

## Restless nights? Nocturnal activity as a useful indicator of adaptability of shelter housed dogs

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### ABSTRACT

Shelter dogs face the challenge of adapting to a kennel environment. Individual differences in adaptation are known to exist. Resting patterns might be indicative of adaptability to such a novel environment, but need to be evaluated for its usefulness, like every potentially physiological and behavioural parameter. Here, we evaluated nocturnal activity patterns of dogs as indicators of adaptability to novel environments. We measured nocturnal activity (3-axial accelerometer, the Actical®) and two physiological stress parameters, i.e. urinary cortisol/creatinine ratio (UCCR) and body weight in 29 dogs relinquished to a shelter (SD group) in the first two days after intake at the shelter (n = 29), after a 12-day habituation period in the shelter (n = 28) and >6 weeks post-adoption (n = 17). A control group of 29 pet dogs kept at home (CPD group), matching the SD group characteristics, was also assessed for its nocturnal activity and UCCR. Linear mixed model analysis, t-tests and Friedman tests were used to analyse the data.

The main findings are: 1) the SD group exhibited higher nocturnal activity (total activity counts, activity duration and number of rest bouts) the first two nights after intake than on night 12, with decreasing inter-individual variances. Compared to the CPD group they showed higher nocturnal activity on night 1 (all  $p < 0.001$ ) and night 12 (all  $p \leq 0.001$ ) except for total activity counts on night 12. We found no 'first-night effect', where sleep is disturbed during the first night; nocturnal activity in the shelter did not significantly differ between nights 1 and 2 in the shelter. 2) In line with literature findings, SD group UCCRs were higher shortly after intake than after a 12-day habituation period and after adoption, and higher than in the CPD group. 3) An interaction was found between weight class and both nocturnal activity and UCCR levels: in their first days in the shelter, smaller dogs showing higher levels than larger dogs. 4) Dogs in the SD group lost, on average, 5% of their body weight between intake and the two-week habituation period.

In conclusion, nocturnal activity, as measured by an accelerometer, may be a valid parameter to monitor adaptability of dogs to a kennel environment. Monitoring nocturnal activity in this way can be a useful and cost-effective additional indicator for assessing dog welfare.

### 1. Introduction

Each year, animal shelters provide a temporary home to many dogs: approximately 20,000 in the Netherlands and 129,000 in the UK (Clark et al., 2012; Heijst et al., 2015). Being placed in a shelter abruptly changes a dog's environment and routine. Its welfare can be seriously compromised by stressors such as high noise levels and disrupted routines (Tuber et al., 1999; Coppola et al., 2006). Adequate adaptation to the demands of the prevailing environmental circumstances is essential

for an animal's welfare. Adaptation is mediated through stress responses (Koolhaas et al., 2006), reflected by physiological measures like urinary cortisol/creatinine ratio (UCCR, e.g. Stephen and Ledger, 2006), body weight loss (Rooney et al., 2009), and by behaviour (Beerda et al., 2000; Stephen and Ledger, 2005). Stress responses in shelter dogs may last several days (Hennessy et al., 1997) or even weeks (Stephen and Ledger, 2006), with high individual variability. Therefore, both physiological and behavioural measures need to be evaluated over time (Juster et al., 2012) to enable insight into individual reactivity and coping capacity (i.

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e. adaptability), which is necessary for reliable welfare assessment (Ohl and Van der Staay, 2012).

Changes in sleeping or resting activity have not yet been investigated for their role in monitoring the welfare of dogs in shelters over time. Sleep can only be reliably determined by measuring brain activity. In humans, the quality and amount of sleep is described as a valuable parameter for assessing coping success with stressful stimuli. This is known to be affected by daytime events (Vandekerckhove and Cluydts, 2010). In dogs, sleep physiology is affected by preceding positive and negative emotional stimuli, with, after negative stimuli, a redistribution of the duration of sleep stages (Kis et al., 2017). Recovery to normal resting patterns and normal frequency or duration of sleep episodes have, in other species, been suggested as important indicators of adaptation to environmental changes (horses, cattle, sheep, pigs: Ruck-erbusch, 1975; calves: Hänninen, 2007). Monitoring resting patterns may therefore provide a useful additional tool for assessing adaptability of dogs to novel housing conditions such as in shelters.

Dogs are polyphasic sleepers with a diurnal resting pattern (Zanghi et al., 2012; Woods et al., 2020) and an average sleep cycle length in a range of situations of about 16–20 min (Adams and Johnson, 1993). In an observational study aimed to describe in-shelter night-time behaviour, shelter dogs awoke and moved into a different lying position every 48–50 min (Haupt et al., 2019). Sleeping patterns in dogs have previously been measured using polysomnography with implanted or surface attached scalp electrodes, where resting behaviour has been specified using non-invasive accelerometry (Zanghi et al., 2013) and using behavioural observations (e.g. Owczarczak-Garstecka and Burman, 2016). As measuring brain activity to identify sleep, the ‘gold standard’, is not yet possible in freely moving shelter dogs, accelerometry and behavioural observations are mainly used to address resting patterns. Accelerometry is, however, unable to detect ‘quiet wakefulness’, e.g. lying inactive but awake, a measure of restlessness suggested for other animals (Paquet et al., 2007; Meagher et al., 2013). To identify resting behaviour, accelerometry is thus combined with video observations.

In shelter dogs, higher proportions of resting during the day have been found to indicate improved welfare, as it was associated with less repetitive behaviour, more positive judgements in a judgement bias task and increased time spent ‘relaxed’ as coded across days by shelter staff (Owczarczak-Garstecka and Burman, 2016). Shelter dogs have also been found to be more active from ~0:00–18:00 h than pet dogs kept at homes (Hoffman et al., 2019) and to have longer bouts of resting during a short-term period of 1 or 2 nights of fostering stay at a home than they had in the shelter the nights before and after the fostering stay (Gunter et al., 2019). Changes in resting patterns in dogs have not yet been studied during the early habituation period in the first weeks after the dogs enter the shelter, which is when to evaluate how a dog adapts to new environments. To evaluate resting patterns as an indicator of adaptability of dogs to a shelter environment, we investigated 1) whether resting patterns were disturbed during this first habituation period, and if so, 2) whether disturbed resting patterns showed recovery during this first period, and 3) how resting patterns related to physiological measures of the stress response for better validation of activity as indicator of adaptation to the shelter environment. As shelter routines differ between shelters, we did this during the night, when dogs are least disturbed by human interference and shelter dogs tend to rest most (Hoffman et al., 2019). We also compared resting patterns and physiological measures of the shelter dogs to that of a control group of pet dogs in their own homes, to evaluate whether the resting patterns were disturbed in the shelter dogs.

We hypothesized that nocturnal activity of shelter dogs as measured by an accelerometer would be disturbed compared to the pet dogs, but would recover after two weeks of habituation showing adaptation in most of the dogs. We also expected that this activity would be related to the dogs’ UCCRs, as a physiological measure of the stress response. Furthermore, we expected the UCCRs to be higher in shelter dogs than in a control group of pet dogs.

## 2. Material and methods

### 2.1. Subjects & housing

#### 2.1.1. Shelter dogs (SD group)

Data were collected between May and September 2016 in the largest animal shelter of the Netherlands (Animal Shelter DOA in Amsterdam). Twenty-nine dogs were recruited for this study, for demographics per dog see supplementary Table 1. As most shelter dogs were mixed breeds or ‘look-a-likes’ instead of purebreds, dogs were breed labelled by visual appearance as judged by an experienced shelter employee, based on dominant morphological breed characteristics described by the Fédération Cynologique Internationale (FCI). Breed labelling in shelter dogs is highly unreliable (e.g. Voith et al., 2009; Gunter et al., 2018). However, we chose to assign a breed label to the dogs with the aim to match a control group of pet dogs based on body conformation and especially body weight, as this might influence accelerometer output (Brown et al., 2010b; Cheung et al., 2014), and within breeds body weights are relatively uniform. Dogs were assigned to three age classes (Lit et al., 2010): young adult (1–4 years), adult (5–7 years) and senior (8–13 years). The age of the dogs was either provided by previous owners or the dog’s official passport, or estimated by the shelter veterinarian during intake assessment by evaluating the teeth and overall condition.

