

**PROVIDING ENRICHMENT IN
CAPTIVE AMPHIBIANS AND REPTILES:
IS IT IMPORTANT TO KNOW THEIR COMMUNICATION?**



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1. Introduction

1.1. Captive keeping of wild animals

The relationship between man and animals has its beginnings long ago in prehistoric times and has changed immensely since then. Kisling (2001) stated: *As early humans developed into a socially and intellectually unique species, they intensified their dominion over other species while distancing themselves from other species through the development of socially elaborate cultures.* (p. 5)

Domesticating animals were the first attempts at keeping wild animals (Kisling, 2001) and a step towards *Homo sapiens* dominating other species by means of intellect. Humans started domesticating animals from the 10th to the 7th millennium B.C. for economic reasons (Petzold, 2008). Keeping animals as living meat resources changed the lifestyle of humans enabling them to become less dependent on hunting and gathering. As complex production-based human societies evolved into urbanized civilizations, royalty and some wealthy individuals prospered, often starting collections of wild animals (Kisling, 2001). From the 3rd to the 2nd millennium B.C., humans in the ancient Orient kept wild animals for religious reasons, and these were the earliest roots of “zoo biology” in a very broad sense: the keeping of wild animals that are not domesticated (Petzold, 2008). The first zoos date back to the 2nd millennia B.C. and they no longer served religious functions, rather demonstrated the power of a ruler to his subjects and competitors. Yet, zoological gardens have existed only in highly developed human cultures and were founded solely when a culture was flourishing (Grzimek, 1958). Between the late 1700s and early 1800s, a growing number of wealthy people caused a shift away from royal and private menageries (Kisling, 2001). The first zoo as we know it today was founded in Paris in 1793. At that time, zoos were mostly recreational, and the animals were kept for the amusement of the public. In 1907, Carl Hagenbeck (1844-1913) developed the first exhibits without bars for zoos at his Tierpark at Stellingen, Germany (Polakowski, 2001). The new exhibit design allowed displaying animals in a more naturalistic, less threatening environment, than behind iron bars (Murphy and Iliff, 2004). The idea of that more natural exhibit design was developed much earlier by a French naturalist named Bernard-Germain-Étienne de la Ville-sur-Ilion, Comte de Lacepède (1756–1825). For Lacepède, it was obvious that more naturalistic exhibits would serve the animals as well as the public, as visitors would have the opportunity to “have a pleasant experience and learn about animals in a non-threatening way” (Murphy and Iliff, 2004). Nowadays the enclosures are designed to be more like natural habitats for their inhabitants instead of just cages (Hediger, 1968).

In the 20th century, the function of zoos slowly changed and the tasks of public zoos expanded by focusing more on the conservation of species, research and the education of the public (Sandford, 1984). Hence, modern zoos are much more than luxury establishments and have an important function for men and animals alike (Hediger, 1970). Zoos have become sanctuaries for endangered species in need of conservation measures (Van Bemmelen, 1986). The need for the conservation of species has become increasingly urgent because of the massive declines in biodiversity in the last decades.

1.2. Biodiversity declines

Biodiversity is “the number and variability of genes, species, and communities in space and time” (Sepkoski, 1997). The stability of biodiversity plays an important role in our ecosystem. The most utilitarian reason for valuing biodiversity highly is its direct and indirect economic value, but biodiversity should also be preserved simply because of its beauty (Osborne, 2001). As humans take up more space and resources, the biodiversity of our planet shrinks at an alarming rate (Banks and Birkett, 1991). The spread and development of the human population comes at a cost, as more animals disappear from the earth (Pimm et al. 1995). Many conservation biologists speak of the “biodiversity crisis” when referring to the rapid and accelerating loss of species and habitats (Collins and Crump, 2009). Since the turn of the 20th century the rate of extinction has accelerated from about one species per year through the 1960’s to as much as possibly one per day in the 1980’s (Sandford, 1984). In the 1990’s, the extinction rate was estimated from 5.5 species per day to as high as 150 species per day

(Sepkoski, 1997). The International Union for Conservation of Nature (IUCN), the world's oldest and largest global environmental organization, estimates that nowadays the extinction rate is between 1,000 and 10,000 times higher than the natural extinction rate that would occur without human influence (IUCN, 2007; Pimm et al. 1995). The IUCN mentions several reasons for these accelerating declines in biodiversity: habitat destruction and degradation, over-exploitation (extraction, hunting, fishing, etc.), pollution, disease, invasion of alien species and global climate change (changes migratory patterns, coral bleaching), the threats that endanger many animals all over the world (IUCN, 2007; Millennium Ecosystem Assessment, 2005).

The need for conservation programs is apparent when looking at the biodiversity declines. There are several ways for zoos to approach those problems and to fight the causes and the effects of the high extinction rate. Approaches to the problems are captive breeding, including seed banks, and reintroduction into the wild (Dodd and Seigel, 1991), programs that many modern zoos pursue.

1.3. Reptiles and amphibians

Reptiles and amphibians are also affected by the massive declines in biodiversity. They suffer severely under changing living conditions and many species are threatened by extinction. In most modern zoos nowadays, reptile and amphibian houses are an inherent part of their animal collection, and efforts are made to conserve those animals for the eyes of future generations.

Reptiles and amphibians are a group of vertebrates that are distinctive for their ectothermal lifestyle, which is quite different from the endothermal lifestyle of mammals and birds. They rely on external heat, like solar radiation, for their body heat (Maruska, 2001). When keeping reptiles and amphibians, attention must be paid to this physiological aspect to ensure their good health. Because they do not need to maintain a constant body temperature, reptiles can survive long periods without food (McMahan, 2001). Keeping reptiles in captivity can be quite a challenge and it is vital to know the natural history of these animals to be able to keep them mentally and physically healthy. Until the 1960's, reptiles and amphibians were hardly ever kept in suitable social situations, and behavioral and environmental cues were oftentimes ignored (Murphy and Xanten, 2007).

The class Amphibia (amphibians) contains frogs and toads, salamanders and newts and a much smaller group of caecilians. The class of living reptiles, Reptilia (reptiles), is comprised of four major groups: Crocodylia (crocodiles and alligators), Squamata (snakes and lizards), Testudines (turtles and tortoises) and a small group of Sphenodontida (tuatara) (Chiszar and Smith, 2001). There are over 6,000 species of amphibians (Stuart et al. 2008) and almost 10,000 species of reptiles (Uetz, 2000; Uetz, 2012) currently recognized, but many are still being discovered. The latest update of the IUCN Red List of Threatened Species shows that 30% of all amphibians assessed and 28% of all reptiles assessed are *threatened* (Fig. 2). 1 % of both reptiles and amphibians assessed are already *extinct* (Fig. 1).

