



Dogs do not exhibit avoidance behavior in response to the smell of snakes

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Abstract: Dogs possess the ability to recognize potential dangers associated with snakes primarily through their visual system, rather than relying on their olfactory system. Previous studies have predominantly focused on the odors of dangerous snakes in the American continent, which do not overlap with the areas of the domestication process. Consequently, it is unlikely that present-day American dogs have developed innate responses to local snakes due to limited historical exposure. The objective of the current study is to conduct experiments involving dogs and the odor of venomous snakes distributed in regions where the ancestor wolves of dogs coexisted and where domestication occurred. For this purpose, we have specifically chosen three species from the *Vipera* genus. The study involved 40 domestic dogs with an average age of 34.6 ± 26.7 months. Each dog participated in a single one-minute trial and was randomly assigned to either the vipers or control conditions. The trials took place in a room containing the owners, the apparatus, and two water bowls. Nonparametric Mann-Whitney tests were utilized to analyze the data. The results indicated that male dogs exposed to the smell of vipers interacted with the apparatus for a longer duration compared to the control odor group. They also remained in the apparatus zone for an extended period. However, no statistically significant results were observed for females and other variables in both sexes, including behaviors directed towards the owner and the door, exploration, passivity, and stress signals. The findings of this study suggest that dogs do not exhibit discrimination between the scent of vipers and the control odor. Consequently, it supports the notion that dogs may be unable to avoid snakebites when they frequent areas populated by venomous snakes.

Key Words: Dog, Snake, Viper, Olfactory system, Domestication.

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Introduction

Snakes represent a significant threat to dogs, as indicated by scientific reports on the epidemiology of snake-related injuries around the world (Adhikari et al., 2018; Alcoba et al., 2022; Barr, 1984). According to the cited papers, mortality rates exceeding 10% have been reported depending on the species of snake involved.

Dogs appear to possess the ability to recognize potential danger associated with snakes by utilizing their visual system. Research by (Siniscalchi et al., 2010) suggests that dogs primarily focus on the left visual hemifield when observing a black silhouette of a snake against a white background. This preference can be attributed to the neural pathways from the lateral visual field of each eye, which predominantly project to the contralateral side of the brain (Rogers & Andrew, 2002). The utilization of the left visual hemifield to inspect the snake silhouette aligns with the specialization of the right hemisphere in dogs for expressing intense emotions such as fear and engaging in escape behaviors, as previously demonstrated in canine species (Siniscalchi et al., 2022). However, snakes are frequently elusive and can be difficult to spot, especially when hiding in grass or vegetation. In such cases, the olfactory sense becomes crucial in promptly alerting dogs to the presence of danger.

According to (Weldon & Fagre, 1989), the scent gland secretions of the western diamondback rattlesnake (*Crotalus atrox*) do not appear to be aversive to dogs. Additionally, a study comparing dogs' responses to various odours, including southern Pacific rattlesnakes (*Crotalus oreganus helleri*), rosy boa (*Lichanura trivirgata*), mouse (*Mus musculus*), and snail (*Cornu aspersum*), found that dogs showed a higher level of interest in the scent of rattlesnakes compared to rosy boa or snail, but less so than mouse. However, no evidence was found to suggest that the odor of rattlesnakes elicited unpleasant emotional states in dogs (Mulholland et al., 2018). These findings provide a potential explanation for the reported high rates of rattlesnake envenomation in dogs, as dogs may be induced to investigate rattlesnake odors without experiencing fear. The findings of the mentioned papers were somewhat unexpected, considering that dogs are known to rely on their olfactory sense to exhibit innate fear responses. In a study by Samuel et al. (2020), it was observed that dogs spent less time in the presence of predator scents and exhibited an increased heart rate compared to their baseline heart rate when exposed to the scents of brown bears and lynx. Considering that the dogs in the study had no prior exposure to the scents of lynx and bears, these results imply that dogs inherently possess the ability to detect potential threats through their olfactory-based fear response. The question arises as to why dogs display fear responses towards potential predators but have not developed similar responses to the scent of snakes, despite the potential threat they pose. One possible explanation could be attributed to the fact that previous studies focused on dangerous snakes from the American continent (Weldon and Fagre, 1989; Mulholland et al., 2018), where the population of wolves was not involved in the domestication process. American dogs belong to a monophyletic lineage that likely originated in Siberia and subsequently migrated to the Americas alongside humans. Following the arrival of Europeans, native American dogs experienced a significant decline, resulting in minimal genetic influence in modern dog populations (Leathlobhair et al., 2018). It is plausible that an aversive effect of predator odors may only exist when the predator and prey have a long evolutionary history in parallel, leading to a genetic predisposition in the prey species to avoid the odors of sympatric predators (Stoddart, 1980). Therefore, it is unlikely that the current American dogs have developed innate responses to local snakes due to their limited exposure and evolutionary history with them.

