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Why do river otters scent-mark? An experimental test of several hypotheses

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We tested several alternative hypotheses about the function of scent marking by the North American river otter, *Lontra canadensis*. Otters may mark at latrine sites with spraints (faeces) to (1) signal species identity, (2) advertise their reproductive status, (3) establish and maintain territories, and (4) communicate social status and identity to group members. Olfactory preference tests were conducted at the Alaska Sealife Center in Seward, Alaska, on a group of 15 wild-caught male otters in February 1999. We found that male otters investigated otter scent more than sealion faeces. The male otters also showed a preference for male scent over the scent of anoestrous females. No preference for the scent of unfamiliar males, compared with the scent of familiar males, was observed, and no preference for the scent of close kin was detected. However, an investigation of dominant relationships of the captive otters showed that dominant males spent more time investigating male scent than did subordinate males. Thus, spraints deposited at latrine sites may function to communicate social status of males.

Among carnivores, olfactory signals often play a key role in intraspecific communication (Beauchamp et al. 1976; Gorman 1980; Brown & Johnston 1983; Kranz 1996; Molteno et al. 1998). In different species and under different ecological conditions, chemical cues found in urine, faeces and scent gland secretions can serve as reliable signals to transmit information about the relative health, sex, social status or reproductive state of an individual (reviewed by Brown & Macdonald 1985), as well as maintain social organization (Jorgenson et al. 1978; Brown 1979; Macdonald 1985; Gese & Ruff 1997; Sillero-Zubiri & Macdonald 1998). Compared with what is known about scent-marking patterns in carnivores (who marks, how often, and where), an understanding of the function of scent marks (what message is transmitted) is more limited.

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North American river otters, *Lontra canadensis*, are an ideal model species for investigating the function of scent marking because they are prolific in their use of olfactory signalling and they have a flexible social organization (Blundell et al. 2002a, b). Although river otters forage in aquatic environments, they establish and frequently visit prominent terrestrial sites at specific locations throughout their home ranges (Durbin 1989; Kruuk 1992, 1995; Testa et al. 1994; Ben-David et al. 1996, 1998; Swimley et al. 1998; Rostain 2000). At these sites, known as latrines, otters deposit spraints (faeces), urine and anal gland secretions ('jellies'; Bowyer et al. 1995; Ben-David et al. 1998; Rostain 2000). Because most sites are not associated with either feeding or denning (Bowyer et al. 1995), and otters rarely defecate in the water (M. Ben-David, unpublished data), latrines probably play a dominant role in communication and social interactions of *L. canadensis*.

Like many species of carnivores, *L. canadensis* shows flexibility in social organization (Beckel 1982; Melquist & Hornocker 1983; Rock et al. 1994; Rostain 2000; Blundell et al. 2002a, b). The degree of sociality varies between as well as within habitat types (Melquist & Hornocker 1983; Erickson & McCullough 1987; Rostain 2000; Blundell et al. 2002a, b). Different social groupings have been

described for *L. canadensis* (e.g. bachelor groups, mixed adult groups, extended family groups and groups with helper animals), with group organization changing seasonally (Beckel 1982; Melquist & Hornocker 1983; Rock et al. 1994; Rostain 2000; Blundell et al. 2002a, b). Thus, it is likely that communication in this species is also flexible and serves several functions.

There is considerable evidence that, among carnivores, odours are involved in sexual communication (Beach & Gilmore 1949; Doty & Dunbar 1974; Dunbar 1977, 1978; Macdonald 1979; Wells & Bekoff 1981; Gorman & Trowbridge 1989; Gese & Ruff 1997; Ding-Zhen et al. 1998; Molteno et al. 1998). In both the Eurasian otter, *Lutra lutra*, and in *L. canadensis*, marking at latrine sites occurs throughout the year. None the less, some evidence that marking is greater during the mating season exists (Humphrey & Zinn 1982; Robson & Humphrey 1985; Macdonald & Mason 1987; Rostain 2000). Indeed, one hypothesis for the function of scent marking in otters proposes that this behaviour is related to the advertisement of reproductive status of males and females (Kruuk 1992).

