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Snake Reciprocation in Terms of Classical Conditioning: Scent and Auditory Stimuli

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Abstract

This study explores the classical conditioning of a snake in order to understand and interpret the physiological responses and behavior exhibited regarding experimental design. The conditioning process involves scent and auditory stimuli where the snake is exposed to running water sounds and a mouse scent. The purpose of this research study is to examine how the snake will later associate the scent of the mouse to the sound and how it would react when sound is the only stimuli administered. The type of snake species used was a young Brook's Kingsnake. The snake was placed in a test tank and the experiment was split into four conditioning sessions and one additional session throughout four days of the week to test the outcome. The first session consisted of familiarization with foreign objects and new placement. The following sessions are followed by a series of conditioning the snake to the sound with the mouse scent. The results indicate that the length of conditioning affected the snake. The results could have been more satisfactory if it was shortened to about four or five tests conducted through two sessions in about two days instead since it was twice as long. This is showed based on the second day's data where the snake was most responsive to the stimuli (Average tongue flicks = 102, duration spent near stimuli = 3 minutes, and speed reaction was moderate).

Keywords: Classical Conditioning, Animal, Stimulus, Snake

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The biological adaptations of snakes have led them to sense their surroundings through sensory functions like the flicking of their tongue. Snakes have forked tongues and its structure helps them to identify chemical gradients used as indicators of information in the environment. It serves its role in terms of "smelling" which gives them a sense of direction to navigate their surroundings. Their tongues lack receptors for taste and smell, but the different chemicals aid in relaying different electrical signals to the brain. Snakes have a three-dimensional way of smelling and use their neural circuitry which is part of their vestibular system in the vestibular nuclei of the brainstem to compare the strength of the signal or motion from a smell or sound generated from each direction of its body in order to evoke an impulse for it to act on and discern the orientation that it is coming from (Durso, 2014). Then it can be said, snakes flick their tongue to stimulate their sensory system for them to be responsive and acknowledge their local environment.

They are limited in certain aspects in regard to their senses, like hearing, since snakes do not have external ears but hear sounds differently than other animals and human beings. Although they have structural components that are missing, research has shown that even though there is an absence of auditory response, there are accounts of snakes responding to certain levels of vibrations (Hartline & Campbell, 1969). Snakes have fully formed inner ear structures but without eardrums. Sawe's (2019) research study found the following:

> Besides the inner ear structure, snakes have a quadrate bone in their jaws that move in response to vibrations as they slither on the ground. For several years scientists were not sure if the snake could hear airborne vibrations. Recent research has indicated that the bone can also respond to airborne vibrations. It is believed this is possible because of

> spinal nerves that conduct vibrations from the skin. This type of hearing is known as somatic hearing. Just like the ears of other animals, the vibrations are sent through the bones to the inner ear and transmitted to the brain as signals where they are interpreted as sound. (para. 3)

Due to a snake's biological ear setup, it is more sensitive to ground vibrations and can only hear a narrow range of frequencies.

Hertz (Hz) is used in this context to establish if snakes can hear sounds. It is a unit of measuring the frequency of low or high sounds. When there is a sound, snakes can recognize low-frequency airborne vibrations and ground vibrations that vary between 50Hz and 1,000Hz. Auditory pitch by air but not low enough in sound range, the snake will not react. Other studies suggest that in the range of 200Hz and 300Hz, and even between 80 Hz and 160 Hz, is the peak sensitivity of the snake. There are many studies, but much is not understood about the exact hearing range of a snake. Although based on the level of frequency compared to the lowest sound of an organ pipe (between 20Hz and 25Hz), and highest note on a piano (4,100 Hz), it is deduced that the snake can only hear low sounds (Sawe, 2019). Although snakes have a limited hearing range, they rely on other senses like flicking their tongue to process odor molecules in the air.

