Journal of Zoology

Journal of Zoology. Print ISSN 0952-8369

Substrate selection for urine spraying in captive wildcats

J. Ruiz-Olmo[1], A. Such-Sanz[2] & C. Piñol[3]

1 Government of Catalonia, Ministry of Agriculture, Livestock, Fisheries, Food and Natural Environment, General Directorate of Environment and Biodiversity, Barcelona, Spain
2 Government of Catalonia, Ministry of Agriculture, Livestock, Fisheries, Food and Natural Environment, Forestal Catalana, SA, Barcelona, Spain
3 Government of Catalonia, Ministry of Agriculture, Livestock, Fisheries, Food and Natural Environment, C.F. Vallcalent, Lleida, Spain

Keywords
olfactory communication; marking site selection; common juniper; urine marking behaviour; intraspecific communication; Volatile Organic Compounds.

Correspondence
Angel Such-Sanz, Forestal Catalana, SA, Government of Catalonia. Ministry of Agriculture, Livestock, Fisheries, Food and Natural Environment – Sabino de Arana, 34, 1°, 1ª- 08028 Barcelona, Spain. Tel: +34 934110926; Fax: +34 934914859
Email: asuch@gencat.cat

Editor: Andrew Kitchener

Received 19 April 2012; revised 12 December 2012; accepted 16 January 2013

doi:10.1111/jzo.12025

Abstract

This study documents the urine spraying behaviour of wildcats, *Felis silvestris*. Urine spraying is considered a short term visual mark and the main form of scent marking by felids. When urine spraying, a wildcat raises up its tail and ejects backwards a spray of urine against a prominent object of its surrounding environment. The selection of a urinating substrate should maximize the communicating value of the mark, but the factors that influence this selection are poorly understood. We hypothesized that urine spraying marks are not placed randomly, but that wildcats select marking post based on traits that enhance the effectiveness of the scent mark, by maximizing their detectability, diffusion or persistence. This study shows that wildcats select common juniper, *Juniperus communis*, to spray their urine mark on not because of the physical traits of the plant, but based on the species. The effectiveness of an olfactive mark has to do with the degradation and oxidation of its chemical components. The common juniper has a high concentration of volatile organic compounds (VOC) with antioxidant activity. We therefore suggest that wildcats can recognize the VOC composition of different plants, and based on its VOC, select those plants which could enhance the olfactory effectiveness of the mark. Thus, the recognition of volatile compounds in the surrounding environment should be important in the marking behaviour of wildcats.

Introduction

Marking behaviour plays an important role in the spatial ecology of carnivorous species (Gorman & Trowbridge, 1986; Palomares, 1993; Seiler, Krüger & Festetics, 1994), including cats living in the wild (Stahl & Leger, 1992) as well as in captivity (Law & Tatner, 1998). Marking behaviour plays a role in shaping the spatial structure of the cat populations, providing information on the general condition of the animal even if it is not present (Gosling, 1982; Natoli, 1985; Kitchener, 1991; Stahl & Leger, 1992; Asa, 1993; Case, 2003), in regulating the movement of animals inside their home range (Benhamou, 1989; Artois *et al*., 2002) or in reproductive activity (Mellen, 1993; Piñeiro *et al*., 2012).

Cats make different forms of olfactory marks, such as urine spraying, squat urination, tree scraping associated with a urine depot and faeces deposition (Corbett, 1979; Leyhausen, 1979; Stahl, 1986; Kitchener, 1991; Stahl & Leger, 1992; Feldman, 1994; Bradshaw & Cameron-Beaumont, 2000). However, urine spraying is considered the main form of scent marking by felids (Feldman, 1994). Although the communication patterns of several large cats have been studied in the field, patterns of cats smaller than 20 kg in body mass have not been widely studied, mainly because of their nocturnal activity, densely vegetated habitat, wide-ranging movements and often wariness (Mellen, 1993). However, there are a few studies on the marking behaviour of wildcats, *Felis silvestris*, the most important among them being those of Corbett (1979) in Scotland and Piñeiro & Barja (2012) in Spain. Captivity offers some advantages over wild conditions when studying the marking behaviour, such as controlled variables, proximity and handling of the animals (including physiological samples and veterinary care) and larger sample sizes (Asa, 1993; Gittleman & McMillan, 1995). However, the experimental conditions of captive animals differ from those of wild animals. Consequently, the response of captive animals could be different from the response of those in the wild, particularly when the captive environment is very artificial (Gittleman & McMillan, 1995; Barja & de Miguel, 2004). Thus, wild animals could select different marking posts or make the decision based on other criteria than captive ones.

From an evolutionary point of view, marking behaviour should optimize the efficiency of the mark in transmitting messages to conspecifics. This can be achieved by marking strategies that increase the diffusion of the scent message (Barja, 2003), the predictability/conspicuousness of the mark (Gosling, 1982) or the controlled release and duration of the validity of the message (Asa, 1993).

