The Effect of *Canis Latrans* Urine on Foraging Habits of Prey Animals

BIOS 35502: Practicum in Environmental Field Biology

Andrea M. Newlands

Advisor: Dr. Walter P. Carson

Abstract

Prey species heavily rely on their sense of smell to assess the risk associated with foraging. Thus, the use of predator scents has the potential to serve as a natural repellent to granivores and herbivores. In this experiment, I placed trays of black oil sunflower seeds mixed in sand in front of camera traps. Trays contained a cup with a cotton ball that was sprayed with either 0%, 50%, or 100% coyote (*Canis latrans*) urine. Trays were changed daily, and leftover seeds were massed to measure the amount of foraging. In the same habitat but distanced away from the camera traps to avoid disturbance, I placed stakes that had been soaked in a saline solution and sprayed them with a concentration of *Canis latrans* urine. There was no significant difference in seed removal between the different concentrations, however there was a significant relation between number of days since the beginning of the experiment and the seed removal across all sites, suggesting habituation. There was not enough response on the saline stakes to accurately draw conclusions. While this study contributes results on habituation and seed removal, further experimentation is necessary to investigate the effect of urine concentration on specific species as well as gather stronger data using saline stakes.

Key words: coyote, *Canis latrans*, scent, prey, foraging, urine, concentration

Introduction

Throughout history, prey have relied on their senses, especially smell, to help them evade predators. Previous studies have considered the effect different aspects of smell can have on rodents in a laboratory setting. Takahashi et al. (2005) observed fear behavior such as freezing, avoidance, or the release of stress hormones in rodents when exposed to differing amounts of cat urine, finding that larger amounts of urine led to more and longer exhibitions of fear behavior. This study raises the question of how concentration, rather than amount, of predator urine can affect prey behavior.

Many small prey animals such as rodents, snakes, rabbits, and deer fawns find a common predator in *Canis latrans* (Coates et al. 1998). One way prey judge the danger associated with a scent based on the diet of the animal. Carnivore urine contains more metabolites composed of sulfur leftover from protein digestion than herbivores. When the diet of *Canis latrans* changes from meat to fruit, the repellency of their urine decreases (Nolte et al. 1994). In addition, the presence of higher 2-phenylethyamine (PEA), a chemical in urine that elicits innate responses in the rodents, levels in carnivores also serves as a signal to prey. Ferrero et al. (2011) observed avoidance in rodents in response to PEA and measured the PEA concentration in nineteen carnivores and nineteen noncarnivores. Eighteen of the nineteen carnivores had PEA levels higher than 2 μM, with *Canis latrans* having a PEA level of 10 μM. In contrast, none of the sampled noncarnivores had PEA levels of above 2 μM and PEA was undetectable in eleven of the nineteen.

Despite the lower likelihood of *Canis latrans* targeting an adult deer, white-tailed deer (*Odocoileus virginianus*) show significant foraging reduction when presented with *Canis latrans* urine but is greater when confronted with the scent of larger predators like bobcats (Swihart et al. 1991). This study was conducted during the winter, but deer may show increased caution while fawns are being born. A multipart study investigating the use of predator scents as effective repellents, howeverm found that *Canis latrans* was more effective than other predator scents, including bobcat, in repelling black-tailed deer (*Odocoileus hemionus columbianus). Canis latrans* urine was also among the most highly effective repellents of snow-show hares (*Lepus americanus)* (Sullivan et al. 1985).

Avoidance behavior in prey as a response to predator scents can serve as a natural repellant, especially when small animals become pests. Wire mesh has been proven more effective than commercial repellants in protecting plants from deer herbivory (Kochenderfer & Ford 2008). Wire may look unsightly, however, making a homeowner more likely to rely on a repellent. A repellant tested in the study, Deer Away, claims to repel small animals as well. While Deer Away is a food-grade product, it is not registered for use on edible crops because the repellent is formulated to last extended amounts of time, making it difficult to wash off. Predator scents only repel herbivores for up to seven days, most likely because the repellent compounds evaporate out, but by placing the scent in a slow-release device, it is could be possible to use coyote urine as a long-term repellent to herbivore pests (Sullivan et al, 1985).

Like mosquito repellent, an 100% concentration may be unnecessary depending on the user's situation. This study will investigate the extent to which different concentrations of *Canis latrans* urine serve as reliable repellents. In addition, the results of the study can show how particular prey species determines whether it is safe or not depending on the strength of the predator's scent. We hypothesize that stronger concentrations of coyote urine will yield fewer signs of prey presence.

