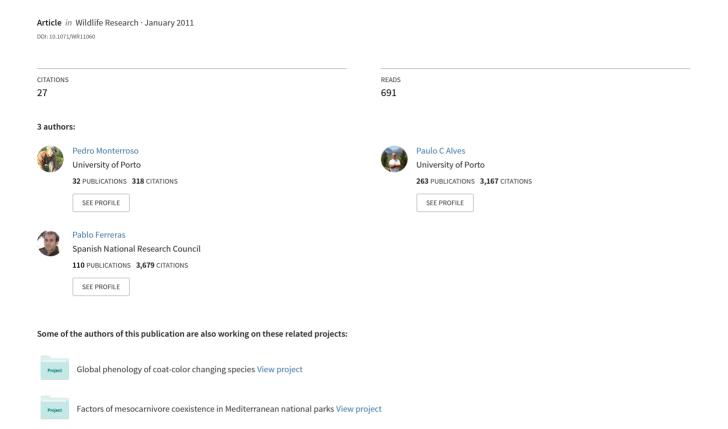
Evaluation of attractants for non-invasive studies of Iberian carnivore communities



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Abstract

Context. The estimation of population parameters for mammalian carnivore species is a challenging task because of their low densities and large home ranges, which make detection probabilities very low. Several factors, such as the species abundance, habitat structure or the use of an attractant affect carnivore detection probabilities; however, attractants are the most easily manipulated. Some previous research suggests that the use of effective attractants can significantly increase detection probabilities.

Aims. To assess the effectiveness of several attractants for Iberian carnivores, and to evaluate their usefulness for non-invasive survey methods.

Methods. The responses of seven carnivore species to six potential attractants were evaluated through cafeteria-like experiments with captive specimens. A selectivity index was applied to assess the relative attractiveness of each tested substance. The enclosure tests were followed by field trials with camera-trapping, using the most promising attractants for field evaluation of their efficiency.

Key results. Enclosure trials revealed that lynx urine was the most effective and generalist attractant because it successfully attracted six of the seven species tested. Rubbing behaviour was also induced in the greatest number of species by lynx urine. Field tests using a combination of lynx urine and valerian extract solution induced investigative behaviours in over 50% of all detection events in all species, with the exception of the Eurasian badger.

Conclusions. No single attractant is effective for all species. Nevertheless, a combination of lynx urine and valerian solution should efficiently attract the majority of species present in Iberian carnivore communities. Furthermore, some species exhibit a rubbing behaviour when they come in contact with the attractants. Regardless of the generalist efficiency of the lynx urine, other tested substances revealed promising results for single-species monitoring.

Implications. Our results provide a baseline for selecting attractants in survey and monitoring programs that focus on carnivore species. The rubbing behaviours exhibited by several of the species tested suggest the use of these attractants could improve the efficiency of field studies that rely on rub-pads for the collection of biological samples.

Additional keywords: attractant effectiveness, behavioural response, efficacy, Iberian carnivores, population monitoring, species detection.

Introduction

The definition of suitable management and conservation programs for wildlife strongly depends on an accurate assessment of target-species distribution, population size and trends (Williams *et al.* 2002). In the case of carnivore species, which occur in particularly low densities and have large home ranges, these parameters are especially difficult to estimate (Wilson and Delahay 2001; Long *et al.* 2008). The inconspicuous

habits along with human phobia of many carnivore species make the use of direct and invasive field methods laborious and sometimes subject species to unnecessary disturbance (Ballenberghe 1984; Michalski *et al.* 2007). For these reasons, non-invasive methods are broadly applied to estimate carnivore distributions (Moruzzi *et al.* 2002), abundance (Mondol *et al.* 2009) and population trends (Travaini *et al.* 2010). Among these, some require an active search of the species presence, whereas

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others (e.g. scent stations, hair-snaring and camera-trapping) rely on natural animal movement for data collection (Wilson and Delahay 2001; Long *et al.* 2008). The use of attractants that stimulate the investigative response of the target species has been reported to significantly increase the detection probabilities of carnivores (Hunt *et al.* 2007; Schlexer 2008; Thorn *et al.* 2009). Therefore, the use of attractants should generally be incorporated into sampling methods, which will increase the reliability of resultant data and allow for more robust estimates of population parameters (Mackenzie and Royle 2005; Long *et al.* 2008).

Attractants used in carnivore studies can be clustered into the following three groups (Schlexer 2008): baits – food items or other substances that attract an animal by appealing to its sense of taste or smell, usually intended to be consumed (Roughton 1982; Zielinski et al. 2005); lures - substances that attract an animal via sense of smell, sight or hearing (Harrison 1997); and natural attractants - objects in the existing environment, that are regularly used by animals as a part of their behavioural repertoire. Scent marks such as anal-gland secretions, urine or faeces can be included in both of the latter attractant types, and play an important role in the communication among sympatric competitors (Ralls 1971; Schlexer 2008). Because scent marks can remain effective for long periods of time, they are used by mammals to avoid aggressive encounters between competitors by allowing for spatial or temporal segregation, the assessment of competitive ability and the establishment of dominance relationships (Ralls 1971; Gosling and McKay 1990). Previous studies assessed the effectiveness of attractants, especially in North America and Australia (e.g. Fagre et al. 1983; Phillips et al. 1990; Clapperton et al. 1994; Edwards et al. 1997; McDaniel et al. 2000 among others); however, nearly all (\approx 90%) of these evaluations focus on canid or felid species such as coyotes (Canis latrans; Fagre et al. 1983; Phillips et al. 1990) and red foxes (Vulpes vulpes; Saunders and Harris 2000; Miguel et al. 2005) or feral cats (Clapperton et al. 1994; Edwards et al. 1997). To our knowledge, no study has focussed on the effectiveness of attractants for entire carnivore communities, with the exception of the study of Andelt and Woolley (1996), which targeted a mammal community of urban mammals in Colorado (USA). In addition, the few scientific studies on the efficiency of attractants for carnivores have yielded conflicting results (Schlexer 2008). Hence, carnivore attractants are still selected mostly on the basis of tradition (Schlexer 2008).

