Lack of Response to Olfactory Lures Among Mammals in Riparian Habitat in Southern Wisconsin

RYAN J. JACQUES[1, 2, 3], JOSHUA M. KAPFER[1], and B. D. ESHELMAN[1]

[1]Department of Biological Sciences, University of Wisconsin-Whitewater, Whitewater, Wisconsin 53190 USA
[2]Current address: Arthur Temple College of Forestry and Agriculture, Stephen F. Austin State University, Nacogdoches, Texas 75962 USA
[3]Corresponding author: ryan.j.jacques21@gmail.com

Jacques, Ryan J., Joshua M. Kapfer, and B. D. Eshelman. 2016. Lack of response to olfactory lures among mammals in riparian habitat in southern Wisconsin. Canadian Field-Naturalist 130(1): 15–21.

Non-invasive mammal surveys often employ olfactory stimuli on the assumption that they will attract mammals and increase the success of monitoring projects. However, information on the effectiveness of scent lures is variable and often relies on data generated from mammal tracks or sign, which can be challenging to quantify. Therefore, we sought to determine whether certain olfactory stimuli are more effective than others at eliciting a response from mammals along riparian corridors in southern Wisconsin, using camera traps to monitor response to four scents and a control of de-ionized water. We recorded the number of times each species responded as well as the length of time spent investigating scents (Muskrat gland, mink gland, Red Fox urine, crayfish oil) and the control. We recorded 2812 passes by 16 mammal species during our 12-month study, with Virginia Opossum (*Didelphis virginiana*), Woodchuck (*Marmota monax*), Eastern Gray Squirrel (*Sciurus carolinensis*), Eastern Fox Squirrel (*Sciurus niger*), Raccoon (*Procyon lotor*), Coyote (*Canis latrans*), and White-tailed Deer (*Odocoileus virginianus*) observed on cameras most frequently. However, 72% of observations involved mammals passing through without investigating any scents or the control, and among the 28% of observations where mammals did investigate, we found no significant differences in their response to specific scents or the control or in response by season. Further analysis revealed no significant differences in the time mammals spent investigating individual scents or the control. The lack of response suggests that factors other than scent may have attracted wildlife to our stations. Although under some circumstances olfactory attractants may increase the level of response to monitoring stations, we suggest that attraction to our stations was largely a behavioural response to novel stimuli in the environment.

Key Words: Mammals; Virginia Opossum; Woodchuck; Eastern Gray Squirrel; Eastern Fox Squirrel; Raccoon; Coyote; White-tailed Deer; riparian habitat; camera traps; olfactory stimuli; scent; lures; attractants; behavioural response; Wisconsin

Introduction

Olfaction is an important mechanism by which mammals sense their environment (Vaughan *et al.* 2010). Although it can play a critical role in food acquisition, it is also important for communication via scent cues in urine, scat, and gland secretions. Past studies have investigated mammal response to olfactory stimuli or have used scent lures to attract mammals for other objectives. These studies include assessments of urban mammal and nuisance mammal response to food scent (e.g., Andelt and Woolley 1996; Campbell and Long 2008), response of prey to predator scent (e.g., Gorman 1984; Sullivan *et al.* 1985; Caine and Weldon 1989; Swihart *et al.* 1991; Russell and Banks 2007), studies of kin recognition (Johnston 2003) and territoriality (e.g., Arnold *et al.* 2011; Shivik *et al.* 2011), and population estimates (e.g., Mowat and Strobeck 2000; Mowat and Paetkau 2002).

Despite a significant amount of research, the response of mammal species to various olfactory stimuli remains poorly understood. This could be due to the lack of uniformity in methods employed and, thus, difficulty in comparing results across studies. Nonetheless, as indicated by Shumake (1977), assessing the effectiveness of olfactory cues to enhance the success of surveys and wildlife management strategies is important and can improve the efficacy of research efforts. Likewise, Schlexer (2008) noted that the use of lures in wildlife management will be handicapped until quantifiable testing of traditional mammal attractants supplants anecdotal conjecture. Thus, studies that quantify the response of mammals to various olfactory cues are potentially valuable to wildlife managers and researchers.

