



## **EXTINCTION OF THE ELECTRICAL PHOBIC AVERSION: SINGLE CASE DESIGN IN A CD1 MOUSE**

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### **Abstract:**

Fear, like other emotions, can be transferred in animals if it is conditioned to a specific stimulus. Instrumental conditioning shows the sound as a primary stimulus and a secondary stimulus of aversion that produce the phobic behavior, and the extinction process is established by a frequency of the primary sound stimulus. The purpose of this study is to demonstrate the extinction of electrical phobic aversion in a single case of a CD 1 mouse through the pre-experimental design, where in the first phase the acquisition of phobia occurs through conditioned stimulus such as sound and electric current. In the second phase, the extinction process by removing the secondary stimulus. According to the objective, the result of this intervention was the extinction in 70% of the phobic aversion to electric shocks in the CD1 strain through conditioned stimulus. The experiment lays out the first clues of the responses manifested by the mouse in each trial, and this information will be useful to know the frequency and duration of these phobic behaviors in the stage of acquisition and elimination of the phobia.

**Keywords:** Instrumental Conditioning, Conditioned Stimulus, Phobia Extinction, CD1 Mouse.

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## 1. INTRODUCTION

Behaviorism is a current of psychology that has accompanied and profoundly marked the history of science and occidental society in the 20<sup>th</sup> century. It is based on the concept that only behavior, as a measurable phenomenon, can be studied. An orientation that offers a therapeutic model based on the symptom resolutions and behavior modification based on conditioning and thawing techniques, where positive behavior is associated with positive reinforcement and vice versa.

From an overall perspective, the historical contribution related to current mechanisms of extinction to electrical phobic aversion in a CD1 mouse is explained. In this unique case, we return to the most significant contributions of orthodox and radical behaviorism as a base for evaluating experimental animal behavior. In other matters, psychology recognizes two types of conditioning: classical conditioning (CC) and operant conditioning (OC). The CC (also called Pavlovian) was demonstrated first by Ivan Pavlov (Pérez-Acosta & Cruz, 2003); while the contributions of the CO were made by the American psychologist Burrhus Frederic Skinner (Perez Alvarez, 2021). As for classical conditioning, Ivan Pavlov's experiments had a great influence, as well as several of its principles: acquisition, extinction, discrimination, and generalization. The classical conditioning method is based on the stimulus-response association. This procedure associates an unconditioned stimulus, which produces a response, with an unconditioned one. After several trials, the conditioned stimulus can produce a response like the unconditioned stimulus. (Núñez Cansado et al., 2015)

In this order of ideas, it is mentioned then that the behaviorist discipline studies human and animal behavior. For Watson, behavior manifests itself in the physical and chemical phenomena of the organism that are learned throughout its life or through conditioned experiments (Pellón, 2013). The Albert case is an experimental phobia model by Watson and Rayne (Digdon et al., 2014), which was based on Pavlovian conditioning. He explains that fear, as well as other emotions, could be transferred to other elements if it was conditioned to a specific stimulus. In this sense, the operant conditioning model proposes that organisms can change their behavior according to consequences (Remmelink et al., 2015). A reinforcing stimulus is associated with operant behavior; if the stimulus is presented different pattern will produce a different pattern of behavior. On the other hand, among

these historical postulates, it is worth mentioning Edward Tolman and Edward Thorndike. Tolman proposes "purposeful behaviorism" that approaches behaviorism from the concepts of intention and purpose (Pautassi & Godoy, 2003), instead Thorndike developed instrumental conditioning through experiments related to the behavior and learning of cats. The author focuses his interest on the prize as a reinforcing element that connects stimulus and response (López Cruz, 2021). That is, learning is reinforced as it is repeated. However, B.F Skinner is the most relevant author to date in the study of operant conditioning from radical behaviorism. His experimental contribution in pigeons or other animals was developed in an environment free of distractions called Skinner's box or other forms with similar mechanisms, in which he could have control of the conditions in which food was provided to animals reinforcing behavioral control. Skinner is ultimately the one who leads psychology to the treatment of behavior. Where learning is established under a series of contingencies product of the emission of operant conditioning (Quiroga, 1995).