The Iberian carnivore community consists of 15 native and one introduced species. Despite the importance of carnivores in Iberian natural ecosystems, there is still a lack of knowledge regarding the distributions and population trends of many carnivore species in Portugal and Spain. In fact, three species have recently been classified as 'data deficient' in Portugal by the latest national red book revisions (Cabral *et al.* 2005) and distribution maps of several species are incomplete (Palomo *et al.* 2007). In the present paper, we evaluate the responses of seven carnivore species present in the Iberian ecosystems to six potential attractants. Our research objectives were to evaluate which attractants are more efficient for each species and to identify combinations of attractants that are effective for the widest range of carnivore species in the Iberian carnivore communities.

Materials and methods

Enclosure facilities, animals and attractants tested

The enclosure tests were conducted in two zoological facilities that harbour autochthonous species of Iberian vertebrate fauna. The Cañada Real Open Center (CROC) is located 48 km west of Madrid (Spain), and the Parque Biológico de Gaia (PBG) is located 10 km south of Porto (Portugal). The species tested at the CROC were red fox (1F), European wildcat (Felis silvestris; 1M and 2F) and Iberian wolf (Canis lupus signatus, 3M and 2F). Common genet (Genetta genetta; 1M and 1F), stone marten (Martes foina, 1M), Eurasian badger (Meles meles, 1M and 1F) and polecat (*Mustela putorius*; 8 individuals of unknown sex) were tested in the PBG. All individuals of the same species from each facility were kept in the same enclosure. Because of logistic constraints, individual marking of the tested specimens was not possible; therefore, we were incapable of assigning behavioural responses to specific individuals. All animals included in the tests were treated in compliance with guidelines outlined by animal ethics committees in Spain and Portugal, as part of the project CGL2009-10741.

The tested attractants were selected on the basis of their traditional use in carnivore studies, and included the following: Collarum Canine Bait (Wildlife Control Supplies, East Granby, Connecticut, USA), a commercial canid-specific attractant; valerian-extract solution, containing valeric acid found in urine and anal-sac secretions of coyote and fox (Saunders and Harris 2000), and described as a felid-specific attractant (Childers-Zadah 1998; Raal et al. 2007); fatty acid scent (FAS), a mixture of seven volatile fatty acids found in fermented egg (Roughton 1982), commonly used as a generalist carnivore attractant in North America (Roughton and Sweeny 1982); lynx (Lynx lynx) urine (obtained from captive specimens (1M and 1F) kept in the CROC); red-fox urine, obtained from captive red foxes (2M and 2F), held at Castilla La-Mancha University facilities; and a homogenised solution of stone-marten excrements (obtained from the captive specimen held at PBG). The urine and excrement solution used to test as attractants were frozen on collection, and kept frozen until the day they were used in the enclosure and field trials.

Experimental procedure

All attractants were tested simultaneously, in a cafeteria-like experiment (Rodgers 1990; Saunders and Harris 2000). The lures were included in a plastic tube ($\emptyset = 1$ cm; depth=3 cm) filled with cotton wool, which was sprayed with 3 mL of attractant. The plastic tubes were attached horizontally to wooden stakes, with the tube mouth facing outwards at a height of ~30 cm above ground. Six wooden stakes, each with a different attractant, were placed inside the enclosures, maintaining a distance of no less than 70 cm from each other. Tests were conducted between December 2008 and January 2009.

Each of the tested animals was exposed to the attractants for 3 h, during a period they were known to be active (as assessed by the facility keepers), namely during the morning for the species present at CROC and after sunset for the species present at PBG. By focusing the trials on periods of each specimen's activity, their response to the attractants was

expected to be maximised. All animal movements were recorded by a video digital camera, model CAMCOLBUL2DC (Velleman, Gavere, Belgium), set so that it could include all six attractants in the frame area. Artificial illumination was used in the enclosures tested during night-time.

We considered that an animal had an investigative response whenever at least one of three behaviours, namely sniff, lick/bite and/or rub, was observed towards a specific attractant. Each individual response was adequately classified as one of the predefined behaviours and its intensity (time spent exhibiting that behaviour) was registered.

Statistical analysis

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Because preference indexes are affected by individual variation, and individual identification of the tested animals was not possible because of logistic constraints, data were standardised to allow for comparisons among species and experimental treatments. The standardisation was performed by using the following equation:

$$SIT = \frac{\frac{\sum IIT}{NI}}{TP},$$

where SIT is standardised investigation time, IIT is individual investigation time (in seconds), NI is number of individuals in the captive trial and TP is trial period (in hours).

To evaluate the preference for a specific attractant in detriment of the others, we applied the modified Ivlev's selectivity index (Ivley 1961), adapted by Jacobs (1974, hereafter JSI). This index is broadly applied in ecological studies to evaluate resource selection, whether the resources are food items (Toft 1980) or habitat types (Palomares et al. 2000). Here, we used this index to evaluate the selection towards attractants, considering that all of them were equally available to the tested animals. The total time spent investigating the attractants was considered as the time the animal was predisposed to investigate the lures set in the enclosure; therefore, the availability for each attractant was considered to be $1/6 \times \sum$ (time interacting with attractants). The significance of the difference between the obtained index value and zero (i.e. no selection) was evaluated by bootstrap resampling (100 replicates) (Manly 1997) and by recalculating the JSI for each bootstrap sample. We then determined the average index, standard deviation and 95% confidence intervals for each attractant and species. We considered an attractant as positively selected whenever the 95% CI of the JSI was positive and did not overlap zero. These attractants scored '+1'. Because the main purpose of the present work was to evaluate the carnivores' relative preference for attractants, and because with the applied experimental design, we could not evaluate behaviour of independent species towards each of them, we did not consider 'avoidance' as a possible outcome. For that reason, those attractants that obtained 95% CI with negative values and those that overlapped zero were aggregated into the score '0'. The sum of the scores of each attractant for all the tested species was considered as an overall measure of performance (OMP), and used to rank their efficiency for the Iberian carnivore community.