Hence, our objective is to conduct experiments involving dogs and snakes in regions where wolves, the ancestors of dogs, coexisted and where the domestication of dogs occurred. For this purpose, we have chosen three specific species of vipers: (i) *Vipera aspis*, which is widely distributed in various habitats and is locally abundant (Saint Girons in 1989). According to the International Union for Conservation of Nature (IUCN), *V. aspis* has a broad distribution primarily across southern and eastern Europe; (ii) *V. ammodytes*, which can be found in southwestern Europe; (iii) *V. berus*, which has the most extensive distribution across Eurasia, extending all the way to the far east (Cui et al., 2016). The selection of these three viper species is based on their distribution patterns, which align with the regions where dog domestication took place. This correlation is substantiated by compelling genetic and archaeological evidence obtained from studies conducted on modern and ancient dogs and wolves. Collectively, these findings consistently indicate that Eurasia was the primary geographical region where the process of dog domestication occurred (Leonard et al., 2002; Thalmann et al., 2013; Wang et al., 2016).

Our anticipated outcomes are as follows: if dogs perceive snakes as a threat, we hypothesize that they will exhibit avoidance behavior towards the apparatus. In such cases, the attachment r within the dogs will be activated, leading them to seek greater proximity with their owner for safety and reassurance (Topál et al., 1998; Prato-Previde et al.; 2003; Mariti et al., 2013; Scandurra et al., 2016). On the contrary, if dogs view snakes as potential prey, we expect them to display heightened interest in the apparatus and engage in olfactory exploration. This experiment aims to provide valuable insights into how dogs perceive and respond to venomous snakes, shedding light on their instinctual behaviors and underlying cognitive processes.

Material and methods

The study included a total of 40 domestic dogs (*Canis familiaris*) residing in Naples, Italy. The sample comprised dogs of diverse breeds, with 40.0% being female and 60.0% male. These dogs were kept as household pets and lived in close association with humans. The recruitment of participants was conducted through a combination of personal contacts and online platforms. The age of the dogs ranged from 4 to 84 months, with a mean age of 34.6 ± 26.7 months. Prior to the study, it was determined through owner interviews that none of the dogs had any previous exposure or experience with snakes.

The research focused on three species of vipers: *Vipera aspis*, *V. ammodytes*, and *V. berus*. All these snake species were housed at the “Zoo delle Maitine” located in Campania, Italy.

To collect snake odours, a clean cardstock (typically employed for rearing purposes) sample was placed inside the glass case where the snakes are kept for a duration of 7 days. Snakes were not fed during this period to prevent any potential contamination of the cardstock with food odors. The cardstock was able to absorb the odor of the snakes as well as their physiological excretions.

After the 7-day period, the cardstock samples were carefully retrieved using disposable gloves. These samples were then sent to the LabEC (Laboratory of Canine Ethology, University of Naples Federico II, Italy) and stored in a freezer at a temperature of -80°C until they were ready to be used in the experiment.

