

Short communication

# Preliminary observations on the urinary behaviour of female Jack Russell Terriers in relation to stage of the oestrous cycle, location, and age

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## Abstract

In female canids, including domestic dogs (*Canis familiaris*), scent-marking with urine is thought to convey information on reproductive state, yet little is known about how urinary behaviour changes across specific stages of the oestrous cycle. We measured urinary behaviour (proportion of directed urinations) of 10 intact Jack Russell Terriers across anoestrus, proestrus, and oestrus during walks in familiar and novel environments. Females ranged in age from 1.3 to 8.7 years. We assessed stage of oestrus using vaginal cytology, behaviour, and physical signs. Proportions of directed urinations were higher during proestrus and oestrus than anoestrus, and were higher in older females than younger females. Our findings indicate that in female Jack Russell Terriers, scent-marking with urine advertises reproductive state and continues to develop in adulthood. Additional data are needed to determine if these findings generalize to female dogs of other breeds and mixed breeds.

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## 1. Introduction

Olfactory communication by canids occurs through the application of urine, feces, and secretions from anal or interdigital glands (Eisenberg and Kleiman, 1972). The act of strategically placing a chemical mark is called scent-marking, and directing urine at an object in

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the environment is the most important criterion for distinguishing scent-marking with urine from simple elimination (Kleiman, 1966). Although more commonly performed by males, urine marking is also displayed by female canids (Kleiman, 1966; Wirant and McGuire, 2004).

Urine marks of female canids likely convey information about reproductive state and may function to establish and maintain breeding relationships (Fox and Cohen, 1977; Rothman and Mech, 1979). A higher frequency of urine marking during the breeding season has been reported for female wolves (*Canis lupus*; Ryon and Brown, 1990), coyotes (*Canis latrans*; Gese and Ruff, 1997), bush dogs (*Speothos venaticus*; Porton, 1983), and bat-eared foxes (*Otocyon megalotis*; Nel and Bester, 1982). Data on scent-marking and reproductive state in female domestic dogs are contradictory and, surprisingly, more limited than those for wild canids. Beach and Gilmore (1949) appear to be the first authors to hypothesize that female dogs urinate more frequently when in heat; they provided no data on this topic and did not cite work of others. Sprague and Anisko (1973) studied urinary behaviour in beagles, and some females were in heat during observations. These authors do not present data on urinary behaviour and reproductive status, but state that there were no obvious differences between oestrous and non-oestrous females in postures or in the proportions of females that urinated frequently or infrequently. In her study of scent-marking in several species of canids, Kleiman (1966) stated that female dogs scent-marked only around oestrus, but presented no data to support this claim. Finally, Wirant and McGuire (2004) found that 60% of urinations by spayed and anoestrous intact females were directed at objects in the environment; females in proestrus and oestrus were not included in their study. In reviewing the literature on oestrus and marking in mammals, Johnson (1973, p. 525) described available data for canids (Beach and Gilmore, 1949; Kleiman, 1966) and stated, “There appears to be no full investigation of marking frequency and the oestrous cycle in any species. However, there are a number of statements on this topic which are typical of the state of much of the marking behaviour literature in that they are not accompanied by supporting data and show some disagreement.” We did not find any data on changes in urinary behaviour across the oestrous cycle in domestic dogs and believe that Johnson’s statements still apply. Thus, while it has been stated that the urinary behaviour of female dogs changes across the oestrous cycle (e.g., Dunbar, 1977: p. 479; Houpt, 2005: p. 167), published data still are lacking.

In this study, we examined urinary behaviour of female Jack Russell Terriers, of different ages, in relation to specific stage of the oestrous cycle. We used vaginal cytology, physical signs, and behaviour to determine reproductive status. We chose Jack Russell Terriers to build on our earlier results with this breed. We predicted that if scent-marking conveys information on reproductive status, then proportions of directed urinations would be higher during proestrus and oestrus than during anoestrus. Given our earlier findings with Jack Russell Terriers (Wirant and McGuire, 2004), we also predicted that proportions of directed urinations would be higher in novel environments and by older females.

## 2. Methods

### 2.1. Animals

We observed 10 intact Jack Russell Terriers during anoestrus, proestrus, and oestrus. Eight of the dogs came from one kennel and two from a second; they were housed in either the homes of their owners or in kennels attached to the home. All females had daily interactions with female (spayed or intact) and male (neutered or intact) Jack Russell Terriers; the exception being when they were in proestrus and oestrus, and the owners did not want to breed them. Three females were bred during the study.