Field tests

After the enclosure tests, we selected a combination of attractants for field trials that promoted a significant investigative response on the maximum number of species.

The field tests were performed in two distinct areas in the Iberian Peninsula, with Mediterranean pluviseasonal continental bioclimates (Rivas-Martínez et al. 2004). These included the Guadiana Valley Natural Park (GVNP), located in southern Portugal, and the Cabañeros National Park (CNP), located in central Spain, in the region of Castilla La-Mancha. The natural vegetation in the GVNP was dominated by the *Myrto communis—Quercetum rotundifoliae* series with other subserial stages (Costa et al. 1998), whereas the vegetation in the CNP was dominated by the *Pyro-Quercetum rotundifoliae* series and other subserial stages (Rivas-Martinez 1981).

The sampling design in each study area followed a grid-sampling scheme, composed by 1-km² grid squares. Camera traps, model Leaf River IR5 (LeafRiver OutDoor Products, Taylorsville, Mississippi, USA), were placed on every other vertex of the grid squares, resulting in a sampling grid of ~1.4 km (which corresponds to the distance between diagonal grid nodes). A circular area of 250-m radius surrounding each grid node was inspected for carnivore paths before placement of the camera trap. The final location of camera traps corresponded to areas of easy access and potentially good detection probability within the mentioned buffer. The distance (mean \pm s.d.) between neighbouring camera stations was of 1203 ± 231 m at GVNP and 1220 ± 238 m at CNP. Camera traps were maintained in the field for a minimum period of 28 days and were inspected for battery or card replacement every 7–10 days.

Attractants were placed in the field at a distance of 2-3 m from the camera traps. The selected attractants were deployed in separated, perforated plastic tubes supported by a wooden stake, at a distance of 10-15 cm from each other and ~ 30 cm above the ground. A volume of 5 mL of each attractant was sprayed into a cotton gaze held inside each plastic tube. Attractants were rebaited every 7-10 days.

The GVNP was sampled from 27 July to 6 September 2009 and the CNP was sampled from 24 September to 28 October 2009. We chose this season for the field trials because it corresponds to the time when the offspring of most medium-sized carnivores from that year become independent (Blanco 1998). Therefore, we would expect a higher number of contacts than during the breeding season.

We considered a series of photographs of the same species within a 30-min interval as dependent events (Kelly *et al.* 2008). Therefore, only detections of the same species separated in time over 30 min were considered for this analysis, to reduce the possibility of the same animal being captured more than once in the same camera trap. Because the field trials were included in a carnivore-community research project, which required a constant and balanced effort of the entire study areas, we could not apply traditional 'control *v*. treatment' experimental protocol during field trials. Nevertheless, despite being set close to one another, the observed animal behaviours (such as sniffing, rubbing or marking) elicited by each of the attractants could be unambiguously identified from the photographs and were registered. The proportion of each observed response over the

total detections for each species was calculated as an index of attractant efficiency.

Results

Captivity tests

A total of 21 h of enclosure tests revealed distinct strengths in the behavioural responses among the species and attractants evaluated. Lynx urine scored the highest of the six attractants evaluated, because it was effective for six of the carnivore species tested (OMP=+6). Only the stone marten did not spend significantly more time investigating lynx urine than what would be expected by chance.

The Collarum attractant was the second top-scored attractant (OMP = +4). This substance stimulated a significant investigative behaviour on the Iberian wolf, European wildcat, Eurasian badger and red fox. FAS effectively attracted the Iberian wolf, genet and stone marten (OMP = +3). The remaining attractants were effective for less than half of the species tested (OMP = +2, +1 and 0, for the valerian solution, red-fox urine and stone-marten excrements, respectively; Fig. 1).

As for the species responses, the Iberian wolf, European wildcat and genet revealed a significant interest for half of the substances they were exposed to (n=3; Fig. 1). The Eurasian badger, the polecat and the red fox investigated two of attractants significantly more than expected by chance. The stone marten revealed a significant interest only for FAS.

The strength of the responses towards the elected attractants also varied among species (Table 1). Because of the high range of strength of responses observed for the different species and attractants, data were summarised with the median and the geometric mean, which reduced the effect of extreme values. The Iberian wolves and genets exhibited the strongest responses

to the positively selected attractants. Each individual of these species spent, on average, between 38.0 (±3.8, s.d.) and $43.8 (\pm 9.9) \,\mathrm{s} \,\mathrm{h}^{-1}$ (Iberian wolves) and between $69.2 (\pm 23.6)$ and 92.7 (\pm 22.8) s h⁻¹ (genets) investigating them. Their overall investigation times were also the highest of all species (Table 1). The average intensity of responses by red foxes towards the positively selected attractants was 21.1 (\pm 7.1, s.d.) and $34.0 (\pm 7.4) \,\mathrm{s} \,\mathrm{individual}^{-1} \,\mathrm{h}^{-1}$. The summarised responses of this species revealed an intermediate response towards the attractants (Table 1). The overall strength of responses of European wildcats, stone martens, Eurasian badgers and polecats were all below 4 s individual⁻¹ h⁻¹ (geometric mean, Table 1). However, the European wildcat did not spend any time at all investigating stone-marten excrements, but revealed intermediate investigation strengths towards the positively selected attractants $(10.4 \pm 2.2 \text{ to } 18.3 \pm 9.4 \text{ s individual}^{-1} \text{ h}^{-1};$ Table 1). The stone marten was only significantly more attracted towards the FAS than expected by chance, with a moderate response $(22.7 \pm 5.5 \text{ s individual}^{-1} \text{ h}^{-1})$. The Eurasian badger and the polecat displayed the weakest responses, with investigative responses below 10 s individual⁻¹ h⁻¹ towards the positively selected attractants (Table 1).