An animal can gain an advantage over conspecifics by denying them access to resources such as food or mates. Territorial spacing and associated scent-marking patterns have been observed in several mustelids (e.g. European badger, *Meles meles*: Kruuk 1978; Kruuk et al. 1984; stoats, *Mustela erminea*: Erlinge 1977; Erlinge et al. 1982; ferrets, *Mustela furo* L.: Powell 1979; Clapperton 1989; and weasels, *Mustela nivalis*: Erlinge 1974). In otters, territoriality and mutual avoidance have been reported in several cases both in freshwater and marine environments (Kruuk & Hewson 1978; Gosling 1982; Hornocker et al. 1983; Melquist & Hornocker 1983; Trowbridge 1983; Gorman & Trowbridge 1989; Kruuk 1992), but the ubiquitous distribution of latrines on the landscape has raised questions about the connection between scent marking and territoriality in these carnivores (Kruuk 1992, 1995). As an alternative to territoriality, Kruuk (1995) suggested that scent marking may signal the use and depletion of food patches and facilitate mutual avoidance on a small spatial–temporal scale.

Alternatively, it is possible that scent marks of otters communicate group membership, much as odours do among clan members of *M. meles* (Kruuk et al. 1984). In Prince William Sound (PWS), Alaska, coastal river otters frequently occur in social groups (Rock et al. 1994; Testa et al. 1994; Blundell et al. 2002a). These groups, which are composed mainly of males, cooperatively forage on schooling pelagic fish (Blundell et al. 2002a). Blundell et al. (2002b) observed that social males leave their group and make long breeding migrations before the arrival of schooling pelagic fish. Thus, members may be separated from their group for varying periods (Blundell et al. 2002a, b), and reuniting of groups may be facilitated by scent marking at latrines. Furthermore, because group associations may change seasonally, olfactory cues could provide a means to re-establish identity among familiar individuals.

Intragroup communication through scent marking may also include transmission of information on social hierarchy (Kruuk 1972). Although our knowledge of social hierarchy in otters is limited to observations on captive animals (M. Ben-David, unpublished data), it is likely that scent marking at latrines may facilitate the establishment and maintenance of such hierarchy because actual fights between otters are rare (Kruuk 1995). Furthermore, if otter groups are kin based, scent marking may facilitate the recognition of relatives.

To examine the function of scent marking by *L. canadensis*, we tested the following predictions on captive, wild-caught, male otters using preference tests. (1) If spraints signal species identity (Endler 1993; Kappeler 1998), we predicted that male otters would spend more time investigating conspecific spraints than scent from a sympatric species, the Steller sealion, *Eumetopias jubatus*. (2) If spraints are a reproductive signal, we predicted that adult male otters would spend more time investigating female sprint than male sprint, especially before or during the mating season. (3) If spraints are a territorial signal, we predicted that an otter would spend more time investigating the sprints of unfamiliar conspecifics than familiar conspecifics, because an unfamiliar animal would probably pose a greater threat than a cooperating group member or a well-known neighbour (Temeles 1994). (4) If spraints are used as an intragroup form of communication, we predicted that male otters would spend more time investigating familiar male sprint than unfamiliar male sprint. (5) If spraints signal relatedness in otters, there should be a positive relationship between time investigating them and degree of kinship.

METHODS

Study Animals

We conducted olfactory preference tests on 15 wild-caught adult male river otters that were live-captured in western PWS, Alaska in spring 1998. Those animals were trapped from the same population under investigation by Blundell and colleagues (Blundell et al. 2002a, b), for a companion study (Ben-David et al. 2000, 2001). Only adult males were used in that study because of the need to avoid separating females from young that could have been sequestered in natal dens, and because males were less likely to experience intrasexual aggression (Blundell et al. 2002a, b). Animals were held in captivity at the Alaska Sealife Center (ALSC) in Seward from May 1998 to March 1999. Animals were housed together in a 90-m^2 area with one large and four smaller salt-water pools. This area could be sectioned into two separate enclosures (one containing the large pool and the other containing the four pools) by closing a sliding gate. Totes with freshwater were placed throughout the enclosure. Otters were fed frozen fish, supplemented with live prey, vitamins and minerals (for more details see Ben-David et al. 2000, 2001; Ormseth & Ben-David 2000). All animals were released back into the wild at the end of the study (Ben-David et al. 2002).