## 2.2. Experimental procedures

All behavioural observations occurred during individual 15-min walks of the dogs between August 2003 and August 2004. The first author (SCW) walked each female on an 8.3 m leash during anoestrus, proestrus, and oestrus. During each stage of the oestrous cycle, half the walks occurred within the female's familiar environment and half occurred in novel environments. We defined "familiar environment" as the area in which the female eliminated on a daily basis; typically, this was the yard outside the home. Sites designated as "novel" were sites such as conservation areas that the dog had visited no more than twice the previous year. Walks occurred in the morning and then again in the late afternoon. The order of the locations (familiar or novel environment) in which a dog was walked was counterbalanced across days. Owners did not permit dogs to go outside the home or kennel until after AM and PM walks were completed; at least 5 h elapsed between walks on a given day. During each walk we recorded the total number of urinations and whether each urination was directed within 20 cm of a conspicuous object (Ranson and Beach, 1985). We defined proportion of directed urinations as number of directed urinations/total number of urinations and used this as our dependent variable in all analyses.

We used the sterile culturette method of vaginal cytology to determine stage of oestrus (Holst, 1985). After each afternoon walk, we lightly restrained each female with a slip lead on a grooming table and gently swabbed her vagina. We rolled the swab across a microscope slide and stained the smear using JorVet Dip Quick (Jorgensen Laboratories, Inc., Loveland, Colorado, 80538 USA). We viewed each slide after it had air-dried, and classified stage of oestrus using criteria presented in Holst (1985). All slides were also independently classified by a veterinarian. Our classifications differed for two of 129 slides; we jointly reviewed the two slides and agreed on a stage.

Anoestrus observations began when females showed no signs of proestrus or oestrus (vulval swelling, vaginal discharge, touch sensitivity and tail flagging). Owners contacted us when they observed signs of proestrus (vulval swelling and reddish vaginal discharge) and we began our proestrus observations. Observations continued through the period of oestrus and ended when either vaginal cytology indicated that the female was in dioestrus or when 10 walks were completed. All stage classifications based on behaviour and physical signs were confirmed through vaginal cytology.

Complete data for one dog consisted of 30 observations, 10 walks during each of three stages during a single oestrous cycle. Due to scheduling difficulties with an owner or an oestrous stage of shorter than expected duration, we were not able to obtain complete data for every dog. Total observations per dog ranged from 10 (this dog was adopted by a new owner after observations during anoestrus, but before observations during proestrus and oestrus) to 30 with a median of 27.5.

## 2.3. Statistical analysis

We computed descriptive statistics (Minitab Version 14.12, Minitab, Inc., State College, PA, 16801, USA) for the proportion of directed urinations by stage of the oestrous cycle, location, and age. We used random-effects logistic regression (STATA: Statistics/Data Analysis, Version 8.2, STATA Corporation, College Station, TX 77845, USA) to model the proportion of directed urinations as a function of age, stage of oestrous cycle, location and interaction terms. Two indicator variables represented stage of the oestrous cycle (proestrus, oestrus); anoestrus was treated as the baseline from which the other two stages varied. The indicator variable for location was coded to indicate the novel environment. Observations were clustered by dog and dog was treated as a random intercept in the model.

## 3. Results

Descriptive statistics suggest that proportions of directed urinations were higher during proestrus and oestrus than anoestrus, increased with age, but did not differ with location walked (Table 1). In the random-effects logistic model for proportion of directed urinations, the two

Table 1

Median proportion of directed urinations in 10 female Jack Russell Terriers by stage of oestrous cycle, location (familiar or novel) in which walks took place and age of dog

Variable	Number of dogs	Number of observations <sup>a</sup>	Median	IQR <sup>c</sup>
Stage				
Anoestrus	10	92	0.40	0.75
Proestrus	9	82	0.61	0.49
Oestrus	9	76	0.67	0.48
Location				
Familiar	10	127	0.50	0.75
Novel	10	124	0.60	0.87
Age				
1.3	1	21	0.25	0.50
1.7	1	10	0.00 <sup>b</sup>	0.00 <sup>b</sup>
2.0	2	57	0.25	0.67
3.0	2	60	0.55	0.42
4.7	1	27	0.50	0.75
5.7	2	54	0.87	0.38
8.7	1	22	0.73	0.24

<sup>a</sup> Number of times the 10 dogs were walked, in total.

<sup>b</sup> This dog was only observed during anoestrus because she was adopted by a new owner before observations during proestrus and oestrus could be conducted.