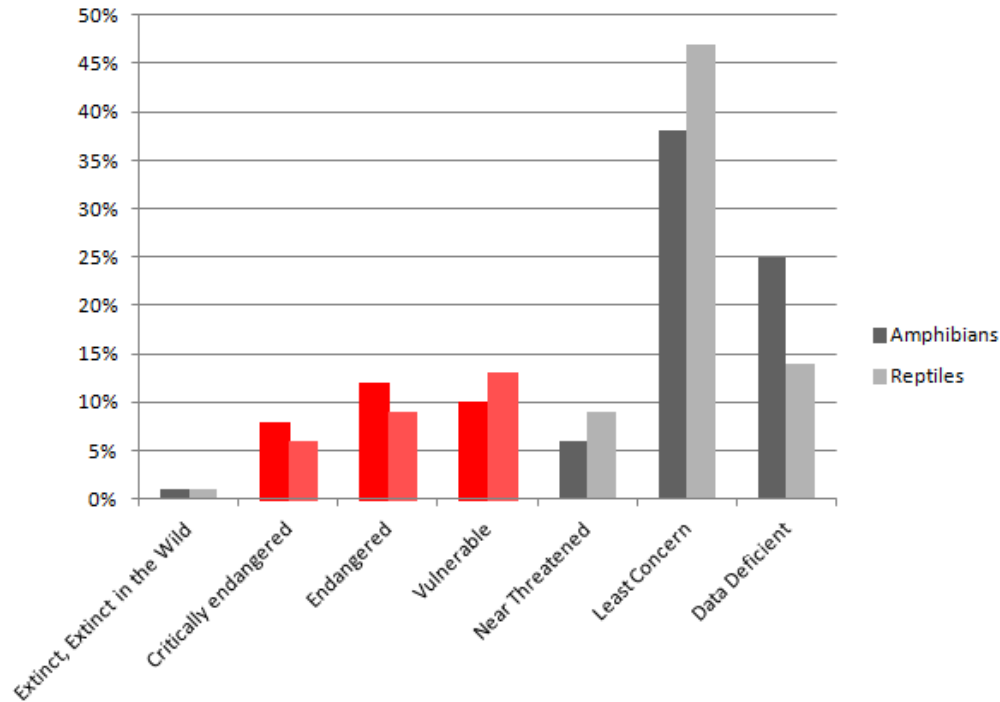


Fig. 1. Estimated percentages of reptiles and amphibians in different Red List Categories designated by the IUCN. Species that are critically endangered, endangered or vulnerable are categorized as threatened (marked in red). Plotted from the data of the IUCN (IUCN, 2009).

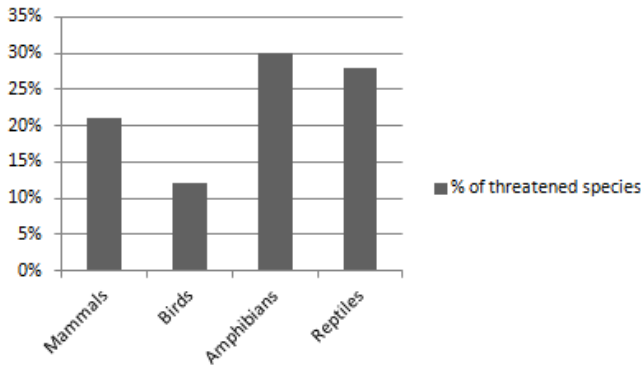


Fig. 2. In comparison to other large animal groups like mammals and birds, reptiles and amphibians are especially vulnerable. Plotted on the data of the IUCN (Extinction Crisis Continues Apace, 2009).

The reasons for the declines in reptiles and amphibians are numerous. The wild populations shrink due to habitat destruction, arrival of invasive species, chemical contaminants, infectious diseases, over-collecting, human consumption, and reptile skin trade (Murphy and McCloud, 2010). In the past, herpetological collections consisted of wild caught animals. Captive bred reptiles and amphibians were typically unavailable (Murphy and Xanten, 2007). Nowadays though, the quantity of wild-caught amphibians and reptiles is considerably smaller, due to the loss of biodiversity and the public's perception about animal protection.

Since reptiles and amphibians have very different lifestyles from humans and other mammals, their captive keeping requires different approaches from the keeping of, for example, mammals. It is essential to know about the animals' lifestyle to be able to keep and possibly breed them. Modern herpetoculture, the keeping of reptiles and amphibians, has been studied since the late 1800 as the

Austrian scientist and father of modern herpetoculture, Johann von Fischer (1850-1901), started publishing his works about maintenance and behavior of captive herpetofauna (Murphy, 2005). A fundamental part of an animal's lifestyle is the way they communicate with each other and their environment. In this thesis, two of the major communication channels of reptiles and amphibians – visual and chemosensory – will be discussed in respect to their importance in keeping captive animals. Each communication channel will then be further examined by looking at four species of reptiles and amphibians which use one of these channels. Further, an example of how to use the knowledge about communication channels will be given at the end of each chapter.

1.4. The site of the study

The observations and experiments described in this paper were conducted at the Reptile Discovery Center (RDC), Smithsonian National Zoological Park, Washington D.C., USA, one of the world's most renowned reptile houses. The RDC developed a public education approach that puts a strong emphasis on the conservation of reptiles and amphibians. In 1992, the former reptile house was transformed into a Reptile Discovery Center with interactive modules, teaching visitors about topics like anatomy, communication, feeding, reproduction, social behavior, and ecology/conservation (Doering et al. 2001). In addition to that, several volunteers serve as instructors, teaching the visitors about the animals displayed, and answering their questions (Murphy and Xanten, 2007). That interactive approach has proven to slow visitors down and to make them spend more time at the exhibits. It also proved to increase the visitor's knowledge about the animals displayed and generally improved their feelings towards reptiles and amphibians (Doering et al. 1994). Today, the collection of the Smithsonian National Zoo's collection includes 69 species of reptiles, totaling 218 specimens, and 16 species of amphibians, numbering 222 animals (as of May 1, 2012).

The RDC team emphasizes the importance of educating visitors on research, and conservation of endangered species. They also value highly the best possible maintenance of captive herpetofauna and have introduced several enrichment methods to keep their animals healthy and active.

2. Enrichment

2.1. Enrichment for captive animals

Until thirty years ago most zoo animal displays were designed with little reference to the animals' native habitats and the physical and behavioral needs derived from these native environments. Physical and behavioural problems of captive animals were not uncommon and only the effects, not the causes of those problems, were treated (Coe, 1997). It was also widely ignored that species other than humans had a high degree of awareness and could suffer if deprived (Bostock, 2001). In recent years though, zoo workers have changed their exhibits and increasingly adapted them to the needs of the animals. Nevertheless, captive keeping is often harshly criticized and rejected by some animal activists. Some animal lovers argue that the continuing existence of zoos can be ethically justified only if zoos guarantee the welfare of their animals (Wickins-Drazilova, 2006). Modern zoos are rightly concerned with the welfare of their animals, but it is quite difficult to assess the welfare of an animal when we can only observe the animal's behaviours (Bostock, 2001). The design of their exhibits, however, is only one factor contributing to the overall welfare of zoo animals. It is a challenge to assess the welfare of each individual animal and to define the meaning of welfare. The typical criteria for measuring animal welfare in zoos are physical health, lifespan, and reproduction. However, other criteria like natural and abnormal behaviour should also be used to assess the welfare of zoo animals (Wickins-Drazilova, 2006). Therefore, to measure animal well-being, both physiological and behavioural measures must be considered. It is also widely believed that the display of behaviour similar to wild conspecifics is the best indicator for well-being (Bostock, 2001).

Undoubtedly, animals that are kept in captivity have to face different environments and different circumstances from their wild conspecifics. Such change ultimately affects their behaviour and alters how they spend their time. The time wild animals spend searching for food is free time to zoo animals because they are fed on a regular basis. Also significant is predation risk, which influences activity patterns and habitat and is most commonly detected through changed foraging behaviour (Mandelik, Jones, and Dayan, 2003). To keep animals healthy, active, and promote natural behaviours, it is therefore vital to give them the opportunity to live in conditions similar to their wild habitat and to perform activities that are natural to them.

A tool that can be used for ensuring the welfare of captive animals is *environmental enrichment* (also known as *behavioural enrichment*). Environmental enrichment is a method used to improve or enhance environments and care for zoo animals in the context of their behavioural biology and natural history (Shepherdson, 2001). It is a tool that allows captive animals to display a wide range of natural behaviours (Hernandez-Divers, 2006) by designing the exhibits to each species' unique needs and providing natural stimuli for the animals. It covers a multitude of innovative techniques that are aimed at providing adequate social interaction or simply keeping the animal occupied (Shepherdson, 2001). One enrichment approach can be to change the type of food or alter its manner of presentation, aiming to increase time spent foraging and feeding to levels similar to those observed in the wild (Forthman and Ogden, 1992). Other stimuli can be spices that are introduced to the enclosure, or the fur of a prey animal, to increase foraging behaviour and curiosity (Hernandez-Divers, 2006; Shepherdson, 2001). Enrichment application requires considering each species' unique communication, social structure, and feeding strategies (Forthman and Ogden, 1992). The appropriate enrichment items must be carefully selected for each species and for specific individuals. Nevertheless, an enriched enclosure could be inherently more dangerous than a sterile enclosure if proper care is not taken. Incorrect enrichment application can cause injuries or even cause death in some cases (Hare, Rich, and Worley, 2008). Therefore, great attention must be paid to the use of enrichment items.