Dogs were excluded for study participation if they were affected by a physical health condition, as determined by a veterinarian during the shelter intake assessment, for example joint problems that could cause pain during movement. Dogs were also excluded if they showed high levels of anxiety- or aggression-related behaviour as assessed by the caretakers during shelter-intake, as those might negatively influence rehabilitation chances or pose a risk for the researchers. Furthermore, dogs were excluded if they were younger than 1 or older than 13 years of age, as development of the HPA axis and circulating cortisol levels may be influenced by age (Palazzolo and Quadri, 1987), or if they were housed in pairs, as social housing may influence activity and cortisol levels (e.g. Dreschel and Granger, 2005).

By Dutch law, a quarantine period of seven days is obligatory when the health and vaccination status of the relinquished or stray dog is unknown. Stray dogs must be kept for 14 days to allow the owners to collect their dog.

Dogs were housed individually in kennels with a glass-fronted indoor and bar-fronted outdoor enclosure of ~5 m<sup>2</sup> each, separated by a hatch for free access into both enclosures. Kennels were only accessible to staff and volunteers for caring for the dogs as described below, and for the researchers of this study to collect data. They were not accessible to members of the public, who were not able to walk near the kennels and view the dogs, conforming with shelter procedures. Kennels were cleaned in the morning between 8:30–12:30 every day. Dogs were fed twice daily, at ~8:30 h and ~16:00 h, with special veterinary diets when needed for dogs e.g. with potential food allergies. Additional food enrichment (e.g. food puzzles, bones) was provided daily and water was available *ad libitum*. Dogs could roam freely on a play field for about one hour per day, preferably with other dogs. Fully vaccinated dogs were walked for ~30–45 min every day or every other day.

Data collection took place seven days a week, as variation in shelter routines including weekends was negligible in this shelter. For the data collection, dogs were handled as minimally as possible by two women with prior dog-handling experience (first author and one research assistant), as human contact can greatly influence stress levels of shelter dogs (e.g. as little as 15 min, McGowan et al., 2018). Dogs otherwise followed the normal routine of the shelter, which includes handling by staff members and volunteers during the moments described above.

After adoption, which occurred between two weeks and 15 months after admission to the shelter, new owners were contacted by the researchers by telephone. New owners of 17 of the 29 shelter dogs (= 60 %) agreed to participate. Based on detailed instructions, owners

collected a urine sample of the dog >6 weeks after adoption (median: 10.5 weeks, range: 6.5–28 weeks). This post-shelter time frame of >6 weeks was chosen as the behaviour of the dogs can change during the first weeks after adoption (e.g. ‘honeymoon period’, [Stephen and Ledger, 2007](#)) and it might take weeks for their personality to come out.

2.1.2. Matched control pet dogs (CPD group)

A control group of 29 healthy pet dogs (CPD group) kept at their own homes were also recruited to compare nocturnal activity and UCCR with the SD group. Dogs in the CPD group were recruited via the website of the Faculty of Veterinary Medicine (Utrecht University), social media and by contacting dog professionals. Dogs were excluded if they were housed with another dog in their homes.

Dogs in the CPD group were matched with characteristics of individuals of the SD group based on the criteria breed, sex, neuter status and age class (see supplementary Table 1), for the following reasons: body weight and age may influence accelerometer measures ([Brown et al., 2010b](#); [Siwak et al., 2002](#); [Cheung et al., 2014](#)) and activity and sleeping patterns of dogs ([Takeuchi and Harada, 2002](#); [Zanghi et al., 2012](#)). Further, neutered males appear to show higher UCCR responses to kennelling than entire males and females ([Part et al., 2014](#)) and neutering may result in changed activity levels ([Garde et al., 2016](#)).

Because daily owner routines affect the activity of dogs (e.g. [Siwak et al., 2002](#); [Dow et al., 2009](#)), owners were instructed to follow their normal routine with their dog during the measurement period and the majority of activity measurements in the CPD group were performed during the workweek, unless owners had no ordinary week-weekend schedules, i.e. had to work during weekends.

2.2. Data collection

2.2.1. Timeline (moments of measurements)

Data were collected from the SD group during three periods (for moments see [Fig. 1](#)):

- I) On the first two days and nights in the shelter (day 1 and 2 after intake): accelerometer data, urine samples, video recordings and body weight;
- II) After a habituation period in the shelter (day 12 after intake): same data collection as above;
- III) Post-adoption (>6 weeks after adoption): urine sample.

A habituation period of 12 days was chosen as dogs were not moved to other kennels during this period and therefore adaptation to the shelter environment was least influenced by changes in the environment. On the day of intake at the shelter (day 0), no data was collected, as most dogs arrived in the afternoon. The first moment of data collection occurred at 0:00 h during the first full day in the shelter.

For comparison with the SD group, the following data were collected for the CPD group:

- I) Urine sample 1 (day 1 chosen by owners and researchers);
- II) Urine sample 2 (day 12) and accelerometer data for two consecutive nights.

2.2.2. Accelerometry

Accelerometry was used to measure the activity of the dogs during the night. An accelerometer is a device with sensors that measure acceleration and can therefore indirectly measure both activity and inactivity (e.g. [John and Freedson, 2012](#); [Clarke and Fraser, 2016](#)). In dogs, accelerometry-determined sleep-wake bouts correlate well with polysomnograph hypnograms, the gold standard for measuring sleep macrostructure ([John et al., 2000](#)).

In this study, the 3-axial Actical® (Philips Respironics Actigraphy, Mini Mitter Division, Bend, OR) was used, as this accelerometer has frequently been used in many other scientific dog studies (e.g. [Hansen et al., 2007](#); [Brown et al., 2010a, b](#); [Michel and Brown, 2011](#); [Cheung et al., 2014](#); [Woods et al., 2020](#)). The raw output of this accelerometer consists of activity counts per defined time period (epoch). For a detailed description of the Actical® mechanisms see [Hansen et al., 2007](#). The accelerometer was protected in armoured metal Actical® cases of 29 × 37 × 11 mm and attached with duct tape to the dog’s soft regular harness. A ventral location for the accelerometer (i.e. the chest) was chosen as this appears the most convenient and has been used in previous studies ([Hansen et al., 2007](#); [Preston et al., 2012](#)). To allow for habituation, the harness and accelerometer were worn both during the day and the measurement night. Owners of the dogs in the CPD group were asked to log the times that the household was asleep on a form provided by the researchers (see supplementary form 1) to identify the quiet night-time in the house.

For each dog, the Actical® was set for a 15 s epoch time and sampling rate of 32 Hz. Actical® software (Philips Electronics N.V. version 3.10) was used to read out the data as .csv files. Using these files in Microsoft Excel, activity measures were calculated for the night period: 0:00–4:00 h for the SD group and 1–5 h after owner sleep onset for the CPD group. The timeframe of 0:00–4:00 h was chosen for the SD group as this is a natural dark period during all seasons and the urban environment around the shelter was relatively quiet during this time slot. For the CPD group, we assumed 1–5 h after bedtime of their owners would be the quietest time as activity of pet dogs is largely controlled by the owners ([Randler et al., 2018](#)). In addition to evaluating the cumulative number of activity counts within this time frame, we registered every epoch as inactive (0 counts/15 s epoch) or active (>0 counts/15 s epoch). The calculated activity measures contained information on: 1) total activity (*total activity counts*, which is the summed counts over the 4-h recording time), 2) total length of activity (*activity duration* in minutes) and 3) how often the dogs were active or at rest (*number of rest bouts*). In addition, the longest bout of uninterrupted rest (*maximum duration of rest bouts* in minutes) was calculated.