Materials and Methods

The Site

The University of Notre Dame Environmental Research Center (UNDERC) is located between the border of Wisconsin and Upper Michigan and is approximately 12 square miles. I divided a map of UNDERC into a 1 mile by 1 mile grid and identified areas of maple forest in nine of these sections as test sites.

The Experimental Procedure: Seed Trays

At each test site, I set up a trail camera in front of a tray filled with sand and 5 grams of black oil sunflower seeds. In the trays, I placed a paper cup with a cotton ball sprayed ten times with a concentration of coyote urine (0%, 50%, or 100%). The same brand of spray bottle was used between concentrations to control for the amount sprayed. The concentration was assigned to each site randomly. Trays were changed daily while cotton balls and cups were changed every other day. After bringing in the trays from the day before, I massed the remaining seeds to determine the effect the concentration had on foraging behavior. The trail cameras helped determine the type of animals visiting the site and number of visitations. A visitation was defined as an animal being on the tray, but no more than one visitation was counted every five minutes. If a tray was visited but no pictures were found on the camera, the animal was assumed to be a mouse or a chipmunk, depending on the amount of shells left in the tray.

The Experimental Procedure: Salt Stakes

Wooden stakes (1.5cm X 3.5cm X 60cm) were soaked in a 90% saline solution for 24 hours then dried in a drying over for at least 96 hours. I placed a wooden stake in the ground at least 10 meters away from each camera trap but in the same maple forest. The stakes were sprayed with its site's assigned concentration of coyote urine ten times and resprayed every four days to reduce the possibility that repeated visits would discourage wildlife from visiting the stake. I removed a stake once a foraging event had occurred. The amount of foraging was determined by using a known volume of modeling clay to fill in foraged parts of the stakes then calculating the difference between the original and leftover volume of clay.

Statistics

I conducted a Kruskal-Wallis test in MYSTAT to determine the significance of seed removal compared between each coyote urine concentration. A Kendall rank correlation was run in R Studio to show the relationship time had with seed removal. Both tests were evaluated at $\alpha=0.05$.

Results

The Wilcoxon test for normality showed that differences in seed removal was not normally distributed, and the data was unable to be transformed into a normal distribution. There was no significant difference between the urine concentration and the amount of seeds foraged (mean ± SE; 0%, 3.67±0.399; 50%, 4.25±0.306; 100%, 4.34±0.313; $p=0.381$, Figure 1), but there was a highly significant correlation between the time since the beginning of the study and seed removal (tau=0.284, $p=1.67\times10^{-4}$, Figure 2).

Only two of the nine stakes experienced foraging, both by hare. Of the original 225 cm^3, 30 cm^3 of the 0% concentration stake was consumed compared to 14 cm^3 of the 100% concentration stake.

The most numerous animals attracted to the seed trays were chipmunks and mice (forty-three and three visitations recorded on camera respectively, with many more evident from seed removal and seeds left in trays daily). Pine squirrels (*Tamiasciurus hudsonicus)* and snow-shoe hares (*Lepus americanus)* were also prevalent (twenty-one and seven visitations respectively). Five black bears (*Ursus americanus*) were found to interact with the seeds trays, three of which being a mother with two cubs. A *Odocoileus virginianus* and hermit thrush also visited the trays.

Discussion

Our results did not support our original hypothesis that higher concentrations of coyote urine would result in less foraging and more avoidance of an area. These results also contradict past studies (Takahashi et al. 2005; Nolte et al. 1994; Ferrero et al. 2011; Swihart et al. 1985; Sullivan et al. 1985). Possible explanations for prey species not responding to predator scent include low efficacy predator odors, individual differences in sensitivity to predator odors, and a mismatch between the predator scent used and the target prey species (Apfelbach et al. 2005). This contradiction may also be because this was a field study. Not restricting access to seed trays to specific species can lead to an inaccurate measure of seed removal since calculating the removal for one species was difficult. The human scent left at the sites and the novel aspect of the trays and cameras could also have overridden any caution the prey would have to the urine. In addition, the repetitive placement of the seeds over an extended period may have habituated the animals near the site, making them less cautious. One observation of habituation is a pine squirrel that was briefly seen near but not on a tray for two days before regularly visiting the tray. Takihashi et al. (2005) found that habituation had a significant effect on decreasing freezing behavior, risk assessment, and the amount of times the rat investigated the origin of predator scent. They found no effect on number of days exposed to the scent and avoidance behavior, likely because rats that avoided the scent would not habituate to it. The effect of *Canis latrans* urine concentration on prey foraging may be further studied by focusing on a specific species in a laboratory setting. This can provide more accurate data related to a single species, and using an individual for only one trial can prevent habituation from interfering with results.