Since psychology emerged as a science, numerous conditioning studies have been presented through experimental designs in laboratories, where the researcher controls the variables and the random assignment of the subject to different conditions (Balluerka & Vergara, 2002). The reduction of phobias has shown efficacy through therapeutic interventions in the behavioral field, such as assisted exposure therapy, cognitive-behavioral therapy, and virtual reality. Most of the recognized works are related to exposure with cognitive techniques. (Capafons Bonet, 2001)

About studies related to the effectiveness of the behavioral program, there is that of Mateu-Martínez and others (2013) whose results also show the need to prevent social rejection and other pathologies in the school environment. The work of Sánchez Román and others (2006), whose objective was to evaluate programs to decrease anxiety in medically ill patients showed that psychoeducational intervention programs decrease anxiety levels. Furthermore, Amodeo's research (2016) shows a decrease in panic attacks and anxiety levels when undergoing treatments through interoceptive exposure and physical exercise.

It is relevant to consider that, through experimental designs with operant and classical conditioning, there is the study that the therapy of exposure and extinction of fear would be reinforced by a series of exercise prior to training

(Jacquart et al., 2017), where laboratory rats were conditioned to fear manipulation therapies. In relation to the aversion technique responds to positive punishment, through aversive stimulation presented in vivo a behavior can be reduced or extinguished. It can be presented through a small electrical stimulus. (Fernandez, 2004)

CD1 mice maintain the characteristic of genetic creation for the use of multipurpose model of experimentation, their strain code is 022, the use of CD1 mice supports objectivity in the performance and development of relevant scientific contributions in the clinical psychological field; in other matters, it maintains only a cycle of 12 months of life. The CD 1 strain corresponds to albino mice for laboratory use and we work with them following the bioethical aspects that support the welfare of the animal. They have developed the senses of smell, taste and vision (Vargas et al., 2018). At this point, researchers such as Pulido and Gallardo (2017), Orozco and others (2017), Pulido and Manzano (2019) experiment with rats and observe their response in a defined space-time. While Santana and others (2020) designed an experiment to observe the behavior and response of animals in free conditions and with head restraint. Finally, an article related to the regulations and ethics in experiments with laboratory rats during classes at a university was considered so that animals are not subject to sacrifice or mistreatment (Reivan-Ortiz, 2020), also article 146 literal 4 of the Organic Code of the Environment in Ecuador (2017) guidelines on the use of animals for research. Haga clic o pulse aquí para escribir texto.

Instrumental conditioning details a primary stimulus and a secondary aversion stimulus that form phobic behavior, and using a continuous reinforcement program leads to extinction. The purpose of this study is to demonstrate the extinction of electrical phobic aversion in a unique case of a CD1 mouse through the pre-experimental design, where in the first phase the acquisition of phobia occurs through conditioned stimuli such as sound and electric current. and in the second phase the process of extinction through the withdrawal of the secondary stimulus.

## 2. METHOD

### Design

The study has a two-phase pre-experimental design. Phase one is aimed at acquisition: with the

independent variable electrical stimulus and sound; and the dependent variable fear. The second phase extinction is formed by the independent variables in the absence of electrical stimulation and presence of sound and the dependent variable fear. The nomenclature is as follows:

Formula:  $x_1 - x_2 = A - E$ . F = single case

$x_1 = sound (treatment - intervention)$

$o = observation - analysis of experimental animal behavior$

$x_2 = electric shock (treatment - intervention)$

*difference = Phobic elimination (subtracts final vs initial value)*

*Biological unit of analysis*

An albino mouse, male, CD1 strain, 25 grams, 12 weeks old. The animal comes from the National Institute of Research and Public Health of Ecuador.