The use of plants as defecating and urinating substrates is common in many carnivores, including wolves, *Canis lupus* (Barja, de Miguel & Bárcena, 2005; Barja, 2009); red foxes, *Vulpes vulpes* (Macdonald, 1979) or tigers, *Panthera tigris* (Smith, McDougal & Miquelle, 1989). In these cases, the selection of the plant depends on its physical characteristics: height, width, foliage, size and species. It also depends on the plant's surrounding environment (Smith *et al*., 1989; Barja, 2009). For example, if a specific plant is conspicuously different from the surrounding plants, this increases the predictability of the mark, and hence its communicating value (Roberts, 1997; Barja, 2009). The chemical composition of the urine mark can also influence its communicating value. For example, the presence of lipids in the urine can slow down the release of its volatile compounds, thus prolonging the release time of the chemical signal (Asa, 1993). Cats can distinguish the freshness of a mark and give more attention to fresh ones (De Boer, 1979). Any physical or chemical characteristic that prolongs the validity of a mark should favour the selection of a substrate to be marked (Barja, 2003; Barja, de Miguel & Bárcena, 2005). However, it has been proved that cats can recognize the volatile organic compounds (VOC) composition of some plants, such as cat nip, *Nepeta cataria*, through its main olfactive system (Hart & Leedy, 1985). Also it is known that common juniper has a high proportion of VOC with antioxidant activity (Wei & Shibamoto, 2007; Misharina, Terenina & Krikunova, 2009).

The objective of this study is to identify the characteristics of a plant that favour its selection as a substrate for urine spraying by captive wildcats. The hypothesis proposed here is that wildcats select marking posts based on traits that enhance the effectiveness of the scent mark, by maximizing their detectability, diffusion or persistence. Then, we predict that wildcats will select conspicuously taller, wider or infrequent plants; plants more exposed to the wind because its location or elevation; or finally, plants which increase the durability of the mark because of its dense foliage or its high concentration of VOC with antioxidant activity.

Methods

Studied animals and enclosures

The work was carried out at the Centre de Fauna Salvatge de Vallcalent (CFSV), in Lleida, Catalonia, Spain. CFSV had one installation dedicated to holding wildcats in captivity for their reproduction. This installation comprised of six facilities, which were radially disposed from an inner hexagon designed for the management of the animals (Fig. 1).

Each facility had an outside enclosure of 207 m^2 and six to eight wooden boxes orientated towards the inner hexagon where the wildcats could hide from humans (0.12 m^3 per box). The cats were fed *ad libitum* with mice, rat, rabbit, quail and chicken.

The facilities were environmentally enriched to meet the ecological requirements of the species (Gittleman & McMillan, 1995). As part of this enrichment, a total of 95 plants of 16 different plant species (trees and bushes) – without including grass and other annual plant species – were planted covering all of the six enclosures. The number of individual plants by species ranged from 1 to 15 (Mean ± standard deviation (SD) = 5.94 ± 4.17, $n = 16$) and by facility from 14 to 19 (X ± SD = 15.83 ± 1.94, $n = 6$). The number of plant species by facility ranged from 10 to 14 (X ± SD = 12.17 ± 1.47, $n = 6$) (Table 1).

Figure 1 General view of the installation for wildcat reproduction in CFSV where the study was carried out. The figure shows the radial distribution of the six facilities from the inner hexagon.

The CFSV kept 14 wildcats in total (four males and 10 females). Each of the six facilities held a group of several individual wildcats simultaneously, and each group remained in the same facility and with the same members (the groups of wildcats were fixed, without rotation of individuals between facilities), throughout the study period. Therefore, each facility represents a permanent group of wildcats during the study. Two of the facilities had one female each and no males. The other four facilities had one male and two females each. The cats, which came from different areas of the Iberian Peninsula, had been kept separately upon their arrival or from birth. Hence, no genetic or learning effects were expected.

Data collection

The facilities were surveyed every morning from April to June of 2005 to ascertain if any plant was marked with urine spraying by wildcats at least once during the study. In Vallcalent, most of the births of wildcats occur in March and April (per. obs.). In the wild, during the sensitive period at the end of pregnancy, the wildcats show a high physiological stress level, and human contact during this period increases the stress (Piñeiro *et al*., 2012). By carrying out the surveys after most of the kittens had been born, we minimized the disturbance to females during this sensitive period.

To avoid pseudoreplication of information, the presence of a mark on a plant was recorded only the first time it was detected, regardless of how many times the plant was urinated on during the study period.