The most surprising species to visit the trays were the *Ursus americanus*. Because there was no significant difference in seed removal, it is unknown why the bears were attracted to the seed trays. Possible lures could have been seeds, the novel tray and camera, human scent, or *Canis latrans* scent. While scents or seeds may have initially lured the *Ursus americanus* to the site, differences in picture angles as the bears moved the cameras shows they later showed interest in the cameras. Further studies may investigate the role of novelty in holding the attention of *Ursus americanus* by using novel items and recording how long the bears remained at the site.

Type of scent may also influence behavior. *Vulpes vulpes* and *Procyon lotor* were both effective in repelling gray squirrels (*Sciurus carolinensis*), but there was no significant difference between the two scents (Rosell 2001). It may be interesting to compare a smaller predator such as *Vulpes vulpes* to a larger predator such as *Canis latrans*.

Almost all sites had many days where only chipmunks or mice visited them, allowing us to see the high level of seed removal of these individuals. I could see how much the mice had been able to eat by assessing the amount of seed shells left in the tray. Trays were set out in the morning, allowing chipmunks to feed on the seeds first during the day, leaving the mice to forage the leftovers at night. Because most if not all seeds were removed from visited trays, we can use the amount of shells to estimate the amount of seeds removed by chipmunks. Although these amounts were qualitatively assessed, there was a marked difference between a site that saw high chipmunk visitation versus no chipmunk visitation. Use of seed trays and the comparison of shell remains could be used in future studies as a method of locating chipmunk populations.

In addition, while sites were placed in maple forests as a control for habitat, it does not consider differences in population densities of different species across these sites. Future studies should combat this by placing a control and experimental condition within the same habitat but far enough away that they would not influence each other.

With two concentrations of stakes having only one foraged stake each, we were unable to run statistics on the data. However, the stakes with the 0% solution displayed twice as much foraging than the 100% solution, showing a relation between the concentration and amount of consumed stake. By setting out more stakes for a longer period, more data could be collected, thus leading to stronger results.

As society and demand for food, especially chemical-free food, grows, finding natural, effective repellents is paramount to success. A generalist predator such as *Canis latrans* has much potential to serve as one of these repellents. By understanding the influence predator cues have on prey foraging behavior, advancements can be made in making such repellents. Furthermore, investigating the effect of concentration of predator scent on foraging behavior can result in the manufacture of not only effective, but efficient repellents.