It is difficult to clearly define enrichment because a simple feeding could be considered a type of enrichment. Generally though, the aim of enrichment is to increase the overall welfare of animals in captivity and to promote species specific behaviours. Another goal of enrichment is to reduce boredom or stereotypic behaviours, such as pacing and self-mutilation (Novak and Drewsen, 1989; Sawisgood and Shepherdson, 2005), and consequently to reduce stress. Stereotypic behaviours often occur when a captive animal has extended exposure to an ecologically relevant problem that it cannot solve. Such ecologic problems include finding a mate, hunting for food, and escaping from human contact (Shepherdson, Brownback, and James, 1989). The frustration caused by the animal's inability to carry out these behaviours often leads to stereotypic patterns. Moreover, when animals are kept in captivity with food and other valuable resources readily available and are no longer dependent on foraging, "learned laziness" can occur (Engberg et al. 1972). The animals stop trying, realizing their behaviours are no longer instrumental in bringing about biologically significant outcomes (Chiszar, Murphy, and Smith, 1993).

The forms and functions of enrichment are manifold, which makes it impossible to mention them all. There are virtually no limits to the possibilities of what can be done as enrichment, though the creativity of animal keepers and the input of staff are needed to develop new methods on a regular basis.

2.2. Reptile and amphibian enrichment

Enrichment literature often focuses on primate and other mammal enrichment, overlooking the enrichment of reptiles and amphibians. To be able to provide enrichment, it is essential to know the species-specific natural histories in order to assess aspects of wellbeing (Warwick, 1995). The ectothermic nature of amphibians and reptiles makes it challenging to adapt mammal enrichment

activities to their needs. Having to control their body temperature by means of an external source, ectothermal animals are not constantly active like endotherms. For a long time it was therefore believed that reptiles and amphibians are unintelligent and do not have the ability to learn. While brains of reptiles do seem universally smaller than those of equal sized endotherms, neither in social behavior nor in learning ability is there inferiority of reptiles to birds and mammals (Burghardt, 1997a; Jerison, 1973). Studies of reptile behavior have been sadly neglected as compared to birds and mammals (Burghardt, 1977).

3. Communication

3.1. Communication channels

Crucial to understanding communication channels is the definition of *communication*. Allwood (1976) gave following definition: *The term communication is used in many different ways. In its widest sense, it is used to designate the sharing or transference of any phenomenon whatsoever between two entities. E.g. in physics, one speaks of 'communicating vessels' or of the 'communication of power to a machine' (p. 48).* More precisely, communication refers to communication between two individuals (e.g. see Fig. 3) and can be defined as “the cooperative transfer of information from a signaler to a receiver” (Alcock, 1998). Thus, successful communication involves at least two individuals: a sender and a receiver. The sender and receiver are not only emitting or receiving information, but are typically capable of some degree of consciousness or intentionality. Consequently, if a male frog calls, but its call is not received by another specimen, communication has not occurred.

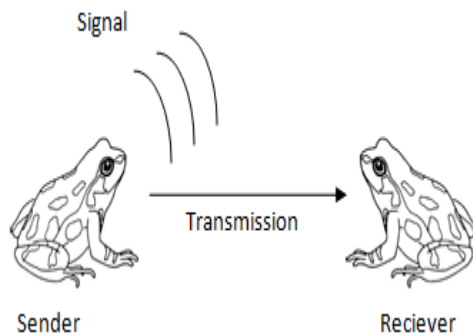


Fig. 3. Communication between two frogs.

There are different information transmission channel forms (referred to as *communication channels* here). Those communication channels are the senses that a creature uses to receive information, like olfaction and vision. The function of animal communication is the possibility to make decisions based upon the behavior, physiology or morphology of others, through the use of signals (Endler, 1993). The functions can include finding or courting a potential mate, or protecting a territory (Ryan and Rand, 1993; Johnstone, 1996).

The sender and the receiver are most often of the same species (intraspecific communication), but successful communication is also possible between two different species (interspecific communication) (Marler, 1967). The latter is obviously more difficult since different species use similar or identical gestures for different messages. A good example for interspecific miscommunication is experienced by many new dog owners who try to train their new pet. It can be very challenging to understand non-human communication, but it is not impossible to make sense of animal signals (Marler, 1967). Human-canine communication is also a good example for effective interspecific communication; humans and dogs can work cooperatively together, e.g. to herd other animals (McConnell and Baylis, 1985).

The senses most utilized depend on the lifestyle and habitat of an animal, which ultimately influences or limits their interspecific communication. Animals that live in environments without any light, for example, sometimes completely lack vision as a sense. The Blind Cave Fish (*Astyanax mexicanus*) lives in pitch-dark and has adapted to caves and lost its eyes and pigmentation (Dowling, Martasian, and Jeffery, 2002). Specialization and diversification is key to the survival and success of species in highly competitive environments and is the cause of a diverse fauna (Pianka, 1977). Different survival strategies and adaptation to different habitats requires the use of different senses. Consequently, well-developed senses that play an important role in the lifestyle of an animal are therefore also used in communicating with others.

3.2. Differences between human and other animal communication

Being mammals, humans and other mammals have very similar ways of transferring information. Humans mostly use verbal and visual signals for communicating, just like other mammals with acute vision and hearing. But our senses are also oftentimes significantly inferior to those of many animals (Schauber, 2008; Smutzer, Sayed, and Sayed, 2006).

Nevertheless, human communication through language is unique and well developed. Words are given arbitrary definitions and the sophistication of grammar permits the creation of an infinite number of messages (Wilson, 1982; Morton, 1983). No other species communication system is known to be as complex, but animals also use a wide variety of ways to communicate, each as diverse as the number of species that exist.

Some animals absorb information from their environment in different, unique ways that are not available for humans. Sharks and rays, for example, use an electrosensory mechanism to locate objects. They are sensitive to weak electric fields emitted by prey, and they use these bioelectric fields to detect the position of the prey (Kalmijn, 1971). Another prey-locating sense used by animals like vampire bats and many snakes is infrared detection. Snakes use their “pit organs” as radiant heat detectors. They help the snakes detect, locate and apprehend their warm-blooded prey and warn them of the presence of warm-blooded predators (Campbell et al. 2002).

The two communication modes that will be discussed in this thesis are more familiar to humans: visual and olfactory communication. Nevertheless, vision and smell are used differently among species. It is necessary to know those differences when keeping certain species in captivity so as to provide successful enrichment and consequently ensure the animals’ welfare.

3.3. Reptile and amphibian communication

Reptiles and amphibians generally rely on visual, acoustical, tactile and chemical signals, with animals often using multiple channels simultaneously (Vitt and Caldwell, 2009; Johnstone, 1996). Being two diverse groups of animals, reptiles and amphibians require varied approaches to successful captive keeping and enrichment. When approaching enrichment possibilities with such a vast number of different species, the natural history, and therefore communication channels of each should be studied intensively beforehand.

4. Visual Communication

Visual communication generally occurs in reptiles and amphibians with both good vision and color vision, which suggests that color is used in communication (Zug, Vitt, and Caldwell, 2001). It is most commonly used by diurnal animals, since it relies on the reflection of light (Blair, 1968; Brattstrom, 1974). Many reptiles are sexually dimorphic in coloration, which further suggests the importance of color in gender and species identification (Zug, Vitt, and Caldwell, 2001).

Visual communication uses body movements or flashing of a body part with a characteristic color or shape. In amphibians and reptiles, limb movements, headbobs, pushups, rapid shuttling movements,

and open mouth threats compose the most common signals (Zug, Vitt, and Caldwell, 2001; Stamps, 1977). Many studies on the social behavior of lizards have proved them capable of a variety of behavioral postures, sequences, and sociality that goes beyond that found in some mammals or birds (Brattstrom, 1974). Head-bob patterns can mean different things for different lizard species and different contexts like fear (Stamps and Barlow, 1973) or subordination (Clarke, 1965). The efficiency of a visual communication system is partially dependent on the number of different signals that can be generated and how those types of signals are related to each other.