The results of past research on wildlife response to scent lures are variable. A number of authors have reported high wildlife visitation rates at scent lure locations. Monterrosso *et al.* (2011) reported that “investigative behavior” of olfactory attractants by some Iberian carnivores occurred frequently (in up to 75–81% of detections). Several studies have reported that Coyotes (*Canis latrans*) also often responded to particularly attractive lures: 41.8% (Turkowski *et al.* 1983) and 45.7% response (Martin and Fagre 1988). Mowat and Strobeck (2000) successfully obtained Grizzly Bear (*Ursus arctos*) hair samples at 48–73% of scent/bait stations sampled. However, a number of authors have reported that response to scent lures was low for a variety of species, including Coyotes (16–20% average visit rate per night of scent station deployment; Roughton and Bowden 1979) and weasels (8% site visitation; Mowat and Paetkau 2002), as well as several mammal species in Louisiana (4.6–11.8% annual visitation rates; Linscombe *et al.* 1983) and Florida (1–27% 2-year visitation rates; Conner *et al.* 1983).

Within-study variability has also been found in the response of different species to olfactory lures. Chamberlain *et al.* (1999) reported that scent lures were effective for Coyotes and Gray Foxes (*Urocyon cinereoargenteus*), but less so for Bobcats (*Lynx rufus*). Variation in response to olfactory lures was recorded among species detected (and type of lure used) during an Iberian carnivore community study (Monterroso *et al.* 2011). Furthermore, dissimilarity both within and across studies was reported in the response to scent lures by species in similar taxonomic groups. For example, Portella *et al.* (2013) found that no felids visited scent/hair snare stations deployed in southern Brazil, while Weaver *et al.* (2005) found this method effective for Ocelots (*Leopardus pardalis*) in southern Texas. Mowat and Paetkau (2002) used fish oil to attract mustelids to hair sampling stations, and found it was effective for American Martens (*Martes americana*) but less effective for smaller weasel species (Long-tailed Weasel [*Mustela frenata*] and Ermine [*M. erminea*]).

Although past attempts to quantify wildlife response to olfactory lures exist, much of this research has analyzed data generated from tracks or wildlife sign. Unfortunately, such data can be of limited value if track quality is poor or tracks are from species that are difficult to identify from sign (e.g., carnivores; Zielinski and Schlexer 2009). However, camera traps, used increasingly in wildlife biology (O'Connell *et al.* 2011), can generate more definitive data for species identification than tracks, scat, or other sign. Further, camera traps have advantages over traditional trapping methods, in that they are generally less invasive and can be particularly effective for studying cryptic species (Wearn *et al.* 2013). They also reduce risk to researchers, as there is no direct contact with wild animals. Camera traps with the ability to record video footage are especially useful for investigating questions related to wildlife behaviour, although, to date, few studies have used camera traps to quantify the response of wildlife to scent lures or to gauge the intensity of response by measuring time spent investigating lures (e.g., Batter 2011; Monterroso *et al.* 2011; Juslin 2011; Portella *et al.* 2013).

Given these knowledge gaps, our objective was to exploit advancements in camera trap technology, including video capability, to investigate whether specific olfactory cues elicit a response among mammals along riparian corridors. We also hoped to assess potential differences in the intensity of that response and whether variation occurred by species or season. To meet our objective, we employed common food, gland, and urine scents that we believed mammals in the riparian habitat would naturally encounter. We hypothesized that mammals in riparian habitats would exhibit a stronger response to deployed scents than a control (i.e., they would respond more often to scents and spend more time investigating scents than the control). We further hypothesized that among the scents offered, some would elicit a stronger response than others, and that this response would vary by species and by season.