Collection of Spraints

Spraints for olfactory experiments were collected from 10 known individuals (seven adult males and three adult females) from four zoos (Oakland Zoo, California, Washington Zoo, Oregon, Woodland Park Zoo, Washington, and Coyote Point Museum, California) and from the 15 captive male otters housed at ALSC. Faeces from one male and two female Steller sealions were collected at ALSC. All samples were collected between November 1998 and February 1999. Husbandry personnel in all zoos collected fresh faecal samples into small ziploc freezer bags, labelled them, and immediately placed them in a −18°C freezer. Because otters have an anal gland, any spraint piles containing anal sac secretions (e.g. jellies: odiferous mucus that is variable in coloration) were not collected. Special care was taken to collect only faeces, not urine or jellies. We transported spraints from the zoos to Seward, Alaska on dry ice.

Preference Tests

We conducted all preference tests between 8 and 20 February 1999. Prior to the start of each experiment, we thawed faecal samples for 0.5–2 h, depending on sample weights. We presented 5 g of faeces in 250-ml glass mason jars with gauze placed on top and duct tape wrapped around the base of the jar. We included this later step to assure that the test animal did not see the spraints inside (control for visual stimulus). To minimize transfer of human scent, we wore disposable latex gloves while handling spraints and containers (Johnston & Robinson 1993). We washed glass containers with hot water and Ajax dish soap after each trial and allowed them to air-dry overnight. Jars were placed in the centre of two 1-m circles approximately 1 m apart at the same distance (1 m) from the salt-water pool. To control for position effects, jars were switched between tests. All tests were conducted blind.

In all preference tests, we enticed the focal animal into the large pool area by simulating the procedures we employed when offering the otters live prey (Ben-David et al. 2000). After the focal animal entered the pool area, it was separated from the rest of the group by closing the gate between the two sections of the enclosure. We observed focal animals from a room adjacent to the outdoor holding facility and test area. Data were recorded with Newton Message Pad 2100 and Ethoscribe software (Tima Scientific, New Brunswick, Canada), and with a remote camera located in the test area. A microwave transmission system transmitted video images to the observation room. We scored tapes with a frame-by-frame analysis using a Zenith VCR and TV systems.

Each 5-min test began when the experimental male approached within 1 m of the stimulus from any direction. We recorded all investigatory behaviours and any scent marking performed by the otter within 1 m of the stimulus. Descriptions of these behaviours (i.e. sniff air, sniff ground, sniff jar) are listed in Table 1. We considered an animal to be sniffing the stimulus when its head was directed towards or at a 90° angle from the stimulus. The duration (in seconds) of each behaviour was recorded. For each preference test, each focal animal was used only once to avoid pseudoreplication, but several animals were used in more than one test. To ensure that prior experience with the procedure did not affect the subsequent behaviour of the animals, we randomized the order of the tests (i.e. each animal was presented with a different order of the tests). Of the 15 males, three individuals refused to enter the enclosure on different trials and thus were not included in our tests.

Stimuli in the preference tests varied with the hypothesis being tested. To test the hypothesis that spraints signal species identity, we gave seven male otters a choice between spraints from an unfamiliar male otter and Steller sealion faeces. To test the hypothesis that scent marks serve in male–female communication and that males recognize potential sexual partners, we gave each adult male otter ($N = 12$) a choice between spraints of an unfamiliar anoestrous adult female and unfamiliar adult male. All preference tests were done directly before the mating season in Alaska, and thus, all males were in reproductive condition with testes fully descended into scrotal sacs. To test whether spraints are used as a territorial signal or in intragroup communication, we gave each adult male otter ($N = 12$) a choice between spraints of a familiar adult male (one of the other captive otters in our group) and an unfamiliar adult male. The spraint of the group member in that test was randomly selected.

Determination of Social Hierarchy

To evaluate the effects of social hierarchy on intragroup communication, we determined the position of the focal animal and the familiar animal (from which the spraint was collected) in the social hierarchy of the group. We determined the position of each of the 15 captive male otters in the social hierarchy through behavioural observations made from a room adjacent to the otter enclosure. We conducted daily observations from 18 January to 7 February 1999, between 0800–1100 and 1600–1700 hours in association with feeding to avoid potential bias that could have been caused by inactivity of several individuals. Whereas all individuals were active during feeding time, most, but not all, were inactive during the rest of the day.

We randomly drew a list of focal animals each day. Each focal animal was observed for 30 min, unless that individual was not active for the first 5 min of the observation period. In those cases, the observer switched to the next focal animal on the list. We recorded all social interactions. These included touching, grooming, social rubbing, sharing food, playing, play wrestling (Beckel 1991), pawing, pushing, biting and fighting (Table 1). We also recorded the direction of the interaction (i.e. whether initiated by the focal animal or its counterpart), and whether the interaction was brief or extended.