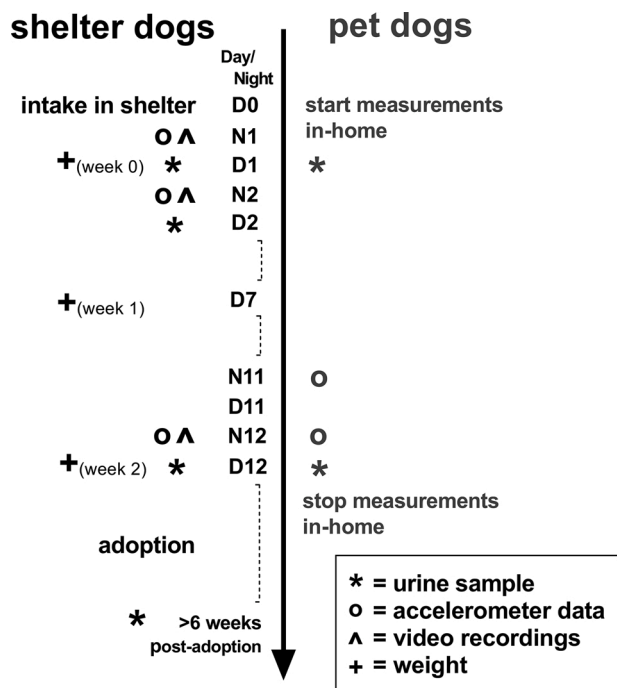


Fig. 1. Timeline with moments of measurements for shelter dogs (SD group) and pet dogs (CPD group). N = night, D = day. Symbols in the figure represent different types of data sampling: \* = urine sample, o = accelerometer data, ^ = video recordings, + = weight.

### 2.2.3. Urinary cortisol/creatinine ratio (UCCR)

UCCR levels were evaluated to determine arousal levels during the night preceding urine collection, as a physiological measure of the stress response (e.g. [Stephen and Ledger, 2006](#)). Morning urine was collected because while urine reflects cortisol levels up to 24 h retrospectively, cortisol levels may fluctuate over the day ([Beerd et al., 1999](#); [Schatz and Palme, 2001](#); [Rooney et al., 2007](#)).

Shelter dogs were taken from their kennel by a researcher or caretaker to the nearest greens for urine collection between 8:23 and 11:20 h with a median of ~ 9:10 h on all days. Naturally voided urine samples were collected by a ladle, which was rinsed with water and dried with paper cloth every time before and after use. The urine was transferred from the ladle to a vial (polypropylene tube, 5 mL, 75 × 13 mm, Sarstedt AG & Co) with a disposable pipette. If dogs were hesitant to naturally void urine and had voided urine in their kennels before urine collection, urine was collected off the kennel floor with the pipette if the urine was not visually contaminated by other faeces or water. These urine samples were most likely voided in the hours before 8:00 h. In a pilot study, urine samples collected from a kennel floor 2, 4 and 6 h after disposal showed no significant difference in UCCRs compared to the same direct ladle-collected urine sample (from two dogs), therefore floor samples were included for analysis. In total, 8/74 (= 11 %) analysed in-shelter urine samples were collected from the kennel floor. All SD group urine samples were frozen by -20 °C ([Hiby et al., 2006](#); [Rooney et al., 2007](#)) within 45 min (median = 10 min), and transferred to -80 °C within two weeks, until analysis.

For dogs in the CPD group and post-adoption in the SD group, owners were given written instructions and an instruction video in which the collection of morning urine with a ladle or a clean plastic/glass container was explained. Owners in the CPD group collected the morning urine of their dogs between 6:30 h and 12:30 h (median = 8:01 h). In the SD group, owners collected the >6 weeks post-adoption morning urine between 7:00 h and 11:00 h (median = 8:30 h). Owners preserved the urine in their own freezer (-10° to -20 °C) until the experimenters collected the urine within 2 weeks of sampling and transferred the vials to -80 °C storage, until analysis.

At the veterinary diagnostic laboratory of the Faculty of Veterinary Medicine at Utrecht University, the Netherlands, all samples were analysed for cortisol with a Radio-Immuno-Assay (RIA: [Rijnberk et al., 1988](#)) and for creatinine using addition of picric acid and spectrophotometry with Jaffé calculation. Samples were not extracted prior to analysis. The UCCR was calculated as: cortisol (nmol/l):creatinine (µmol/l) x1000 = ratio x10<sup>-6</sup>.

### 2.2.4. Body weight

We evaluated body weight changes in the shelter dogs, as body weight loss has been suggested to be sign of compromised welfare in dogs as a result of high stress levels (e.g. [Rooney et al., 2009](#)). All shelter dogs were weighed on the same scale in the shelter (AllScales® Europe) by the veterinarian or the researchers on days 1, 7 and 12 (week 0, 1 and 2). Dogs were assigned to five weight classes (body sizes): <10 kg, 10–20 kg, >20–30 kg, >30–40 kg and >40 kg ([Hennessy et al., 1997](#); [Brown et al., 2010b](#)).

### 2.2.5. Behavioural observations

The resting and activity behaviour of the dogs in the SD group was observed during the night from 0:00–4:00 h, in addition to accelerometer recordings.

The dogs were monitored using a video system with infrared night vision camera's (PRO 2-bullet camera system 2B03 P, BASCOM, Nieuwegein, The Netherlands, with 4 cameras) positioned permanently on camera stands in front of both the inside and the outside kennel. For technical reasons, only 11 dogs ended up with video material for all three nights (night 1, 2 and 12, see supplementary Table 1 for information on these dogs). Videos were randomized in Excel to allow blind scoring for the night in the shelter. Night videos were observed by two

observers using The Observer XT 12 (Noldus Information Technology), with inter-observer Cohen's Kappa reliability of 0.76 – 0.98 (substantial agreement, [Landis & Koch, 1977](#)) for four videos with different dogs and intra-observer Cohen's Kappa reliability of 0.99 (almost perfect agreement).

The ethogram ([Table 1](#)) was composed of behavioural patterns reflecting activity and resting behaviour and were based on the literature of dog activity studies ([Schipper et al., 2008](#); [Part et al., 2014](#); [Owczarczak-Garstecka and Burman, 2016](#)). Practical applicability of the ethogram for the observers in this study was evaluated during pilot observations, i.e. to identify behaviours that were visible on the in-kennel videos and could reliably be scored by the observers.

### 2.3. Data analysis and statistics

Data were stored and cleaned in Microsoft Excel® (Microsoft Corporation). Statistical software program RStudio (version 1.0.136 – ©RStudio, Inc.) was used to perform linear mixed model analysis with the package 'nlme' ([Pinheiro et al., 2020](#)), Friedman tests with the package 'rstatix' and Spearman correlation with the package 'Hmisc'. Graphs were created in Graphpad Prism (version 8.3.0 – ©GraphPad Software, LLC).

Relative changes in body weight were calculated for week 1 (day 7) and week 2 (day 12) based on the weight in week 0 (day 1) = 100 %.

A Spearman rank correlation for non-parametric variables was calculated for time of urine sample collection and UCCR levels for the in-shelter measurements.

Outcome variables were evaluated for normality by performing Shapiro-Wilk tests and visual inspection of boxplots and quantile-quantile plots of the data. The variables UCCR levels, nocturnal total activity counts, activity duration, number of rest bouts and maximum duration of rest bouts were all right-skewed and therefore natural log-transformed before t-tests and inclusion in mixed models and back transformed for interpretation. Back transformed (exp) log model values resulted in ratios, with a ratio <1 meaning a lower value and >1 a higher value than the reference mean.

