

# Something Stinks: the Effect of Predator Scent Cues on the Behavior of Small Rodents

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## Abstract

Organisms are strongly affected by density interacts as well as predator-prey interactions. Both can cause evolutionary and behavioral shifts, with predator-prey interactions creating strong trait-based responses in the prey species. Because predation plays such a key role in prey survivorship and fitness, it is important for the prey to be able to recognize and avoid predators in their ecosystem. Direct and indirect predator cues are used to assess risk in the environment and as a guideline to alter prey behavior. Olfactory cues are one important direct predator cue that is used commonly by mammalian prey. This experiment exposed *Peromyscus maniculatus* to two different predator scent cues (*Vulpes vulpes* urine and *Coluber constrictor foxii* feces). These were run along side a third control treatment that used only water. I found no significant difference between the times mice spent near the cue and near the non-cue control. There was also no difference between the times spent near the cue between the three treatments of urine, feces, and control. There was no difference in behavior based on individual mouse characteristics such as sex or sexual maturity either. I, therefore, suggest that the assumption under which most ecology operates, that prey will avoid direct predator cues, should be examined more closely in regards to *P. maniculatus*.

## Introduction

Predation is a very important interaction in biological systems, causing many behavioral and evolutionary shifts in organisms (Spencer et al. 2014). It consists of the negative influence of a carnivore or carnivores on prey species and can be a

factor of natural selection. Mammalian prey species are under predation pressures not only from other mammals but also from reptiles, fish, birds, and even amphibians (Wasko et al. 2013).

In ecosystems, animals are pressured by both competition within their own and similar species as well as the direct negative effect of predation. Responses to both density and predation help to form the life strategy of an organism. In many cases, the presence of predation can help to relax the pressures of competition between individuals and reduces the density-based responses (Werner and Peacor 2003). The presence of predation has a larger effect than simply reducing the population size. Prey seeks to avoid predation and thus trait-based responses in phenotype, behavior, and development also develop due to this interaction (Preissier et al 2005). These response are often stronger and longer-lasting than density and can push species towards behavioral and physical changes evolutionarily.

The environmental structure of a habitat can have a strong impact on the predator-prey interactions of the ecosystem (Spencer et al. 2014). The bunchgrass prairie of the National Bison range has heterogeneous habitat structures with shrub islands and invasive species interrupting the bunchgrass and causing a variation in the cover presented to the prey animals. This differing matrix of habitat can affect how prey species perceive the predator cues around them and cause behavioral modifications such as habitat avoidance and foraging reduction (Jones and Dayan 2000).

From the prey's side of the predator-prey interaction, recognition of the presence of the predator is crucial. Many studies have shown that mammalian prey species prefer foraging areas that avoid indirect predator cues such as open ground, moonlight, and forest edges (ie. Orrock et al. 2004). Because of the need to manage predation risk, direct predator cue recognition such as vocalizations, visual cues, and olfactory cues is also thought to be an essential part of small mammal behavior (Sivy et al. 2010). In mammalian systems, olfactory recognition plays a key role in risk management and signaling (Wasko et al. 2013). Prey responses to scent cues and recognition of predators can include reduced foraging behavior and general activity, decreasing trapping success and encounters in the wild (Spencer et al. 2014). In a study by Osanda et al.'s (2013), juvenile mice exhibited avoidance and freeze behavior when near wolf urine cues. Despite the studies that have been performed regarding these hypothesized behaviors, there is still much to be learned about the affects different types of predators and predator cues on mammalian prey behavior (ie. Starke III and Ferkin 2012, Wakso et al. 2013).

With many studies seeking to demonstrate a behavioral shift in rodents exposed to direct predator cues such as urine, feces, and chemical markings, there has been a bias in the experimentation towards one type of predator group. Mammalian predation and predator cues have been the most studied group interaction with reptilian predation on rodents being of a lesser consideration. Snake predation is, however, very prevalent among many ecosystems (Wasko et al. 2013). With snakes often being a more consistent rodent threat than mammalian

predators, it is important to understand how rodents respond to reptilian predation as well as mammal predation.

The experiment conducted this summer sought to compare rodent (*Peromyscus maniculatus*) prey response to direct predator cues from both a mammalian predator (*Vulpes vulpes*) and a reptilian predator (*Coluber constrictor foxii*). My hypothesis was that if *P. maniculatus*, there will be a greater response to the reptilian predator cue than the mammalian predator cue. I also hypothesized that males would be less affected by the scent cues than females and that adults would be less affected than juvenile mice.