In 70.5 h of observation, we observed each focal animal an average of nine 30-min periods (range 5–15 periods). In later analyses of social hierarchy, we pooled all negative

Table 1. Description of river otter scent marking and social behaviours scored in this study

Behaviour	Description
Sniff air	Otter was inside the 1-m radius area around the jar. The animal's head was directed towards or at a 90° angle from the stimulus, but there was no contact with either jar or ground. Slight bobbing of the head and flaring of nostrils was observed when animal was facing the camera.
Sniff ground	Otter was inside the 1-m radius area around the jar. The animal's head was directed towards or at a 90° angle from the stimulus, and touching the ground.
Sniff jar	Otter was inside the 1-m radius area around the jar. The animal's head was directed towards or at a 90° angle from the stimulus, and touching the jar.
Scent marking	The animal was depositing urine or a sprint. This behaviour was usually accompanied by a raise and flicking of the tail.
Touching	Two or more animals were in contact with each other's head or shoulder without moving.
Grooming	Two or more animals gently gnawed each other's head and shoulders.
Social rubbing	Two or more animals were in full body contact and were moving and rubbing on each other. No biting or gnawing occurred.
Sharing food	Two or more animals fed on the same fish together. No aggressive behaviour was associated with eating.
Playing	Two or more animals chased each other on land and in water. No body contact occurred.
Play wrestling	Two or more animals rolled together on land and in water. Full body contact but no aggression occurred.
Pawing	An animal pushed another with an outstretched paw. Pawing was usually accompanied by a high-pitched screech.
Pushing	An animal pushed another with the shoulder or rump.
Biting	An animal pushed and bit another with bared teeth. No other body part was involved.
Fighting	Two or more animals rolled together on land and in water, while biting each other and screeching. Open wounds may occur.

interactions (pawing, pushing, biting and fighting) as well as all positive ones (touching, grooming, social rubbing, sharing food, playing and play wrestling) to reduce the dimensionality of the data. To establish social hierarchy based on these interaction matrices, we used the web version of the Dominance Structure Applet (Java), which produces estimates for dominance ranks based on paired comparisons with the Batchelder–Bershad–Simpson (BBS) method (Jameson et al. 1999). Because negative interactions were rare among the captive otters (82 instances of 1464 observed interactions, which included three instances of biting and one fight), we used the same program with positive interactions, and compared the inverse of the resulting linear hierarchy with that based on the negative interactions. Where the two did not agree (only one case), we consulted our original data and resolved the inconsistency based on comments and notes related to the interactions.

Determination of Relatedness

To evaluate the effects of kinship on intragroup communication, we determined the relatedness of the focal animal and the familiar animal (from which the sprint was collected) with DNA analysis. To establish relatedness among the captive otters, we extracted DNA from frozen blood samples collected from the 15 males during their capture in spring 1998. Concurrently, we extracted DNA from 95 otters captured in a companion study in the same area (Blundell et al. 2002a, b). Using a library of nine polymorphic microsatellite loci, we were able to obtain individual microsatellite fingerprints for each of the river otters studied. Amplifications of DNA microsatellites were done in a GeneAmp PCR System 9600 (Perkin-Elmer) thermocycler and resolved on an ABI 373S Automated Sequencer with GS350 TAMRA run as an internal size standard in each lane. Alleles were sized in base pairs and

analysed using ABI GeneScan 3.1 and Genotyper 2.1 software (Blundell et al. 2002b). The coefficient of relatedness (R) between dyads for all 110 otters was calculated with the program Kinship (Version 1.2; Queller & Goodnight 1989; Queller et al. 1993; Goodnight et al. 1994) to avoid potential biases that could result from the small sample size of the captive animals.

