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DO EURASIAN BEAVERS SMEAR THEIR PELAGE WITH CASTOREUM AND ANAL GLAND SECRETION?

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Abstract—The scent-matching hypothesis postulates that scent marks provide an olfactory link between a resident owner and his territory, and that this enables intruding animals to recognize the chance of escalated conflicts. However, it is unclear if Eurasian beavers (*Castor fiber*) mark their own pelage with castoreum from their castor sacs (i.e., the same material used in territorial marking); and/or if beavers mark their pelage with anal gland secretion (AGS) from the anal glands to waterproof the pelage and to act as a "living-scent mark". Chemical analysis (gas chromatography and mass spectrometry) of hair samples from 22 live-trapped beavers revealed that castoreum compounds were not present in any samples, AGS compounds were found from 3 animals (13.6%) around the cloaca, and the compound squalene was found in all the samples. Beavers may release castoreum directly into the water when it meets an intruder. Thereby, the "scent mark" in the water can provide an olfactory link between a resident owner and his territory. Squalene, in contrast to AGS, may be essential for keeping beaver pelts water-repellant.

Key Words— Beaver, *Castor fiber*, anal gland secretion, castor sacs, castoreum, pelage, GC-MS, hair, squalene, Norway.

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INTRODUCTION

The scent-matching hypothesis postulates that scent marks provide an olfactory link between a resident owner and his territory, and that this enables intruding animals to recognize the chance of escalated conflicts. If the smells match, then the animal that the intruder has met must be the territory owner. If the hypothesis were true, one would expect owners to (1) mark where intruders are most likely to encounter marks; (2) mark themselves with the substances used to mark the territory; (3) make themselves available for scent matching by intruders; and (4) remove or replace marks of others (Gosling, 1982).

All family members of Eurasian beavers (*Castor fiber*), except kits less than five months old, participate in marking the territory at scent mounds close to the water's edge (Wilsson, 1971; Rosell, 2002). Predictions 1, 3, and 4 for the scent-matching hypothesis have all received some support for the Eurasian beaver (Rosell, 2002). However, prediction 2 needs to be further clarified. Castoreum (primarily a mixture of secondary metabolites from urine) i) is the main scent signal used in the defenseof beaver territories during winter (Rosell and Sundsdal, 2001). However, the anal gland secretion (AGS) may function to waterproof the pelage. Excision of the anal glands seems to reduce the ability of the pelage to repel water (Walro and Svendsen, 1982). Prohibition of autogrooming in Eurasian beaver produces similar results (Wilsson, 1971). AGS may also act as a chemical messenger in the water territory sensed through close range or contact with the animal (Rosell, 2002).

The aim of this study was to investigate and to search for characteristic chemical compounds from the castor sacs and the anal glands in the beaver pelage in an attempt to clarify if (1) beavers mark themselves with castoreum from their castor sacs i.e., the same material used in territorial marking; and/or if (2) beavers mark themselves with AGS from the anal glands to waterproof the pelage and to act as a "living-scent mark".

METHODS AND MATERIALS

Study Area and Animals. Twenty-two beavers were live-trapped (15 males and 7 females) from 9 different territories during June 2 to August 18, 2000, by using landing nets in the municipalities of Nome and Sauherad in Telemark County, Norway. Beavers were from different age-classes (1->6 years old), weight varied from 3-24.5 kg, and they were trapped between 2100-0230 hr (see Rosell and Hovde, 2001 for further details about handling, sex, and age determination).

Sample Collection. Hair samples were collected from 3 different places on the beavers: 1) at the back just above the scaly tail, 2) around the cloaca, and 3) on the belly. Samples were collected using clean scissors. I cut off hair onto a sheet and thereafter stored it in a 100 ml glass vial. After each sampling, the scissors were cleaned with alcohol and dried with a propane gas burner.

I collected castor sacs and anal glands from 60 animals shot locally during the normal hunting season from 28 January–6 May 1997–99 and used the castoreum and AGS from these animals as the basis for comparison with possible castoreum and AGS compounds present in the hair samples. See Rosell and Sundsdal (2001) for a review of types of compounds found in castoreum and AGS. All samples were stored at -20°C until chemically analyzed (for further details see Rosell and Sundsdal, 2001).