Visual communication is best known from iguanian lizards (Blair, 1968; Marler, 1968), but occurs in many other reptiles and amphibians. Visual displays are most often directed at specific individuals, but assertion or advertisement displays can be performed by territorial males who want to reinforce their territorial status to other males or to attract females in sight (Zug, Vitt, and Caldwell, 2001). Oftentimes, visual communication are unseen by the addressee, and the efforts of the sender remain unnoticed.

4.1. Veiled Chameleon - *Chamaeleo calytratus* (Duméril & Bibron, 1851)

4.1.1. Species description

The Veiled Chameleon, also known as the Yemen Chameleon, is native to the dry plateaus, grasslands, inland river valleys, and rocky steppes of southwestern Saudi Arabia and Yemen (Fig. 4) (Badger, 2006; Sprackland, 2010; Bowmaker, Loew and Ott, 2005), but it also arrived as an invasive species in the United States. Populations have become established at the Southern Gulf Coast of Florida and Hawaii (Anderson and Carpenter, 2011; Kraus and Duvall, 2004; Krysko, Enge, and Wayne King, 2004; Sprackland, 2010). It generally prefers tropical and subtropical habitats (Anderson and Carpenter, 2011). As for many chameleons, it is a tree dwelling species, but juveniles generally prefer to live in high grass (Gibbons, Green, and Mills, 2009).



Fig. 4. Distribution of the Veiled Chameleon in its original range.

Veiled Chameleons (Fig. 5) are relatively large lizards and reach a total body length of 45-62 cm. The body of both male and females is greenish in color and bears a few bright colored bands, which can be green, yellow, orange, black, white, or blue in color (Krysko, Enge, and King, 2004). The animals can change their body color, but do not adapt it to match their background, like some other chameleons. Color change is rather a physiological response to emotion and is also dependent on temperature and activity of the lizard (Sprackland, 2010; Krysko, Enge, and King, 2004). Males are larger than females, brighter in color, and they have a crown-like structure on their heads, which is known as a casque. Their eyes are on independent turrets and can look in two different directions at the same time (Gibbons, Green, and Mills, 2009). Another striking feature is their long projectile tongue that shoots forward to entrap the prey and recoils back into its mouth (Zug, Vitt, and Caldwell 2001; Perkins, 2010).



Fig. 5. Male (right) and female (left) Veiled Chameleon. Photograph courtesy of Smithsonian's National Zoo, Matthew Evans.

Veiled Chameleons' diet consists primarily of insects (Bowmaker, Loew, and Ott, 2005). Both males and females are strictly solitary and communicate over distances of several meters for territorial interaction and courtship behavior. A Veiled Chameleon can be quite aggressive to unwanted approaches of conspecifics of either sex and will attack with vigorous bites. This is especially observed in non-recipient females (Sprackland, 2010; Neèas, 1999; Bowmaker, Loew and Ott, 2005). Veiled Chameleons primarily use visual communication, including body movements, head jerking, and color pattern changes (Kelso and Verrell, 2002), but low frequency vibration may serve as another mode of communication (Barnett, Cocroft, and Fleishman, 1999; Hill, 2001).

The Veiled Chameleon is not threatened by declining populations according to the IUCN (Anderson and Carpenter, 2011).

4.1.2. Courtship displays

There is a broad range of signals that occur during courtship and are used between reproductive male and female Veiled Chameleons (Krebs and Davies, 1993). Male signals differ from female signals and many of these displays exhibit some aspect of male 'quality' (Karsten et al. 2009). The courtship displays are used to coordinate the time of copulation, so that successful reproduction can occur. The quality of courtship displays may also allow choosing between potential mates (Bradbury and Vehrencamp, 1998). Resting males and females exhibit different color patterns described by Kelso and Verrell (2002): *"The male-specific color pattern consists of a pale green or beige background, vertical yellow bars on the sides of the body, and dark markings along the front of the casque, around the eyes, and running horizontally along the body. Males alter the relative intensities of these colors depending on social context. Females vary their color patterns according to both social context and, it is believed, stage in the reproductive cycle [...]. Resting females exhibit an olive green background with brown markings and often a light-colored lateral stripe."* (p. 497)

During courtship, the color patterns change, to signal reproductive readiness or rejection. The color changes are usually accompanied by several different postures. The male's behavior patterns can be: 'non-courtship' behavior patterns, 'courtship/precopulatory' behavior patterns, and 'copulatory' behavior patterns. Female behavior patterns vary from receptive to non-receptive females.

4.1.3. Trial: Encounter of a male and a female

To determine how the Veiled Chameleon uses visual communication, an introduction of a male Veiled Chameleon to the enclosure of a female chameleon was conducted and videotaped. What was observed is only one of many different outcomes that could have happened.

Female Chameleon:

In the beginning of the observation, the female's body color is a dark green with dark markings and vertical bars on the side. There are also some light yellow lateral markings. As the male starts approaching the female, her first response is a slight change in color from an olive green to a darker complexion. As the male continues to approach her, she darkens even more until the dark green turns into almost black. Ultimately she threatens him, by compressing her body laterally, raising her occipital flaps and then gasping her mouth wide open. The rejection posture sequence is completed by her rocking from side to side, which Cuadrado (2000) calls the 'crazy dance'. Normally, the female would then attempt to bite the male, but the observation was stopped at that point for the sake of the health of the animals

Male Chameleon:

The first response of the male as he spotted the female was the brightening of his colors, by intensified its coloration and the contrast between dark and light markings. As the male approached the female, it flattened its body (lateral compression), extended its gular area (throat), and tightly curled its tail. It stopped and held that position for some seconds and then kept on approaching the female.

The behavior of the male is according to non-courtship behavior (Ethogram: Appendix: I.a).

4.1.4. Discussion

The visual communication of Veiled Chameleons is quite complex and consists of sequences of posture and color changes. To the untrained eye, it is not always obvious what the posture and color changes mean, but it is quite important for the husbandry of captive Veiled Chameleons. For instance, to avoid injuries when trying to breed this species, it is indispensable to be aware of the meaning of their postures. When thinking about possibilities for enrichment of captive Veiled Chameleons, it is apparent that visual enrichment is suitable for this species. The feeding of live crickets is one option to stimulate the animals and give them the opportunity to hunt actively.

4.2. Green Crested Basilisk - *Basiliscus plumifrons* (Cope, 1875)

4.2.1. Species description

The Green Crested Basilisk, also known as Plumed Basilisk or Jesus Cristo Lizard, is native from southern Mexico to northern Colombia (Fig. 6) (Badger, 2006; Sprackland, 2009; Capula, 1989; Pianka and Vitt, 2003). Its habitat is limited to hot, humid rainforests where it prefers to dwell alongside streams (Sprackland, 2009; Sprackland, 2010). The Green Crested Basilisk is a medium to large sized lizard (Hirth, 1965; Pianka and Vitt, 2003) that got its name from its bright green color that is topped with white and black bands on its back and neck. It can grow up to a total length almost 1 m (Sprackland, 2009; Nicholls, 2001). Green Crested Basilisks are sexually dimorphic, males having a crest on the back and tail, large plumes on their heads, along with a dewlap, an extendable flap of skin under the jaw (Fig. 7a). The generally smaller females have only one, smaller crest on the head (Fig. 7b) (Nicholls, 2001; Lewis and Grant, 2009; Sprackland, 2009).

The name Jesus Christ lizard comes from their ability to run across water. When forced to flee, it drops into water and runs across the stream (Capula, 1989; Pianka and Vitt, 2003). They are able to stay on the surface because the toes of their hindfeet are edged with large scales. When the foot strikes the water, the scales are forced outward, and in a snowshoe-manner, the foot does not sink deeply into the water (Hsieh, Lauder, and Wake, 2004; Laerm, 1974). The lizard runs quickly enough to avoid sinking (Badger, 2006). It can reach speeds of approximately 2.14 m/s (Laerm, 1974). Green Crested Basilisks are omnivorous, feeding on small vertebrates and invertebrates, such as rodents, insects, and frogs (Sprackland, 2010; Sprackland, 2009) as well as flowers and fruit (Capula, 1989; Hirth, 1963).