The rubbing behaviour was rarely exhibited, except by the Iberian wolf and the genet (Table 2). For this reason, the JSI could not be applied to this behaviour. Nevertheless, some indications can be obtained from the animals' rubbing responses. Although the Iberian wolf exhibited rubbing behaviour for all attractants, this behaviour was more intense towards FAS (24.5 s individual $^{-1}$ h $^{-1}$). Genets also displayed a generalist rubbing behaviour; however, the intensity of these responses was stronger towards FAS, lynx urine and red-fox urine. The red fox rubbed on Collarum, lynx urine and red-fox urine; however these responses were very weak (<2 s individual $^{-1}$ h $^{-1}$). Both

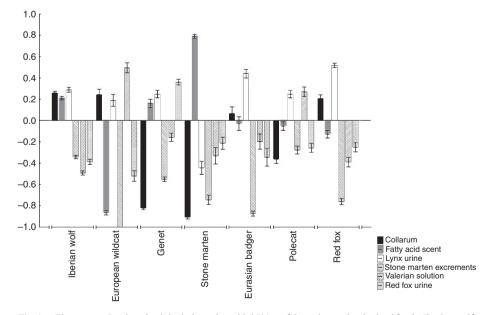


Fig. 1. The average Jacobs selectivity index value with 95% confidence intervals, obtained for the Iberian wolf, European wildcat, genet, stone marten, Eurasian badger, polecat and red fox towards each of the tested attractants during the enclosure tests in Cañada Real Open Center, Spain, and Parque Biológico de Gaia, Portugal, between December 2008 and January 2009.

Table 1. Mean investigation time

The mean $(\pm s.d.)$ investigation time (s individual⁻¹ h⁻¹) the Iberian wolf, European wildcat, genet, stone marten, Eurasian badger, polecat and red fox spent investigating attractants in enclosures in Cañada Real Open Center, Spain, and Parque Biológico de Gaia, Portugal, between December 2008 and January 2009. Zero values were replaced by the value 0.001 for the calculation of the geometric mean

Species	N	Collarum	FAS	Lynx urine	Stone-marten excrements	Valerian solution	Red-fox urine	Median	Geometric mean
Iberian wolf	5	41.1 (±6.0)	38.0 (±3.8)	43.8 (±9.9)	14.3 (±2.2)	10.3 (±2.4)	13.3 (±3.8)	26.15	22.62
European wildcat	3	11.1 (±3.8)	$0.6 (\pm 0.5)$	10.4 (±4.4)	$0.0 (\pm 0.0)$	18.3 (±9.4)	$2.7 (\pm 1.7)$	6.55	1.23
Genet	2	$6.0 (\pm 3.2)$	69.2 (±23.6)	$78.8 (\pm 24.0)$	16.5 (±4.2)	39.7 (±13.1)	92.7 (±22.8)	54.45	35.46
Stone marten	1	$0.4 (\pm 0.3)$	22.7 (±5.5)	$2.8 (\pm 2.0)$	$1.1 (\pm 1.0)$	$3.8 (\pm 2.9)$	4.3 (±2.1)	3.3	2.78
Eurasian badger	2	$3.1 (\pm 1.6)$	$2.6 (\pm 1.5)$	5.4 (±2.0)	$0.2~(\pm 0.2)$	$2.0 (\pm 1.3)$	1. 6 (± 1.2)	2.6	1.77
Polecat	8	$2.0 (\pm 0.8)$	$3.5 (\pm 1.0)$	$5.7 (\pm 1.7)$	$2.3 (\pm 0.8)$	$6.3 (\pm 2.6)$	$2.5 (\pm 1.1)$	3	3.36
Red fox	1	21.1 (±7.1)	11.9 (±3.8)	$34.0 (\pm 7.4)$	$2.3 (\pm 1.5)$	$7.3 (\pm 3.6)$	9.6 (±3.8)	10.75	10.55
Median		6	11.9	10.4	2.3	7.3	6.95		
Geometric mean		5.45	8.73	14.00	0.83	8.26	8.37		

Table 2. Mean rubbing time

The mean (±s.d.) rubbing time (sindividual⁻¹ h⁻¹) the Iberian wolf, European wildcat, genet, stone marten, Eurasian badger, polecat and red fox spent investigating attractants in enclosures in Cañada Real Open Center, Spain, and Parque Biológico de Gaia, Portugal, between December 2008 and January 2009

Species	N	Collarum	FAS	Lynx urine	Stone-marten excrements	Valerian solution	Red-fox urine	No. of attractants with rubbing responses
Iberian wolf	5	3.6 (±0.8)	24.5 (±3.5)	1.8 (±0.7)	5.0 (±1.2)	1.3 (±0.4)	3.9 (±1.3)	6
European wildcat	3	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	15.7 (±9.4)	$0.0 (\pm 0.0)$	1
Genet	2	$0.7 (\pm 0.6)$	57.1 (±23.0)	69.9 (±23.9)	$7.6 (\pm 3.2)$	22.4 (±12.4)	74.2 (±23.4)	6
Stone marten	1	$0.0~(\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	0
Eurasian badger	2	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.2 (\pm 0.2)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	1
Polecat	8	$0.0~(\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$3.7 (\pm 2.5)$	$0.0 (\pm 0.0)$	1
Red fox	1	1.4 (±1.1)	$0.0 (\pm 0.0)$	$1.1 (\pm 0.8)$	$0.0 (\pm 0.0)$	$0.0 (\pm 0.0)$	$0.3~(\pm 0.3)$	3
No. of species with rubbing responses		3	2	4	2	4	3	

European wildcats and polecats displayed rubbing behaviours towards only one attractant, the valerian solution; whereas the Eurasian badger rubbed only against the lynx urine. The stone marten was the only species that did not rub on any of the tested attractants.

Field tests

Although the combination of FAS attractant+lynx urine was effective for all species tested during the captivity trials (Fig. 1), yielding a joint OMP score of '+7', the combination of lynx urine+valerian solution induced rubbing behaviour in a greater number of species (n = 6; Table 2), suggesting a greater efficiency. Furthermore, previous field experience with lynx urine revealed its effectiveness for the attraction of the stone marten (Monterroso 2006). For these reasons, the combination of attractants selected for the field trials was lynx urine+valerian solution.