Data Analysis

To test whether otters spent more time investigating conspecific scent in the interspecific preference test, and because we had relatively small sample sizes, we used a Wilcoxon paired-sample test (Zar 1999). Similarly, we used the same test to determine whether male otters showed more interest in the scent of females, and whether otters investigated unfamiliar scent more than familiar odours. To evaluate whether position in the social hierarchy influenced the time an otter investigated the scent of a group member versus an unfamiliar male, we determined whether the owner of the familiar scent was dominant over or subordinate to the focal animal. We then used a Friedman test for randomized block experimental design to investigate whether dominant animals spent more time investigating familiar scent than unfamiliar odours compared with animals that were subordinate to their group member (familiar scent). We then used Wilcoxon (familiar versus unfamiliar) and Mann–Whitney U (dominant versus subordinate) tests, as surrogates for multiple comparisons, to determine where differences in exploration time of scent occurred (Zar 1999). In addition, we calculated the difference in exploration time of familiar and unfamiliar male scent for each focal animal. We then explored the effects of relatedness (independent variable) on the difference in exploration time of familiar and unfamiliar male scent (dependent variable) with linear regression (Zar 1999).

RESULTS

Interspecific Preference

Male otters discriminated between the scent of conspecifics and that of Steller sealions and were more attracted to the scent of conspecifics. Otters spent significantly more time investigating the odour of unfamiliar male otters ($\overline{X}\pm SE=24.9\pm5.7$ s) compared with the odour of Steller sealions (15 ± 3.5 s; Wilcoxon paired-sample test: $T^+ = 27$, $T^- = 1$, $N = 7$, $P = 0.03$).

Intersexual Preference

Male otters also discriminated between the scent of the same and opposite sex. They spent significantly more time investigating odour from unfamiliar adult males (12.7 ± 2.3 s) than from unfamiliar adult females (7.9 ± 1.3 s; Wilcoxon paired-sample test: $T^+ = 63$, $T^- = 3$, $N = 11$, $P = 0.004$).

Familiarity and Dominance

We were able to rank six males as subordinate and six males as dominant to the familiar animals whose odour they were presented during the familiarity preference tests. Our analysis indicated a significant difference in exploration time of familiar and unfamiliar scent by dominant and subordinate male otters (Friedman test: $\chi^2_3 = 12.25$, $P = 0.007$; Fig. 1). This difference resulted from differences in behaviour of dominant and subordinate animals (Fig. 1). Compared with subordinate animals, dominant ones spent more time investigating both unfamiliar male scent (Mann–Whitney U test: $U = 2.0$, $N_1 = N_2 = 6$, one-tailed $P = 0.005$) and familiar scent (Mann–Whitney U test: $U = 5.5$, $N_1 = N_2 = 6$, one-tailed $P = 0.021$). No difference in investigation time for either dominant or subordinate animals was detected between familiar and unfamiliar male scent (Wilcoxon paired-sample test: dominant: $T^+ = 16$, $T^- = 5$, $N = 6$,
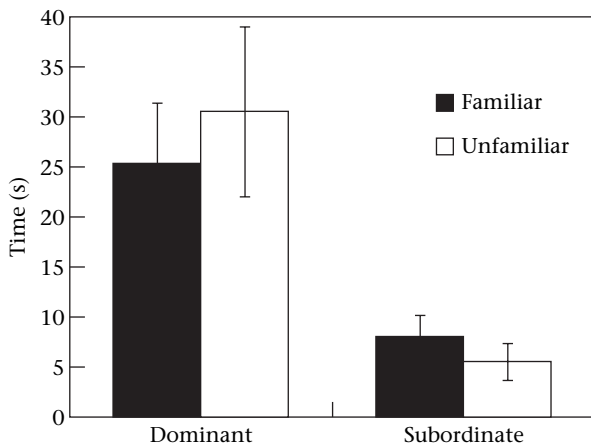
$P = 0.345$; subordinate: $T^+ = 4$, $T^- = 11$, $N = 6$, $P = 0.244$; Fig. 1).

Effects of Kinship

The average value of relatedness coefficients among the 15 male river otters in our experiment was 0.2 (range -0.031–0.75), and was comparable to the average value of relatedness of otter groups in PWS (Blundell et al. 2004). Linear regression revealed that there was no relationship between relatedness and the time an otter investigated familiar and unfamiliar male scent (linear regression: adjusted $R^2_{11} = -0.008$, $F_{1,10} = 0.911$, $P = 0.36$; Fig. 2).

Overmarking Behaviour

The otters in our experiments rarely overmarked the spraints of other individuals. Although otters marked in 13 cases (of 24 trials) only three occurred within the 1-m radius of the jars. In these three cases, one otter marked over a female spraint, one marked over an unfamiliar male spraint, and one marked over a familiar spraint.