Like all iguanid lizards, Green Crested Basilisks heavily use visual communication signals (Purdue and Carpenter, 1972). Although visual communication clearly plays an essential role in the

lizards' communication, chemical communication may also play an important role, in as much as licking of substrate is commonly observed. Substrate licking may be used to pick up chemical information from their environment (DeFazio et al. 1977; Bissinger and Simon, 1979).

The Green Crested Basilisk's conservation status is unknown as is not assessed by the IUCN Red List of Endangered Species, but they are popular pets and therefore commonly found in many pet stores.

4.2.2. Head bob displays

Like nearly all lizards, Green Crested Basilisks are territorial and retain that behavior in captivity. Territory ownership plays a very important role for male Green Crested Basilisks' reproductive success. If a male can defend a territory with key resources, like water sources, and a constant supply of food, females will be attracted. Males communicate by using a range of visual displays like head bobs and nods, dewlap extensions, push-ups, tail slapping, and mouth opening or patrolling displays to advertise their territories (Nicholls, 2001).



Fig. 6. Distribution of the Green Crested Basilisk.

Fig. 7. Male (top) and female (bottom) Green Crested Basilisk. Photograph courtesy of Smithsonian's National Zoo, Jessie Cohen (top), Meghan Murphy (bottom).



Especially prominent are the Green Crested Basilisks head bob displays which appear very different for other lizard species. The Green Crested Basilisks uses head bobs, throwing their heads up in a straight line. Nicholls (2001) described: “A *head bob* is an action when the head is initially thrown up to an almost vertical position, followed by a short pause then by a series of individual bobs decreasing in amplitude (8-14 bobs in total). Head bobs are of a slower and more determined pace and they tend to be more pronounced than head nods. A *head nod* is the same action as a head bob but there are more individual bobs in the same amount of time and the initial and final nods have smaller amplitude when compared to head bob “(p. 100). Head bobs and nods are considered as aggressive behavior and are usually directed to other males. The dominant male is likely to display with the highest frequency (Nicholls, 2001). Still, without other competitors around, head bobbing is very common and may be directed towards the female to affirm the male’s dominant position

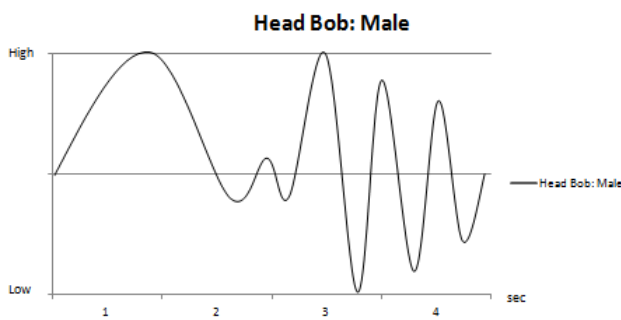


Fig. 8a. Length and relative amplitude of a common head bob observed in the male Green Crested Basilisk at the National Zoo.

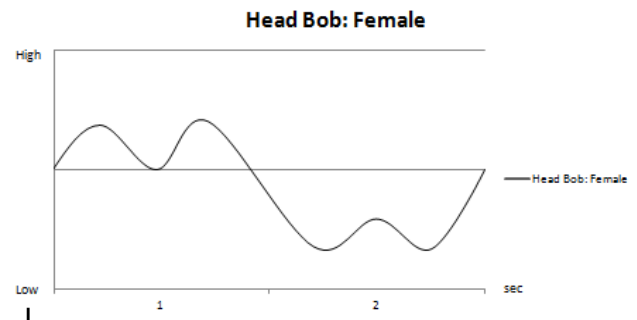


Fig. 8b. Length and relative amplitude of a common head bob observed in the male Green Crested Basilisk at the National Zoo.

4.2.3. Observation: Interactions between male and female

As for the Green Crested Basilisks at the National Zoo, it was mostly the male that could be observed bobbing his head, but the female was frequently seen head bobbing too (Video: Green Crested Basilisk). The male’s head bobs are much more pronounced, longer and more complex (e.g. see Fig. 8a) than the female’s head bobs (e.g. see Fig. 8b)

The male was observed head bobbing either towards the female or while patrolling his territory. When he was out of her sight, it was often observed that he would extrude his tongue to lick the substrate. The combination of undirected head bobbing and tongue extrusion leads to the conclusion that that behavior is part of a patrolling performance. Although there was no other male present, he would still patrol around his territory, with the aim to defend it. The only times she was seen bobbing her head was in response to his initial head bobs in her direction.

4.2.4. Discussion

Since Green Crested Basilisks are very territorial, it can be challenging to keep more than one male in an enclosure, but it is certainly possible to keep them in larger groups with more males. As long as there are enough good territories available (adequate water area and food), aggressive

encounters can be reduced to a minimum. The social contact to conspecifics can certainly be seen as a kind of enrichment, but the animals can profit from other types of visual enrichment.

4.3. Grand Cayman Iguana - *Cyclura nubila lewisi* (Grant, 1940)

4.3.1. Species description

The Grand Cayman or Blue Iguana is a subspecies of the Cuban Iguana (*Cyclura nubila*) (Alberts and Hudson, 2001) and is native to the Grand Cayman Island in the Caribbean (Burton, 2004b). It was formerly widespread in dry and coastal habitats throughout the island but now are found only in the High Rock-Battle Hill area to the east and south of the Queen's Highway (Fig. 9). These primarily terrestrial iguanas prefer to live in dry, rocky areas and at beaches (Sprackland, 2010; Sprackland, 2009).

The Grand Cayman Iguana is the largest native land animal on Grand Cayman with a total length of 1.5 m and weighs as much as 14 kg (Blue Iguana Recovery Program, 2012a; Gibbons, Green, and Mills, 2009; Sprackland, 2009). The Grand Cayman Iguana is also known as the Blue Iguana, owing to the striking blue color (Fig. 10) it obtains during breeding season (Blue Iguana Recovery Program, 2012b; Burton, 2004a). Grand Cayman Iguanans can live up to 50 years or more (Gibbons, Green, and Mills, 2009).

Although they possess sharp teeth, the Grand Cayman Iguanans are strict herbivores and live off leaves, berries, and flowers (Burton, 2012; Sprackland, 2010). For most of the year Grand Cayman Iguanans live a solitary life and defend their territory against every intruder. Male iguana territory stretches over a much larger range than a female's territory (Goodman and Echternacht, 2005). As it is common for iguanas, Grand Cayman iguanas mainly use visual signals, and to a much lesser extent chemical and tactile signals, in social communication. It is also known that iguanas strongly react to colors, which is probably the reason for the blue color of males during the breeding season (Burton, 2004a). The bright blue colors may signal to females that the male is a strong and high-quality mate they should choose for reproduction.

The conservation status of the Grand Cayman Iguana is listed as *critically endangered* by the IUCN (Burton, 2004b). The remnant wild population was estimated between 10–25 individuals in 2002 and is expected to be extinct in the next 5–10 years (Burton, 2004b; Gibbons, Green, and Mills, 2009; Goodman and Echternacht, 2005). The major threat to these unique animals is the spread of feral dogs and cats on the island. Another factor in their rapid decline is habitat loss due to deforestation (Burton, 2012; Burton, 2004b; Blue Iguana Recovery Program, 2012a).



Fig. 9. Distribution of the Grand Cayman Iguana.



Fig. 10. Male Grand Cayman Iguana. Photograph courtesy of Smithsonian's National Zoo, Jessie Cohen.

4.3.2. Head bob displays

Members of Iguanidae are known for its elaborate visual communication, and especially for their head bob patterns. Iguanids are territorial lizards and establish social hierarchies when confined to a limited space (Hunsaker and Burrage, 1969). Both female and male Grand Cayman Iguanas defend their territories vigorously against intruders of the same or the other sex outside of breeding season. Only during breeding season in early May will females allow males close to them (Blue Iguana Recovery Program, 2012b). Before an attack is launched, aggressive displays like head bobbing challenge the opponent and prompt him or her to retreat.

