# International Journal of **Biological** and Natural Sciences

# CHEMICAL COMMUNICATION IN REPTILES OF THE ORDERS TESTUDINES, CROCODRYLIA AND RYNCOCHEPHALIA

# Julio César Castañeda Ortega

Biology course, Universidad Veracruzana Xalapa, Veracruz Mexico Orcid: 0000-0003-2663-9155

## Moisés Aguilar Valencia

Degree in Biology, Universidad Veracruzana Xalapa, Veracruz Mexico

## Viviana Domínguez Ochoa

Degree in Biologu, Universidad Veracruzana Xalapa, Veracruz Mexico

## Benito Hernández Castellanos

Biology course, Universidad Veracruzana Xalapa, Veracruz Mexico Orcid: 0000-0001-6475-5232



All content in this magazine is licensed under a Creative Commons Attribution License. Attribution-Non-Commercial-Non-Derivatives 4.0 International (CC BY-NC-ND 4.0). **Abstract:** Reptiles are organisms that rely heavily on chemical senses. As a result, they have developed various types of chemical signals, such as pheromones and kairomones. These signals play a crucial role in ecological interactions within ecosystems. Turtles, tuatara, and crocodiles are examples of reptiles that have evolved specific adaptations in their chemosensory organs.

These adaptations allow them to carry out various behaviors, such as courtship, mate selection, and prey discrimination. In the case of turtles and crocodiles, they can perceive chemical traces in different media, such as water and air, with the olfactory epithelium being their main sensory organ; However, the tuataras, despite having a vomeronasal organ, their adaptations are different from the specialists of this organ (snakes and lizards) since rhynchocephalians do not oscillate their tongue to detect chemical traces, they do so through a direct channel, these characteristics being determining factors. for the conduct displayed by the members of the aforementioned orders themselves.

**Keywords:** Reptiles, pheromones, kairomones, turtles, crocodiles, tuataras.

# INTRODUCTION

Reptiles interact with the environment and other organisms through sensory systems; these can detect physical and chemical stimuli from the environment. Among the physical sensesaretouch,sight,hearing,proprioception, among others. Likewise, there are senses responsible for detecting chemical stimuli. The chemical signals released by vertebrates for orientation are called semiochemicals, which serve as intermediaries in interactions between organisms and have the capacity to affect their physiology and behavior. In the chemical communication of reptiles, two types of semiochemicals are involved: pheromones and kairomones (Le Master et al., 2001). When chemical signals are emitted and detected by organisms of the same species, they are called pheromones, which are very important for intraspecific interactions of organisms, such as reproduction (Stockhorst & Pietrowsky, 2004). When the chemical signals emitted by one species are detected and used by an organism of a species other than the one that emitted the signal and it is used for its benefit, they are called kairomones.

The main benefits obtained by detecting kairomones are finding potential prey, as well as detecting and avoiding contact with a potential predator (Muller-Schwarse, 2006).

Chemical signals are compounds used by organisms as a source of information to interpret environmental stimuli, although they did not evolve for this function in the sender (Lima, 2022). Chemoreception is defined as the ability of living beings to detect and differentiate certain substances. volatile and non-volatile chemicals existing in the surrounding environment (Castañeda-Ortega, 2008). Chemoperception is one of the oldest mechanisms used by organisms to communicate and learn about their environment (Amo & Bonadonna, 2018). Chemical trails allow reptiles to select an appropriate habitat, since when odorants disperse in the environment it provides them with information about the presence of conspecifics or possible predators (Martín et al, 2021). Reptiles use chemical signals to evaluate characteristics such as sex, mating history, body and population condition (Richard et al, 2020).

The use of chemical traces has important implications in different ecological processes such as predation, where predators usually tend to use chemical signals to recognize and locate their prey, since these allow the indirect transfer of information between transmitter and receiver ( Raya-García, 2016). Secretions from the skin, those from the cloaca, femoral glands and excreta play a very important role in their chemical communication (López & Martín, 2004). Predators use a variety of signals to recognize and locate appropriate prey. These signals that allow prey recognition have an important impact, since the recognition of these traces can mediate interspecific interactions, community dynamics and other fundamental ecological processes such as chemosensory behavior in predatory reptiles (Clark & Ramírez, 2011). The detection of chemical traces also allows reptiles to select a potential partner, which is essential, since their qualities will determine the quality and probability of survival of their offspring (Richard et al, 2020).