<sup>c</sup> Interquartile range, defined as the distance between the first and third quartiles of the data, i.e., IQR = Q3–Q1 where Q3 is the third quartile and Q1 is the first quartile.

Table 2

Random-effects logistic regression model for proportion of directed urinations for 10 female Jack Russell Terriers in relation to stage of the oestrous cycle, location (familiar or novel) in which walks took place and age of dog

Proportion	Coefficient	95% CI	<i>P</i> -value
Proestrus	1.55	(0.66, 2.43)	0.001
Oestrus	1.30	(0.47, 2.13)	0.002
Location	–0.06	(–0.74, 0.63)	0.875
Age	0.85	(0.25, 1.46)	0.005
Constant	–2.55	(–4.71, –0.40)	0.020

indicator variables for stage of oestrus cycle and age of dog were significant (*P*-values 0.001, 0.002, and 0.005, respectively) (Table 2). The location variable was not significant (*P*-value 0.875) (Table 2). No interactions were significant; thus, we dropped them from the final model.

#### 4. Discussion

Our finding that proportions of directed urinations were higher during proestrus and oestrus than during anoestrus in Jack Russell Terriers is consistent with the general hypothesis that scent-marking functions in female canids to advertise reproductive state. Our results agree with some but not all previous observations for domestic dogs. Kleiman (1966) observed that dingoes and New Guinea singing dogs only marked with urine during the period before oestrus and during oestrus itself. Although we found increased marking by Jack Russell Terriers during proestrus and oestrus, we also found substantial marking during anoestrus. Our results differ from those of

Sprague and Anisko (1973) who observed no difference in the urinary behaviour of oestrous and non-oestrous beagles, although they suggested that such differences were likely. Finally, most studies of wild canids have found increased frequency of scent-marking by females during the breeding season (Nel and Bester, 1982; Porton, 1983; Ryon and Brown, 1990; Gese and Ruff, 1997); our results are consistent with these reports. Limitations of our study include a small sample ( $n = 10$  dogs) from only two kennels with somewhat related dogs of a single breed.

We assessed reproductive condition of female dogs using daily vaginal cytology, a technique that was feasible given our use of companion animals. In most studies of wild canids, the reproductive state of females was assessed through behavioural observations and knowledge of breeding seasons (Rothman and Mech, 1979; Gese and Ruff, 1997). Daily monitoring of changes in vaginal cytology and urinary behaviour allow for more precision when examining the relationship between reproductive state and urinary behaviour. Nevertheless, the finding of increased scent-marking with urine during periods of reproductive activity in female canids appears robust across methodologies.

We predicted that the proportion of directed urinations would be higher in novel environments, but found no significant difference when females were walked within familiar environments (median proportion directed urinations, 0.5) or novel environments (0.6), although the data were in the direction predicted. Bekoff (1979) found that free-ranging female dogs urine marked more within novel environments. We previously found that spayed and anoestrous intact females directed a larger proportion of their urinations when walked in novel environments, although spayed females showed a much greater tendency to do so than did anoestrous intact females (Wirant and McGuire, 2004). Thus, the effect of location may be weak in intact females and we would have needed a larger sample size to detect it in this study.

In our study, proportions of directed urinations were higher in older females than in younger females. We found the same pattern for spayed and anoestrous intact female Jack Russell Terriers (Wirant and McGuire, 2004). In contrast, a longitudinal study of the development of urinary behaviour of beagles revealed that between the ages of 4 and 14 months, urinations directed at targets in the environment dramatically increased in males, but increased only slightly in females (Ranson and Beach, 1985). The present study and our previous work included females of a greater range of ages (this study: from 1.3 to 8.7 years; Wirant and McGuire, 2004: from 4 months to 11 years) than did the study by Ranson and Beach (1985), and directed urinations were most apparent in females 3 or more years of age, an age older than the oldest dogs observed by these authors. The age-related data presented for Jack Russell Terriers in this study and in our previous study (Wirant and McGuire, 2004) are preliminary and cross-sectional in nature. A long-term longitudinal study of urinary behaviour is needed to fully understand how urinary behaviour changes with age in female domestic dogs.

## 5. Conclusion

Proportions of directed urinations in female Jack Russell Terriers were higher during proestrus and oestrus than anoestrus; these are the first published data to demonstrate changes in urinary behaviour across the oestrous cycle for domestic dogs. Proportions of directed urinations were also higher in older than younger females, suggesting that urinary behaviour continues to develop even in adulthood. Inclusion of additional breeds and mixed breeds in future studies is necessary to determine whether our findings with Jack Russell Terriers apply generally to domestic dogs.

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