For all the plants in the facilities, the following variables were recorded: the species of the plant; its maximum height

Table 1 Characteristics of the plants used as urine mark substrates for wildcats in the study. Abbr. is the abbreviation used for each plant species, and *N* is number of plants. *H* is the maximum height measured in centimetres from the soil to the top of the plant. *W* is the maximum width of the plant measured in cm. Mean and standard deviations are given for these measures. The numbers in the shape and foliage density columns represent the number of plants in each shape and foliage density categories. Shape: El, Elongated; Te, Tree; Cy, Cylindrical; Co, Conical; Cr, Crown; Sp, Spherical; C, Cake; Br, Branched; Tr, Triangular. Foliage density: VL, Very low; L, Low; M, Moderate; H, High; VH, Very high

Abbreviation	Common name	Scientific name	*N*	*H* *X* (SD) Cm	*W* *X* (SD) Cm	Shape	Foliage density
Au	Strawberry tree	*Arbutus unedo*	10	*127*(62.65)	*69.3*(21.01)	6Br, 3Cr, 1Sp	1VL, 3L, 2M, 1H, 3VH
Bs	Box wood	*Buxus sempervirens*	14	*92.29*(19.44)	*70.36*(15.66)	10Sp, 1Cr, 2Tr, 1El	2VL, 3L, 2M, 3H, 4VH
Bv	Barberry	*Berberis vulgaris*	6	*51.5*(20.76)	*42*(25.07)	2Br, 3Cr, 1Sp	1L, 2H, 3VH
Cb	Southern catalpa	*Catalpa bignonioides*	1	*144*(-)	*62*(-)	1Br	1VL
Ea	Russian silverberry	*Eleagnus angustifolia*	7	*78.29*(27.97)	*61.57*(19.94)	4Cr, 2Br, 1Sp	2VL, 2M, 1H, 2VH
Ej	Japanese euonymus	*Euonymus japonicum*	2	*65*(21.21)	*33*(8.49)	2Br	1M, 1H
Jc	Common juniper	*Juniperus communis*	7	*125.14*(52.93)	*37.86*(10.21)	4El, 2Co, 1Sp	7VH
Jp	Phoenicean juniper	*Juniperus phoenica*	2	*13*(4.24)	*15*(0.00)	2El	1VL, 1L
Oe	Olive	*Olea europaea*	6	*270.5*(43.11)	*260.67*(32.04)	6Te	2L, 1M, 1H, 2VH
Pg	Pomegranate	*Punica granatum*	6	*60.83*(11.43)	*19.17*(7.36)	5Br, 1Sp	2VL, 1L, 1H, 2VH
Ps	Blackthorn	*Prunus spinosa*	3	*62*(24.33)	*25*(12.29)	1El, 2Br	1VL, 2VH
Ra	Butcher's broom	*Ruscus acculeatus*	6	*49.5*(10.56)	*56.5*(11.15)	2C, 3Sp, 1Br	3VL, 3VH
Rc	Dog rose	*Rosa canina*	2	*32*(11.31)	*20*(14.14)	1Sp, 1Br	1VL, 1VH
Ro	Rosemary	*Rosmarinus officinalis*	15	*32.87*(7.63)	*17.67*(5.69)	3C, 1El, 2Br, 2Cy, 7Sp	13VL, 1L, 1VH
Sj	Austral cordgrass	*Spartina junceus*	6	*104.17*(15.33)	*56.17*(20.41)	1El, 1Sp, 4Br	2VL, 1L, 3VH
Tb	Common yew	*Taxus baccatta*	2	*99*(5.66)	*52.5*(13.44)	1El, 1Br	2L
Total			95	*88.91*(64.92)	*59.20*(58.48)	29Br, 11Cr, 27Sp, 2Tr, 6Te, 11El, 2Cy, 2Co, 5C	29VL, 15L, 8M, 10H, 33VH

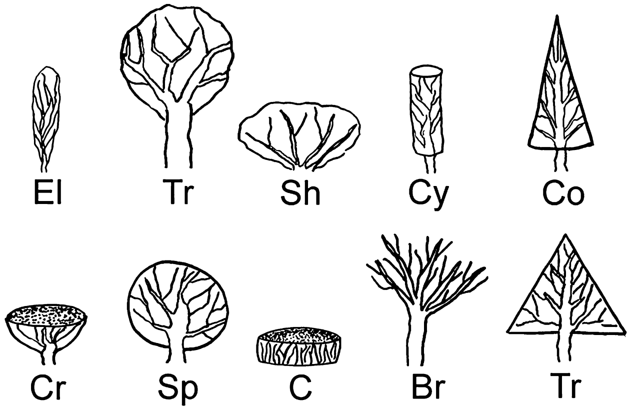

Figure 2 The different shape types of plants used as urine mark substrates for wildcats in the study. El: Elongated; Te: Tree; Cy: Cylindrical; Co: Conical; Cr: Crown; Sp: Spherical; C: Cake; Br: Branched; Tr: Triangular.

Figure 3 A wildcat displaying the rubbing behaviour on a repeatedly urinated common juniper plant. The dried leaves at the bottom of the plant are a consequence of the action of the fatty acid in the urine.

(measured in centimetres from the soil to the top of the plant); maximum width (measured in centimetres across the widest part of the plant); foliage density [a categorical measure based on visual estimation that ranges from one (very low density) to five (very high density)]; shape of the plant [assigned to one of the following categories (Fig. 2): elongated, tree, cylindrical, conical, crown, spherical, cake, branched, triangular]. Although in the wild there should be a general association between the species and the shape variables, the necessary pruning of the plants carried out in the CFSV to meet the space and management requirements meant that the association was not as strong as could be expected in the wild.