A head bob by a Cayman Island Iguana is an action where its head is quickly thrown up to an almost vertical position, often immediately followed by a quick bob with decreasing amplitude (1-2 bobs in total). The display structure between males and females does not differ notably.

Grand Cayman Iguanas often combine those head bobs with push up displays and open mouth posturing. In comparison, head bob structures by Grand Cayman Iguanas look quite different to other lizards' head bob displays. Comparison to the head bob displays of Green Crested Basilisks, many differences are observed. The comparison between the two species demonstrates how the same visual communication tool is used differently by two different species of lizard.

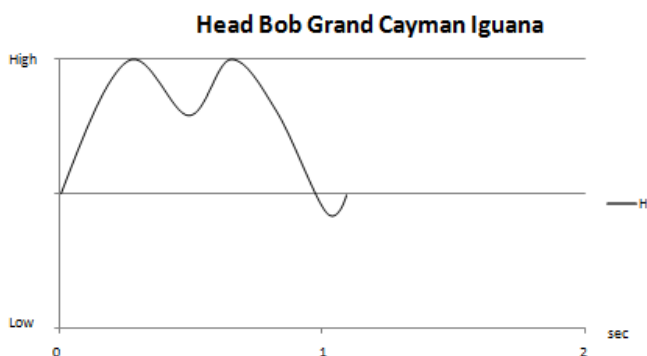


Fig. 11. Head bob sequence diagram for a Cayman Island Iguana. The line in the diagram denotes the relative length and amplitude of the head during a display sequence. The model is based on observations of the Grand Cayman Iguanas at the National Zoo.

4.3.3. Observation: Interactions between male and female

The Grand Cayman Iguanas at the National Zoo are a male and a female that are kept in one enclosure. Since Grand Cayman Iguanas are quite territorial, a lot of aggressive displaying between the male and the female could be observed. Head bobbing most often occurred when one of the iguanas

deemed that its personal space was intruded by the other. After an exchange of several individual head bobs, the intruder either left voluntarily or was chased away. Grand Cayman head bobs can include between one and two individual bobs and are much shorter and less complex than the head bobs of male Green Crested Basilisks (e.g. see Fig. 11, 8a, and 8b). They were often accompanied by open mouth postures and push up displays.

Similar to the male Green Crested Basilisks, the male Grand Cayman Iguana also spontaneously displayed as he moved around his activity range, independent of social encounters. These displays, called “assertion displays” (Stamps, 1977), can be interpreted as a “declaration of the lizard’s presence to any other lizard in the immediate area, an aggressive gesture for others to ‘keep their distance’” (Carpenter, 1967).

4.3.4. Discussion

There are a lot of similarities between the two lizard species, Grand Cayman Iguanas and Green Crested Basilisks. Both species heavily rely on visual communication and make extensive use of head bobbing displays. However, the head bob sequences of the two species look fundamentally different. Although both species use head bobbing as a way of communicating, it is necessary to study the context in which the head bobs occur for each species. That way, keepers can better interpret the meaning of displaying sequences in particular situations. When aggression happens between lizards sharing an enclosure, staff can intervene and prevent possible injuries. It is therefore vital to know what the visual displays of each lizard mean when keeping them in captivity with conspecifics.

Not only can the social activities of the lizards be monitored better, but visual enrichment can be incorporated in the daily lives of captive animals. As iguanas have excellent vision and heavily react to color, the caretakers can make use of that knowledge by trying different tools that stimulate the animals. Incorporating colorful and moving enrichment items can certainly offer opportunities to add excitement to the daily routine.

4.4. Cuban Crocodile - *Crocodylus rhombifer* (Cuvier, 1807)

4.4.1. Species description

The Cuban Crocodile inhabits only a restricted area in Cuba and the adjacent Isle of Pines (Fig. 12). It favors freshwater habitat such as swamps, marshes, and rivers and rarely swims in salt water (Ralph de Sola, 1930). It is considered the most terrestrial of all crocodylians (Trutnau and Sommerlad, 2006).

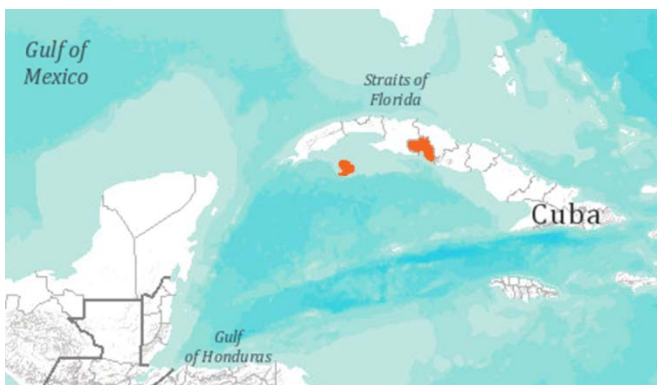


Fig. 12. Distribution of the Cuban Crocodile.



Fig. 13. Cuban Crocodile. Photograph courtesy of Smithsonian's National Zoo, Meghan Murphy.

It is a small crocodylian species and averages a total length of 2-2.5 m to a maximum of 5 m (Steel, 1989). The Cuban Crocodile is sexually dimorphic in size and color, with the female being usually larger and lighter in color than males. Adults have a grey to black coloration with a sulfur yellow sprinkling and a light belly without markings (Fig. 13) (Trutnau and Sommerlad, 2006).

The prey of the Cuban Crocodile is very diverse and consists of all sorts of mollusks, insects, crustaceans, fish, birds, and small mammals (de Sola, 1930; Trutnau and Sommerlad, 2006).

Crocodylians, including the Cuban Crocodiles, display remarkable social behaviors. No other reptile group demonstrates the amount of care and protection of their offspring (Chiszar and Smith, 2001). Crocodylians are the most vocal of the reptiles and appear to have a sophisticated range of auditory signals (Burghardt, 1977; Gans and Maderson, 1973), but visual signals, like a wide range of postures, also play an important role in crocodylian communication (McMahan, 2001). They have fairly good eyesight and their elliptical pupil has great light gathering ability (McMahan, 2001), which suggests good night vision. Generally, visual signals, often in combination with auditory signals, are widespread in crocodylian communication. Visual signals prevail in short-distance communication, whereas auditory cues are primarily used in long distance interactions (Zug, Vitt, and Caldwell, 2001). Head slaps atop the water's surface are one example of the visual signals used by crocodylians (McMahan, 2001).

According to the IUCN, the Cuban Crocodile is a *critically endangered* species (Targarona et al. 1996). Its restricted habitat and range make it a very vulnerable species. Additionally, humans have hunted these ancient reptiles to near extinction for their skin (McMahan, 2001) and hybridization with American Crocodiles now threatens the small population of Cuban Crocodiles. The total population size is estimated to be 4,000 (Targarona et al. 1996).

4.4.2. Training

Crocodiles are a common sight in zoos, but their size and strength make them dangerous captives. Due to those difficulties, zoo staffs have to come up with creative ways to handle these dangerous animals. Training has been utilized advantageously with captive mammal collections for decades. These training methods are only now becoming increasingly popular tools for herpetology keepers. There are many purposes of training animals, but they all try to alter an animal's behavior or reactions (e.g., fear or aggression). Leache (2001) noted: "Animal training may be viewed in terms of animal-learning process. Animal learning can be defined as a more or less permanent change in an animal's response or behavior potential resulting from experience or practice. (p. 1257)

Different concepts can be used for training animals, such as classical conditioning (teaching animals to know its own name), operant conditioning (using tools like a whistle or a target) and habituation (establishing a daily routine) (Leache, 2001).

Training can be one approach in reducing the stress associated with veterinary procedures, handling, or transporting crocodylians. Knowing their communication channels is a premise for successful training because the trainer needs to communicate his or her intentions to the animal. Training can be used to ease many necessary daily husbandry procedures and to help simplify stressful events like blood draws.