This study examines how this snake species would behave if this were to hypothetically occur (be conducted) in its natural habitat. If the snake senses the scent of a prey in a certain location, and they continuously smell it in that place, then snakes will regularly approach that place and associate it as a source to find food and food signifies survival. Natural instincts activate and will induce the snake to search in that area until it loses interest because of the outcome of not receiving food.

Classical conditioning is a learning process that occurs through associations between a neutral stimulus (sound) and an unconditioned stimulus (mouse scent) (Cherry, 2019). No reward is provided to the snake in order not to involve an approach with operant conditioning and to make the experiment more realistic since snakes might not receive food for more than a week in the wild and additionally analyze how much it can continue to be interested by just a scent. Therefore, the purpose of this study is to condition the snake to come to the sound without the scent of food and in terms replacing the association of scent means food to sound means food. The hypothesis of the experiment predicts that the snake would show an increase in physiological responses and respond to the sound in place of the mouse scent after conditioning.

Method

Subjects

The subject of the experiment is a Brook's Kingsnake (Lampropeltis getula brooksi). They are known to be non-venomous. The snake used was born in a secured area where snakes are held captive and not in a natural environment. It is a subspecies of the Florida Kingsnake and part of the Colubridae family. It lives in a plastic container that fits its relatively small size. The snake is around a year old. It has black with yellow “specks” on and throughout its scales. The colors range from light to dark. The underside is a pale yellow to white.

The geographic location of the Lampropeltis getula brooki (speckled kingsnake) is found in Florida especially in the southern parts. They are found in farmlands, cypress swamps, oak hammocks, and everglades (ItsAaronRose, 2021, para 1).

Design

The type of research that was conducted was a quasi-experimental design involving behavioral conduction of classical conditioning. There was manipulation of the conditioning of the snake, but no random assignment was done. The snake chosen was based on being easily accessible and manageable. The experiment involved one independent variable, the conditioning, and four different levels or conditions which were: a device without sound, a device with sound, mouse scent on cotton swabs, and a device emitting sound placed with the scent on the cotton swabs. The dependent variables involved the speed of response of the snake, the approach to the source of sound measuring the length of time spent near the device, and the number of tongue flicks made in the area closest to the device and in its direction.

The experiment was split into four conditioning sessions and one additional session throughout four days of the week to test the outcome. The first session consisted of familiarization with foreign objects and new placement and one conditioning test was conducted at the end. The following sessions are followed by a series of conditioning the snake to the sound with the scent. Each session was done once a day while the last day had two sessions. The sessions were conducted for a period of one hour each in the same week in consecutive days. The sessions, each during conditioning, were split into three tests and each test was timed for exactly 5 minutes. The spacing done between each test was reduced further on and varied in duration. This design was most suitable based on lab hours and scheduling.

Procedure

The snake was set in a special area, a test tank, made for it to be able to move within its parameters. For this experiment, natural scents were used in a comparative test on odor and sound stimuli recognition. Artificial fragrances were not used to avoid irritation and sensitivity. The snake was presented with a type of odor, in this case a mouse odor. A mouse odor would

stimulate the snake's palate because mice are part of the snake's diet. The snake would smell the scent and approach it. Then, when presented with the scent in addition to the snake's reaction, a sound will also resonate. The sound will emit slight vibrations and the device will be placed in the test tank at the side of the wall and on the ground since snakes cannot hear or recognize airborne sounds unless in terms of specific frequencies and are more responsive to vibrations. The earphones will be connected to the headphone jack of the mobile device and the sound will play from it. The earbuds will be placed down inside so that the sound can be felt but not as strongly if coming from a speaker or originally from the phone. The type of sound that will be used is running water sounds. Nothing too strong that has bass in order not to frighten the snake.

Data Analysis

There are three measurements used to assess the snake's physiological responses before, during, and after conditioning. They are the number of tongue flicks, speed of reaction, and duration. The number of tongue flicks is observatory and is counted based on how many times the snake flicked its tongue in close range to the stimuli (used for sensing surroundings and showing interest). The speed reaction is rated on an ordinal scale using the terms slow, moderate, slightly fast, and immediate to distinguish the speed of response of the snake when the items are first added. The duration is the amount of time the snake spent in close proximity to the stimuli.

