1 referring to the day of admittance to the shelter). Cortisol levels peaked on day 17 and steadily declined thereafter, although a high degree of individual variation was found, with cortisol levels peaking sooner in some dogs. Cortisol levels in kennelled dogs were significantly higher on all days except d 31 than the baseline measures taken from 20 dogs in their home environments. There were no differences between cortisol levels in male and female dogs on each day of sampling and there was no significant linear correlation between age and cortisol levels. The results are discussed in relation to stress management and the welfare of kennelled dogs.

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In recent years, many rescue shelters have incorporated the ‘no-kill’ philosophy into their dog re-homing policies [1]. Consequently, rather than being euthanased, many healthy, adoptable dogs spend prolonged periods of time in kennels prior to being re-homed. Although the mean duration of confinement at many shelters is less than one month (e.g. RSPCA, personal communication N. LaHive), still some individual dogs are confined for many months or years (e.g. The Dogs’ Trust, personal communication C. Laurence). Consequently, the welfare of dogs that are kennelled for extended periods is of interest and significance to humane organisations.

The detection of poor welfare can be established, in part, through the assessment of stress responses [2,20]. Previous studies involving behavioural [27] and physiological [10,12] measures have suggested that dogs experience anxiety and fear

almost immediately upon admittance into rescue shelters. Factors including separation of the dog from its owner, handling by unfamiliar kennel staff, novel surroundings and a change in husbandry routine are likely to contribute to the behavioural and physiological indicators of stress that are observed in the short-term [3,11,12].

Behavioural correlates of stress have also been observed in dogs kennelled for longer periods [4,6,15]. For example, kennelled dogs were observed to display behaviours associated with frustration and depression eight weeks following admittance to a shelter [28]. At this stage, stress responses are likely to occur because dogs are deprived of adequate opportunities to engage in species-specific behaviours such as social interactions with other dogs and people [14].

Cortisol levels determined from urine, saliva, blood and faeces have been used previously as measures of the physiological stress response in kennelled dogs [2,5,9,17,25]. Hennessy et al. [10] reported that plasma cortisol levels were initially elevated during the first 3 days upon arrival to the rescue shelter but then declined thereafter. Although cortisol

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levels then decreased 8 weeks following this initial elevation, ACTH levels did not significantly decrease once cortisol levels declined, suggesting that dogs remain stressed for longer than cortisol levels alone would indicate [13].

There are two important points to consider before these results are generalised across the entire shelter dog population. Firstly, fearful and aggressive dogs were omitted from Hennessy's study [10]. Since fearful behaviour is associated with elevated cortisol levels [16], it is feasible that fearful dogs that take longer than non-fearful dogs to habituate to the kennel environment would also display elevated cortisol levels for a longer period. Subsequently, the peak in urinary cortisol in a sample that excludes fearful dogs may be a conservative estimate compared with the entire shelter dog population. Secondly, various studies have suggested that the cortisol sampling technique itself can cause cortisol levels to increase. For example, Hennessy et al. [11] observed an increase in cortisol levels 20 min after a vein-puncture procedure, which was attributed to the unaccustomed shelter dogs finding the blood sampling technique to be stressful. Similarly, Kobelt et al. [18] reported increases in plasma cortisol 3-min after blood collection, and increases salivary cortisol 4 min after saliva collection in dogs. Since dogs can learn to anticipate the collection of blood and saliva, these cortisol changes may occur prior to sample collection.

The present study determines urinary cortisol levels in a random sample of shelter dogs kennelled over a 31-day period, with the aim of evaluating the physiological stress response using a non-invasive technique. The technique of collecting naturally voided urine for the analysis of urinary cortisol is advocated as less stressful and more humane [2], although no studies have yet determined changes in urinary cortisol levels in shelter dogs from the time they are admitted. We also assess sources of individual variability in this stress response, indicating how susceptibility to stressors may differ by gender and age. This information will provide humane organisations with a clearer understanding regarding the welfare implications for dogs during their first month in kennels.

1. General method

1.1. Subjects

Shelter sample: Subjects were selected using a random sampling. The sample consisted of 81 dogs that were all relinquished by their owners to Battersea Dogs Home at Old Windsor, UK. The dogs included 57 males (34 neutered, 23 entire) and 24 females (12 neutered, 10 entire), ranging in age from 7 months to 11 years (median age 3.5 years). Fifty three of the dogs were eventually re-homed with new owners, 10 were transferred to other rescue shelters to receive rehabilitation, 2 to specialist breed rescue shelters, 1 to detection dog training, and 15 were euthanased for health or behaviour reasons.

Re-homed sample: In order to gain baseline cortisol levels for comparison with levels measured in kennelled dogs, urine was collected from 20 re-homed dogs at least 6 months following adoption from the shelter. The re-homed sample included

12 males (7 neutered, 5 entire) and 8 females (6 neutered, 2 entire) ranging in age from 6 months to 12.5 years (median age 18 months).

1.2. Husbandry and housing

Intake kennels: Dogs were given a health check by a veterinary nurse within 20 min of entry into the shelter. This helped to ensure that dogs included in the study were physically healthy and so unlikely to be suffering from Addison's Disease, where cortisol is under-produced, and Cushing's Syndrome, characterized by the over-production of cortisol [21]. Dogs were then housed singly in an intake kennel block (10.7 m² split into an indoor 2.8 m² and outdoor area 7.9 m²) and were not on view to the public. During their stay in the intake kennels, dogs were exercised in a grass-covered compound for approximately 20 min per day (weather permitting). Members of kennel staff spent an additional 15 min per day interacting with the dogs.

Dogs were subjected to a behavioural assessment, and those identified as healthy and fit for adoption were transferred within 2 and 25 days (mean \pm SEM: 8.4 \pm 0.6) to the rehoming kennels. The rate at which dogs were transferred to the re-homing kennel was dependent on the kennel space available and was at the discretion of the staff.

Re-homing kennels: Dogs were again housed singly in kennels (total area 9.5 m², split into an indoor 4.6 m² and outdoor area 4.9 m²). Dogs had auditory and visual contact with other dogs, but were not able to have physical contact with dog in adjoining kennels. Here, dogs were available for viewing by the public for 5 to 6 h a day, 6 days a week. Dogs received 30 min walks 2–3 times per week, and were also exercised daily for 20 min in a fenced, grass-covered enclosure.

All dogs were fed twice daily: once before 10:00 h and again before 16:00 h. Water was available ad libitum. Dogs were confined to their indoor kennels between 17:00 and 08.00 h. A security night light was left on in kennels.

1.3. Urine collection and hormone determination

1.3.1. Urine collection

Urine samples were collected from kennelled dogs on d 2, 5 and 10 following admittance into the shelter and every 7 days for a maximum of 31 days, or sooner if the dog was removed from the shelter. Urine was collected each morning at approximately 08.00 h. Dogs were sampled as they progressed through kennels, however some dogs did not provide samples on specific days, either because they had already urinated in their kennel, or the experimenter was unable to capture the urine. The collection procedure consisted of releasing the dog from the indoor kennel where it had been confined in overnight and walking it on lead to a grass paddock until the animal urinated. The urine sample was captured directly by the experimenter using a "Uripet" (National Veterinary Supplies) which is a plastic vessel that enable the urine to be immediately transferred to a vial (5 ml Bijous, Fisher Scientific). Samples were chilled in an ice-box within 5 min of collection and frozen

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