Study Area

The response of mammals to olfactory stimuli at experimental locations was monitored for 12 months (May 2012 to May 2013) at five locations in Jefferson and Walworth Counties in southern Wisconsin, USA. The landscape associated with the five monitoring locations contained a mix of open canopy and lightly wooded habitats along perennial streams. The proportion of open and wooded land cover varied by location, although was similar across specific sites where we deployed camera traps. Associated streams varied in the velocity of water flow and substrate composition, although all streambeds possessed a mix of cobble and silt. We conducted our experiments on private rural land with limited human activity to reduce the potential for camera trap theft. We also chose study sites that would allow us to construct our monitoring stations within 3 m of a stream and immediately adjacent to obvious wildlife activity (i.e., animal trails). We followed the recommendation of Sargeant *et al.* (1998) and separated our experimental monitoring locations by more than 2 km to reduce the likelihood of recording the same individuals at multiple sites; our study locations were separated by 5–26 km. Major highways and roads also separated sites from each other, further decreasing the likelihood of individual animals traveling between them.

Methods

Experimental Set-up

Each experimental monitoring location included a camera trap to record mammal response to four scent lures: crayfish oil, Red Fox urine, Muskrat gland, and mink gland, along with a control of de-ionized water. All scents were purchased from a commercial vendor (F&T Fur Harvester's Trading Post, Alpena, Michigan, USA). We chose to use commercially purchased scents because research suggests that some mammals respond more strongly to aged versus fresh urine (Price 1977), and we were most interested in assessing scent lures that were easily obtainable by researchers. We placed 15–20 mL of each scent or the control individually into 50-mL vials. We capped each vial with a screw top lid, punctured with an equal number of holes to allow scent dispersal, but to reduce the likelihood of scent dehydration. Each vial, the top of which was visible to any passing mammal, was seated in a 30.5-cm length of polyvinyl chloride (PVC) pipe that was separated from adjacent pipes by 60 cm and staked perpendicular to the ground, which raised scents above ground level. Raising the scents made it easier to determine whether a response was elicited, as passing animals typically lifted their head to investigate scent vials. A single camera trap (Ltl Acorn 5210A, Old Boys Outdoors, Stone Mountain, Georgia, USA) with infrared flash, set to record 15-s video clips when triggered, was locked in a steel security box and lag-screwed to a tree 2.5 m from scent and control vials.

We visited experimental monitoring locations about once a week to replace and rotate scent and control vials, swap camera memory cards, and assess battery life. On arrival, we removed old scent and control vials from PVC tubes and replaced them with fresh vials. To control for possible location bias associated with the order in which the scent and control vials were deployed in front of camera traps, we also rotated vials one position clockwise roughly every 7 days. We randomly determined the original order of scent and control vial deployment in front of camera traps at the start of the study. We kept this order consistent across all monitoring locations and followed the same vial rotation order throughout the study. We wore nitrile gloves in the field when handling vials and camera traps to reduce the transfer of human scent to survey equipment.

On removal from the field, vials were washed with laboratory detergent (Alconox, Inc., White Plains, New York, USA) and rinsed with de-ionized water for later reuse. Care was taken to ensure that a given vial was used to hold only a single scent type or the control throughout the study to avoid incidental transfer of scent and cross-contamination of stimuli.

Video Scoring and Data Analysis

We uploaded video clips about once a week, immediately after returning from monitoring locations in the field. We then reviewed video footage and recorded the responses of all detected mammals to scent and control vials. For every occasion that an individual mammal passed in front of the camera trap, we recorded species and date and counted the number of times it responded to a scent or the control. If multiple animals passed through simultaneously, the response of each was scored separately. We also recorded passes that did not include any response to a scent or the control. This allowed us to quantify both response to olfactory stimuli and the number of times individuals passed through the monitoring location without responding. We then used a standard stopwatch to determine the amount of time (to the nearest 0.1 s) individual animals spent investigating a scent or the control.

We considered a "response" to be the movement of an animal's head toward a vial elevated on a PVC pipe (i.e., the individual's nose was clearly in contact with or immediately adjacent to a specific scent or the control). Because we could not accurately identify individual animals of the species detected, we quantified the number of responses during single "pass events," rather than quantifying responses by individually identified animals. We arbitrarily considered passes to be unique events if they were separated by at least 2 minutes.