In reptiles there are three chemoperceptive systems: taste, smell or the main olfactory pathway, and the vomeronasal or accessory olfactory pathway, the latter two related to the perception of odors. Olfactory or chemical communication has numerous advantages for vertebrates over other types of communication. It can be used when visual and auditory signals are difficult to detect, such as in the dark, underground or in dense vegetation (Castañeda-Ortega, 2008). The ability to detect odorous stimuli in reptiles has been studied mainly in organisms belonging to the order Squamata, however it is not exclusive to this order, since it is also present in Ryncochephalia, Testudines and Crocodrylia.

# **ORDER RYNCOCHEPHALIA**

Rhynchocephalia is a lineage of lepidosauromorph reptiles, this order is the sister taxon of Squamata and is represented by the genus Sphenodon. The genus has only one species Sphenodon punctatus, with two subspecies. which survives on 32 islands off the coast of New Zealand (Bever & Norell, 2017). It has a weakly developed vomeronasal organ (Rieppel et al., 2008), with a low density of vomeronasal chemoreceptor cells (Gabe & Saint Girons, 1976).

Unlike members of the order Squamata, the tuatara does not oscillate its tongue (Cooper et al., 2001), and the vomeronasal canal does not open to the oral cavity as in Squamata but to the nasal cavity, suggesting that the products Chemicals sampled lingually cannot reach the vomeronasal organ (Bellairs, 1984). A study carried out with Sphenodon punctatus (figure 1) reported that it had similar behavioral responses to Hemidactylus frenatus when faced with the same series of stimuli. Spending more time in the area with the smell of the prey (worm) than with the pungent stimulus and control. Both launched bites only toward the prey stimulus and showed similar attack latency. However, only geckos showed tongue extrusion when faced with the smell of prey (Besson et al, 2009).



Figure 1. *Sphenodon punctatus* It has a functional vomeronasal organ that allows it to detect the scent of its prey.

# **ORDER TESTUDINES**

A relatively little studied group is that of turtles, however, it is known that they have both systems and both the main olfactory and the vomeronasal systems and are well developed (Endres, 2013). Turtles are capable of detecting odorants in both air and water; the chemical traces perceived can be used as a source of information for navigation during migration, as well as determining the proximity of land masses (Southwood et al., 2007). *Podocnemis unifilis* (figure 2) uses the chemical traces of conspecifics present in the water to orient itself, with juvenile organisms increasing their activity in response to the chemical traces of adult conspecific females. The perceived traces allow them to form aggregations and orient themselves during migration. (Ibáñez & Vogt, 2015).

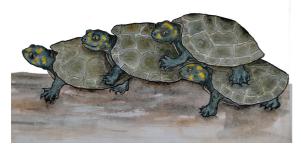


Figure 2. In freshwater turtles such as *Podocnemis unifilis, chemoperception* is important in both sexual and agonistic social interactions.

This ability may be adaptive depending on the life stage of the turtles. For example, because predators are more abundant in coastal areas, juvenile turtles that are carried to the coast by ocean currents could benefit by quickly moving away from the coast when detecting the scent of these potential predators; On the contrary, adult organisms that migrate to nesting areas in different areas or continents could benefit from the ability to perceive odorants in the air of terrestrial vegetation, the soil or the excrement of seabirds to determine the proximity to the nest. that are on the mainland (Endres, 2013).

In several species of turtles, feces are used alone or in combination with glandular secretions, as a source of chemical components. Most turtles have several specialized glands in their skin, such as Rathke's glands, or chin glands that secrete chemical compounds into the environment (Muñoz, 2004). It has been reported that the odor secreted by the Rathke glands in adult specimens of *Chelonia mydas*  produced a behavioral alteration in juvenile conspecifics.

Upon contact with this odor, juveniles significantly reduce their activity until they become immobile (Kitayama et al., 2020). For their part, turtles of the genus *Sternotherus* use the secretions of the axillary gland as an anti-predatory mechanism; however, in intraspecific communication and during the reproductive season, the glandular contents serve as sex recognition signals through which the males that courting identify the females (Muñoz, 2004).

For their part, Kelley & Mendonça (2021) report that secretions from the chin gland together with the presence of a visual stimulus produce changes in behavior in both females and males of *Gopherus polyphemus*; Turtles of both sexes spent more time interacting and performed a greater number of behaviors in front of a turtle model treated with the odorous stimulus in relation to the interaction time and behaviors displayed in front of a model inoculated with distilled water. Which suggests that, along with the presence of a visual stimulus, the presence of chemical stimuli is necessary to participate in social behaviors.