Five linear mixed effects models were fit, one for each outcome variable: body weight proportional change, UCCR and three nocturnal activity measures: *total activity counts*, *activity duration* and *number of rest*

**Table 1**  
Ethogram of in-shelter activity and resting behaviour. Modified from mentioned references.

Behaviour	Description	References	Scoring
Recumbent head down	The dog's abdomen is touching the ground with its dorsal, caudal or lateral side whilst legs are extended forwards, curled close to the body or laid to one side. Eyes may be open, closed or not visible.	<a href="#">Owczarczak-Garstecka and Burman (2016)</a>	Duration
Recumbent head up	As above, with its head up, eyes can be open or closed.		Duration
Stationary	Sit: hindquarters in contact with the ground and front legs extended; or stand: four feet in contact with the ground and legs fully, or almost fully, extended.	<a href="#">Part et al. (2014)</a> ; <a href="#">Schipper et al. (2008)</a>	Duration
Movement	Dog moves around the enclosure (e.g. walking, running, mobile exploration). Ambulates at any speed.		Number

bouts. Fixed effects were added to each model: 'day' (UCCR)/'night' (activity measures)/'week' (body weight), 'kennel history' (no/unknown/yes), 'body weight class' (<10 kg, 10–20 kg, >20–30 kg, >30–40 kg, >40 kg), 'age class' (1–4 yrs, 5–7 yrs, 8–13 yrs), 'sex' (male/female), 'neuter status' (no/unknown/yes) and 'reason for admission to the shelter' (relinquished/stray). 'Day', 'night' or 'week' was included as a factor and not treated as continuous in the model. Interactions between 'day', 'night' or 'week' and one of the other main factors were added in the start model when visual inspection of boxplot graphs revealed potential interactions. Full models were tested with a random effect for 'dog ID' (individual identity) and/or various correlational and variance structures (with autoregressive model of the order 1 (AR1) correlation structure or weights) to test the best fit. With the best fitting model structure, explanatory variables were dropped based on a backward selection approach, using the Akaike information criterion (AIC) to determine the best model fit with maximum likelihood estimation. Restricted maximum likelihood estimation was used for the final model. Models were evaluated by visual inspection of the residuals (normality and constant variance).

UCCR levels and activity data of the SD group and CPD group were compared using t-tests with corrected p-value for multiple comparisons (on log-transformed data) for the following comparisons: I) CPD group day/night 1 & 12, II) CPD group day/night 1 & SD group >6 weeks post-adoption and III) CPD group day/night 1 & SD group day/night 1.

Behaviour data, even after transformation, was not normally distributed. Therefore, a non-parametric repeated measures Friedman test was used for each activity behaviour with post hoc pairwise comparisons for all three nights (1, 2 and 12) using paired Wilcoxon signed-rank test with Bonferroni adjusted p-values.

To evaluate the relation between nocturnal activity and UCCR, a Spearman's rank correlation for non-parametric variables was calculated for nocturnal *activity duration* and UCCR the next morning, for the in-shelter measurement days (1, 2 and 12).

## 2.4. Ethical note

This study involved no invasive procedures on the participating dogs. The Animal Welfare Body of the Utrecht University concluded that the study does not meet the definition of an animal experiment as defined in the Dutch Experiments on Animals Act and Directive 2010/63/EU, as the animals would encounter minimal levels of discomfort. All owners agreed and volunteered to participate in this study and signed informed consent for participation and publication of the results, conforming to the General Data Protection Regulation in the Netherlands. The participating shelter consented to the study.

## 3. Results

### 3.1. Demographics

The SD group had the following characteristics: a mean age of 4.5 years (range 1–13 years); males (n = 19; 6 neutered, 13 entire) and females (n = 10; 3 neutered, 5 entire, 2 unknown status). Dogs in this group were admitted to the shelter either relinquished by their owners (n = 20) or found as strays (n = 9). Of the relinquished dogs, 7 were known to have been kennelled in e.g. boarding kennels, kennel in backyard etc., before, 10 were kennelled for the first time and 3 had an unknown kennel history. Of the stray dogs, 1 was known to have been kennelled before and 8 had an unknown kennel history. Shelter dogs were assigned to weight classes <10 kg (n = 9), 10–20 kg (n = 0), >20–30 kg (n = 9), >30–40 kg (n = 9) or >40 kg (n = 2).

The CPD group had the following characteristics: mean age 5.1 years (range 1–13 years); males (n = 17; 9 neutered, 8 entire) and females (n = 12; 9 neutered, 3 entire).

### 3.2. Nocturnal activity (accelerometer results)

Nocturnal activity data from the total 29 dogs in the SD group was available for 24 dogs on night 1, 28 dogs on night 2 and 25 dogs on night 12. From the CPD group, data was available for 28 dogs on night 1 and 29 dogs on night 12.

#### 3.2.1. SD group

For nocturnal *total activity counts* data, the best fit was a mixed model with only random intercept for 'dog ID' and no other structures. For nocturnal *activity duration* and nocturnal *number of rest bouts*, the best fit was a model with random intercept for 'dog ID' and a variance model to allow different variances for the separate nights. Model results for the SD group data showed that for the *total activity counts*, *activity duration* and *number of rest bouts* an interaction between 'night' and 'weight class' significantly explained variability. For all three nocturnal activity parameters, 'age class' and 'neuter status' were also included in the best fitting models, with an interaction of 'night' and 'age class' for *number of rest bouts*. For *activity duration* and *number of rest bouts*, 'kennel history' was also included. No significant association with 'sex' and 'reason for admission to the shelter' was found for any of the three nocturnal activity parameters. The focus in this paper, as the effects of 'age class', 'neuter status' and 'kennel history' seem smaller and this information was often unknown or guessed, is mainly on the effect of 'night' and 'weight class' on the activity parameters.

Regarding the effect of 'night' and 'weight class', all nocturnal activity parameters used in the mixed models significantly decreased over time from night 1 and 2 to night 12 in the shelter showing a start of recovery for some individuals (Table 2, Fig. 2). Significantly higher estimates of *total activity counts*, *activity duration* and *number of rest bouts*, especially during the first nights, were found in lighter dogs (<10 kg) than in heavier dogs (>30–40 kg and >40 kg, Table 2, Fig. 3).

Regarding the other factors included in the final models, the estimates and confidence intervals are stated in supplementary Tables 2–4. Regarding the effect of 'age class' on all activity parameters, younger dogs seemed more active than older dogs, but this difference was smaller on night 12. Neutered dogs were less active than intact dogs (males & females combined). Dogs with a known history in kennels were active for a longer duration than dogs with no kennel history, although confidence intervals for the dogs with kennel history were relatively large. For *number of rest bouts*, dogs with an unknown kennel history were less active than dogs with a known history, although this difference was relatively small.

Variances in activity between dogs decreased from night 1 to night 12 in the SD group and were larger than the variance in the CPD group (Fig. 2). Original values (mean and standard deviation) are displayed in supplementary Table 5.