In addition, for each plant it was recorded whether it had been urinated on or not. The urinated plant was identified by the presence of a strong smell or by a characteristic pattern of dead leaves (Fig. 3). For each urinated plant, the height at which it was urinated on was recorded. If a plant was urinated on at least once during the study period, it was given the value 1 and otherwise 0.

Table 2 Results of the model selected by the Wald stepwise forward logistic regression performed with all data combined. It shows the estimated coefficient of the variable (*B*) with standard error (SE), the Wald statistics, the degree of freedom (d.f.) and their *P*-value (*P*). Correct percentage is the percentage of plants correctly assigned to the category urinated or non-urinated using this model

Variables	*B*	SE	Wald statistics	d.f.	*P*	Correct percentage
Step 1						
Species			1.1229931	15	1.000	
Au	19.01	28420.86	0.00	1	0.999	94.70
Bv	0.00	32817.54	0.00	1	1.000	
Bs	20.29	28420.86	0.00	1	0.999	
Cb	0.00	49226.22	0.00	1	1.000	
Ea	0.00	32226.19	0.00	1	1.000	
Ej	0.00	40193.07	0.00	1	1.000	
Jc	42.41	32226.19	0.00	1	0.999	
Jp	0.00	40193.07	0.00	1	1.000	
Oe	0.00	32817.54	0.00	1	1.000	
Ps	0.00	36691.10	0.00	1	1.000	
Pg	0.00	32817.54	0.00	1	1.000	
Rc	0.00	40193.07	0.00	1	1.000	
Ro	0.00	30256.30	0.00	1	1.000	
Ra	0.00	32817.54	0.00	1	1.000	
Sj	0.00	32817.54	0.00	1	1.000	
Constant	–21.20	28420.86	0.00	1	0.999	

Statistics

To study the variables related to the use of plants as urine marking substrates, the forward Wald stepwise logistic regression was used. The following variables were used as predictors: species, shape, maximum height, maximum width and foliage density. Among these, plant species, shape of plant and foliage density were converted to dummy variables. To study in more detail the plant selection as a urine substrate for the wildcats, the Jacob's selection index was calculated separately for each of the six facilities considering the variables retained by the logistic regression model, using the formula D = (r-p)/(r + p-2rp), where *r* is the proportion of the urinated plants at a facility made by a plant species and *p* is the proportional abundance of that species of the total plants at each facility. The Jacob's index ranges from −1, meaning maximum avoidance, to 1, meaning maximum preference. Jacob's index of 0 indicates that there is no selection of the resource (Jacobs, 1974). To check if the mean Jacob's index of the six facilities differs statistically from 0, we used Student's *t*-test (Palomares *et al*., 2001; Hayward *et al*., 2006).

We compared mean maximum height and mean maximum width between marked and non-marked plants using the Mann–Whitney *U*-test with Monte Carlo simulation. All the tests used in the study were two-tailed. The level of significance for every statistical test was $P < 0.05$. For all the statistical analyses, SPSS 15 software was used (SPSS Inc., Chicago, IL, USA).

Results

In the six facilities, 12 individual plants in total were urinated on at least once during the study period, ranging from one to four plants per facility ($X \pm$ SD = *2* ± 1.26, $n = 6$).

The results show that captive wildcats select the plant on which to make their urine mark based on species criteria. Of the marked plants, 58.33% were common junipers, 33.33% box wood and the rest 8.33% strawberry trees. We found that 100% of the common junipers ($n = 7$), 28.57% of the box wood ($n = 14$) and only 10% of the strawberry trees ($n = 10$) were marked. The model selected in the logistic regression analysis only includes the plant species variable and the constant. This model indicates that for a plant, belonging to one of these species increase the probability to be urinated on (Table 2).

The model retained by the logistic regression analysis, which include the species and the constant, allows for more accurate predictions than the null model, which only includes the constant (*Omnibus* $X^2 = 48.82$; d.f. = 15; $P = 0.000$). It allows us to correctly ascribe a plant as urinated or not urinated in 58.3% of the actually marked plants, and in 94.7% of the total plants, by just knowing if the plant belongs to one of these three species.

The logistic regression model fits the data well (Hosmer–Lemeshow $X^2_4 = 0.000$, $P = 1.000$) and explains an important part of the variability of the data (R^2 Nagelkerke = 0.756).

The results of the Jacob's index analysis shows that common juniper and strawberry trees have a mean Jacob's index (D) statistically different from 0: juniper was selected (*D* ± SD = *0.90* ± 0.12, $n = 6$, $t_5 = 18.610$, $P < 0.001$) and strawberry was avoided (*D* ± SD = *–0.78* ± 0.53, $n = 6$, $t_5 = -3.636$, $P = 0.015$) as a urinating substrate. Box wood shows a mean negative Jacob's index (*D* ± SD = *–0.21* ± 0.88, $n = 6$) but not significantly different from 0 ($n = 6$, $t_5 = -0.583$, $P = 0.585$). The remainder of the species, with a mean Jacob's index of –1, were avoided as a urinating substrate (Fig. 4).