Although chemoperception plays a predominant role in recognizing the environment, the interaction with other sensory systems can help improve some behaviors such as migration. For example, research carried out in Chelonia mydas suggests that odorants in water and air alone they are insufficient to guide turtles in the sea to locate nesting areas, so the detection of magnetic signals is also necessary. It is suggested that the latter can take the turtles in the vicinity of an island, but normally would not allow them to be located; for this last step, they will be required to be guided by chemical signals perceived both in the water and in the air (Endres et al. al., 2016). With

respect to habitat selection, work carried out with Chrysemys picta reported that this reptile is capable of discriminating between chemical signals from conspecifics that lived in the same pond and the smell of conspecifics from other ponds, with females preferring to stay longer in the pond. water with the smell of their own pond than in the water that had the smell of other ponds, this was not the case with males who showed greater exploratory behavior (Quinn & Graves, 1998).

Chemical trails also make it easier to select a potential mate; by analyzing the response of Chrysemys picta males towards chemical traces exuded by females and males of conspecifics. In a pond divided into three sections (one inoculated with male odor, another with female odor traces, and a third without odor stimuli as a control), a male specimen of C. picta was released. Analyzing the time that the males stayed in each of the three areas of the pond, it was found that the males stayed the longest in the ponds where the odorous stimuli of the females were found and they spent the least amount of time in the control area. In the case of male odor, these did not produce a change in the exploratory behavior of the turtles (Becker & Thomas, 2020).

Similar results were found with Mauremys leprosa, which in a sexual context was able to detect and discriminate chemical signals released by its conspecifics and modify its use of space in response to them. The males preferred to stay in ponds that had been inoculated with the odor of females with a greater immune response, while the females preferred water with the odors of larger males. However, males avoided pools of water inoculated with the odor of larger males and remained in water inoculated with the odor of smaller males, presumably to avoid agonistic interactions (Ibáñez et al., 2012). Another work with Mauremys leprosa reports that outside the mating season, both males and

females avoid interacting with the chemical traces of their conspecifics of the opposite sex; however, during the mating season, males interact significantly more in inoculated ponds. with chemical signals from females and avoid water with chemical signals from other males. Unlike females, which during the mating season prefer to stay in ponds inoculated with chemical signals from other females, avoiding spending a long time in ponds inoculated with the smell of males (Muñoz, 2004).

# ORDER CROCODYLIA

Crocodiles hunt in both terrestrial and aquatic environments, so they detect chemical substances in both air and water. The vomeronasal organ is absent, however, they have a well-developed main olfactory organ (Hansen, 2007). It is believed that some organisms with scavenging habits such as Crocodylus palustris and C. niloticus depend on olfactory trails to detect carrion, which can reach a distance of between 450 and 700 meters respectively around the swamp. Dietary imprinting has also been observed in Alligator mississippiensis fed in captivity with otter meat, which showed greater foraging behavior (Weldon and Ferguson, 1993).

A study carried out by Weldon et al (1990) reports that juveniles and adults of A. mississippiensis (figure 3) use chemoperception to locate food such as carrion and wounded prey both underwater and on land. In the experiment, both juveniles and adults were able to identify packages containing remains of prey (Procyon lotor) both on land and in water. Likewise, a group of juveniles placed in tanks partially filled with water exhibited behaviors such as lateral head movements and mouth openings only to the aqueous meat extract compared to the control. While the juvenile group tested in an olfactometer showed more gular pumping

when faced with the smell of beef than against the distilled water control. Gular pumping has been associated with the uptake of odorants during foraging (Hansen, 2007).

Scott & Weldon (1990) carried out experiments with adult specimens of Alligator mississippiensis; where they were presented with four types of odorous stimuli (beef, otter, Crotalus atrox meat or an aqueous extract of beef) placed in paper bags with 24 holes through which the odor emerged. As a control, bags that only contained paper were used. When presenting each of the four stimuli vs. the control, more attacks were reported on the bags with beef and inoculated with the aqueous extract, which suggests that they detected chemical substances transported by water or air from the materials that were presented to them.

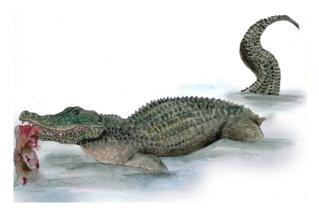


Figure 3. Although the vomenasal organ is absent in members of the order Crocodylia, crocodiles have the ability to detect scent trails of potential prey.