Each cardstock sample was subsequently cut into squares measuring 6cm x 6cm. The squares belonging to the *Vipera* genus were combined together, while squares from unused cardstock served as the control group. These cardstock squares were then employed in the testing procedure of the study.

The experiment was conducted in a 3.7 m x 2.9 m ethological room, which was divided into four distinct zones using adhesive tape as boundaries. The first two zones, labeled A and B, represented the door zones, and were enclosed within a perimeter of 50 cm x 100 cm each. The third zone, named the owner zone, had a perimeter of 100 cm x 120 cm, and contained the owner’s chair positioned with its back against the wall. Lastly, the fourth zone, the apparatus zone, housed the apparatus and was situated in front of the owner’s zone, with a perimeter measuring 50 cm x 100 cm. Additionally, two water bowls were placed in the corners adjacent to the apparatus zone (Fig. 1).

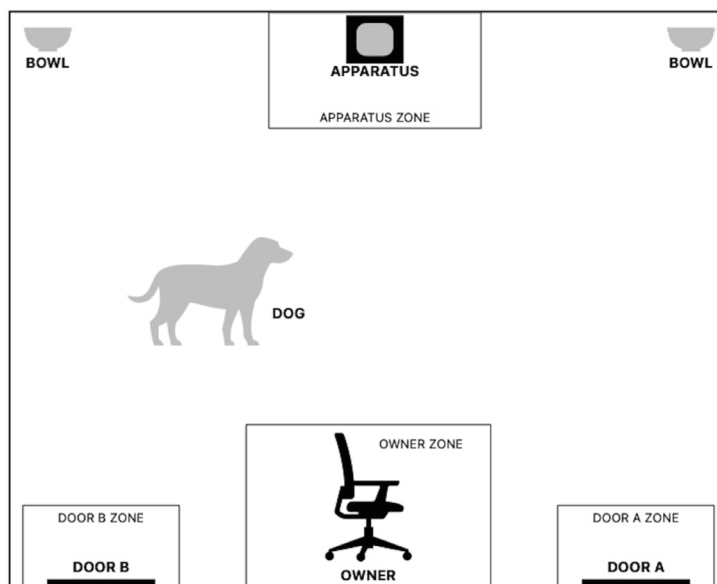


Figure 1. Ethological room.

For odor dispersion, two distinct apparatuses were employed to prevent contamination: one for the vipers and another for control purposes. Each apparatus consisted of a curved tube with a perforated plate at the bottom, allowing for the insertion of odor samples (Fig. 2). The ethogram adopted is in Table 1.

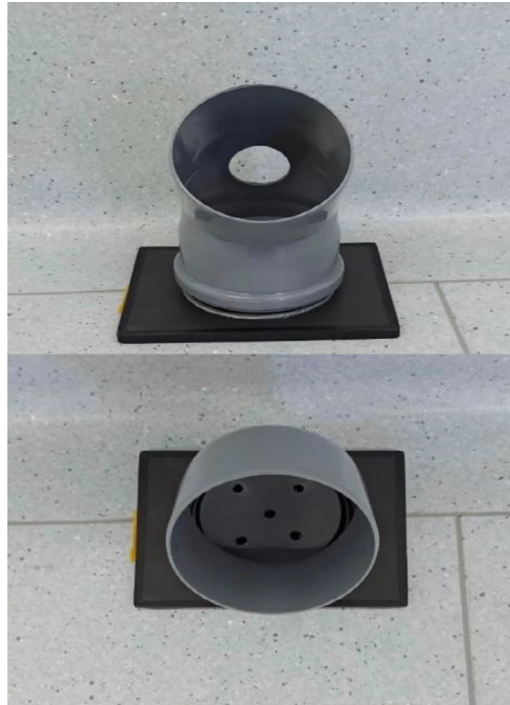


Figure 2. Apparatus

Before starting the test, the handler and the dog were welcomed by the laboratory staff outside the laboratory. The necessary information about the dog was recorded through a questionnaire, and a bowl of water was provided *ad libitum*, ensuring that the dog could drink freely. This was done to ensure that drinking behavior during the test was not driven by thirst and could be recorded as stress behavior. During this phase, detailed explanations and instructions regarding the procedure were provided, without disclosing the specific objective of the study. Subsequently, a familiarization and exploration phase took place. The handler and the dog, while on a leash, entered the ethological room through door A and exited through door B, and vice versa. The dog was given the liberty to freely explore the environment during this phase.