In the particular case of the Cuban Crocodiles at the National Zoo, the training method used is called *operant conditioning*, in which animals modify their behaviors in response to outcomes created by their actions (Ramirez, 1990). Positive reinforcement is used during training to condition animals to perform desired behaviors on request. The animal learns to attach a certain benefit to performing certain behaviors on cue.

The long-term goals of this crocodile training are to weigh and draw blood without restraint. A more immediate goal is name recognition for each animal. Knowing that crocodylians use

combinations of visual and vocal signals to communicate, the training can be adapted to the animals' natural communication behavior.

4.4.3. Operant conditioning

Before the training could begin, the animals had to be conditioned to a whistle. The whistle is a bridge, as it is initially paired with a food reward, to condition the animal to associate that noise with a food reward (Forthman and Ogden, 1992). Once that association was established, the training could begin (Video: Cuban Crocodile part 1-3).

Cuban Crocodile: Name recognition training:

First, the animals are separated and only one animal is trained at a time. The trainer calls the animal by its name, squats down, and gives it one minute to respond, by approaching the trainer. If the animal complies, the trainer blows a whistle and feed the crocodile. At the National Zoo the animal is given one minute to respond to the trainer's request. Once the behavior is completed and the animal is rewarded, the trainer then moves and repeats that procedure. Once the animal has learned a behavior, which is defined as responding to the trainer's call within the designated time frame 85 to 90 percent of the time, the trainer can then begin to mold the behavior. For example, shortening the time required for the animal to complete the behavior. If an animal has successfully mastered a behavior and for any reason is not responding, the trainers will not attempt to continue training the animal. If the behavior is not sufficiently learned as stated above, and the animal is still learning the behavior, the trainer may try to elicit the correct behavior by repeating the cue. If there is still no response from the animal, the training session is ended and another attempt can be started one hour later. In case the animal gives an aggressive response such as such as lunging, biting bars, hissing, tail slapping, the training session is ended and another attempt can be started in two hours.

The name conditioning forms the basis for further training like targeting and stationing.

Cuban Crocodile: Target training:

Target training teaches the animal to touch, follow, or go to a target. Many things can be used as targets, as long as they are safe to use and the animal can perceive them. At the RDC, dowel rods with a $\frac{3}{4}$ inch diameter served as the target.

The trainer calls the animal by its name, squats down, and gives it one minute to respond, by touching the target with its snout. If the animal approaches the trainer, she or he will whistle and feed the crocodile. Then the trainer moves and repeats that procedure.

Nile Crocodile Stationing:

"Stationing" derives from target training and tells the animal where it is expected to go and stay during a training session (Ramirez, 1990). The animals at the RCD are not trained to perform that behavior yet, as their training status is not advanced enough for this elaborate training method. The video shows an example how a different crocodilian species, a Nile Crocodile (*Crocodylus niloticus*), at the Bronx Zoo, NY, was to perform stationing behavior for a blood draw. The animal was trained to touch the station, in this case a black Frisbee that is attached to a broomstick, with the anterior portion of its snout with its mouth closed.

Since the animals can distinguish between colors, the target sticks and stations are specific to each individual animal. The animal is trained to touch the station until the station is removed.

In this video, the area where the animal was stationed was confined by two wooden posts. These posts provided a non-invasive barrier that prevented the crocodile from flipping around quickly toward the animal keepers. As soon as the animal touches the station, the trainer whistles and then feeds the

animal. This behavior allows the veterinarians to go inside the enclosure and draw blood without sedating the dangerous animal.

The station helps the animal to feel more secure during the unfamiliar procedure, because it indicates a clear and easy way to understand behavior (Hellmuth et al. 2012).

4.4.4. Discussion

Basic trained behaviors such as targeting and stationing are outstanding approaches to position and manipulate an animal in a safe and for the animal stress-free manner. Veterinary procedures, such as blood draws can be performed easier and without the use of sedatives or heavy restraint. Once the animal understands the behavior, targeting and stationing can become a tool to train many other behaviors, such as stepping onto a scale (Hellmuth, et al., 2012).

A combination of visual and auditory signals is used for training, which accords with the natural communication habits of crocodylians. It is also an excellent way to strengthen the relationship of keeper and the animals, through positive experiences that the animals attach to the specific person. In addition to that, the training can be viewed a form of enrichment to the animals, since it engages them in physical exercise, and mental stimulation through the learning process.

5. Olfaction or Chemical Communication

Olfaction or chemoreception relies on the presence of odors and chemical releasers, called pheromones (Karlson and Buenandt, 1959). Those can be divided into two subcategories: volatile (nasal) or surface adherent (vomeronasal). The odors used for communication between two animals are derived from glandular secretions (Zug, Vitt, and Caldwell, 2001) and are secreted either as liquids or as air born odors (Wilson, 1968). Pheromone based communication was probably the first manner of communication to arise during the evolution of single-celled organisms; receptors were used to locate other organisms that served as food (Shorey, 1976). The advantage of chemical communication over other modes of communication is that odors can be smelled for a long time, even long after the sender of the message is gone (Wilson, 1968). Human olfactory abilities are slight in comparison to those of many animals (Smutzer, Sayed, and Sayed, 2008). While humans have only about 350 functional olfactory receptor genes out of 1,000, mice have about 1,100 functional olfactory receptor genes out of 1,300 (Zozulya, Echeverri, and Nguyen, 2001).

Vomeronasal communication is a type of olfaction not accessible to humans because we do not possess the necessary organs to interpret odors through this means. Zug, Vitt, and Caldwell (2001) described the process of vomeronasal olfaction like this: Odor chemicals are picked up by the tongue or the surface of the head and transported to the nasal sac in amphibians and the roof of the mouth in reptiles, and then ultimately to the vomeronasal organ (p. 223). The vomeronasal organ is also called the Jacobson's organ (Fig. 14). Some salamanders can also detect chemical signals through unspecialized receptors in their skin.

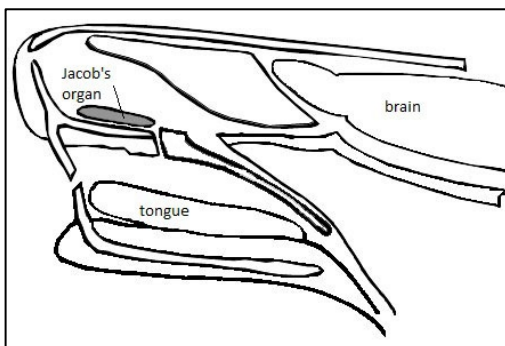


Fig. 14. Position of the vomeronasal (Jacobson's Organ).

Chemical communication's uses include individual or species recognition, territorial marking or sexual stimulation. For reptiles and amphibians, chemical communication has been studied most intensely in salamanders and skinks; but it is used widely by all members of the autarchoglossan clade (skinks, snakes and their relatives) (Zug, Vitt, and Caldwell, 2001).

5.1. Eastern Hellbender - *Cryptobranchus alleganiensis alleganiensis* (Daudin, 1803)

5.1.1. Species description

The Hellbender is widely distributed in fast flowing rivers and creeks in much of the eastern United States (Fig. 15). They are found from southern New York to northern Alabama and extreme northeastern Mississippi, and westward to central and southern Missouri and northern Arkansas (Petranka, 1998). The Ozark Hellbender (*Cryptobranchus alleganiensis bishopi*) is a western subspecies of the Hellbender and is found in central Missouri and northern Arkansas (Weehler et al. 2003).

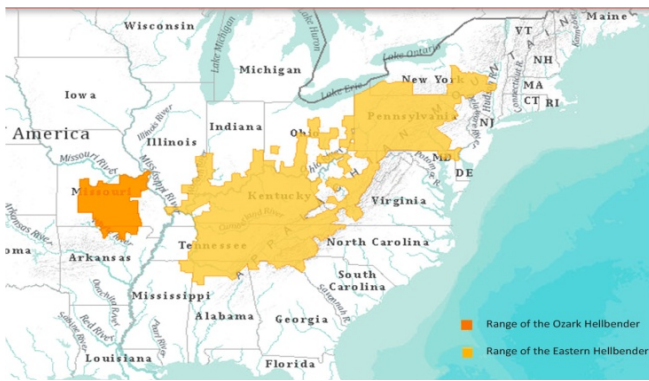


Fig. 15. Distribution of the Eastern Hellbender and its subspecies, the Ozark Hellbender.