#### CONCLUSIONS

Chemical signals are the product of the organisms' need to communicate in longlasting and considerably effective ways. When living in aquatic environments, crocodiles and turtles make the most of the olfactory epithelium, thanks to which they can communicate with the environment through the medium. whether aquatic or through the air. Similarly, the tuatara underdeveloped the vomeronasal organ, of which they have fewer nerve endings than their sister group, the latter being specialists in this organ; However, studies suggest that despite the difference between the capacity of the secondary olfactory organ, the behaviors displayed are similar, which indicates an evolutionary route of the species.

However, the scarcity of Sphenodontia individuals leads to a clear lack of information on the chemical communication of other non-scaly reptiles, leaving us with proven similarities but not well-studied mechanisms.

Turtles, on the other hand, are organisms that diversify in numerous environments, which is why they use chemical signals that can assertively elucidate their environment. Living in water or in the desert, these means make chemical stimuli have a direct relationship in the behavior of individuals, crocodiles on the other hand use chemical traces mainly in the water or in the air to discriminate between different stimuli to search for food.

The lack of chemical communication studies in these organisms presents solid lines of research, which could determine the morphophysiological relationships and the response to chemical stimuli, providing a better overview of the ecology and inter and intraspecific relationships of the aforementioned orders, since They are persistent organisms today with adaptations that have made them endure in a world where there are specialists in traces and chemical communication.

## REFERENCES

Amo, L. & Bonadonna, F. Editorial: The Importance of Olfaction in Intra- and Interspecific Communication. **Front. Ecol. Evol.** 6:71. 2018.

Becker, J. T., & Thomas, R. B. Male response to female chemical signals in Painted Turtles (*Chrysemys picta*). Transactions of the Kansas Academy of Science. 123(3-4), 453-459. 2020.

Bellairs, A. A. Closing address with comments on the organ of Jacobson and the evolution of Squamata, and on the intermandibular connection in Squamata. **Symposium of the Zoological Society of London.** 52:665–683. 1984.

Besson A. A., Thierry A, Boros E., Allen K., Bradley S., Norrie C., & Cree A. Evidence of Food Chemical Discrimination in Tuatara (O. Rhynchocephalia): Comparison with a Gekkotan Lizard (O. Squamata) **Journal of Herpetology**. 43(1):124-131. 2009.

Bever, G. S., & Norell, M. A. A new rhynchocephalian (Reptilia: Lepidosauria) from the Late Jurassic of Solnhofen (Germany) and the origin of the marine Pleurosauridae. **Royal Society Open Science**. 4(11), 170570. 2017.

Castañeda-Ortega, J. C. **Respuestas antidepredatorias de las serpientes** *Thamnophis proximus* **y** *Nerodia rhombifera* **ante olores de mamíferos.** [Tesis de maestría] Instituto de Neuroetología Xalapa, Veracruz, Mexico. 2008.

Clark, R. W., & Ramirez, G. Rosy boas (*Lichanura trivirgata*) use chemical cues to identify female mice (*Mus musculus*) with litters of dependent young. **The Herpetological Journal.** 21(3), 187-191. 2011.

Cooper Jr, W. E., Ferguson G. W., & Habegger J.J. Responses to animal and plant chemicals by several iguanian insectivores and the Tuatara, *Sphenodon punctatus*. Journal of Herpetology. 35:255–263. 2001.

Endres, C. S. Perception of Airborne Chemosensory Cues by Sea Turtles. [Tesis de doctorado], The University of North Carolina at Chapel Hill. 2013.

Endres, C. S., Putman, N. F., Ernst, D. A., Kurth, J. A., Lohmann, C. M., & Lohmann, K. J. Multi-modal homing in sea turtles: modeling dual use of geomagnetic and chemical cues in island-finding. **Frontiers in Behavioral Neuroscience.** 10, 19. 2016.

Gabe, M., & H. Saint Girons. Contribution á la morphologie comparée des fosses nasales et de leurs annexes chez les lépidosoriens. **Mémoires du Muséum National d'Histoire Naturelle.** Nouvelle Serie A 98:1–87. 1976.

Hansen, A. Olfactory and solitary chemosensory cells: two different chemosensory systems in the nasal cavity of the American alligator, *Alligator mississippiensis*. **BMC Neuroscience**. 8, 1-10. 2007.