DISCUSSION

Our results suggest that olfactory signals in *L. canadensis* probably communicate species and sexual identity. Pinniped faeces may not appear to be an obvious choice for the species identity tests, but in PWS, Alaska, coastal river otters and sealions interact frequently, especially when foraging on schooling fish (Blundell et al. 2002a). These interactions could on occasion be detrimental to otters (Blundell et al. 2002a). Thus, investigation of sealion faeces would be adaptive for coastal river otters. That
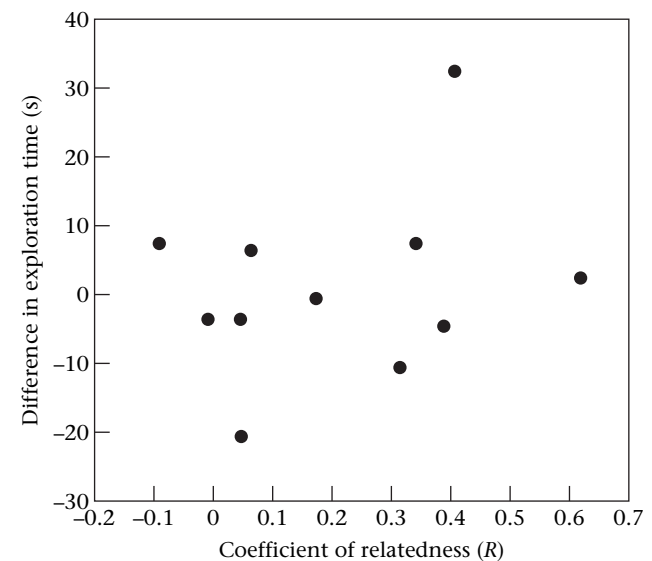

Figure 1. Mean $\pm$ SE time (s) spent by dominant ($N = 6$) and subordinate ($N = 6$) male river otters investigating spraints of familiar versus unfamiliar male otters.

Figure 2. Relationship between coefficient of relatedness and the difference in time spent by 12 male river otters exploring scent of unfamiliar versus familiar male otters.

otters spent significantly more time investigating faeces of conspecifics than those of sealions provides evidence that olfactory cues are important for intraspecific communication. This is supported by the observation that, in a marsh population of river otters in Florida, otters visit conspecific scent more than the scent of mink, *Mustela vison* (Humphrey & Zinn 1982). We recognize, however, that additional tests in which faeces of other mustelids are presented to river otters would better explore the role of scent marking in species identity.

Our tests indicated that, outside the breeding season, male otters explored the spraints of males more than those of anoestrous females. It is possible that otters may rely on urine or anal gland secretions rather than spraints to signal reproductive state. In a captive *L. lutra* female, urinary oestradiol levels peaked every 36 days over a 2-year period (Trowbridge 1983), suggesting that reproductive status is transmitted through urine. Gorman et al. (1978) found that a pair of captive *L. lutra* deposited anal sac secretions with a periodicity similar to that of the female oestrous cycle. Our use of spraints that were devoid of either urine or jellies may have limited our ability to test the reproductive status hypothesis.

Alternatively, the scent of an oestrous female may be so distinct that only a cursory exploration by males is necessary to alert them that the spraint belongs to an oestrous or anoestrous female. Under such conditions, experimental males in our study may have found the unfamiliar male scent more important to investigate. Although we conducted our experiments at the onset of the breeding season in Alaska and all males had descended testicles, female spraints from other zoos were collected several months before we began our experiments. It is likely that we would have found different patterns in the investigation of scent marks by our captive males if we had offered them the scent of an oestrous female.

That males investigated the scent of unfamiliar males more than that of unfamiliar anoestrous females indicates that olfactory signals play a role in social communication because, in coastal *L. canadensis*, males are more social than females (Blundell et al. 2002a). In our tests, dominant males spent more time than subordinate ones investigating the odour of other males irrespective of familiarity. Why were dominant male otters more interested than subordinates in the scent of other individuals? Field studies of carnivores have shown that dominant animals mark more, and often overmark subordinate scent (Rails 1971; Erlinge et al. 1982; Gosling 1982; Kruuk et al. 1984; Macdonald 1985; Gorman 1990; Gosling & McKay 1990; Sliwa & Richardson 1998). Therefore, it is likely that a dominant animal would tend to monitor scent-marking behaviours of other individuals more than a subordinate animal. Odours have been found to reflect dominance relationships of other mustelids. In *M. meles*, dominant males mark at a higher frequency than other members of a clan (Kruuk et al. 1984). Dominant males also squat-mark other clan members more frequently, especially females in oestrus (Kruuk et al. 1984). Dominant male *M. furo* L. mark more than subordinate males, and typically overmark odours of subordinate males (Clapperton 1989). In *M. erminea*, both dominant