No differences were found between marked and non-marked plants in their maximum height and maximum width

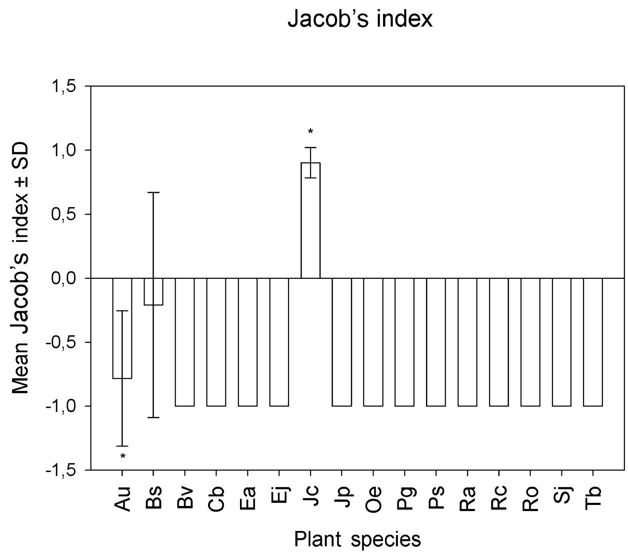

Figure 4 Selection of different plant species as urine mark substrates by wildcats. The figure shows the mean value of Jacob's index across the six facilities for each plant species, with standard deviations. Au, Strawberry tree *Arbutus unedo*; Bs, Box wood *Buxus sempervirens*; Bv, Barberry *Berberis vulgaris;* Cb, Southern catalpa *Catalpa bignonioides;* Ea, Russian silverberry *Eleagnus angustifolia*; Ej, Japanese euonymus *Euonymus japonicum*; Jc, Common juniper *Juniperus communis*; Jp, Phoenicean juniper *Juniperus phoenica*; Oe, Olive *Olea europaea*; Pg, Pomegranate *Punica granatum*; Ps, Blackthorn *Prunus spinosa*; Ra, Butcher's broom *Ruscus aculeatus*; Rc, Dog rose *Rosa canina*; Ro, Rosemary *Rosmarinus officinalis*; Sj, Austral cordgrass *Spartina junceus*; Tb, Common yew *Taxus baccatta*. *Signifies that the mean Jacob's index is statistically different from 0 for that plant.

($U = 355.5$, $n_1 = 12$, $n_2 = 83$, $P = 0.110$ and $U = 461.0$, $n_1 = 12$, $n_2 = 83$, $P = 0.684$, respectively). The mean (± SD) height at which wildcats urinated was 56.67 ± 14.54 cm ($n = 12$, range: 25–75 cm).

Discussion

Our observations at the CFSV indicated that wildcats selected the common juniper as the substrate on which to spray their urine marks.

From an evolutionary point of view, marks (urine and excrements) are a limited resource for animals (Gosling, 1982; Barja & de Miguel, 2004). Consequently, the marking behaviour should evolve to maximize the efficiency of the mark (Gosling, 1982; Vila, Uriós & Castroviejo, 1994). For example, the animal can use a conspicuous substrate to enhance the probability of a mark being detected (predictability) or to increase the active field of the mark (increasing diffusion), or the animal can use a substrate that regulates the release and durability of the mark (lengthening diffusion).

Depositing a mark in a conspicuous substrate enhances its visual efficiency, since the message is more likely to be detected by intruders, but it also can enhance its scent efficiency through a more extensive diffusion of the message by the wind (Barja & de Miguel, 2004; Piñeiro & Barja, 2012). The use of a conspicuous substrate has been documented in many carnivores such as wild (Vila *et al*., 1994; Barja *et al*., 2005; Barja, 2009) and captive (Barja & de Miguel, 2004) wolves, wild and captive otters, *Lutra lutra* (Reuther *et al*., 2000); red foxes, *V. vulpes* (MacDonald, 1979); Iberian lynxes, *Lynx pardinus* (Robinson & Delibes, 1988), and wildcats (Corbett, 1979; Piñeiro & Barja, 2012). For example, wildcats deposit their faecal marks in plants with greater diameters and visually conspicuous, increasing the probability of detection of the mark by both competitors and potential mates (Piñeiro & Barja, 2012). Although in general faecal marks have a stronger visual component than urine mark, in captivity, repeated urine mark over the same plant can cause a pattern of dead leaves, as we can see in the Fig. 3, influencing the visual component of the urine mark.

Both visual efficiency and wind diffusion of the mark should be an advantageous strategy in the wild, but they are likely to be irrelevant in captivity where the available space and environmental complexity is reduced. Conspicuity of a mark post depends on its size, shape, location or relative abundance in the surrounding environment. In the study of Piñeiro & Barja (2012), wildcats select herbaceous and woody plants, more conspicuous plants and those with greater diameter. In contrast, they do not select plants based on their height. The authors argue that this could be because these factors enhance the visual component and the latency of the faecal mark.