### Instruments

The Skinner box to be used is a classic model with current systematic software redesign (Raspberry Pi 3) that allows analog and digital operation with monitor and keyboard for execution, has a video camera on top, three integrated audio speakers, has a metal structure with coverage and cardboard extension for electrical system later, attached we find alternating current analog voltmeter box, in its lower internal part we find a grid connected with electricity with CD1 mouse adjustment, no light or lever elements will be used for food or water, we conclude describing in its front double compartment with manual access ascending glass window.

### Procedure

#### Acquisition:

The CD1 mouse was removed from the animal facility and placed in an experimental box. This step was carried out three days before the start of the tests, to get used to the environment. The rodent was then fed into Skinner's experimentation box for the final test.

The acquisition phase of phobic aversion was tested in three trials. In the first, conditioning of a primary sound stimulus and a secondary stimulus of electrical aversion with continuous operational execution of 5 seconds was performed. It was rehearsed three times, with an interval of 2 and 4 minutes. It is in the third test, where the level of acquisition of experimental phobia was reached (Table 1).

Table 1. Trial of the phobia acquisition phase

Essay N°	Amperage	Observation	Answer	Time
1	1 mA	Direct	phobia 5 sec	2 min
2	2 mA	Direct	phobia 5 sec	4 min
3	n1	Direct	x1 - x2= 5 sec	0
4	0	0	0	0

**Extinction:**

In the second phase corresponding to the extinction process, the primary sound stimulus was established with the withdrawal of secondary

electric current stimulus. Five trials were tested with an interval of 5 seconds, in the later there is an interval of 30 seconds depending on the final trial 5.

Table 2. Phase 2 trial

Trials	Level	Observation	Answer	Time
1	n1	x1	x1 - ext. 5 sec	5 sec
2	n2	x1	x1 - ext. 5 sec	5 sec
3	n3	x1	x1 - ext. 5 sec	30 sec
4	n1	x1	x1 - ext. 5 sec	15 sec
5	n1	x1	x1 - ext. 5 sec	0

Extinction of experimental phobia = 45 sec.

**Data analysis**

The study of the data was done by direct observation of the behavioral behaviors of the animal. We analyze the acquisition of fear through overt aversion behaviors and the extinction of fear through the suppression of associated behaviors.

**3. RESULTS**

For the results it was observed that the frequency, duration, intensity and target behavior and observable behaviors of the acquisition, the frequency was given by the number of trials, in this case are 4. Four levels of rank were established to

verify the level at which the acquisition occurs.

The duration corresponded to the established time of sound and current for classical operant conditioning, a duration of 5 seconds was established for conditioning.

The intensity of the secondary stimulus was determined at 70 volts based on a 4-level parameter where level three was selected, which corresponds to 70% of the current chip's intensity.

It was observed that through the three trials and the intervals, aversive behavior was acquired at a level of phobia that manifested itself with the behaviors stipulated in Table 3.

Table 3. Behaviors observed during the phobic acquisition phase

Essay N°	Observed results
1	Avoidant behavior and flight, squealing, piloerection, jumping, spinning,
2	Avoidant behavior and flight, squealing, jumping, spinning piloerection Defecation, urination
3	Avoidant behavior and flight, squealing, jumping, spinning, piloerection Defecation, urination, nervous tic, inhibition of movements, muscle contraction, contraction of the tail mechanism, crouching in a corner, hair loss, inhibition of vision, does not detect colors, hunched body, global tremor of limbs, daze.

Between the process of phobic aversion and extinction there was a 3-minute break.

**Extinction of phobia**

Five tests were conducted, where the primary variable of sound and at the same time the elimination of electrical aversion were established, with a frequency of 5 seconds of the primary sound stimulus. The intervals are 5 seconds between test 1 and 3. After the third trial there is an interval of

30 seconds, which continues a fourth trial with an interval of 15 seconds to reach the last, where the extinction response is evidenced subject to the experimental behavioral analysis of phobia reversal.

The process of extinction occurred from the beginning because it did not receive the

aversive current. This secondary stimulus disappeared in 70% in the fifth trial where the hyperaesthetic decrease is evident. Table 4 shows

the behavior of rodents during the elimination of experimental phobia.