During field tests, we detected eight carnivore species on both study areas, six of which were evaluated during the enclosure tests, whereas the following two were not: the Egyptian mongoose (*Herpestes ichneumon*) and the least weasel (*Mustela nivalis*). Overall, 472 carnivore detections were obtained, 126 in GVNP and 346 in CNP. All species, except the Eurasian badger, displayed interactive behaviours (sniffing, biting or marking) towards some of the lure attractants on more

than 50% of the detections (Table 3). The highest scores were obtained by the red fox, the wildcat, the stone marten and the Egyptian mongoose, which interacted with the attractants on at least 70% of the detection occasions.

Of the two available attractants in the field tests, lynx urine obtained higher proportion of interactions for all species, except for the least weasel and the Egyptian mongoose, which interacted more with the valerian solution than with lynx urine.

Discussion

Despite the small sample size available for the enclosure tests, the results suggest that none of the tested attractants alone is significantly more efficient than the others for all carnivore species tested in our study. The lynx urine was the most efficient attractant for the majority of species, because only the stone marten did not spend more time than expected by chance investigating it. The Eurasian lynx does not occur naturally in the Iberian Peninsula; however, it co-occurs elsewhere with most of the carnivore species present in Iberian ecosystems (e.g. wolf, red fox, European wildcat, stone marten, Eurasian badger and polecat) (Mitchell-Jones *et al.* 1999). Where it occurs, the Eurasian lynx is a top predator, known to kill smaller carnivores (Palomares and Caro 1999). In the Iberian Peninsula, its congener, Iberian lynx (*Lynx pardinus*), is

Table 3. Field-trial carnivore responses

The responses exhibited by the red fox, European wildcat, stone marten, polecat, least weasel, Eurasian badger, genet and Egyptian mongoose towards valerian extract and lynx urine during field trials in Guadiana Valley Natural Park Portugal and Cabañeros National Park, Spain, July-October 2009

Species	GVNP	No. of detections CNP	Total	Proportion of investigative behaviours over all detections	Proportion of attractant-specific investigative occasions over all occasions with investigative behaviour	
					Lynx urine	Valerian solution
Red fox	41	263	304	0.75	0.69	0.25
European wildcat	22	4	26	0.81	0.67	0.14
Stone marten	16	42	58	0.72	0.52	0.17
Polecat	6	0	6	0.67	0.50	0.00
Least weasel	2	0	2	0.50	0.00	1.00
Eurasian badger	12	16	28	0.18	0.80	0.20
Genet	9	21	30	0.53	0.69	0.19
Egyptian mongoose	18	0	18	0.78	0.29	0.57
Mean (±s.d.)				0.62 (±0.21)	0.52 (±0.26)	0.32 (±0.32)

sympatric with all species tested (Palomo et al. 2007), being superior competitor to most of the mesocarnivores, often killing them (Palomares and Caro 1999). Several studies on carnivores suggest that individuals can identify odours from a competitor species (Erlinge and Sandell 1988; Harrington et al. 2009), even when it has never come in contact with them before (Harrington et al. 2009). The lack of avoidance and the rubbing behaviour exhibited by several species in enclosure tests, and the frequent investigative behaviour towards lynx urine from most Iberian carnivores observed in field trials suggest that the predator's scent promotes investigative and scent-marking behaviours from other carnivores. This finding is in accordance with Harrington et al. (2009), who found little support for an avoidance of otter (Lutra lutra) odour by American mink (Mustela vison). Similarly, Howard et al. (2002) found that coyotes and bobcats (Lynx rufus) were attracted to each other's faeces. These two species are known to react negatively to each other (Wilson et al. 2010), and therefore this attraction to the faeces of the competing species could be the result of investigative processes that allow for the employment of adequate behavioural strategies for coexistence (Wilson et al. 2010). Our data suggest that the presence of lynx scent in the 'familiar' surroundings of captive and free-living Iberian carnivores must be understood by the animals as the presence of a competitor or a threat, which induces an investigative behaviour and even scent marking of their own. This was observed in red foxes, which urinated and rubbed against the scent, and in stone martens and genets that defecated on it (P. Monterroso, pers. obs.).

The attractiveness of valerian extract on cats has been referred by other authors (Raal et al. 2007; Klar et al. 2009; Jerosch et al. 2010), although its effectiveness has never been assessed. Our results from the enclosure tests comply with the suggestion of these previous authors because it induced not only a significant investigative response from wildcats, but it also promoted a strong rubbing behaviour. Such a response to valerian scent is traditionally known and has resulted in its use in field studies for hair snaring (Djabalameli 2005). Similar behaviour is found in other felid species towards another plant extract, the catnip (Nepeta cataria; Edwards et al. 1997; Harrison 1997; McDaniel

et al. 2000). Interestingly, our field tests revealed that wildcats showed more interest in lynx urine than they did in valerian solution. Edwards et al. (1997) suggested that the efficacy of scent-based lures may be strongly influenced by seasonal changes in reproductive behaviour, becoming particularly less effective when reproductive behaviour is relatively subdued. Our field trials were performed in late summer, when territoriality is reduced and no reproductive activity is expected to occur (Sunquist and Sunquist 2002). Therefore, it is possible that, in this season, wildcats are more interested in a potential competitor and/or predator within their home ranges than with a reproduction-appealing scent.