Figures

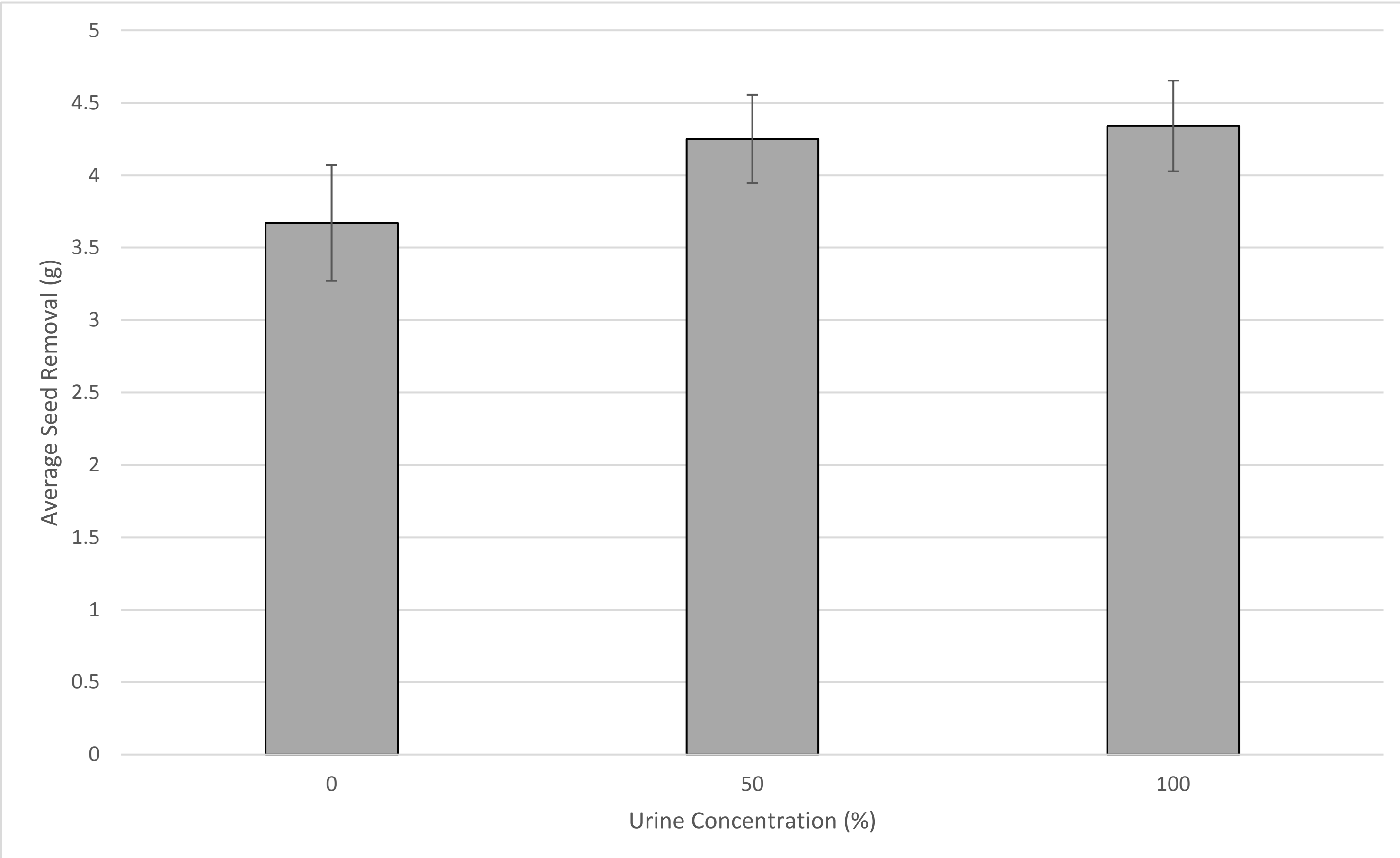

Figure 1: Average Seed Removal at Different Urine Concentrations. A Kruskal-Wallis test comparing the relationship between urine concentration and seed removal revealed no significant difference with a p-value of 0.381.