The test consisted of a single trial lasting for one minute. Dogs were randomly allocated in the vipers or control conditions. During this trial, the owner maintained the dog by using a leash initially. An experimenter then entered the ethological room and positioned the apparatus containing the odour sample to be tested in the center of the apparatus zone. The samples (including control cardstocks) were stored in a fridge at a temperature of -80°C and get out 20 minutes prior to the test, allowing for thawing. Before leaving the room, the experimenter captured the dog's attention by touching the apparatus and making eye contact with the dog. Once the experimenter exited, the dog was released, and the test began when the dog made the first sniff on the apparatus. An additional experimenter observed and documented the procedure from outside the room using a closed-circuit television system equipped with four cameras. Handlers were given specific instructions not to interact with the dogs during the test and were provided with a magazine to avoid making eye contact with the dog. If the dog did not approach the apparatus independently within 10 seconds of being released, the handler had the option to invite the dog to interact with the device.

Table 1. Ethogram

CATEGORIES	BEHAVIORS	DEFINITION
OWNER	Gaze at the owner	From a stationary position, dog turns/lifts its head toward the handler, without approach
	Interaction with the owner	The dog establishes physical contact with the owner, e.g., rubbing, licking, or jumping up
	Approach with the owner	The dog goes to the owner from anywhere in the room
APPARATUS	Gaze at the apparatus	The dog from a stationary position gazing at the apparatus
	Interaction with the apparatus	Any behavior involving the dog being in contact with the apparatus, e.g., nosing, licking, etc.
	Approach with the apparatus	The dog goes to the apparatus from anywhere in the room
DOOR	Gaze at the door	The dog from a stationary position gazing at the door
	Interaction with the door	Any behavior involving the dog being in contact with the door, e.g., nosing, licking, etc.
	Approach with the door	The dog goes to the door from anywhere in the room
EXPLORATION	Visual exploration	The dog from a stationary position gazing at the other target not included in the above categories
	Olfactory exploration	The dog sniffs the target in the room not included in the above categories
PASSIVITY		The dog is lying on the floor and disinterested
STRESS	Stress signals	Includes all behaviors indicating stress (i.e., yawning, scratching, shaking, licking lips, barking, drinking, locomotion without a clear target)
OTHER	Mixed	Includes all behaviors not included in the above categories (i.e., moving toward other targets)

Data analysis

The data distribution was tested using the Shapiro–Wilk test. Based on the data distribution, Mann-Whitney test was used to examine the differences between dogs belonging to the group exposed to the smell of the "viper" and the group exposed to the "control" odor in different categories (whole sample; males vs females; males and females) for several parameters: Approach_Owner; Gaze_Owner; Interaction_Owner; Approach_apparatus; Gaze_apparatus; Interaction_apparatus; Approach_door_A; Gaze_door_A; Interactio_door_A; Approach_door_B; Gaze_door_B; Interaction_door_B; Visual_exploration; Olfactory_exploration; Passivity; Locomotion; Yawning; Shaking; Licking_nose; Barking; Yapping; Panting; Scratching; Drinking.

The statistical analysis was performed using SPSS software, and results were considered statistically significant for $P < 0.05$.

Results

The analysis of the behavioral score revealed that male dogs belonging to the group exposed to the smell of the viper interacted longer with the apparatus with respect to the group exposed to the control odor ($U = 107.500, z = 2.091, P = 0.035, \text{Fig. 3}$) and remained for more time in the apparatus zone ($U = 108.00, z = 2.116, P = 0.035, \text{Fig. 4}$).