### **Materials and Methods**

In this study, three different trials were run in the laboratory. They consisted of a trial with two neutral stimuli, a trial with a neutral and a mammalian predator cue, and a trial with a neutral and reptilian predator cue. The three stimuli used in the experiment were a neutral cotton ball with  $\frac{1}{2}$  teaspoon of water, the feces of a blue racer (*Coluber constrictor foxii*), and  $\frac{1}{4}$  teaspoon of red fox (*Vulpes vulpes*) urine on a cotton ball. All stimuli were placed inside a plastic cup with a lid with six holes. The snake feces came from animals captured on the National Bison Range using snake boards. Snakes were allowed to shed before feeding attempts were made, and the first feces excreted in captivity were discarded. One snake consumed a young *Mus musculus*, and two rounds of feces were able to be collected. These were kept frozen and allowed to thaw before each trial. The fox urine was a preserved amount collected for the use of hunting (F & T Fur Harvester's Trading Post).

The rodent species used in this experiment was *Peromyscus maniculatus*. Rodents were captured using live Sherman traps baited with oats and grass seed mixed with peanut butter in four different locations around the National Bison Range. The mice were weighed, sexed, checked for sexual maturity, and placed in a clean holding box. All lactating females were released immediately without being run. Each individual was used in behavioral trials no more than three days after being caught in the field. All individuals were given access to food and water before and after the trial. In each trial, the stimuli was randomly assigned to a side of the tank and an animal will be randomly selected for one of the three trials. The chamber was marked along the side to denote distance from the stimulus. The neutral zone measured 7 cm total in the center. Either side of the tank was marked at 5cm and 15cm from the outer edge. The tank itself was 50cm by 26cm by 32cm. The sides of the terrariums were covered by cloth dividers so that the mice were unable to see each other. The tanks and cue cups were also cleaned with 10% bleach in between every trial in order to prevent olfactory cues from previous mice from affecting future trials. Each trial was run for thirty minutes with a video camera recording the trial with a five minute acclimation time before hand. The beginning of each round of trials was ten o'clock at night in order to correspond with the nocturnal activity of *P. maniculatus* and extended until all of the mice captured were tested. Analyses will be performed on the basis of how long the individual has spent in the neutral zone, far away from the stimulus, and near the stimulus.

## **Results**

The result from the fox urine, snake feces, and control treatments for the Kruskal-Wallis test for the cue side was not significant with a p-value of 0.5373. The result from the fox urine, snake feces, and control treatments for the Kruskal-Wallis test for the non-cue control side was not significant either with a p-value of 0.4285. The result from the fox urine, snake feces, and control treatments for the Kruskal-Wallis test for the neutral zone was not significant with a p-value of 0.7422 (Figure 1). The Kruskal-Wallis tests run between the sex of the captured individuals for the cue, control, and neutral zones of the tank were also not significant with p-values of 0.6738, 0.7762, and 0.8913. The Kruskal-Wallis tests for the sexual maturity with the cue, control, and neutral zones of the tank were not significant with p-values of 0.3345, 0.4957, and 0.2907.

The  $\chi^2$  contingency table showed that the amount of times mice crossed in between sides of the tank was not significant with a p-value of 0.2659 (Figure 2).

Paired Wilcoxon tests were run between the cue side and control side for the fox urine treatment and found not significant with a p-value of 0.3591. The paired Wilcoxon test for the snake feces treatment was not significant with a p-value of 0.5995. The paired Wilcoxon test for the control treatment was not significant with a p-value of 0.4543.

The paired Wilcoxon test for the sides of the tank was significant with a p-value of 0.0035 (Figure 3). The paired Wilcoxon test for the edge of the tank was also significant with a p-value of 0.0009 (Figure 4).

## **Discussion**

The results of the olfactory predator cue experiment were not statistically significant and do not support the hypothesis that small mammal behavior changes

in the presence of a direct scent cue. There are many different possibilities for this result. An avoidance response in the presence of direct predator cues alone may not be a strong behavior. There may need to be other factors not represented in the study in order for *P. maniculatus* to react strongly to the cue. Though the underlying assumption is that mammalian prey species will want to avoid the predator cue, this may be a mistaken assumption as many other studies have shown non-significant responses to these cues (Wasko et al. 2013; Spencer et al. 2014; Sivy et al. 2010; Starke III and Ferkin 2012). There may also be a particular failure in the laboratory designs being used. In this experiment, there may have been too small an area for the *P. maniculatus* to feel that it could avoid the cue, thus confounding the behavior. The lack of hides may have also affected the ability for the mice to feel they had avoided the cue. Adding hides would link indirect and direct cues and could offer a possible future experiment.

The sexual traits of the capture animals were also analyzed in order to determine whether sexual maturity played a role in predator avoidance behavior in the presence of a direct olfactory cue, but no significant results were found. This means that the sex of the animal may not determine its choice when in the presence of a direct predator cue. The sexual maturity also did not have significant results. Unlike Osanda et al. (2013), the juvenile and adult *P. maniculatus* had no statistically significant behavioral change in the presence of a predator cue.