#### 3.2.2. SD versus CPD group

No significant differences were found in the CPD group between the first and second night of wearing the accelerometer (sample estimated mean difference [ratio] and 95 % confidence interval [CI] for *total activity counts*: 1.13 and 0.55–2.30, for *activity duration*: 1.02 and 0.76–1.37, for *number of rest bouts*: 1.03 and 0.79–1.34, and for *maximum duration of rest bouts*: 0.96 and 0.78–1.18). However, all activity measures for the SD group in night 1 were significantly different from the first night in the CPD group (higher *total activity counts*: sample estimated mean difference [ratio] = 4.77, 95 % CI = 2.32–9.83,  $t[46] = 4.35$ ,  $p < 0.001$ ; higher *activity duration*: sample estimated mean difference [ratio] = 3.75, 95 % CI = 2.54–5.53,  $t[34] = 6.92$ ,  $p < 0.001$ ; higher *number of rest bouts*: sample estimated mean difference [ratio] = 2.97, 95 % CI = 2.18–4.04,  $t[41] = 7.11$ ,  $p < 0.001$ ; and lower *maximum duration of rest bouts*: sample estimated mean difference [ratio] = 0.53, 95 % CI = 0.43–0.67,  $t[43] = -5.54$ ,  $p < 0.001$ ). There was no significant difference in *total activity counts* between night 12 in the SD group and night 2 in the CPD group (sample estimated mean difference [ratio] =

**Table 2**

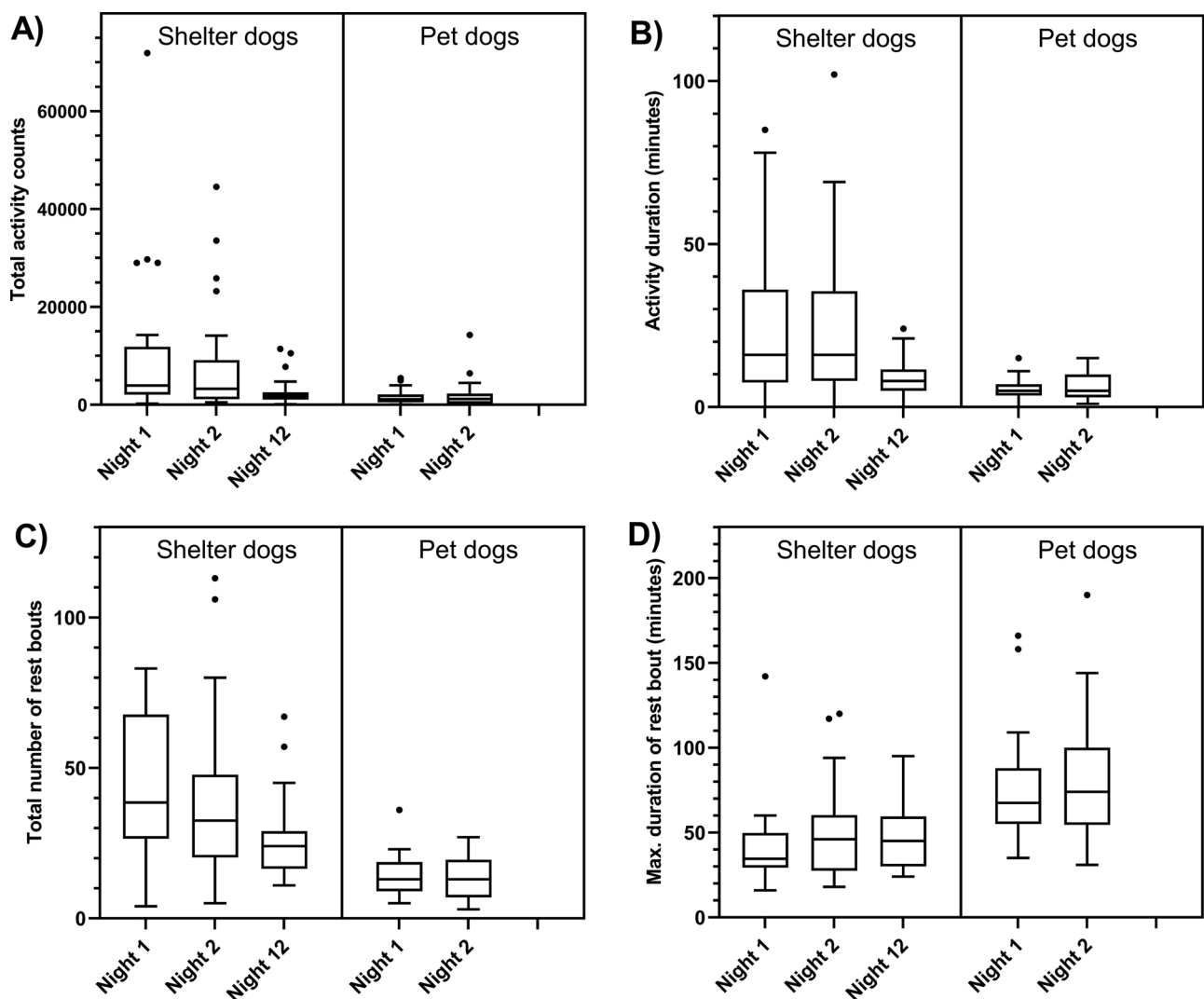
**Model results for nocturnal activity accelerometer measures of the SD group.** Estimated parameter values (EP) and 95 % confidence intervals (CI) of nocturnal activity for night (after intake, 0:00–4:00 h) and weight class. Reference category in the model was weight class >30–40 kg, all other weight class estimates are ratios which expresses the relative size of the estimated means of two conditions.

Parameter	Category	Weight class	Night 1		Night 2		Night 12	
			EP	95 % CI	EP	95 % CI	EP	95 % CI
Total activity counts	Reference	>30–40 kg	3884 <sup>1</sup>	1814 - 8313	0.81 <sup>2</sup>	0.31 - 2.15	0.60 <sup>2</sup>	0.22 - 1.60
		<10 kg	9.78 <sup>3</sup>	3.17 - 30.16	4.26 <sup>3</sup>	1.46 - 12.30	1.24 <sup>3</sup>	0.40 - 3.83
	Estimated difference	>20–30 kg	2.02 <sup>3</sup>	0.69 - 5.88	2.12 <sup>3</sup>	0.77 - 5.80	2.06 <sup>3</sup>	0.73 - 5.83
Activity duration		>40 kg	0.36 <sup>3</sup>	0.07 - 1.85	1.32 <sup>3</sup>	0.26 - 6.74	0.43 <sup>3</sup>	0.05 - 4.02
	Reference	>30–40 kg	24.37 <sup>1</sup>	15.16 - 39.18	0.66 <sup>2</sup>	0.35 - 1.25	0.66 <sup>2</sup>	0.40 - 1.10
		<10 kg	4.62 <sup>3</sup>	2.71 - 7.88	3.78 <sup>3</sup>	1.64 - 8.75	0.85 <sup>3</sup>	0.46 - 1.57
	Estimated difference	>20–30 kg	1.44 <sup>3</sup>	0.86 - 2.40	2.24 <sup>3</sup>	0.99 - 5.07	1.39 <sup>3</sup>	0.77 - 2.51
		>40 kg	0.50 <sup>3</sup>	0.23 - 1.09	1.35 <sup>3</sup>	0.36 - 5.07	0.57 <sup>3</sup>	0.17 - 1.95
Number of rest bouts	Reference	>30–40 kg	65.07 <sup>1</sup>	43.62 - 97.09	0.76 <sup>2</sup>	0.45 - 1.30	0.64 <sup>2</sup>	0.43 - 0.96
		<10 kg	1.99 <sup>3</sup>	1.28 - 3.09	1.85 <sup>3</sup>	0.90 - 3.80	0.63 <sup>3</sup>	0.38 - 1.07
	Estimated difference	>20–30 kg	1.31 <sup>3</sup>	0.87 - 1.98	1.52 <sup>3</sup>	0.77 - 2.99	1.15 <sup>3</sup>	0.71 - 1.86
		>40 kg	0.39 <sup>3</sup>	0.21 - 0.72	1.41 <sup>3</sup>	0.48 - 4.11	0.55 <sup>3</sup>	0.21 - 1.45