males and females mark significantly more than subordinates (Erlinge et al. 1982). Furthermore, it has been shown that scent marks are used to settle conflicts in *M. erminea*, with subordinate individuals avoiding or using submissive vocalizations upon encountering the scent marks of dominant stoats (Erlinge et al. 1982). Thus, similar to other carnivores, differences in the frequency of investigation of scent marks as well as scent-marking behaviour in *L. canadensis* may provide a mechanism to maintain social organization. Establishing and maintaining dominant relationships is a viable way to minimize costs of aggressive interactions among carnivores (Gese & Ruff 1997; Macdonald et al. 1998; Mech 1999).

In contrast to our predictions, however, the captive otters did not spend more or less time investigating familiar versus unfamiliar male spraint. This may be a result of the high flexibility of group composition in *L. canadensis*. Blundell et al. (2002a, b) demonstrated that, in PWS, group composition changes seasonally and between years, with new dispersing males joining established male groups (Blundell et al. 2002b). It is likely that both dominant and subordinate animals frequently encounter familiar and unfamiliar scent and explore them in relation to their social status.

Our finding that relatedness had little influence on results of the familiar versus unfamiliar preference tests indicates that scent marking may not be important in kin recognition in *L. canadensis*. Blundell et al. (2004) showed that, in PWS, there was no association between relatedness of otters and any measure of sociality or spatial proximity. In PWS, cooperative foraging on schooling pelagic fish rather than kinship appears to be the driving force behind group formation in coastal river otters (Blundell et al. 2002a). Thus, kin recognition from scent marks, although possible, may not be important for group interactions.

We expected the captive animals to show high interest and overmark the scent of 'intruding' unfamiliar animals. Because otters did not investigate unfamiliar spraints more than familiar ones and overmarking rarely occurred, we were unable to conclude that scent marking signals territoriality. This result may not be surprising, given the meager field evidence supporting territoriality in male *L. canadensis* (Beckel 1982; Melquist & Hornocker 1983; Ben-David et al., unpublished data). Similarly, most evidence from *L. lutra* indicates that spraints are unlikely to be a territorial signal (Durbin 1989; Kruuk 1995). The use of spraints to signal the use and depletion of food patches and to facilitate mutual avoidance on a small spatial–temporal scale is more difficult to assess with our data, although otters probably investigate more unfamiliar than familiar scent when trying to avoid contact with other animals. This hypothesis clearly merits further investigation.

In conclusion, in our tests of alternative hypotheses for the function of scent marking by river otters, we found that spraints are likely to signal species identity. In intraspecific communication, communication of male social status seems to be a primary function of spraints left at latrine sites. We were unable to conclude that scent marks are used to advertise territorial boundaries. We also

found no evidence for communication of familiarity or kinship. We were unable, however, to rule out communication of reproductive condition during the breeding season because spraints were collected from anoestrous females. We propose that a study investigating the role of spraints in sexual communication would be done by presenting male otters with a choice between the spraints of anoestrous and oestrous females. We also see the need to repeat our experiments with female otters to explore the role of scent marking in the behaviour of that sex. In addition, presenting the same individual with a choice between the familiar odours of a higher-ranking and a lower-ranking group member will better clarify the relation between social status and scent marking. Furthermore, we recommend comparing the frequency of scent marking in dominant versus subordinate animals.

A continuing investigation of olfactory signals in *L. canadensis* and other otter species is an important step towards developing more productive monitoring protocols, because spraints are commonly used to determine the status of otter populations (Humphrey & Zinn 1982; Robson & Humphrey 1985; Mason & Macdonald 1986, 1987; Macdonald & Mason 1987; Reuther 1993; Kranz 1996; Herreman & Ben-David 2001). It is likely that the frequency of spraining by male and female, dominant and subordinate, and social and solitary otters will vary with the transmitted message. Therefore, understanding the function of scent marking may provide valuable information for correcting population estimates based on surveys of spraints and latrines. Because all otter species are currently threatened or endangered in some part of their historical ranges (Mason & Macdonald 1986; Kruuk 1995), understanding the function of scent marking may improve our conservation efforts for these carnivores.

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