The Collarum Canine Bait and FAS showed significant relative efficiency for some species; however, the overall evaluation of these attractants suggests that they are not an adequate choice for the entire Iberian carnivore communities. These attractants can be better used for studies focusing on a limited number of species. As expected, the Collarum Canine Bait could be efficient for canid species, such as the wolf or the red fox. Our results suggest that, in Iberian carnivore assemblages, FAS should be used only in studies focussed on the wolf, the genet and the stone marten, despite being broadly used in the United States in carnivore surveys (Roughton and Sweeny 1982) and being a recommended attractant for canids and temperate felids (Schlexer 2008). The homogenised solution of stone-marten excrements was ineffective for any of the species tested. The stone marten, as other mustelids, uses faeces for scent marking (Hutchings and White 2000; P. Monterroso, unpubl. data). However, scent marking does not occur all the time. Mammals tend to mark when they are both intolerant of, and dominant to, other members of the same species or when they come into contact with scent of competitor species (Ralls 1971; Miguel et al. 2005). The captive stone marten from which excrements where collected exhibited abnormal behaviour during enclosure trials. A possible outcome of the abnormality in this specimen's behaviour might have been non-scent marking of faeces, which could explain the lack of interest displayed by all species towards this substance. Furthermore, as excrements where presented in the form of a solution, there was no visual stimuli, which also affects the scat

attractiveness to other carnivores (Howard et al. 2002). These two factors combined may have been responsible for the lack of interest demonstrated by all carnivores in the homogenised solution of stone-marten excrements. Red-fox urine was only effective for genets, and promoted a strong rubbing response in this species. To our knowledge, no competitive interaction has ever been described involving these two species. We acknowledge the fact that the captive environments in which the tested animals are maintained might, to some extent, influence their behaviour towards interspecific scents. Nevertheless, genets often occur in sympatry with red foxes (Palomo et al. 2007) and therefore a possible subtle interaction might exist between these two species.

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Rubbing behaviour in enclosure tests was exhibited by almost all species, but only towards a reduced number of attractants and very few times. This kind of behaviour is frequent across different kinds of mammals, and serves the purpose of leaving their scent in response to the scent of a stranger (Ralls 1971). This behaviour has been observed in felids (Clapperton *et al.* 1994; Harrison 1997; Thomas *et al.* 2005) and canids (Harrison 2006) and serves as the basis for hair-sample collection in field surveys (McDaniel *et al.* 2000; Thomas *et al.* 2005; Weaver *et al.* 2005; Schmidt and Kowalczyk 2006). Although none of the tested attractants elicited a strong rubbing response from more than two species, the lynx urine and the valerian solution induced this type of behaviour for the largest number of species.

Most evaluations of carnivore attractants involve captive animals and their effectiveness is assessed by exposing the animals to the evaluated substances (Phillips et al. 1990; Harrison 1997; Saunders and Harris 2000); however, fieldtesting is more appropriate because it incorporates environmental factors and population density (Schlexer 2008). Because we could not apply an adequate experimental protocol for our field trials, it is not possible to unequivocally state that the use of attractants provides higher encounter rates than does not using any attractant at all. Nevertheless, our results suggest that the combination of lynx urine and valerian solution elicits investigative behaviours in nearly all target species. These results not only support those provided by the enclosure tests regarding the efficiency of lynx urine for most carnivores, but they also revealed that this attractant might also attract the stone marten and, to some extent, the Egyptian mongoose (not evaluated in captivity trials).

Our findings suggest that using lynx urine as an attractant in non-invasive survey methods would increase detection probability relative to the remaining attractants tested because this substance is actively investigated by most carnivore species present in Iberian communities. Furthermore, our results demonstrate that several of these species (e.g. the wildcat, the wolf or the red fox) exhibit rubbing behaviour in the presence of this attractant, a fact that allows for the use of rub pads to acquire hair samples that could be later used for genetic evaluation, e.g. in mark-recapture studies. Another advantage of this attractant is that lynx specimens exist in most zoological facilities, making it accessible to wildlife researchers. Indeed lynx urine fits the criteria of Fagre et al. (1983), who suggested that an adequate lure should be (1) uniform in quality, (2) high in availability, (3) low in cost, (4) easy to handle and (5) highly attractive to target species. Some factors, such as seasonality, might affect the

composition of the urine samples collected throughout the year, thus compromising Fagres' first criteria. However, urine samples from captive animals generally fulfill these requirements because captive animals are maintained at near constant conditions, regarding feeding and environment, all year long (Howard *et al.* 2002).

The fact that the use of the same lures results in varying degrees of success (Schlexer 2008) highlights the importance of carefully replicating and evaluating attractant studies so as to obtain standardised and consistent patterns of target-species responses. To our knowledge, the present study is the first attempt to evaluate the efficiency of attractants for Iberian carnivore species. Despite the low number of captive animals tested and the seasonal characteristics of field sampling, our tests reveal patterns of relative attractant efficiency for Iberian carnivores, suggesting that for studies that focus on the assessment of carnivore assemblages similar to those present in the Iberian Peninsula, lynx urine should be a preferred lure over markings of smaller species or other commercial lures.

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References

Andelt, W. F., and Woolley, T. P. (1996). Responses of urban mammals to odor attractants and a bait-dispensing device. *Wildlife Society Bulletin* **24**, 111–118.

Ballenberghe, V. V. (1984). Injuries to wolves sustained during live-capture. The Journal of Wildlife Management 48, 1425–1429. doi:10.2307/3801811

Blanco, J. C. (1998). 'Mamíferos de España, 2 Vols.' (Ed. Planeta: Barcelona, Spain.)

Cabral, M. J., Almeida, J., Almeida, P. R., Dellinger, T., Ferrand de Almeida, N., Oliveira, M. E., Palmeirim, J. M., Queiroz, A. I., Rogado, L., and Santos-Reis, M. (Eds) (2005). 'Livro Vermelho dos Vertebrados de Portugal.' 2nd edn. (Instituto da Conservação da Natureza/Assírio Alvim: Lisboa, Portugal). [In Portuguese.]

Childers-Zadah, V. (1998). Use of valerian plant and/or root as a scent attractant for stimulating canines and felines. US Patent 5,786,382.

Clapperton, B. K., Eason, T., Weston, R. J., Woolhouse, A. D., and Morgan, D. R. (1994). Development and testing of attractants for feral cats, *Felis catus* L. *Wildlife Research* 21, 389–399. doi:10.1071/WR9940389

Costa, J. C., Aguiar, C., Capelo, J. H., Lousã, M., and Neto, C. (1998). Biogeografia de Portugal continental. *Quercetea* **0**, 5–56.