We used contingency table analysis to determine whether there were significant differences between the number of responses of each species to individual scent types or the control (Zar 2010). We analyzed only species with expected values of five or more responses, which included Virginia Opossum (*Didelphis virginiana*), Woodchuck (*Marmota monax*), Eastern Gray Squirrel (*Sciurus carolinensis*), Eastern Fox Squirrel (*Sciurus niger*), Raccoon (*Procyon lotor*), Coyote (*Canis latrans*), and White-tailed Deer (*Odocoileus virginianus*). We also used contingency table analysis to determine whether the number of responses to individual scents or the control differed by season. Because count data were not normally distributed and samples were related, a Wilcoxon signed rank test was conducted to determine whether the number of occasions when individuals responded to any of the scents or the control differed from the number of occasions when they did not respond (Zar 2010).

Because time, recorded in seconds, was not normally distributed, we log transformed data to obtain normality and one-way ANOVA (all animals combined) was used to determine if differences existed in time spent investigating any of the four scents versus the control (e.g., did mammals spend more time investigating crayfish oil than the control?). Two-way ANOVA was also performed on the transformed data to compare the amount of time spent investigating scents and the control by each species (e.g., did species A spend more time investigating all scents and the control than species B?). We conducted contingency table analyses in Excel (Microsoft, Redmond, Washington, USA), whereas we conducted Wilcoxon signed rank tests and ANOVA in SPSS (IBM, Armonk, New York, USA).

Results

We recorded 2812 passes by 16 mammal species during our 12-month study (Table 1). In addition, an unidentified rodent (likely a mouse, *Peromyscus* sp.) was frequently detected. Because of its small size, we are not certain whether it reliably triggered the camera trap during all passes and, therefore, it was excluded from our tallies. Some species, such as Raccoon (n = 1241), White-tailed Deer (n = 246), and Virginia Opossum (n = 243), were recorded frequently.

A response was observed in 798 cases for an overall response rate of 28% for all species combined (Table 1). The Wilcoxon signed rank test indicated that the number of passes during which an animal responded was significantly less than the number of passes when no response was recorded (two-tailed test: $t = -2.542$; $P = 0.011$; $\alpha = 0.05$). We arbitrarily eliminated those species that exhibited a response rate below 10% (Table 1). The six species remaining (Eastern Gray and Eastern Fox Squirrels were combined) responded at a rate of 38% and the Wilcoxon signed rank test indicated no difference between passes with a response and those without a response (two-tailed test: $t = -1.599$; $P = 0.110$).

Table 1. All species of mammals photographed by camera traps from May 2012 to May 2013, including the number of responses to at least one scent or control vial and "no response" recorded for passing individuals, Jefferson and Walworth Counties, Wisconsin, USA.

Species	No. passes where response observed	No. passes where no response observed	% response
Virginia Opossum (*Didelphis virginiana*)	61	182	25.10
Eastern Chipmunk (*Tamias striatus*)	1	39	2.50
Squirrels (*Sciurus* spp.)*	40	666	5.67
Woodchuck (*Marmota monax*)	18	57	24.00
Common Muskrat (*Ondatra zibethicus*)	5	4	55.56
Eastern Cottontail (*Sylvilagus floridanus*)	3	21	12.50
Raccoon (*Procyon lotor*)	472	769	38.03
Striped Skunk (*Mephitis mephitis*)	0	17	0.00
American Badger (*Taxidea taxus*)	0	4	0.00
Long-tailed Weasel (*Mustela frenata*)	1	1	50.00
American Mink (*Neovison vison*)	6	39	13.33
Red Fox (*Vulpes vulpes*)	7	14	33.33
Coyote (*Canis latrans*)	51	29	63.75
Domestic Cat (*Felis catus*)	2	57	3.39
White-tailed Deer (*Odocoileus virginianus*)	131	115	53.25
Total	798	2014	28.38

**S. carolinensis* and *S. niger.*

Contingency table analysis of the same six species revealed no difference in the number of responses to individual scents or control vials (χ^2 calculated value = 30.9671; df = 20; χ^2 critical value = 31.410; $\alpha = 0.05$; Table 2). Contingency table analysis also did not show a difference between responses to scent and control vials by season (χ^2 calculated value = 18.4867; df = 12; χ^2 critical value = 21.026; Table 3).