In our case, wildcats did not select plants as a urine substrate based on its width, height, foliage density or shape. In addition, the conspicuousness due to relative abundance and location of the three species urinated in the facilities, were similar. This suggests that in our study the wildcats did not select the substrate to urine spraying based on predictability or increased diffusion of the mark.

Controlling the release and degradation of the VOC of a mark is an advantageous strategy in marking efficiency. Marks could have their own mechanism to control the release and degradation of their chemical message. In that sense, Barja (2003) considers that one of the advantages of excrements over urine marks are that excrements release the VOC slower than urine marks, and therefore lengthen the durability of the mark. Nonetheless, felid urine has an incorporated mechanism which controls the release and durability of its own VOC. For example, domestic cats secrete in their urine large amounts of cauxin, which regulates the production of felinine (Hendriks *et al*., 1995; Miyazaki *et al*., 2008). The felinine decomposes, due to microbial activity and/or oxidation (Hendriks *et al*., 1995), to different sulphur-containing volatile compounds, which give a species-specific odour to the cat urine that acts as a putative pheromone (Miyazaki *et al*., 2006, 2008). Also, the lipids in the feline urine, although small in proportion, slow down the release and increase the longevity of VOC in the urine mark of lions (Andersen & Vulpius, 1999) and tigers (Burger *et al*., 2008).

Interactions with environment could regulate the release and degradation of the mark. Barja (2003) suggests that plants can be advantageous over other substrates because they slow down the release of the VOC and lengthen the durability of the mark. Piñeiro & Barja (2012) suggest that dense foliage coverage slow down the visual and olfactive degradation of the faecal marks. It is possible that the controlled release of the mark could occur by interactions of VOC of the mark with VOC of the surrounding plants. The VOC composition is species-specific for each plant species (Owen, Boissard & Hewitt, 2001), and modulates its interactions with other plants and animals (Llusià & Peñuelas, 2000; Baldwin, Kessler & Halitschke, 2002; Heil, 2008). It has been shown that cats can react to the VOC composition of different plants, such as cat nip, *N. cataria* (Hart & Leedy, 1985; Grognet, 1990); valerian, *Valeriana officinalis*; cat thyme, *Teucrium manum*; and oriental vine, *Actinidia polygama* (Case, 2003). Although we did not measure the VOC of the plants in our study, common junipers have significant concentrations of VOC with antioxidant activity such as α-pinene (Wei & Shibamoto, 2007; Misharina *et al*., 2009; Spinelli *et al*., 2011). Strawberry trees also have antioxidant compounds, although in smaller concentrations (Owen *et al*., 1997; Kivcak *et al*., 2001).

The selection criterion that wildcats used in Vallcalent was the species of the plant, being selected the common juniper, and box wood was used according to its availability. Selection of marking posts based on plant species by a carnivore has rarely been documented (Piñeiro & Barja, 2012).

The pattern of substrate selection by wildcats in our study coincides with the higher theoretical concentration of VOC with antioxidant activity of the common juniper. A possible explanation is that wildcats can detect differences in VOC composition, as shown with the cat nip (Hart & Leedy, 1985), and select plants with higher concentrations of antioxidant VOC, which possibly could slow down the degradation of the urine mark, and therefore enhance its olfactive efficiency. The variation of the visual efficiency of the urine mark depending of the plant species marked possibly influence the marking post selection by wildcats. However, we do not have enough data, and consider that more studies exploring the relationship between the VOC composition of different plant species and the degradation of the scent marks are needed. New studies should consider the VOC concentration, the marking frequency and the time dedicated to mark on each plant species, since these data should give us a better knowledge of marking behaviour of wildcats.

The captive environment is much simpler than the wild, where wildcats have many different species, sizes and differences in relative abundance of plants available to them. Therefore, wildcats could have a different urine marking behaviour in the wild than in captivity (Gittleman & McMillan, 1995). Nonetheless, our results suggest that recognition of plant species is important to marking behaviour in wildcats, possibly through the recognition of their VOC composition.

Acknowledgements

The authors are grateful to all the participants in this study, especially Rocío del Carmen Muñoz Ruiz, José Luis Santiago Fidalgo, Antonio López Fernández, Julián Sánchez Bravo and Yolanda Llop.

References

Andersen, K.F. & Vulpius, T. (1999). Urinary volatile constituents of the lion, *Panthera leo*. *Chem. Senses* **24**, 179–189.

Artois, M., Duchême, M.J., Pericard, J.M. & Xemar, V. (2002). Le chat domestique errant ou haret. In *Encyclopédie des Carnivores de France*: 1–50. Artois, M. & Maurin, H. (Eds). Puceul: Société Française pour l'Etude et la Protection des Mammifères.

Asa, C.S. (1993). Relative contributions of urine and anal-sac secretions in scent marks of large felids. *Am. Zool.* **33**, 167–172.