Ibáñez, A., López, P., & Martín, J. Discrimination of conspecifics' chemicals may allow Spanish terrapins to find better partners and avoid competitors. **Animal Behavior.** 83(4), 1107-1113. 2012.

Ibáñez, A., & Vogt, R. C. Chemosensory discrimination of conspecifics in the juvenile yellow-spotted river turtle *Podocnemis unifilis*. **Behaviour.** 152(2), 219-230. 2015.

Kelley, M. D., & Mendonça, M. T. Mental gland secretions as a social cue in gopher tortoises (*Gopherus polyphemus*): tortoise presence stimulates and maintains social behaviour with chemical cues. Acta Ethologica. 24(1), 1-8. 2021.

Kitayama, C., Yamaguchi, Y., Kondo, S., Ogawa, R., Kawai, Y. K., Kayano, M., Tomiyasu, J. & Kondoh, D. Behavioral effects of scents from male mature Rathke glands on juvenile green sea turtles (*Chelonia mydas*). Journal of Veterinary Medical Science. 82(9), 1312-1315. 2020.

LeMaster, M. P., Moore, I. T. & Mason, R. T. (2001) Conspecific trailing behavior of red-sided garter snakes, *Thamnophis sirtalis parietalis*, in the natural environment. **Animal Behaviour.** 61,827-833.

Lima, L. D. Chemical Cues. En: Vonk, J., Shackelford, T.K. (eds) Encyclopedia of Animal Cognition and Behavior. Springer, Cham. 2022.

López, P. & Martín, J. **Sexual selection and chemoreception in lacertid lizards** En: The Biology of Lacertid lizards. Evolutionary and Ecological Perspectives. Pérez-Mellado, V., Riera, N. and Perera, A. (eds.). Institut Menorquí d'Estudis. Recerca, 8: 119-137. 2004.

Martín, J., Ibáñez, A., Garrido, M., Raya-García, E., & López, P. Chemical cues may allow a fossorial amphisbaenian reptile to avoid extremely saline soils when selecting microhabitats. **Journal of Arid Environments**. 188, 104452. 2021.

Müller-Schwarze, D. (2006) Chemical ecology of vertebrates. Editorial Cambridge University Press. E. U. A. 563 pp.

Muñoz, A. Chemo-orientation using conspecific chemical cues in the stripe-necked terrapin (*Mauremys leprosa*). Journal of Chemical Ecology. 30, 519-530. 2004.

Quinn, V. S., & Graves, B. M. Home pond discrimination using chemical cues in *Chrysemys picta*. Journal of Herpetology. 32(3), 457-461. 1998.

Raya-García, E. Respuesta quimiosensorial y aspectos reproductivos en dos especies simpátricas del género *Conopsis* (Serpentes: Colubridae) [Tesis de maestría] Universidad Michoacana de San Nicolas de Hidalgo. 2016.

Richard, S. A., Bukovich, I. M., Tillman, E. A., Jayamohan, S., Humphrey, J. S., Carrington, P. E., Bruce, W. E., Kluever, B. M., Avery, M. L. & Parker, M. R. Conspecific chemical cues facilitate mate trailing by invasive Argentine black and white tegus. **PLoS One.** 15(8), e0236660. 2020.

Rieppel, O., Gauthier, J.& Maisano J. Comparative morphology of the dermal palate in squamate reptiles, with comments on phylogenetic implications. **Zoological Journal of the Linnaean Society**. 152:131–152. 2008.

Scott, T. P., & Weldon, P. J. Chemoreception in the feeding behaviour of adult American alligators, *Alligator mississippiensis*. Animal Behaviour. 39(2), 398–400. 1990.

Southwood, A. L., Higgins, B. M., Swimmer, Y., & Brill, R. W. Chemoreception in loggerhead sea turtles: an assessment of the feasibility of using chemical deterrents to prevent sea turtle interactions with longline fishing gear. NOAA Tech.Memor. NMFS-PIFSC-10. 2007.

Stockhorst, U., & Pietrowsky, R. Olfactory perception, communication, and the nose-to-brain pathway. **Physiology & Behavior**. 83(1), 3–11. 2004.

Weldon, P. J., & Ferguson, M. W. Chemoreception in crocodilians: anatomy, natural history, and empirical results. Brain, **Behavior and Evolution**. 41(3-5), 239-245. 1993.

Weldon, P. J., Swenson, D. J., Olson, J. K., & Brinkmeier, W. G. The American alligator detects food chemicals in aquatic and terrestrial environments. **Ethology.** 85(3), 191-198. 1990.