There were two tests that did give significant results. The side of the tank (left or right) showed a significant difference with the right side of the tank being preferred. The middle of the tank (the area closest to the other tank) was also

significantly preferred to the outer edges of the tank. There are a few possibilities that could account for these preferences. A lingering scent could remain from past trials despite the tank being cleaned with 10 % bleach between each trial. There might have also been a difference in warmth between sides of the cage, though all both sides of the tanks were an equal distance for the windows. Light does not seem to have been a factor as not only were the tanks equally lighted, but many of the mice were in the front of the tank which has the most lighting. Sound might also be a factor, but both of the tanks were exposed to the same noise level.

There are many possibilities for improvement on this study and future studies. Using different predator species and cues might avoid any confounding element of an olfactory cue. Avian hunting calls could be used in comparison to a mammalian call to examine the affect of auditory predator cues. Comparing interspecies responses would also be an interesting future experiment. *Microtus pennsylvanicus* would provide a good species to juxtapose with *P. maniculatus* because not only is it diurnal as opposed to nocturnal, but *M. pennsylvanicus* is also herbivorous as opposed to granivorous. These factors are more likely to result in differing predator response behavior. In order to avoid a laboratory artifact, it would also be beneficial to run trials in the field. Scent posts could be placed with trail cameras to monitor them. Food trays could also be used near to the posts to analyze the affect of direct predator cues on time spent foraging in small mammals. Lastly, further experiments to understand the significant differences in location in the cage and locations near the edges might give a greater understanding of what behavior is being changed in the experimental chamber.

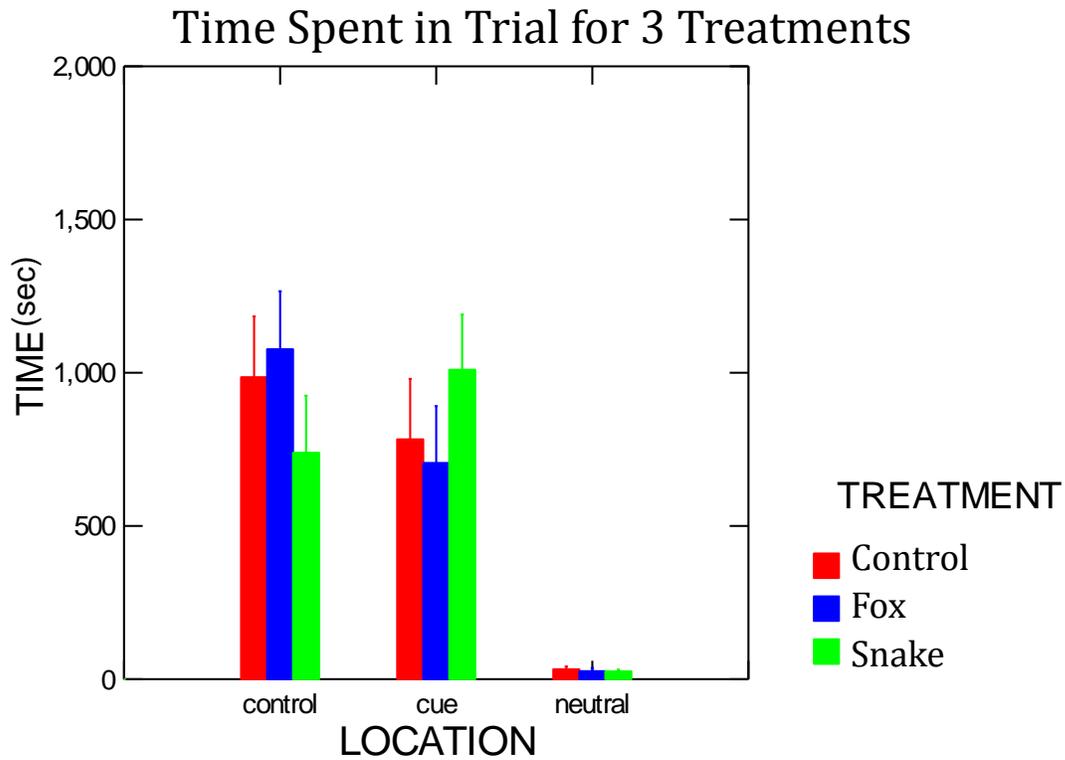
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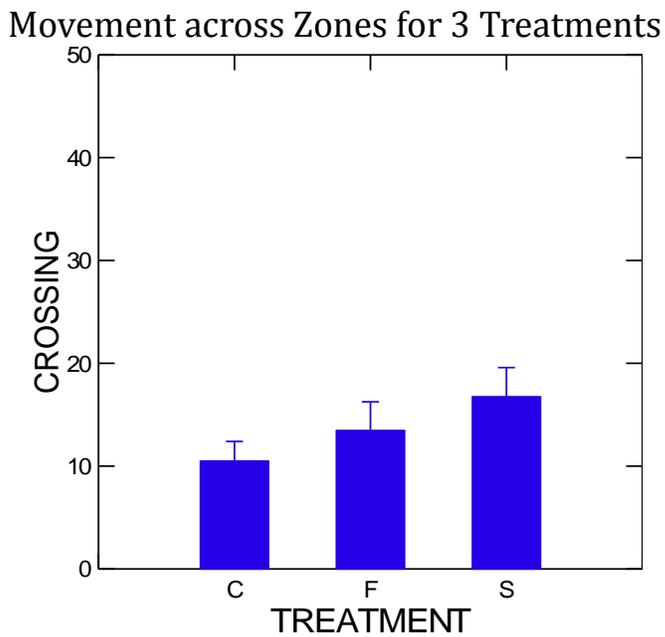
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## Figures