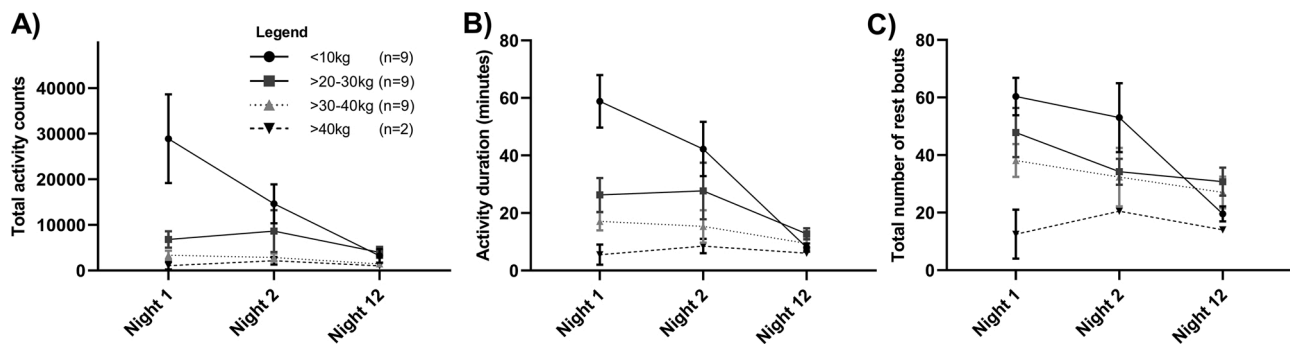
<sup>1</sup> Estimated mean in reference weight class and night 1 (and reference age class and neuter status for all parameters, and reference kennel history for number of rest bouts, see supplementary Tables 2–4).

<sup>2</sup> Estimated ratio of mean of specified night and mean at night 1 in reference weight class.

<sup>3</sup> Estimated ratio of mean of specified weight class and mean in reference weight class at same night.



**Fig. 2. Nocturnal accelerometer results.** Box and whisker (Tukey) plot with median and outliers (dots) for night 1, 2 and 12 in the shelter for the SD group (0:00 – 4:00 h) and night 1 and 2 after the start of wearing the accelerometer for the CPD group (1–5 h after owner sleep onset). A) total activity counts, B) activity duration, C) total number of rest bouts and D) maximum duration of rest bouts during the night.



**Fig. 3. Nocturnal accelerometer results per weight class for the SD group.** A) total activity counts, B) activity duration and C) number of rest bouts during the night (0:00-4:00 h). Means and standard error of the mean (SEM) divided in different body weight classes (in kg) including total sample size per weight class.

2.01, 95 % CI = 1.00–4.07), but the SD group had significantly higher *activity duration* (sample estimated mean difference [ratio] = 1.68, 95 % CI = 1.24–2.29,  $t[52] = 3.40$ ,  $p = 0.001$ ) and *number of rest bouts* (sample estimated mean difference [ratio] = 1.87, 95 % CI = 1.42–2.45,  $t[52] = 4.57$ ,  $p < 0.001$ ) and significantly lower *maximum duration of rest bouts* (sample estimated mean difference [ratio] = 0.60, 95 % CI = 0.48–0.75,  $t[52] = -4.67$ ,  $p < 0.001$ ).

Mainly *activity duration* and *number of rest bouts* of the SD group showed larger interindividual variation than of the CPD group, especially on night 1 and 2 (Fig. 2). Original values (mean and standard deviation) are displayed in supplementary Table 5.

### 3.3. Urinary cortisol/creatinine ratio

There was no correlation between time of the day of urine sample collection and UCCR (Spearman’s  $\rho = -0.06$ ,  $p = 0.59$ ). UCCR data from the total 29 dogs in the SD group was available for 25 dogs on day 1, 22 dogs on day 2 and 27 dogs on day 12. From the CPD group, data was available for 27 dogs on day 1 and 29 dogs on day 12.

#### 3.3.1. SD group

In the SD group, the best fit was a mixed model with random intercept for ‘dog ID’ and a variance model to allow different variances for the separate nights. The factor ‘day’, together with an interaction effect of ‘day’ and ‘weight class’, significantly explained UCCR variability. There were no significant associations with ‘age class’, ‘sex’, ‘neuter status’, ‘kennel history’ and ‘reason for admission to the shelter’.

Regarding the effect of ‘day’, UCCRs were significantly higher at day 1 than day 12 and 6 weeks post-adoption (Table 3, Fig. 4.I). Regarding the interaction between ‘day’ and ‘weight class’, this pattern over time depended on weight class, with lighter dogs (<10 kg) showing higher UCCRs during the first days (Table 3, Fig. 4.II).

Interindividual variation decreased from day 1 to day 12 and was larger in the shelter than 6 weeks after adoption (Fig. 4.I). Original values (mean and standard deviation) are displayed in supplementary

**Table 3**

**Model results for urinary cortisol/creatinine ratios ( $\times 10^{-6}$ ) of the SD group.** Estimated parameter (EP) and 95 % confidence intervals (CI) of UCCR ratios for day (after intake) and weight class. Reference class in the model was weight class >30–40 kg, all other weight class estimates are ratios which expresses the relative size of the estimated means of two conditions.

Category	Weight class	Day 1		Day 2		Day 12		6wks PA	
		EP	95 % CI	EP	95 % CI	EP	95 % CI	EP	95 % CI
Reference	>30–40 kg	3.96 <sup>1</sup>	2.36 – 6.65	0.71 <sup>2</sup>	0.39 – 1.30	0.81 <sup>2</sup>	0.44 – 1.50	0.62 <sup>2</sup>	0.35 – 1.11
	<10 kg	2.51 <sup>3</sup>	1.18 – 5.33	2.09 <sup>3</sup>	1.12 – 3.91	1.60 <sup>3</sup>	0.85 – 3.01	0.93 <sup>3</sup>	0.56 – 1.53
Estimated difference	>20–30 kg	1.67 <sup>3</sup>	0.80 – 3.48	2.11 <sup>3</sup>	1.13 – 3.96	0.63 <sup>3</sup>	0.32 – 1.25	0.65 <sup>3</sup>	0.36 – 1.17
	>40 kg	0.43 <sup>3</sup>	0.13 – 1.37	0.53 <sup>3</sup>	0.15 – 1.82	0.71 <sup>3</sup>	0.25 – 2.04	0.52 <sup>3</sup>	0.21 – 1.27

<sup>1</sup> Estimated mean in reference weight class and day 1.

<sup>2</sup> Estimated ratio of mean of specified day and mean at day 1 in reference weight class.

<sup>3</sup> Estimated ratio of mean of specified weight class and mean in reference weight class at same day.

Table 5.

Correlation tests for UCCR levels and nocturnal *activity duration* showed a weak and not significant positive correlation on night/day 1 (Spearman’s  $\rho = 0.29$ ,  $p = 0.19$ ) and night/day 2 (Spearman’s  $\rho = 0.26$ ,  $p = 0.26$ ), and a weak not significant negative correlation on night/day 12 (Spearman’s  $\rho = -0.22$ ,  $p = 0.3$ , see supplementary Fig. 1).