Results

1. *Before conditioning*

The number of tongue flicks for the headphones without sound was 40, with sound was 80 which is twice the number without sound, and with the soaked cotton swabs that had a mouse's scent was around 150-160 without sound, which was almost double the number of

tongue flicks with just the device emitting sound. The numbers were taken from an observational standpoint. The speed reaction and response of the snake towards the device without sound was slow to moderate, with sound moderate, and with mouse scent alone was slow to moderate. The duration spent in close proximity to the earphones without and with sound was around 2 minutes, and with the mouse scent alone was almost 5 minutes. Each test was timed for exactly 5 minutes.

2. *Behavioral Observations*

When I, the experimenter, seemed to recoil or flinch slightly when first trying to remove the snake from its living space, the snake would sense my reaction and move in a very fast manner away as if also frightened by my behavior towards it. When I approached it directly by not flinching, it stayed still. I can deduce that snakes notice behavior that can make them immediately respond whether to attack an enemy when there is an opening or to flee when sensing danger or are frightened. It also mimicked the behavior of a rattlesnake, shaking its tail, which indicated it felt threatened. This is a natural adaptation for Brooks Kingsnakes. Additionally, it displayed burrowing behavior. It continuously kept trying to dig its head in the wooden floorboard and the sides of the test tank when the sound of running water was playing and when it was not.

On the third day, in the third test, the snake pushed the mouse scent away from the sound and stayed beside the sound. The sound might have been perceived as a way to escape since it slithered up the wall in the direction of the sound. After the sound stopped, it left back immediately to the scent and remained where the scent was.

3. *Conditioning sessions*

Day one: With the combination of trying first the mouse scent without the sound there were 90-100 tongue flicks, its response was slow, and the duration spent near the items was about 3 minutes in total. Then when the mouse scent was paired with the device resonating sound there were 45 tongue flicks, its speed reaction was also slow, and the duration spent was around 2 minutes. The difference was that it approached the source of sound first. Each test was timed for 5 minutes.

The pairing of the mouse scent with the sound was done three times in day two, three, and four. During spacing for each test, the items were removed and then were placed back inside after it was time to condition the snake again.

Day two: There was spacing of time between the first test and the second test and the second test and the third test where it was approximately 10 minutes and then it decreased to 7 minutes respectively. Each test was timed for 5 minutes. The average of the three tests was 102 tongue flicks and speed reaction moderate to spending around 3 minutes in duration at the items and sound.

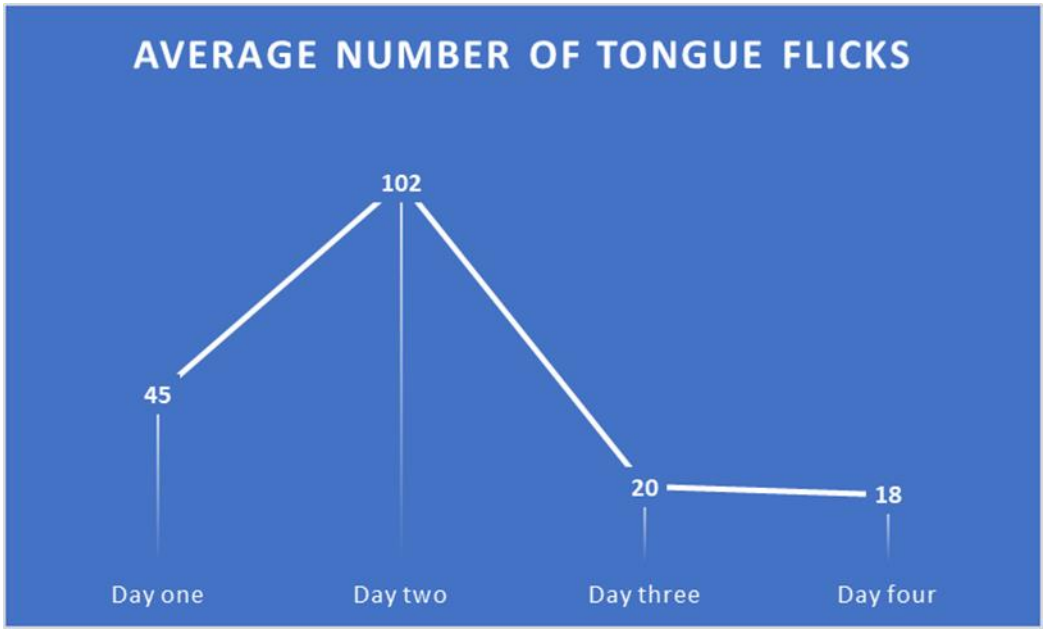
Day three: There was spacing of time between the first and the second and the second and the third where it was approximately 5 minutes each. Each test was timed for five minutes. The average of the three tests was 20 tongue flicks and speed reaction moderate to spending around 2 minutes in duration at the items and sound.