Djabalameli, J. (2005). Hair catching with the aid of the 'Lure Stick'. Proof of wildcat (*Felis silvestris*) existence by genetic analysis. A project by BUND and Bayerisches Landesamt für Umweltschutz. In 'Biology and Conservation of the European Wildcat (*Felis silvestris silvestris*)' seminar. Nienover, Germany

- Edwards, G. P., Piddington, K. C., and Paltridge, R. M. (1997). Field evaluation of olfactory lures for feral cats (*Felis catus L.*) in central Australia. Wildlife Research 24, 173–183. doi:10.1071/WR96013
- Erlinge, S., and Sandell, M. (1988). Coexistence of stoat, *Mustela erminea*, and weasel, *M. nivalis*: social dominance, scent communication, and reciprocal distribution. *Oikos* 53, 242–246. doi:10.2307/3566069
- Fagre, D. B., Howard, W. E., Barnum, D. A., Teranishi, R., Schultz, T. H., and Stern, D. J. (1983). Criteria for the development of coyote lures. In 'Vertebrate Pest Control and Management Materials: Fourth Symposium.' ASTM STP 817. (Ed. D. E. Kaukeinen.) pp. 265–277. (American Society for Testing Materials: Philadelphia, PA.)
- Gosling, L. M., and McKay, H. V. (1990). Competitor assessment by scent matching: an experimental test. *Behavioral Ecology and Sociobiology* 26, 415–420. doi:10.1007/BF00170899
- Harrington, L. A., Harrington, A. L., and Macdonald, D. W. (2009). The smell of new competitors: the response of American mink, *Mustela vison*, to the odours of otter, *Lutra lutra* and polecat, *M. putorius*. *Ethology* 115, 421–428. doi:10.1111/j.1439-0310.2008.01593.x
- Harrison, R. L. (1997). Chemical attractants for central American felids. Wildlife Society Bulletin 25, 93–97.
- Harrison, R. L. (2006). A comparison of survey methods for detecting bobcats. Wildlife Society Bulletin 34, 548–552. doi:10.2193/0091-7648(2006)34 [548:ACOSMF]2.0.CO;2
- Howard, M. E., Zuercher, G. L., Gipson, P. S., and Livingston, T. R. (2002). Efficacy of feces as an attractant for mammalian carnivores. *The Southwestern Naturalist* 47, 348–352. doi:10.2307/3672492
- Hunt, R. J., Dall, D. J., and Lapidge, S. J. (2007). Effect of a synthetic lure on site visitation and bait uptake by foxes (*Vulpes vulpes*) and wild dogs (*Canis lupus dingo, Canis lupus familiaris*). Wildlife Research 34, 461–466. doi:10.1071/WR05110
- Hutchings, M. R., and White, P. (2000). Mustelid scent-marking in managed ecosystems: implications for population management. *Mammal Review* 30, 157–169. doi:10.1046/j.1365-2907.2000.00065.x
- Ivlev, V. S. (1961) 'Experimental Ecology of the Feeding of Fishes.' (Yale University Press: New Haven, CT.)
- Jacobs, J. (1974). Quantitative measurement of food selection. A modification of the forage ratio and Ivlev's electivity index. *Oecologia* 14, 413–417. doi:10.1007/BF00384581
- Jerosch, S., Götz, M., Klar, N., and Roth, M. (2010). Characteristics of diurnal resting sites of the endangered European wildcat (*Felis silvestris* silvestris): implications for its conservation. *Journal for Nature* Conservation 18, 45–54. doi:10.1016/j.jnc.2009.02.005
- Kelly, M. J., Noss, A. J., Di Bitetti, M. S., Maffei, L., Arispe, R. L., Paviolo, A., Angelo, C. D., and Di Blanco, Y. E. (2008). Estimating puma densities from camera trapping across three study sites: Bolivia, Argentina, and Belize. *Journal of Mammalogy* 89, 408–418. doi:10.1644/06-MAMM-A-424R.1
- Klar, N., Herrmann, M., and Kramer-Schadt, S. (2009). Effects and mitigation of road impacts on individual movement behavior of wildcats. *The Journal of Wildlife Management* 73, 631–638. doi:10.2193/2007-574
- Long, R. A., MacKay, P., Ray, J., and Zielinski, W. (2008). 'Noninvasive survey methods for carnivores.' (Island Press: Washington, DC.)
- Mackenzie, D. I., and Royle, J. A. (2005). Designing occupancy studies: general advice and allocating survey effort. *Journal of Applied Ecology* 42, 1105–1114. doi:10.1111/j.1365-2664.2005.01098.x
- Manly, B. (1997). 'Randomization, Bootstrap and Monte Carlo Methods in Biology.' 2nd edn. (Chapman and Hall: Boca Raton, FL.)
- McDaniel, G. W., McKelvey, K. S., Squires, J. R., and Ruggiero, L. F. (2000). Efficacy of lures and hair snares to detect lynx. Wildlife Society Bulletin 28, 119–123.
- Michalski, F., Crawshaw, P. J. Jr, Oliveira, T. G., and Fabián, M. E. (2007). Efficiency of box-traps and leg-hold traps with several bait types for capturing small carnivores (Mammalia) in a disturbed area of southeastern Brazil. Revista de Biologia Tropical 55, 315–320.