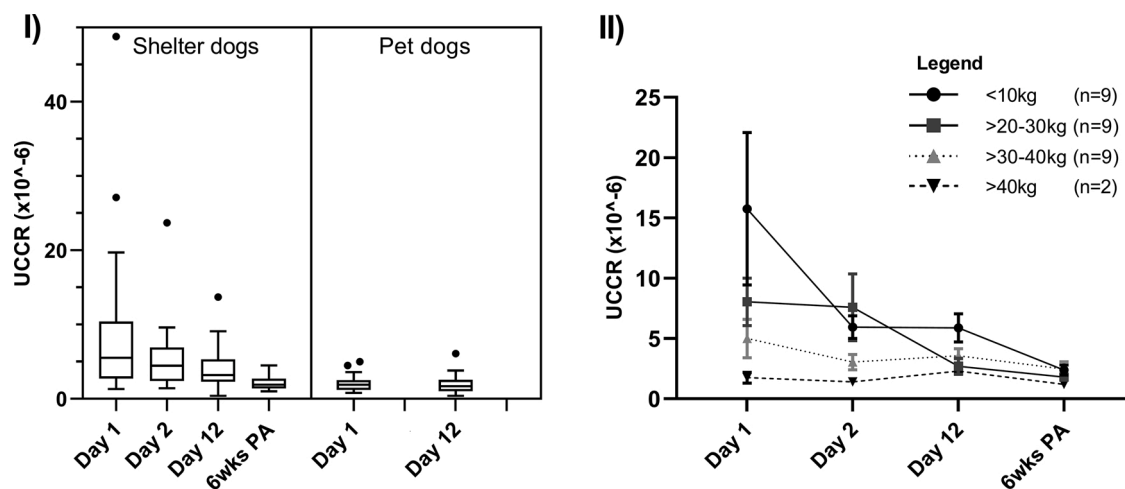
#### 3.3.2. SD versus CPD group

No significant difference was found in UCCRs between CPD group day 1 and 12 (sample estimated mean difference [ratio] = 1.11, 95 % CI = 0.83–1.49), and between CPD group day 1 and SD group post-adoption (sample estimated mean difference [ratio] = 1.11, 95 % CI = 0.81–1.50). UCCRs on day 1 of the SD group were higher than the CPD group day 1 (sample estimated mean difference [ratio] = 3.09, 95 % CI = 2.05–4.65,  $t[37] = 5.60$ ,  $p < 0.001$ ) and UCCRs of the SD group day 12 were higher than the CPD group day 12 (sample estimated mean difference [ratio] = 1.99, 95 % CI = 1.40–2.81,  $t[52] = 4.0$ ,  $p < 0.001$ , Fig. 4).

In-shelter measures of the SD group showed larger interindividual variation than after adoption and compared to the CPD group, especially on day 1 (Fig. 4). Original values (mean and standard deviation) are displayed in supplementary Table 5.

### 3.4. Body weight

For 26 shelter dogs, a proportional body weight change could be calculated, as weight was measured at all three time points. In total, 24 of these dogs lost weight at week 2, 1 dog gained weight and 1 dog did not change in weight. The best fit for proportional body weight change data was a mixed model with only random intercept for ‘dog ID’ and no other structures. Proportional body weight variability was explained by the factors ‘week’ and ‘reason for admission to the shelter’. Compared to week 0, an estimated not significant mean relative weight loss of 1.8 % at week 1 and significant weight loss of 3.8 % at week 2 was found for stray dogs ( $n = 8$ ). For relinquished dogs, an additional 1.9 % weight



**Fig. 4. Urinary cortisol/creatinine ratios (UCCR) results.** I) Box and whisker (Tukey) plot with median and outliers (dots) for the SD group on day 1, 2, 12 in the shelter and 6 weeks post-adoption (PA) and for the CPD group on day 1 and 12 into the study. II) Means and standard error of the mean (SEM) for the SD group divided in different body weight classes.

loss was found on top of the weight loss reported for stray dogs for both weeks (n = 18, see Table 4). There were no associations with other independent factors.

### 3.5. Nocturnal behaviour

In the behavioural observation data, nocturnal activity behaviour (0:00–4:00 h) in the SD group significantly changed over time. Friedman test  $\chi^2$ , p-value and effect size for nocturnal activity behaviour were *recumbent head down*:  $\chi^2(2) = 10.4$ ,  $p = 0.0056$ ,  $W = 0.47$  (moderate); *recumbent head up*:  $\chi^2(2) = 6.55$ ,  $p = 0.0379$ ,  $W = 0.30$  (small); *stationary (sit/stand)*:  $\chi^2(2) = 13.8$ ,  $p = 0.00099$ ,  $W = 0.63$  (large); *movement*:  $\chi^2(2) = 14.4$ ,  $p = 0.00076$ ,  $W = 0.65$  (large); *number of movements*:  $\chi^2(2) = 14.3$ ,  $p = 0.00079$ ,  $W = 0.65$  (large).

Post hoc comparisons revealed that, after Bonferroni adjustments, differences were significant between nights 1 and 12. The percentage of time spent in *recumbent position with head down* was higher on night 12 than night 1 (mean[S.D.]: 92.4[6.7]% versus 74.4[18.9]%,  $Z = 5$ ,  $p = 0.029$ ). On the other hand, the percentage of time *stationary* in night 12 was lower than in night 1 (1.1[2.0]% versus 8.7[7.6]%,  $Z = 66$ ,  $p = 0.003$ ) and the same was found both for the percentage of time in *movement* (0.6[0.7]% versus 4.2[5.7]%,  $Z = 66$ ,  $p = 0.00098$ ) and the *number of movements* (11[13] versus 82[81],  $Z = 66$ ,  $p = 0.012$ ). No significant differences were found between nights 1 and 2 or 2 and 12 for any nocturnal activity behaviour (night 2: *recumbent head down*: 86.9 [10.5]%; *stationary*: 4.5[4.9]%; *movement*: 1.5[1.2]%; *number of movements*: 29[32]). No significant differences between nights for percentage of time spent in *recumbent position head up* was found (night 1: 12.3 [9.5]%; night 2: 6.9[5.1]%; night 12: 6.0[5.5]%).

**Table 4**  
**Model results for proportional body weight change.** With the estimated proportional weight (EPW) in comparison with week 0 (proportion = 1 for all dogs) and 95 % confidence (Wald) intervals (CI) for stray and relinquished dogs.

Category		Proportional body weight	
		EPW	95 % CI
Stray (reference category)	Week 1	0.982 <sup>1</sup>	0.962–1.001
	Week 2	0.962 <sup>2</sup>	0.943 – 0.982
Relinquished		-0.019 <sup>3</sup>	-0.042 – 0.004

<sup>1</sup> Estimated mean proportional body weight for stray dogs on week 1.  
<sup>2</sup> Estimated mean proportional body weight for stray dogs on week 2.  
<sup>3</sup> Estimated mean difference between the proportional body weight of relinquished dogs compared to reference stray dogs on both weeks.

## 4. Discussion

In this study, we evaluated resting patterns by measuring nocturnal activity as an indicator of adaptability of dogs to a shelter environment, during a two-week period of habituation after intake at the shelter.

### 4.1. SD group evaluation

#### 4.1.1. Nocturnal activity of SD's

Shelter dogs had significantly higher nocturnal activity, as measured by the accelerometer, during the first two nights after intake than at 12 nights in the shelter. Behavioural observations supported the accelerometer measures. On night 12, nocturnal activity behaviour was lower and resting behaviour higher than on the first nights. This is shown most clearly by the accelerometer measure activity duration, which provides information on the total amount of rest the dogs were able to achieve, whereas total activity counts and number of rest bouts can also be influenced by the intensity of movements or a high frequency of rest bouts only at specific time points. In addition, differences in activity duration between days were large for most weight classes (except the largest weight class, with only two individuals) and activity duration reflects the percentage of time in *movement*, which had a large effect in our behaviour analysis.

This difference in response over time reflects habituation, a form of non-associative learning (Thompson and Spencer, 1966), which allows for the discrimination between biologically relevant and iterative stimuli (Eisenstein and Eisenstein, 2006) after exposure to a novel environment or situation (Salomons et al., 2010). Here, dogs learned that potentially harmful stimuli in the shelter environment that previously woke them up did not predict a positive or negative outcome and that therefore an alert response was no longer required, allowing them to rest more. Therefore, our results suggest that dogs at least partly adapted to the shelter environment.

Resting patterns can reflect dog's welfare; lack of rest may also contribute to a decrease in a dog's welfare. Interestingly, poor rest quality and quantity induced by sleep deprivation is associated with physical and mental health problems in humans (e.g. Roberts et al., 2009; Luyster et al., 2012). Monitoring resting levels in shelter dogs can be beneficial, since it gives an indication of overall adaptation to the shelter. Also, if dogs can be helped to rest more and better, this can result in faster adaptation to the shelter environment i.e. improved welfare.