One-way ANOVA, which considered the summed response of all species to scent and control vials, did not detect an overall difference in the time spent investigating individual scents or the control ($F = 1.9063$; df = 4, 3980; $P = 0.1065$). Two-way ANOVA showed a significant difference in the amount of time individual species spent investigating all vials ($F = 14.609$; df = 5, 1482; $P < 0.001$), but no difference in time spent

Table 2. Observed (Obs.) and expected (Exp.) values calculated by contingency table analysis to assess species-specific response to control and scent vials. Only the six species that exhibited the greatest response are included.

	Control		Crayfish oil		Muskrat gland		Fox urine		Mink gland		
Species	Obs.	Exp.	Obs.	Exp.	Obs.	Exp.	Obs.	Exp.	Obs.	Exp.	Total
White-tailed Deer (*Odocoileus virginianus*)	66	57.78	67	58.18	59	62.68	51	57.59	61	67.77	304
Raccoon (*Procyon lotor*)	191	183.23	181	184.48	190	198.76	180	182.61	222	214.91	964
Virginia Opossum (*Didelphis virginiana*)	8	17.11	13	17.22	25	18.56	23	17.05	21	20.06	90
Woodchuck (*Marmota monax*)	3	4.75	2	4.78	6	5.15	5	4.74	9	5.57	25
Coyote (*Canis latrans*)	18	22.81	30	22.96	30	24.74	22	22.73	20	26.75	120
Sciurus spp.*	9	9.31	4	9.38	10	10.10	13	9.28	13	10.92	49
Total	295		297		320		294		346		1552

**S. carolinensis* and *S. niger.*

Table 3. Observed (Obs.) and expected (Exp.) values associated with contingency table analysis to test for seasonal response to control and scent vials.

Season	Control		Crayfish oil		Muskrat gland		Fox urine		Mink gland		
	Obs.	Exp.	Obs.	Exp.	Obs.	Exp.	Obs.	Exp.	Obs.	Exp.	Total
Spring	51	60.60	59	61.21	74	66.66	68	60.60	69	71.92	321
Summer	166	150.66	148	152.17	154	165.73	135	150.66	195	178.78	798
Fall	67	68.34	67	69.03	75	75.18	78	68.34	75	81.10	362
Winter	16	20.39	29	20.59	27	22.43	19	20.39	17	24.20	108
Total	300		303		330		300		356		1589

investigating individual scent or the control vials (F = 0.761; df = 4, 1482; P = 0.551). We also detected no significant interaction between the independent variables: species and scent (F = 0.409; df = 2, 1482; P = 0.990). Each species treated the scent and the control equally, but there was a significant difference in the amount of time certain species spent investigating all scent and control vials, collectively. A post-hoc analysis (Tukey's test) revealed that squirrels (*Sciurus* spp.) spent less time investigating all scent and control vials than the others species included in our analysis.

Discussion

Although we hypothesized that mammals would respond more strongly to scent vials than the control vial and that certain scents would elicit a greater response than others, our data do not to support this. Only four of 16 identified mammal species responded to at least one scent or control vial on 50% or more passes through the monitoring locations. Furthermore, our results do not support the hypothesis that response to scents or the control varies significantly by species or season. The longer time spent investigating scent and control vials by most mammals we analyzed compared to squirrels may have been a result of inherently greater curiosity or food response (e.g., Raccoon; reviewed by Davis 1907), stronger territorial response (e.g., Red Fox [*Vulpes vulpes*], Coyote; Allen *et al.* 1999), or minimal neophobia (i.e., fear of novel stimuli).