Day four: There was spacing of time between the first and the second and the second and the third where it was approximately 5 minutes then decreased to 3 minutes respectively. Each test was timed for five minutes. The average of the three tests was 18 tongue flicks and speed

reaction moderate to spending around 2 minutes and 30 seconds in duration at the items and sound.

Figure 1.

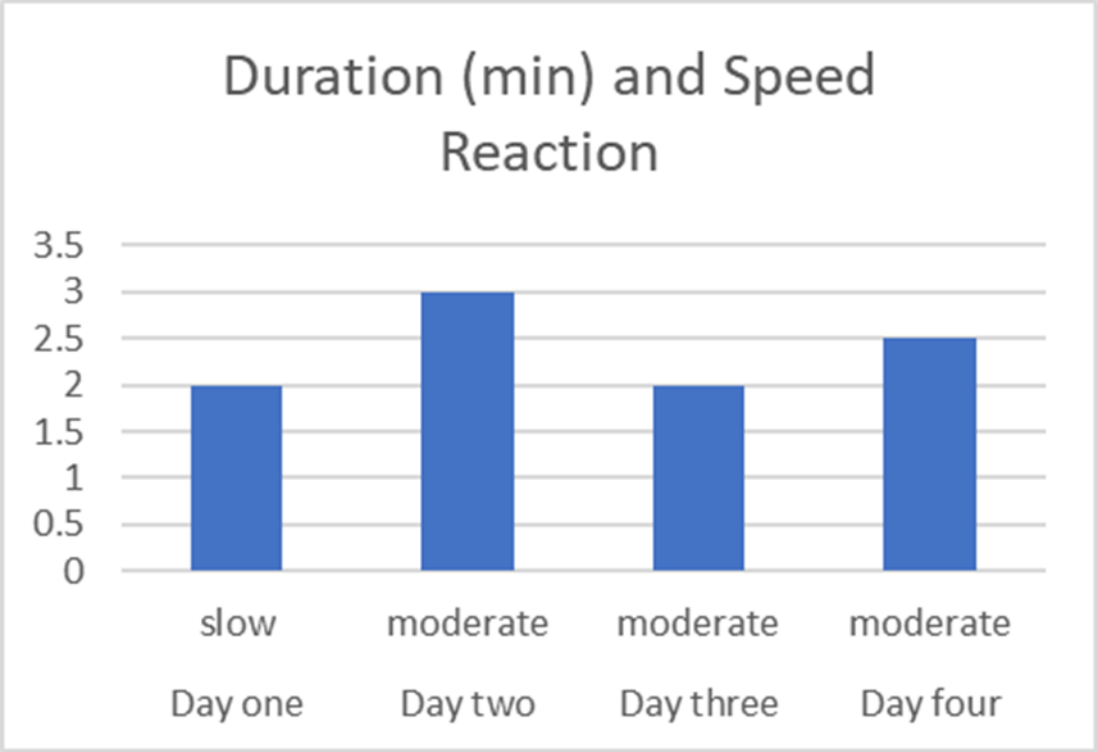
Average Number of Tongue Flicks During Conditioning per Day.