In humans, a first night's sleep in a novel environment might result in disruptions in sleep macro- and microstructure, referred to as the 'first-night effect' (Agnew et al., 1966), and the same might hold true for dogs.



To our knowledge, we were the first to compare nocturnal activity responses between nights 1 and 2 in shelter dogs. We found no significant difference between night 1 and 2, which refutes the idea of a ‘first-night effect’ in dogs in shelters, but points in the direction of a ‘more-than-one-night-effect’.

#### 4.1.2. Urinary cortisol/creatinine ratios of SD's

Shelter dogs had significantly higher UCCRs on the first morning after intake than on day 12, in line with the pattern observed for nocturnal activity, which showed a decrease over time. However, changes in nocturnal activity corresponded with changes in UCCRs on group level but not on individual level, as only weak and non-significant correlations were found.

Similar partial decreases in UCCRs in dogs entering kennels, over days or even weeks, has been observed in other studies, although temporal dynamics (i.e. how UCCR levels change over time) differ between studies (Hiby et al., 2006; Stephen and Ledger, 2006; Rooney et al., 2007). These temporal dynamics, including potential individual differences, could not be examined in the present study due to a time ‘gap’ as data were not collected between day 3–11 in-shelter, precluding this analysis. Future research will explore daily changes in nocturnal activity levels and UCCR over the first weeks in a shelter.

#### 4.1.3. Body weight changes of SD's

Body weight loss was seen in most dogs (92 %) in our SD group during the first two weeks in the shelter, with on average 5% weight decrease. We were unable to standardize or monitor caloric food intake of dogs as we did not want to change shelter enrichment routines, which leaves several explanations that could account for this weight loss. For instance, medical conditions can lead to weight loss (Gram et al., 2017), although none were identified by the shelter veterinarian. A body condition score was not determined at intake, although given that nearly all dogs lost weight in our study, it is very unlikely that weight loss was due to initial excess weight. It remains possible that weight loss is due to an increase in daytime activity during sheltering. Stress may be a most interesting factor, as body weight loss has already been suggested to be stress-related in shelter cats (Tanaka et al., 2012). Relinquished dogs lost more relative weight than stray dogs, which may be due to a lower body condition score of strays at intake or a higher food intake of strays. Daytime activity, food intake and body condition scores should be monitored in future studies, to explore body weight loss in shelter dogs in more detail.

## 4.2. Comparisons between SD and CPD group

### 4.2.1. Nocturnal activity

Especially during the first two nights in the shelter, dogs in the SD group were more nocturnally active than dogs in the CPD group. It seemed that, during the 4-h timeframe, dogs in the CPD group could complete multiple sleep cycles of ~16–20 min (Adams and Johnson, 1993). Dogs of the SD group not only had a higher number of activity counts than dogs of the CPD group during nights 1 and 2, but were also active for a longer period of time and showed a higher number of resting bouts, a possible indication of restlessness. In this group, the lowest maximum duration of rest bouts was 16 min in night 1, 18 min in night 2 and 24 min in night 12, indicating that some shelter dogs might not have been able to complete multiple sleep cycles their first nights.

Overall, our results add to the existing literature suggesting that shelter dogs spend less time resting than pet dogs. Hoffman et al. (2019) found shelter dogs to have higher average activity levels during the five least active hours (~23:16–04:16) compared to pet dogs, which is in line with our findings. Hoffman and colleagues calculated means based on all actigraphy data from the first three days and nights in the shelter up to twelve days. To draw conclusions on adaptive capacity however, measurements need to be taken over time. To the best of our knowledge, we are the first to report changes in nocturnal activity patterns over time

in a shelter.

### 4.2.2. Urinary cortisol/creatinine ratios

Compared to the CPD group, UCCRs were higher in the SD group. This difference vanished 6 weeks after adoption in the SD group. Thus, our findings indicate that after two weeks in the shelter, shelter dogs still had higher cortisol levels than pet dogs. However, it is possible that these SD group levels will decrease further over time, as long-term (>1 year) kennelled working dogs in spatially restricted (<10m<sup>2</sup>) kennels showed lower UCCRs than pet dogs in homes, potentially due to long-term downregulation as a consequence of chronic stress (Hewson et al., 2007).

### 4.3. Interindividual variation and body size differences

Investigating interindividual variation provides information on individual adaptive responses (i.e. welfare). We found interindividual variability in UCCRs and nocturnal activity to be highest during the first two days and nights in the shelter, suggesting that individual dogs responded differently to sheltering mainly during the first few days. Interestingly, this variability was highest in smaller dogs (<10 kg), i.e. there were larger differences between smaller dogs than between larger dogs. Our sample size per breed (or breed type) was too small to control for breed and shelter dog breeds cannot reliably be breed labelled (e.g. Voith et al., 2009), making it impossible to distinguish between body weight versus breed.

A relationship between accelerometer activity measures and body weight has been found before (Jones et al., 2014), with lighter (smaller) dogs showing higher measures than heavier (larger) dogs in a shelter situation. Similarly, a negative correlation between body weight and activity counts was also found in dogs during relatively controlled (i.e. on-leash) movements assuming different activity detection by accelerometers for different body weight classes (Brown et al., 2010b). However, we found activity level differences between body weight classes to be lower on night 12 in the shelter.

Similar relations with body weight have been found for UCCR. A negative correlation between body weight and UCCRs has been described in dogs before (Zeugswetter et al., 2010; Jones et al., 2014; Gunter et al., 2019), although others found no effect of body weight on plasma cortisol concentrations (Hennessy et al., 1997). This negative correlation might be explained, as Zeugswetter et al. (2010) suggested, by the relatively small muscle mass in smaller dogs as creatinine production is proportional to muscle mass (Van den Brom and Biewenga, 1981). However, this theory does not explain our data, as we found an interaction effect between time in the shelter and UCCRs, with higher UCCR levels for smaller dogs in the shelter but not post-adoption, and decreasing UCCRs over time in the shelter.

Our data suggest that smaller dogs show on average a higher and more inter-individual variable stress response mainly during the first days in the shelter compared to larger dogs, potentially due to breed-specific differences. For example, smaller breeds are assumed to be trained and socialised less and are more likely to show fear of dogs and strangers than larger dogs (Arhant et al., 2010; Puurunen et al., 2020) and therefore could experience more stress in a shelter environment.

Moreover, animals can cope differently with environmental stressors and therefore restlessness can also be expressed by lying inactive but awake rather than by active behaviours (Meagher et al., 2013). However, at group level this is unlikely, as our behaviour analysis showed more recumbent head down behaviour on night 12. This undermines a ‘quiet wakefulness’ hypothesis, although we could not reliably observe whether dogs had their eyes open or closed as the head of the dog was often turned away from the camera. Also, we can not exclude the hypothesis that dogs learned that their response (active behaviour) had no effect on the situation, and therefore they showed more passive behaviour during the night.

#### 4.4. Conclusion

Nocturnal activity and UCCRs were higher in shelter dogs than in pet dogs and these levels decreased over time in the shelter. Nocturnal activity, UCCR and resting behaviour during the night suggest disrupted nocturnal resting patterns after intake at the shelter and partly adaptation to the shelter environment within two weeks after intake. We therefore suggest that resting patterns as measured by accelerometers can be a reliable proxy of adaptation of dogs to a shelter environment, as validated by UCCRs and behavioural observations. Because accelerometers are quickly becoming cheaper and more accessible to shelters, nocturnal activity can be a cost-effective additional parameter to monitor adaptability of dogs to a shelter environment.

In the future, daily monitoring of nocturnal activity and UCCR levels during the first period in the shelter is needed to evaluate the temporal dynamics in the recovery of nocturnal resting patterns and to have a better insight into individual differences in adaptation patterns.

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#### Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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#### Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:<https://doi.org/10.1016/j.applanim.2021.10.5377>.

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