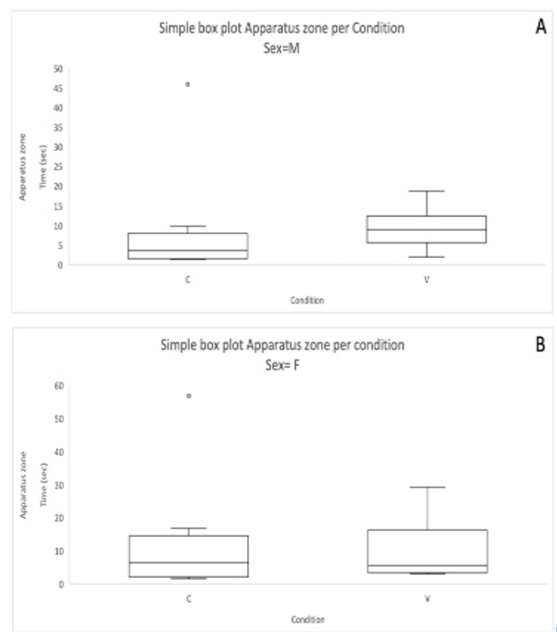
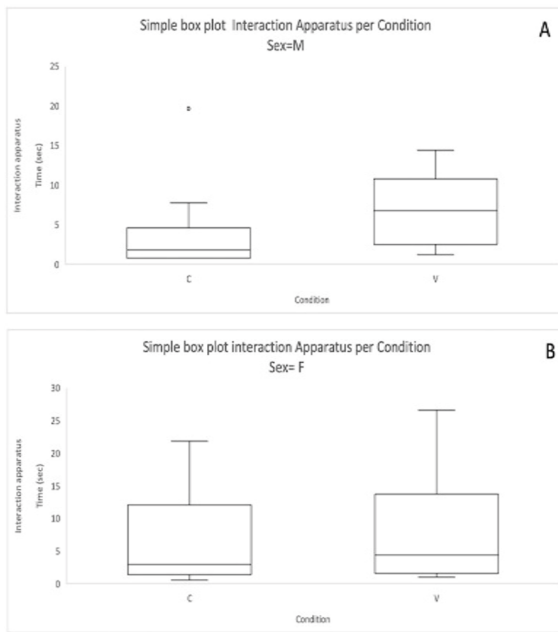


Figure 3. Graphical representation of the physical interaction with the apparatus in males (A) and females (B).

Figure 4. Graphical representation of the time spent in the apparatus zone in males (A) and females (B).

No others statistically significant results were revealed (see Table 2).

Table 2. P-values for Mann-Whitney analyses considering viper odor sniffing conditions vs control odor in different categories: whole sample; males and females.