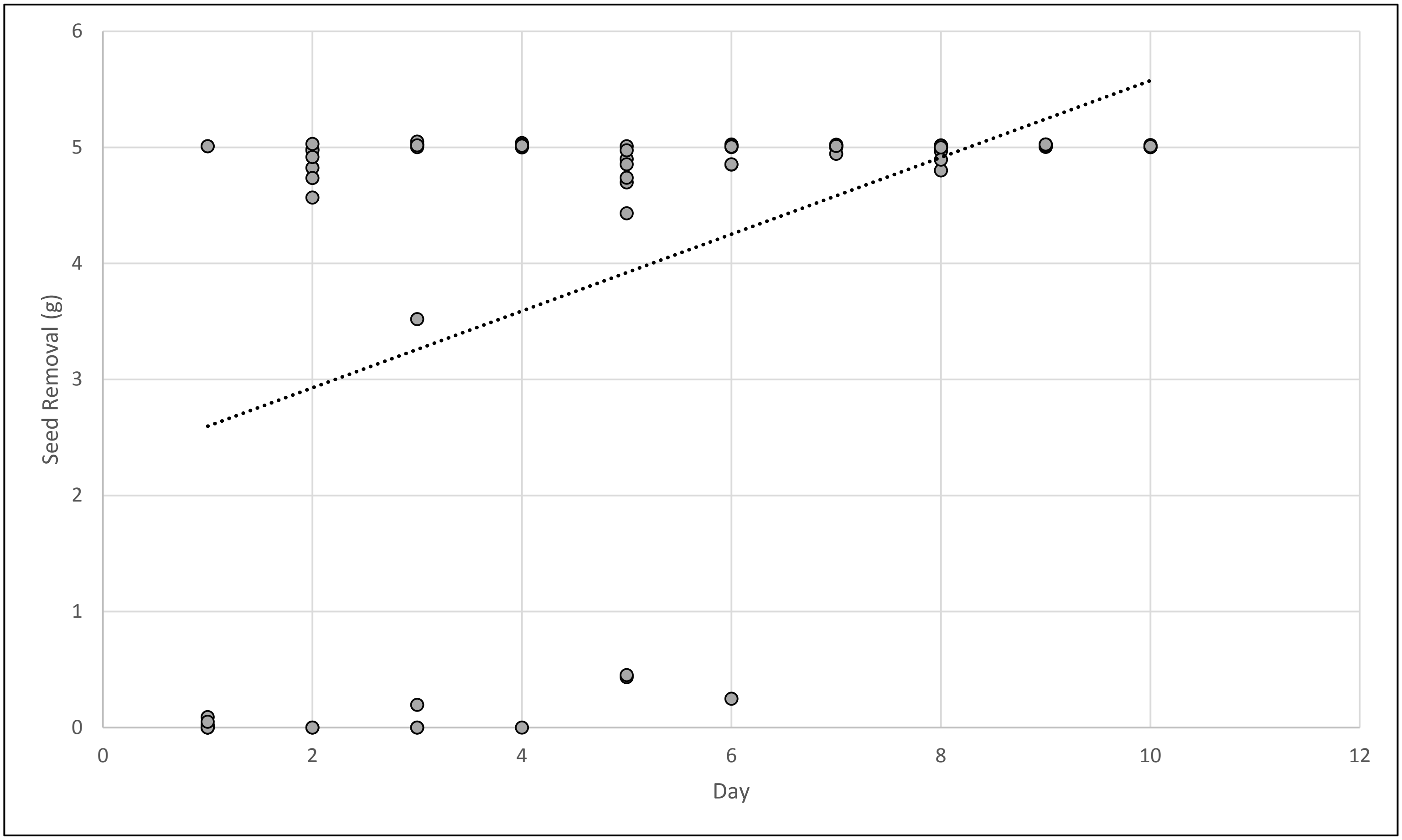

Figure 2: Average Seed Removal Over Days. A Kendall rank test revealed a significant relationship between days since the beginning of the experiment and seed removal across all sites, returning a tau of 0.284 and a p-value of 1.67×10^{-4}.

Acknowledgements

Infinite thanks are given to Dr. Walter P. Carson and Dr. Michael J. Cramer for their advice and support during the planning and completion of this project. Thanks are also extended to Adriana Cintrón, Laura Matthews, Samantha Johnson, and Mariah Lighthall for assisting in field work and data collection. I would also like to thank Hannah Legatzke who made this paper possible. Immense gratitude is given to Rodrigo Rivero for introducing me to UNDERC-East. Of course, thanks are due to the Bernard J. Hank Family Endowment for making this project and my stay at UNDERC possible.

References Sited

Apfelbach, R., Blanchard, C. D., Blanchard, R. J., Hayes, R. A., & McGregor, I. S. (2005). The effects of predator odors in mammalian prey species: a review of field and laboratory studies. *Neuroscience & Biobehavioral Reviews*, *29*(8), 1123-1144.

Coates, S. F., Main, M. B., Mullahey, J. J., Schaefer, J. M., Tanner, G. W., Sunquist, M. E., & Fanning, M. D. (2002). The Coyote (Canis latrans): Florida's newest predator. *WEC124. Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida, Florida*. Accessed June 3, 2017. http://manatee.ifas.ufl.edu/lawn_and_garden/master-gardener/gardening-manatee-style/c/coyotes.pdf

Ferrero, D. M., Lemon, J. K., Fluegge, D., Pashkovski, S. L., Korzan, W. J., Datta, S. R., Spehr, M., Fendt, M. & Liberles, S. D. 2011. Detection and avoidance of a carnivore odor by prey. *Proceedings of the National Academy of Sciences*, *108*(27), 11235-11240.

Kochenderfer, James N., and W. Mark Ford. "Utility of wire cages, tree shelters, and repellants to minimize herbivory to oak by white-tailed deer." 2008. U.S. Department of Agriculture, Forest Service. Accessed June 4, 2017. http://asdevelop.org/wp-content/uploads/2016/09/deercages.pdf

Nolte, D. L., Mason, J. R., Epple, G., Aronov, E., & Campbell, D. L. (1994). Why are predator urines aversive to prey?. *Journal of Chemical Ecology*, *20*(7), 1505-1516.

Rosell, F. (2001). Effectiveness of predator odors as gray squirrel repellents. *Canadian Journal of Zoology*, *79*(9), 1719-1723.

Sullivan, T. P., Nordstrom, L. O., & Sullivan, D. S. (1985). Use of predator odors as repellents to reduce feeding damage by herbivores. *Journal of chemical ecology*, *11*(7), 903-919.

Swihart, R. K., Pignatello, J. J., & Mattina, M. J. I. (1991). Aversive responses of white-tailed deer, Odocoileus virginianus, to predator urines. *Journal of Chemical Ecology*, *17*(4), 767-777.

Takahashi, L. K., Nakashima, B. R., Hong, H., & Watanabe, K. 2005. The smell of danger: a behavioral and neural analysis of predator odor-induced fear. *Neuroscience & Biobehavioral Reviews*, *29*(8), 1157-1167.