Sample Preparation. Hair samples (0.15 g) were extracted overnight in 10 ml toluene. The solutions were filtered in a glass funnel with Schleicher & Schuell 595 paper filter. Thereafter, the hair was squeezed as dry as possible with a glass rod on the filter. The solutions were evaporated to a volume of 0.25 ml in a Heidolph VV 2011 rotary evaporator to concentrate the solution. See Rosell and Sundsdal (2001) for preparation of AGS and castoreum.

Chemical Analysis. One µl of the resulting solution was injected into a Hewlett-Packard 6890 Series II gas chromatograph equipped with a nonpolar HP-5 MS 5% phenyl-methyl-siloxane column (30.0 m × 0.25 mm ID × 0.25 µm film thickness) installed in a Hewlett-Packard 5973 Series mass spectrometer detector with a split/splitless inlet used in the splitless mode (for further details see Rosell and Sundsdal, 2001). The main focus of my study was to determine if the beavers had castoreum and/or AGS in their pelage. Therefore, positive identification of the compounds was not attempted, i.e., comparison with a known standard was not done. However, the compound squalene was positively identified (see results/ and discussion). Rosell and Sundsdal (2001) showed that this method worked well to detect compounds from the castoreum and the AGS in scent marks on snow.

RESULTS AND DISCUSSION

I found no compounds from the castor sacs on the pelage, which suggests that beavers do not mark themselves with castoreum. However, it is well known that beavers urinate into the water, i.e., release castoreum (Wilsson, 1971). They also release castoreum when frightened, e.g., when captured and handled (personal

observation). Beavers may have adopted an alternative strategy, i.e., they may release castoreum directly into the water when they meet an intruder. Thereby, the "scent mark" in the water can provide an olfactory link between a resident owner and his territory, and this will enable an intruding animal to gauge the chance of escalated conflicts.

Compounds from the anal glands were found in 3 of 22 animals (13.6%) but only around the cloaca area (1 (male, 3 years old), 8 (male, >5 years old) and 19 (female, 4 years old) compounds, respectively). This suggests that beavers do neither mark themselves with AGS to waterproof the pelage, nor act as a "living scent mark". Wilsson (1971) was not able to observe that the beaver systematically touched the opening of the anal glands or the area around these during grooming. For the Eurasian beaver, the AGS is a thick paste of a greyish color in females, and is an oily fluid with a whitish or pale straw color in males. For the North American beaver (*C. canadensis*), the AGS of the two sexes show also a consistent difference regardless of age: it is brown and viscous in males, but it is whitish or light yellow and runny in females (see Rosell and Sun, 1999). Obviously, the viscous secretion of the Eurasian female and the North American males would make the pelage sticky and would lessen water repellency of the pelage. The differences in viscosity between the sexes make it also unlikely that the anal glands have been developed to waterproof the pelage.

The sebaceous glands in the skin of mammals produce lipid mixtures (sebum) which coat the skin and hair. A relative minor contribution to this lipid film is made by the lipids arising from the keratinized cells of the epidermis (Lindholm et al., 1981). The results from this study showed that the hydrocarbon squalene ($C_{30}H_{50}$) was positively identified in all the hair samples. Squalene is also found in the skin surface lipids of the North American beaver (Lindholm and Downing, 1980). Therefore, when beavers groom, the pelage is cleaned, the hair shaft is aligned, maintaining maximum loft, and the hairs are probably oiled with the aid of squalene, which reduces water permeability. This could explain why prohibition of autogrooming in Eurasian beaver reduced the ability of the pelage to repel water (Wilsson, 1971). In addition to the natural lipid squalene, the density of the underfur (23 000 cm^{-2} on the ventral and 12 000 cm^{-2} on the dorsal surface), the guard hairs, and the surface tension of the water help to prevent water from penetrating to the skin (Novak, 1987). Thus, squalene, in contrast to AGS, may be essential for keeping beaver pelts water-repellant.

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