- Miguel, F. J., Marques, I., and Monclús, R. (2005). Respuesta de los zorros (*Vulpes vulpes* Linnaeus, 1758) al olor de otros carnívoros. *Galemys* 17, 113–121. [In Spanish with abstract in English]
- Mitchell-Jones, A. G., Amori, G., Bogdanowicz, W., Krystufek, B., Reijnders, P. J. H., Spitzenberger, E., Stubbe, M., Thissen, J. B. M., Vohralik, V., and Zima, J. (1999). 'The Atlas of European Mammals.' (T and AD Poyser Natural History: London.)
- Mondol, S., Karanth, K. U., Kumar, N. S., Gopalaswamy, A. M., Andheria, A., and Ramakrishnan, U. (2009). Evaluation of non-invasive genetic sampling methods for estimating tiger population size. *Biological Conservation* 142, 2350–2360. doi:10.1016/j.biocon.2009.05.014
- Monterroso, P. (2006). Selecção de habitat e análise dos factores que condicionam a presença e abundância de gato-bravo (*Felis silvestris*) no Parque Natural do Vale do Guadiana. M.Sc. Thesis, University of Porto, Porto, Portugal. [In Portuguese.]
- Moruzzi, T. L., Fuller, T. K., DeGraaf, R. M., Brooks, R. T., and Li, W. (2002). Assessing remotely triggered cameras for surveying carnivore distribution. *Wildlife Society Bulletin* 30, 380–386.
- Palomares, F., and Caro, T. M. (1999). Interspecific killing among mammalian carnivores. *American Naturalist* 153, 492–508. doi:10.1086/303189
- Palomares, F., Delibes, M., Ferreras, P., Fedriani, J. M., Calzada, J., and Revilla, E. (2000). Iberian lynx in a fragmented landscape: predispersal, dispersal, and postdispersal habitats. *Conservation Biology* 14, 809–818. doi:10.1046/j.1523-1739.2000.98539.x
- Palomo, L. J., Gisbert, J., and Blanco, J. C. (2007). 'Atlas y Libro Rojo de los Mamíferos Terrestres de España.' (Dirección General para la Biodiversidad – SECEM – SECEMU: Madrid, Spain.) [In Spanish]
- Phillips, R. L., Blom, F. S., and Engeman, R. M. (1990). Responses of captive coyotes to chemical attractants. In 'Proceedings of the Vertebrate Pest Conference'. (Eds L. R. Davis and R. E. Marsh.) pp. 285–290. (University of California: Davis.)
- Raal, A., Orav, A., Arak, E., Kailas, T., and Müürisepp, M. (2007). Variation in the composition of the essential oil of *Valeriana officinalis* L. roots from Estonia. *Proceedings of Estonian Academy of Sciences Chemistry* 56, 67–74.
- Ralls, K. (1971). Mammalian scent marking. *Science* **171**, 443–449. [New Series] doi:10.1126/science.171.3970.443
- Rivas-Martinez, S. (1981). Les étages bioclimatiques de la végétation de la Peninsule Iberique. *Anales del Jardin Botanico de Madrid* 37, 251–268
- Rivas-Martínez, S., Penas, A., and Díaz, T. E. (2004). 'Mapa Bioclimático de Europa, Bioclimas.' (Servicio Cartográfico de la Universidad de León: Madrid, España) Available at http://www.ucm.es/info/cif/form/maps.htm [accessed January 2010]
- Rodgers, A. R. (1990). Evaluating preference in laboratory studies of diet selection. *Canadian Journal of Zoology-Revue Canadienne de Zoologie* 68, 188–190. doi:10.1139/z90-026
- Roughton, R. D. (1982). A synthetic alternative to fermented egg as a canid attractant. *The Journal of Wildlife Management* **46**, 230–234. doi:10.2307/3808425
- Roughton, R. D., and Sweeny, M. W. (1982). Refinements in scent-station methodology for assessing trends in carnivore populations. *The Journal* of Wildlife Management 46, 217–229. doi:10.2307/3808424
- Saunders, G., and Harris, S. (2000). Evaluation of attractants and bait preferences of captive red foxes (*Vulpes vulpes*). *Wildlife Research* 27, 237–243. doi:10.1071/WR99052
- Schlexer, F. V. (2008). Attracting animals to detection devices. In 'Noninvasive Survey Methods for Carnivores'. (Eds R. A. Long, P. MacKay, W. J. Zielinski and J. C. Ray.) pp. 263–292. (Island Press: Washington, DC.)
- Schmidt, K., and Kowalczyk, R. (2006). Using scent-marking stations to collect hair samples to monitor Eurasian lynx populations. *Wildlife Society Bulletin* **34**, 462–466. doi:10.2193/0091-7648(2006)34[462:USSTCH] 2.0.CO;2

Sunquist, M., and Sunquist, F. (2002). 'Wild Cats of the World.' (Chicago University Press: London.)

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- Thomas, P., Balme, G., Hunter, L., and McCabe-Parodi, J. (2005). Using scent attractants to noninvasively collect hair samples from cheetahs, leopards and lions. *Animal Keeper's Forum* 7/8, 342–384.
- Thorn, M., Scott, D. M., Green, M., Bateman, P. W., and Cameron, E. Z. (2009). Estimating brown hyaena occupancy using baited camera traps. South African Journal of Wildlife Research 39, 1–10. doi:10.3957/056.039.0101
- Toft, C. A. (1980). Feeding ecology of thirteen syntopic species of anurans in a seasonal tropical environment. *Oecologia* **45**, 131–141. doi:10.1007/BF00346717
- Travaini, A., Rodríguez, A., Procopio, D., Zapata, S. C., Zánon, J. I., and Martínez-Peck, R. (2010). A monitoring program for Patagonian foxes based on power analysis. *European Journal of Wildlife Research* **56**, 421–433. doi:10.1007/s10344-009-0337-5
- Weaver, J. L., Wood, P., Paetkau, D., and Laack, L. L. (2005). Use of scented hair snares to detect ocelots. *Wildlife Society Bulletin* 33, 1384–1391. doi:10.2193/0091-7648(2005)33[1384:UOSHST]2.0.CO;2

- Williams, B. K., Nichols, J. D., and Conroy, M. J. (2002). 'Analysis and Management of Animal Populations.' (Academic Press: San Diego, CA.)
- Wilson, G., and Delahay, R. (2001). A review of methods to estimate the abundance of terrestrial carnivores using field signs and observation. Wildlife Research 28, 151–164. doi:10.1071/WR00033
- Wilson, R. R., Blankenship, T. L., Hooten, M. B., and Shivik, J. A. (2010). Prey-mediated avoidance of an intraguild predator by its intraguild prey. *Oecologia* 164, 921–929. doi:10.1007/s00442-010-1797-8
- Zielinski, W. J., Truex, R. L., Schlexer, F. V., Campbell, L. A., and Carroll, C. (2005). Historical and contemporary distributions of carnivores in forests of the Sierra Nevada, California, USA. *Journal of Biogeography* 32, 1385–1407. doi:10.1111/j.1365-2699.2005.01234.x

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