Similar to our results, Thorson *et al.* (1998) found that Fox Squirrels (*Sciurus niger*) did not respond to experimentally applied scent (i.e., urine of Red Fox, Raccoons, and White-tailed Deer). However, there is great variability in the response of prey to the olfactory cues of other species, including the cues of predators (reviewed by Swihart *et al.* 1991; Apfelbach *et al.* 2005). This variation appears to be influenced by factors such as the species under investigation and the type of cue available for assessment. Several studies have reported a low rate of mammal response to olfactory lures, which corroborates our findings (Roughton and Bowden 1979; Linscombe *et al.* 1983; Mowat and Paetkau 2002). Others have also failed to detect a preferential response to specific scent lures by species under investigation. For example, little variation in the response of Coyotes to different deployed scents has been reported (Linhart *et al.* 1977; Roughton and Bowden 1979; Bullard *et al.* 1983), a phenomenon also observed in other carnivores during field trials (e.g., Travaini *et al.* 1996; Batter 2011). Yet, conflicting outcomes exist across published results. Some studies have found that Raccoons do not exhibit a preferential response to certain attractants (i.e., scents or bait; Nottingham *et al.* 1989; Kavanaugh and Linhart 2000), while others report that Raccoons respond frequently to specific food scents and bait, such as fruit (Campbell and Long 2008) or fish (Campbell and Long 2007). These latter studies suggest that perhaps the food scent we employed (crayfish oil) was not preferentially attractive enough. It is noteworthy that we found no evidence that the species we analyzed exhibited neophobia toward our deployed scents. This is evident because individuals frequently passed through scent station locations, regardless of whether they responded to the experimental olfactory cues we deployed, and corroborates past findings specific to Coyotes (Windberg 1996).

Although scent lures may help attract some species during surveys, our data indicate that responses may not always relate to specific scent lures. We observed that if an individual responded to one scent in our experimental array, it was also likely to investigate the other scents and the control, but we found that no single scent was more effective than others or the control at eliciting a response. We believe we observed a general behavioural response to the presence of novel stimuli (i.e., the scent stations) rather than an olfactory response. The possibility also exists that mammals were attracted to the general experimental locations we established because of the combination of scents provided, but did not further investigate individual scents on arrival. Unfortunately, our data do not provide an opportunity to address this effectively.

Regardless, the response of individuals to the deployed scent and control vials that we observed was low overall (i.e., mammals passed through without investigating scent or control vials significantly more often than they stopped to investigate). Our findings, coupled with the large variation in the reported effectiveness of scent lures, may challenge the usefulness of olfactory attractants during surveys. Rather than deploying scent lures, it may be more effective to concentrate efforts on selection of high-quality camera locations by identifying appropriate habitat or detecting evidence of a target species through knowledge of its natural history. We found that camera traps with video capability were particularly useful in quantifying responses at stations where attractants were deployed. Our ability to record video clips enabled us to count the number of responses and the duration of time spent investigating a given scent station. This allows for a more sophisticated assessment of a species' interest than counting photographs or tracks associated with scent stations.

Past research supports the assertion that mammals respond to scent lures. However, the effectiveness of mammal attractants (olfactory, auditory, visual) may depend on study context (i.e., target species, study goals, geographic location, prey, and scents naturally present at study sites). Mammals that were not among those that we detected may show a greater response to scent lures or to specific scent types. For example, felids in both captive and field settings will investigate various scent lures (McDaniel *et al.* 2000), including cologne (Thomas *et al.* 2005), although variability in the responses to other types of scents exists (Anile *et al.* 2012). Food bait might elicit a stronger response than scent alone for certain species, as was found for Canada Lynx (*Lynx canadensis*) in Minnesota (Moen and Lindquist 2006). Length of study could also influence visitation rates, as some species could become conditioned to the continued presence of scent lures over time (Robson and Humphrey 1985). The use of olfactory stimuli may be more effective for species that naturally exist in low densities and are otherwise very difficult to detect (i.e., wild felids), compared to the relatively abundant mammal species that we studied. Further research focused on the efficacy of other olfactory attractants and various combinations of attractant methods (i.e., novel visual or auditory stimuli) will aid in filling knowledge gaps.

Acknowledgements

We thank P. Jacobs, K. Karow, D. Karow, J. Boyd (Whitewater Greenhouse, LLC), and the Cushman Family for allowing access to their properties for data collection. We thank A. Harpster for assistance in the field. We also thank the University of Wisconsin-Whitewater Undergraduate Research Program and the Department of Biological Sciences for helping to support this project.

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Received 3 September 2014
Accepted 21 November 2015