Baldwin, I.T., Kessler, A. & Halitschke, R. (2002). Volatile signaling in plant-plant-hervibore interaction: what is real? *Curr. Opin. Plant Biol.* **5**, 1–4.

Barja, I. (2003). Patrones de señalización con heces en el lobo ibérico. *Etología* **11**, 1–7.

Barja, I. (2009). Decision making in plant selection during the faecal-marking behaviour of wild wolves. *Anim. Behav.* **77**, 489–493.

Barja, I. & de Miguel, F.J. (2004). Variation in stimulus, seasonal context, and response to urine marks by captive Iberian wolves (*Canis lupus signatus*). *Acta Ethol.* **7**, 51–57.

Barja, I., de Miguel, F.J. & Bárcena, F. (2005). Faecal marking behaviour of Iberian wolf in different zones of their territory. *Folia Zool.* **54**, 21–29.

Benhamou, S. (1989). An olfactory orientation model for mammals' movements in their home ranges. *J. Theor. Biol.* **139**, 379–388.

Bradshaw, J.W.S. & Cameron-Beaumont, C. (2000). The signalling repertoire of the domestic cat and its undomesticated relatives. In *The domestic cat: the biology of its behaviour*: 67–94. Turner, D.C. & Bateson, P. (Eds). Cambridge: Cambridge University Press.

Burger, B.V., Viviers, M.Z., Bekker, J.P.I., le Roux, M., Fish, N., Fourie, W.B. & Weibchen, G. (2008). Chemical characterization of territorial marking fluid of male Bengal tiger, *Panthera tigris*. *J. Chem. Ecol.* **34**, 659–671.

Case, L.P. (2003). *The cat: its behavior, nutrition, & health*. 1st edn. Iowa: Blackwell Publishing Company.

Corbett, L. (1979). *Feeding ecology and social organization of wildcats* (Felis silvestris) *and domestic cats* (Felis catus) *in Scotland*. PhD thesis, University of Aberdeen, Aberdeen.

De Boer, J.N. (1979). The age of olfactory cues functioning in chemo communication among male domestic cats. *Behav. Process.* **2**, 209–226.

Feldman, H.N. (1994). Methods of scent marking in the domestic cat. *Can. J. Zool.* **72**, 1093–1099.

Gittleman, J.L. & McMillan, G.C. (1995). Mammalian behavior: lessons from captive studies. In *Conservation of endangered species in captivity: an interdisciplinary approach*:

355–377. Gibbons, E.F., Durrant, B.S. & Demarest, J. (Eds). New York: State University of New York Press.

Gorman, M.L. & Trowbridge, B.J. (1986). The role of odor in the social lives of carnivores. In *Carnivore behavioure, ecology and evolution*: 66–67. Gittleman, J.L. (Ed.). London: Chapman and Hall.

Gosling, L.M. (1982). A reassessment of the function of scent marking in territories. *Z. Tierpsychol.* **60**, 89–119.

Grognet, J. (1990). Catnip: its uses and effects, past and present. *Can. Vet. J.* **31**, 455–456.

Hart, B.L. & Leedy, M.G. (1985). Analysis of the catnip reaction: mediation by olfactory system, not vomeronasal organ. *Behav. Neural. Biol.* **44**, 38–46.

Hayward, M.W., Henschel, P., O'Brien, S.J., Hofmeyr, M., Balme, G. & Kerley, G.I.H. (2006). Prey preferences of the leopard (*Panthera pardus*). *J. Zool. (Lond.)* **270**, 298–313.

Heil, M. (2008). Indirect defence via tritrophic intearctions. *New Phytol.* **178**, 41–61.

Hendriks, W.H., Moughan, P.J., Tarttelin, M.F. & Woolhouse, A.D. (1995). Felinine: a urinary amino acid of Felidae. *Comp. Biochem. Physiol. B Biochem. Mol. Biol.* **4**, 581–588.

Jacobs, J. (1974). Quantitative measurement of food selection. A modification of the forage ratio and Ivlev's electivity index. *Oecologia* **14**, 413–417.

Kitchener, A.C. (1991). *The natural history of the wild cats*. 1st edn. London: Academic Press Limited.

Kivcak, B., Mert, T., Demirci, B. & Baser, K.H.C. (2001). Composition of the essential oil of *Arbutus unedo*. *Chem. Nat. Comp.* **37**, 445–446.

Law, G. & Tatner, P. (1998). Behaviour of a captive pair of clouded leopards (*Neofelis nebulosa*): introduction without injury. *Anim. Welfare* **7**, 57–76.

Leyhausen, P. (1979). *Cat behaviour. The predatory and social behaviour of domestic and wild cats*. 1st edn. New York, London: Garland SMTP Press.

Llusià, J. & Peñuelas, J. (2000). Seasonal patterns of terpene content and emission from seven Mediterranean woody species in field conditions. *Am. J. Bot.* **87**, 133–140.

Macdonald, D.W. (1979). Some observations and field experiments on the urine marking behaviour of the red fox, *Vulpes vulpes* L. *Z. Tierpsychol.* **51**, 1–22.