Table 4. Behaviors observed during the generation phase

<i>Trials</i>	<i>Observed results</i>	<i>Remission rate</i>
1	<i>Hypersensitivity due to sound conditioning, decreased flight and escape mechanism, does not jump, does not emit squealing sounds, maintains piloerection, does not perform urination or defecation, maintains muscle retraction</i>	30%
2	<i>Decreases nervous tic, decreased muscle contraction, performs leg movement</i>	
3	<i>Walk without contracting your tail, open your eyes, regain your vision,</i>	
4	<i>Start the parsimonious walk to tranquility, decreases the tremor, does not contract the tail</i>	70%
5	<i>Insinuation of gnawing, nose movement, paw movement, partially decreases sensitivity</i>	

The result was an almost total remission of extinction of aversive phobia with electric current supported by the extinction mechanism, by reducing avoidant behavior or flight.

Table 5. Comparative table of phobia time structuring

<i>Case</i>	<i>Allocation</i>	<i>Preprueba</i>	<i>Posprueba</i>
1	Acquisition	3 fr essays	Phobia Duration in 4 minutes
	Extinction	5 fr essays	Phobia reversal

#### 4. DISCUSSION

The purpose of this study was to demonstrate the extinction of electrical phobic aversion in a unique case of a cd1 mouse by pre-experimental design. In accordance with the objective, this intervention was positive because 70% of the phobic aversion to electric shocks in the CD1 strain was extinguished by conditioned stimulus, based on the behavioral changes that occurred in the acquisition of the aversive behavior and the extinction, which was stabilized with more functional behaviors. The extinction of phobic aversion was measured according to the decrease in behavior; that is, the same responses were not obtained in each trial where the sound, conditioned by the electric current, was reversed.

In the present study, the primary stimulus is sound and the secondary stimulus, the current of electricity. Here the amperage of electricity was 2  $m_A$ , while Chaaya et al., (2019) subjected a group of rats to electric shocks of 1  $m_A$  as the primary stimulus; in the same experiment, another group was conditioned with auditory tones as the primary stimulus for 60 seconds. Stansley et al (2018), also

experienced fear conditioning in mice with electric discharges of 1 s, 0.7  $m_A$  spaced 30 s apart.

During the acquisition phase, the association of sound and electrical charge reflect changes in the rodent's behavior pattern up to the level of phobia. A series of specific behaviors are recorded in each trial as the operational frequency increases which coincides with the study of Orozco and others (2017) where the results of the observational analysis of behavior show orderly changes in Wistar strain rats exposed to experimental manipulations. Regarding the CD1 mouse in this experiment, phobic behavior was manifested from the first discharge of electricity as in the research of other authors. (Poulos et al., 2016) (Navarro, 2021)

After conditioning, the CD1 mouse was subjected to extinction tests. Phobia behaviors disappeared in the fifth trial; which means is achieved in one day like the studies of Quirk and others (2000); and Sidorov and others, (2018) albeit with mice of different strains. Although CD1 does not achieve the total extinction of behavior, something like what happened with the Wistar rats of Navarro's experiments (2021).

In reference to the limitations of the present study, these are related in the first place to the duration of the experiment. The time allocated for the trials was one day, which although it reached an acceptable percentage of extinction of phobic behavior could not be verified if in the days this percentage increased when subjected to more tests.

The other difficulty has to do with the impossibility of comparing the behavior of the mouse with others of the same strain, but of different sex and age; because it was tested only with a twelve-week-old CD1 male since studies such as those of Bisby and others (2021) demonstrate different behavioral phenotypes in adolescent mice and rats.

Being a single-day experiment, it is not possible to verify that the fear process can occur again during the following days through a longitudinal methodology, which has happened.

It would be interesting that future experimental research on the elimination of phobias, through conditioned stimuli, is oriented to comparative analyses of the behavior of CD1 strains with different characteristics, subjected to tests for several days to observe significant differences.

For now, the experiment discussed here raises the first clues to the responses manifested by the mouse in each trial. This information will be very useful when you want to know the frequency and duration of these phobic behaviors in the stage of acquisition and elimination of the phobia.

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