	Frequency			Duration			Latency		
	Sample populatioon	Male	Female	Sample populatioon	Male	Female	Sample populatioon	Male	Female
Approach_Owner	0.789	0.608	0.442	0.893	0.820	0.959	0.421	0.361	0.645
Gaze_Owner	0.893	0.910	0.798	0.893	0.955	0.798	0.810	0.865	0.798
Interaction_Owner	0.768	0.361	0.574	0.573	0.331	0.878	0.830	0.608	0.721
Approach_apparatus	0.768	0.459	0.645	0.768	0.459	0.645	0.789	0.569	0.878
Gaze_apparatus	0.611	0.776	0.721	0.768	0.955	0.645	0.649	0.608	0.798
Interaction_apparatus	0.630	0.277	0.574	0.057	0.035	0.645	1	1	1
Approach_door_A	0.830	1	0.798	0.915	0.910	1	0.789	0.955	0.574
Gaze_door_A	0.421	0.955	0.234	0.390	0.820	0.382	0.649	0.820	0.442
Interactio_door_A	0.748	0.691	1	0.748	0.649	1	0.936	0.910	0.798
Approach_door_B	0.649	0.865	0.505	0.611	0.608	1	0.830	0.910	0.798
Gaze_door_B	0.708	0.691	0.234	0.649	0.776	0.279	0.630	0.691	0.161
Interaction_door_B	0.169	0.392	0.328	0.178	0.392	0.328	0.169	0.459	0.279
Visual_Exploration	0.768	0.776	1	0.611	0.608	0.878	0.789	1	0.878
Olfactory_exploration	0.405	0.733	0.328	0.728	1	0.382	0.573	0.776	0.645
Other	0.789	0.865	0.878	0.789	0.569	1	0.851	0.277	0.279
Passivity	0.469	0.865	0.382	0.555	0.910	0.442	0.469	0.865	0.328
Locomotion	0.810	0.776	1	0.810	0.776	1	0.810	0.776	1
Yawn	0.630	1	0.382	0.611	1	0.328	0.611	1	0.328
Shaking	0.810	0.776	1	0.810	0.776	1	0.810	0.776	1
Licking_nose	0.215	0.277	0.505	0.196	0.252	0.574	0.205	0.119	0.878
Barking	0.789	0.733	1	0.789	0.733	1	0.789	0.733	1
Yapping	1	1	1	1	1	1	1	1	1
Panting	0.789	1	0.721	0.789	1	0.721	0.789	1	0.721
Scratching	0.688	0.649	0.959	0.708	0.608	1	0.611	0.531	1
Drink	0.124	0.186	0.574	0.215	0.531	0.442	0.469	0.119	0.574
Door_A_zone	0.979	0.733	0.721	0.851	0.955	0.878	0.810	0.531	0.798
Door_B_zone	0.668	0.910	0.645	0.915	0.955	0.959	0.592	0.820	0.574
Owner_zone	0.979	0.910	0.798	0.205	0.167	0.645	0.469	0.252	0.878
Apparatus_zone	0.258	0.361	0.574	0.078	0.035	1	1	1	1
Other_zones	0.708	0.955	0.505	0.307	0.531	0.574	0.153	0.093	1

Discussion

The findings of the current study indicate that dogs do not show discrimination between the smell of vipers and the control odor. Additionally, indicators of emotional responses, such as increased interest toward the owner, did not differ across conditions, suggesting that the activation of the attachment system was not influenced by these smells. These results align with previous reports on rattlesnakes (Weldon and Fagre, 1989; Mulholland et al., 2018).

It is worth noting that prey species do not necessarily possess an innate ability to recognize the danger associated with potential predator odors, and even in the presence of sympatric predators, negative results have been obtained (Apfelbach et al., 2005).

The failure to recognize viper odors in dogs could be also attributed to the phenomenon of chemical crypsis, where organisms employ chemical mechanisms to blend in with their environment and avoid detection by potential predators or prey. Viperid snakes, as ambush predators, spend extended periods in immobility, waiting for prey to enter their striking range (Greene, 1997). Despite their dangerous nature, viperids are also subject to predation by several species (Branch, 1998), indicating that chemical crypsis may serve as a protective response to avoid detection (Miller et al., 2015). Nevertheless, it is important to highlight that the observation of males perceiving something in the snake odour doesn't entirely align with or fully support this notion.

On the other hand, visual stimuli, such as snake target pictures, appear to elicit an alarming response in various animal species (LoBue & DeLoache, 2008). In dogs, the preferential use of the left visual hemifield has been demonstrated in the analysis of a black silhouette of a snake on a white background (Siniscalchi et al., 2010). This preference can be attributed to the fact that neural pathways from the lateral visual field of each eye primarily project to the contralateral side of the brain (Rogers & Andrew, 2002). The utilization of the left visual hemifield to inspect the snake silhouette aligns with the specialization of the right hemisphere for expressing intense emotions, including escape behavior and fear, as previously shown in canine species (Siniscalchi et al., 2014, 2022), as well as in other animal models (as summarized in ref. (Siniscalchi et al., 2021)). Therefore, while studies focusing on the visual system support the idea that dogs can recognize snakes as threatening stimuli, data from the olfactory system do not appear to be as effective as visual stimuli.