Mellen, J.D. (1993). A comparative analysis of scent-marking, social and reproductive behavior in 20 species of small cats (*Felis*). *Am. Zool.* **33**, 151–166.

Misharina, T.A., Terenina, M.B. & Krikunova, N.I. (2009). Antioxidant properties of essential oils. *Appl. Biochem. Microbiol.* **45**, 642–647.

Miyazaki, M., Yamashita, T., Suzuki, Y., Saito, Y., Soeta, S., Taira, H. & Suzuki, A. (2006). A major urinary protein of the domestic cat regulates the production of Felinine, a putative pheromone precursor. *Chem. Biol.* **13**, 1071–1079.

Miyazaki, M., Yamashita, T., Taira, H. & Suzuki, A. (2008). The biological function of cauxin, a major urinary protein of the domestic cat (*Felis catus*). In *Chemical signals in vertebrates*: 11: 51–60. Hurst, J.L., Beynon, R.J., Roberts, S.C. & Wyatt, T.D. (Eds). New York: Springer.

Natoli, E. (1985). Behavioural responses of urban feral cats to different types of urine marks. *Behaviour* **94–95**, 234–243.

Owen, S., Boissard, C., Street, R.A., Duckham, S.C., Csiky, O. & Hewitt, C.N. (1997). Screening of 18 Mediterranean plant species for volatile organic compound emissions. *Atmos. Environ.* **31**, 101–117.

Owen, S.M., Boissard, C. & Hewitt, C.N. (2001). Volatile organic compounds (VOCs) emitted from 40 Mediterranean plant species: VOC speciation and extrapolation to habitat scale. *Atmos. Environ.* **35**, 5393–5409.

Palomares, F. (1993). Faecal marking behaviour by free-ranging common genets *Genetta genetta* and Egyptian mongoose *Herpestes ichneumon* in southwestern Spain. *Z. Saugetierkd.* **58**, 225–231.

Palomares, F., Delibes, M., Revilla, E., Calzada, J. & Fedriani, J.M. (2001). Spatial ecology of iberian lynx and abundance of european rabbits in southwestern Spain. *Wildl. Monogr.* **148**, 1–36.

Piñeiro, A. & Barja, I. (2012). The plant physical features selected by wildcats as signal posts: an economic approach to fecal marking. *Naturwissenschaften* **99**, 801–809.

Piñeiro, A., Barja, I., Silván, G. & Illera, J.C. (2012). Effects of tourist pressure and reproduction on physiological stress response in wildcats: management implications for species conservation. *Wildl. Res.* **9**, 532–539.

Reuther, C., Dolch, D., Green, R., Jahrl, J., Jeffereies, D., Krekemeyer, A., Kucerova, M., Madsen, A.B., Romanowski, J., Roche, K., Ruiz-Olmo, J., Teubner, J. & Trindade, A. (2000). Surveying and monitoring distribution and population trends of the Eurasian otter (*Lutra lutra*). *Habitat* **12**, 1–148.

Roberts, S.C. (1997). Selection of scent-marking sites by klipspringer (*Oreotragus oreotragus*). *J. Zool. (Lond.)* **243**, 555–564.

Robinson, I.H. & Delibes, M. (1988). The distribution of faeces by the Spanish lynx (*Felis pardina*). *J. Zool. (Lond.)* **216**, 577–582.

Seiler, A., Krüger, H.H. & Festetics, A. (1994). Reaction of a male stone marten (*Martes foina* Erxleben, 1777) to foreign faeces within its territory: a field experiment. *Z. Saugetierkd.* **59**, 58–60.

Smith, J.L.D., McDougal, C. & Miquelle, D. (1989). Scent marking in free-ranging tigers, *Panthera tigris*. *Anim. Behav.* **37**, 1–10.

Spinelli, F., Cellini, A., Marchetti, L., Mudigere Nagesh, K. & Piovene, C. (2011). Emission and function of Volatile Organic Compounds in response to abiotic stress. In *Abiotic stress in plants – mechanisms and adaptations*: 367–394. Kumar Shanker, A. & Venkateswarlu, B. (Eds). Rijeka, Croatia: InTech.

Stahl, P. (1986). *Le chat forestier de l'Europe* (Felis silvestris *Schreber, 1777*). *Explotation des resources et organisation spatiale*. PhD thesis, Université de Nancy, Nancy.

Stahl, P. & Leger, F. (1992). Le chat sauvage (*Felis silvestris* Schreber, 1777). In *Encyclopédie des Carnivores de France*: 1–50. Artois, M. & Maurin, H. (Eds). Puceul: Société Française pour l'Etude et la Protection des Mammifères.

Vila, C., Uriós, V. & Castroviejo, J. (1994). Use of faeces for scent marking in iberian wolves (*Canis lupus*). *Can. J. Zool.* **72**, 374–377.

Wei, A. & Shibamoto, T. (2007). Antioxidant activities and volatile constituents of various essential oils. *J. Agric. Food Chem.* **55**, 1737–1742.