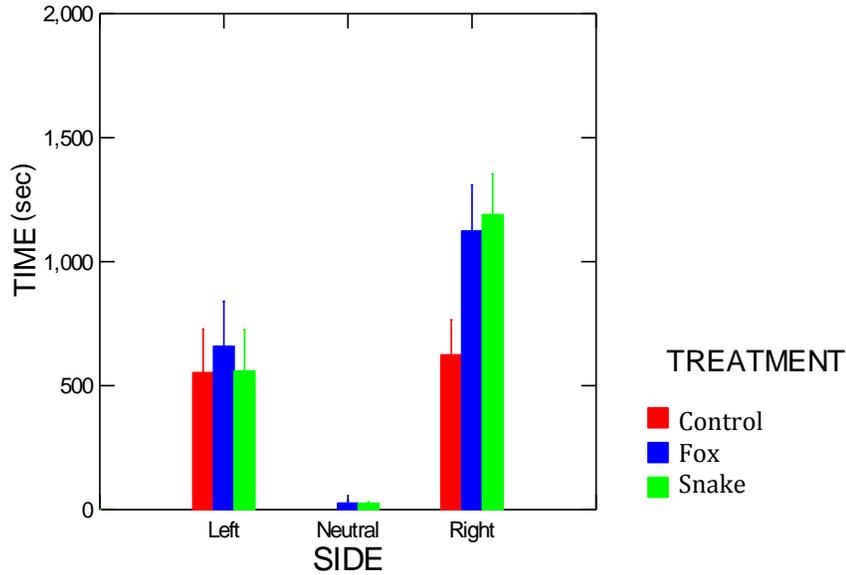


**Figure 1.** The time spent (sec) near the non-cue control, the cue, and in the neutral zone in three predator scent cue treatments



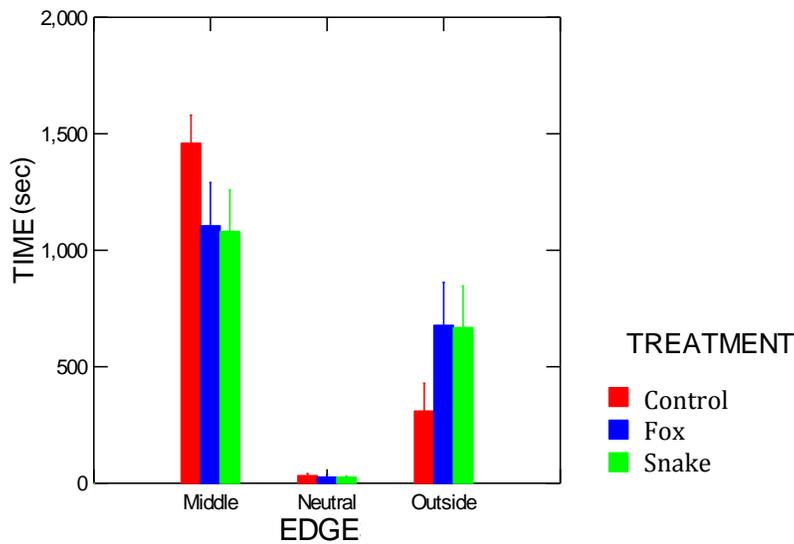
**Figure 2.** The number of times the neutral zone was crossed from the cue zone to the non-cue control zone and vice versa for the control, fox, and snake treatments

### Time Spent on Sides of the Tank for 3 Treatments



**Figure 3.** The time spent (sec) on the left, right, or neutral part of the tank in three predator scent cue treatments

### Time Spent on Edges of the Tank for 3 Treatments



**Figure 4.** The time spent (sec) in the middle of two adjacent tanks, the outside edge of the tank, or neutral part of the tank in three predator scent cue treatments