Note. The figure above displays the mean value of the number of tongue flicks during conditioning for each day.

Figure 2.

Average Duration Spent in Close Proximity to Scent and Sound Device in Terms of Average Speed Reaction During Conditioning per Day.



Note. The bar graph displays the mean speed reaction of the snake when presented with the stimuli and the duration spent near the mouse scent and sound during conditioning in four days.

4. *After Conditioning*

After multiple tries and having the snake repeat the process, the odor would no longer be available and only sound would. When the sound was on the right side of the wall, there was no response. When the sound was placed on the left side of the wall, the snake avoided the device with sound at first then it went to it and stayed near it for around 2 minutes and displayed 20 tongue flicks. The sound being on the opposite wall, it showed a response that was slow, stayed for a duration of 1 minute, flicked its tongue around 30 times. The snake spends almost an equal amount of time with the device with no sound at the left wall and with device emitting sound at the right side of the test tank (2 minutes each) while the amount of tongue flicks was 40 with no sound which was greater than with sound which was around 25 instead.

Discussion

The purpose of this study was to examine the relationship between scent and the snake's physiological responses, and how it could be later interpreted. The experiment was to condition the snake and make the neutral stimulus (the running water sound) into a conditioned stimulus which results in a conditioned response. In the hypothesis, there was a prediction that the snake would show an increase in physiological responses and respond to the sound instead of its food's scent after conditioning. The results indicated that there was a decreased interest over time in the repetitive process and eventually showed that it did not respond as predicted or expected.

This study has limitations. The housing enclosure of the snake could have affected its interest in the items and behavior. The snake resides in a small, enclosed space and the test tank is bigger and has more room for the snake to stretch and explore its surroundings. The snake's active display was most likely due to the change in its setting which gives it the ability to do more roaming than its inhabited space. This would automatically increase its interest because of the environment being foreign and unfamiliar (Hoehfurtner, Wilkinson, Walker, et al., 2021). A less big and wide enclosure replaced by a size almost similar to its housing space would have potentially caused a decrease in interest in its surrounding and an increased focus on the active stimuli present.

The type of snake species used might have affected the outcome of the experiment. A different kind of snake that did not have a burrowing behavior might have resulted in a different response since it focused on trying to hide. The enclosure could have had enrichment where it resembled more of its home space. According to Hoehfurtner, Wilkinson, Nagabaskaran, and Burman (2021), research "found that increased enrichment and enclosure size together resulted

in snakes being more active and visible a greater proportion of time" (p. 6). This would be in contrast to how they would be in a standard enclosure.

This outcome could have resulted from an excessive number of conditioning trials since it was paired together, the scent and sound, 10 times. It appears that conducting the conditioning associations between the previously neutral stimulus paired with the unconditioned stimulus at about 4 to 5 times was enough to condition the snake and make the sound a conditioned stimulus. Although, when it continued to 10 sessions it lost interest because of the lack of reinforcement and finding nothing repeatedly. This could mean that snakes, when searching for prey, will repeatedly revisit an area that has its food's scent for the amount of 4 to 5 times that is in terms of these sessions accounts for two to three times in an hour per day so which would amount to 2 to 3 days in total.

Some differences could make this study appear to be more accurate and valid and have a better outcome. If the study incorporated some level of operant conditioning, the snake could have been more receptive. "Despite the fact that operant conditioning techniques have been applied to a wide array of vertebrate and invertebrate species, there are very few reports of operant conditioning in snakes, and most of those are limited to small species that perform behavioral responses in mazes and/or escape paradigms" (Emer et al., 2015, p. 275). Thus, this conducted study reveals in perspective how many times a snake would approach a location with sound when only smelling scent and not being reinforced.

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