The only significant difference observed in our study was a sex difference, with male dogs exposed to the smell of vipers interacting with the apparatus for a longer duration compared to the group exposed to the control odour. Previous reports have consistently shown that male dogs tend to exhibit bolder behavior (Svartberg, 2002; Strandberg et al., 2005; Kubinyi et al., 2009; Eken Asp et al., 2015; Scandurra et al., 2018). This observation could support the idea that the smell of vipers elicits emotional responses in dogs, allowing males to interact more due to their bolder personality traits. However, even with this difference, it was not sufficient to keep the dogs' noses away from the smell, indicating that a protective response was not activated in both male and female dogs. This conclusion supports the view that dogs are unable to avoid exposure to snakebites when they frequent areas populated by venomous snakes.

Conclusions

Overall our results indicated that dogs do not exhibit discrimination between the scent of vipers and the control odor. Furthermore, male dogs exposed to the smell of vipers interacted with the apparatus for a longer duration compared to the control odor group and remained in the apparatus zone for an extended period. Although preliminarily, these findings support the notion that dogs may be unable to avoid snakebites when they frequent areas populated by venomous snakes.

Conflict of interest statement

The authors declare there to be no conflicts of interest.

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Ethical Statement

The authors declare that all experimental procedures complied with ethical standards under the EU Directive 2010/63/EU for animal experiments.

Authorship Statement

All authors equally conceived of the study, participated in its design and coordination, and helped to draft the manuscript. All authors read and approved the final manuscript.

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I cani non mostrano comportamenti di evitamento in risposta all'odore dei serpenti

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Sintesi

I cani possiedono la capacità di riconoscere i potenziali pericoli associati ai serpenti principalmente attraverso il loro sistema visivo, piuttosto che fare affidamento sul loro sistema olfattivo. Gli studi precedenti si erano concentrati prevalentemente sugli odori dei serpenti pericolosi nel continente americano, che non si sovrappongono alle aree del processo di addomesticamento. Di conseguenza, è improbabile che i cani americani di oggi abbiano sviluppato risposte innate ai serpenti locali a causa della limitata esposizione storica. L'obiettivo del presente studio è stato condurre esperimenti coinvolgendo i cani e l'odore dei serpenti velenosi distribuiti nelle regioni in cui coesistevano i lupi antenati dei cani e dove si è verificato l'addomesticamento. A questo scopo abbiamo scelto appositamente tre specie del genere *Vipera*. Lo studio ha coinvolto 40 cani domestici con un'età media di $34,6 \pm 26,7$ mesi. Ogni cane ha partecipato a una singola prova di un minuto ed è stato assegnato in modo casuale alle vipere o alle condizioni di controllo. Le prove si sono svolte in una stanza contenente i proprietari, l'apparecchio e due ciotole per l'acqua. Per analizzare i dati sono stati utilizzati test non parametrici di Mann-Whitney. I risultati hanno indicato che i cani maschi esposti all'odore delle vipere hanno interagito con l'apparato per una durata maggiore rispetto al gruppo di controllo dell'odore. Inoltre, sono rimasti nella zona degli apparecchi per un lungo periodo. Tuttavia, non sono stati osservati risultati statisticamente significativi per le femmine e per altre variabili in entrambi i sessi, inclusi comportamenti diretti verso il proprietario e la porta, esplorazione, passività e segnali di stress. I risultati di questo studio suggeriscono che i cani non mostrano discriminazione tra l'odore delle vipere e l'odore di controllo. Di conseguenza, ciò supporta l'idea che i cani potrebbero non essere in grado di evitare i morsi di serpente quando frequentano aree popolate da serpenti velenosi.