Fig. 16. Eastern Hellbender. Photograph by Sarah Kuppert.

Hellbenders are the largest salamanders in North America, reaching 70cm (27inches) (Nickerson and Mays, 1973). The body color varies from greenish, yellow-brown to slate grey with very scattered dark spots (Fig. 16) (Petranka, 1998).

The Eastern Hellbender is a relatively long-lived species, with longevity exceeding 20 to 30 years (Weehler, et al., 2003). Morphologically, they are capable of swimming but prefer to slowly crawl on the bottom with their sturdy limbs (Smith, 1907). Eastern Hellbenders prefer rocky, clear creeks and rivers with large rocks for shelter (Hammerson and Phillips, 2008). Being nocturnal animals, they remain hidden under large flat rocks during the day and emerge in the darkness for feeding, mating, and other activities.

The small eyes located on the top of its head can detect light but are not very good at forming concrete images, a common adaptation of nocturnal species. Due to the restriction of its visual abilities, visual communication is to be ruled out as an important communication channel. Vocalization is rare, hence excludes the auditory channel as a main communication channel (Nickerson and Mays, 1973). Since the visual and auditory channels seemingly play a minor role in interspecific communication, olfaction is likely the major communication channel.

At night they forage for food, using their lateral line system, a series of lines of superficial sense organs along their bodies (Wright, 1951), to detect prey animals (Herman, 2000). They are strictly carnivorous, their diet consisting primarily of crayfish. They also eat their own epidermis that they

shed. In captivity, they eat almost any small moving animal (Smith, 1907).

Next to the lateral line detection, olfaction is an important sense for the hellbenders that serves several uses. Aquatic salamanders have very simple and small nasal cavities, but they possess large vomeronasal organs (Zug, Vitt, and Caldwell, 2001). Bergmann and Foldi (2012) mentioned that “chemoreception is well developed in most salamanders and may play an important role in courtship, species recognition, feeding and territoriality”.

All salamanders rely significantly on chemical signals, especially for courtship (Verrell, 1989). They use pheromones to distinguish between species and to locate conspecifics; additionally, odors identify the reproductive status and sex of conspecifics and stimulate sexual activity in females (Houck and Sever, 1994).

The Eastern Hellbender is a primarily solitary species and rather territorial (Hillis and Bellis, 1971). Outside of the breeding season, adults defend rocks that they occupy from other hellbenders and only rarely share them with other individuals (Petranka, 1998). The Eastern Hellbenders that were observed in the experiment were raised in captivity, which ultimately changed their social behavior. The eggs were collected from the Allegheny River water basins and hatched in captivity at the Buffalo Zoo (Bell, MacBlane, and Wheeler, 2010). Being kept in groups of as many as nine hellbenders at a time, they likely became more tolerant of other hellbenders.

The Hellbender is listed as *near threatened* in the IUCN Red List of Threatened Species; it is in significant decline through much of its range owing to widespread habitat loss, (Hammerson and Phillips, 2008). Habitat degradation due to dams, logging mining, and road construction pose a serious threat to hellbenders. Chemical pollutants are probably also detrimental, especially to larvae and eggs. Recreational fishing is another factor that causes injuries and death because these amphibians are commonly used as bait. Some fishermen mistakenly believe that hellbenders are dangerously poisonous and therefore kill them at every opportunity (Hammerson and Phillips, 2008; Nickerson and Mays, 1973). For all these reasons, the number of hellbenders has declined drastically throughout its entire range in the last 20 years (Nickerson and Mays, 1973). It is vital to their survival as a species that the public is educated and that erroneous beliefs are thereby eradicated. Captive raising and head starting programs offer opportunities to inform the public about conservation needs and simultaneously provide opportunities to study the animals and engage in reproduction efforts.

5.1.2. Scent experiment: Methods and procedures

Olfaction plays an important role in the Eastern Hellbenders intraspecific communication, although how keen their sense of smell is remains unknown. Most salamanders use chemical cues for individual, sex and species recognition, which can be derived from pheromones that are produced by specialized glands (Wells, 2007). To get an idea of the olfactory abilities of these aquatic salamanders, we constructed an observational experiment. There were nine individuals housed in two tanks with a single water system. The method used for the experiment was to drip a food scented extract (Earthworm, *Lumbricus terrestris*) into the sump of the aquarium system, and then observe the reaction of the animals, characterized by rapid movement as the salamanders searched for food. The extract was gradually diluted with aquarium water in each subsequent experiment until the salamanders displayed no reaction. The goal was to determine the lowest reaction threshold to the diluted extract, indicated by failure to elicit searching activity, thereby assuming the ratio of extract to water that was undetectable (Methodology: Appendix: II.b) The aquarium system used (Fig. 17) holds 756.098 liters of water (199.74 gallons); the ratio of water to worm extract (0.05 ml) was therefore 1,512,196:1.

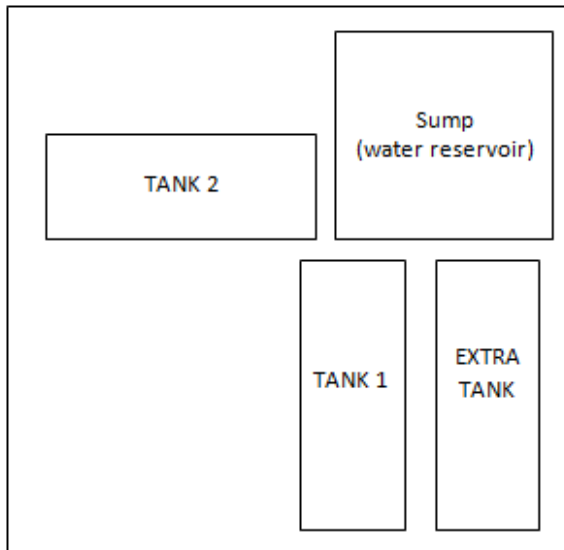


Fig. 17. Measurements of water in the system (in inches): Tank 1: (47 W x 17,375 L x 12,25 H), Tank 2 : (47 W x 17,375 L x 11,5 H), and the extra tank : (47 W x 17,375 L x 12 H), Sump (39 W x 44,25 L x 15 H). All tanks had 6 inch round sponge filters with and airline attached to them. Tank 1 and Tank 2 were the tanks that contained the animals, with nine hellbenders in each tank. Inside the tanks were bulkhead screen, through which the water left the tanks. The tanks also contained PVC pipes that were cut in halves, to serve as hides for the animals. Tank 1, the tank used for the observations contained 4 small caves (1 x 4.5), 1 big cave (4 x 3.5).

In each trial, a 10 min pre-baseline and a post-baseline observation were performed. The observations were conducted during daytime while overhead lights in the observation room were on. Because hellbenders are nocturnal they are usually in a resting state during the day, therefore it was easier to observe the beginning of foraging behavior characterized by increased movement.

The worm extract is made from the liquid residue of cut earthworms. To extract the liquid from the earthworms they were into pieces and placed into a small plastic container. Approximately 1ml of aquarium water was added to the container to express the liquid from the cut earthworms. The extract was then collected with a syringe and further diluted with a specific amount of aquarium water. The ratio aquarium water to extract varied each time the experiment was run according to the desired percentage of extract in one drop. Only one drop (0.05ml) of the diluted extract was then dripped into the sump.

Prior to the introduction of the worm extract, the hellbenders were essentially motionless. Within a few seconds of adding to extract to the sump water, they went into a feeding frenzy, crawling out of their hiding spot and looking for food.

5.1.3. Data analysis and results

Different dilutions were tested to find the water to extract ratio where the animals would not react to the introduced smell (Analysis: Appendix: II.c). At a dilution of 1% in one drop, the animals stopped reacting to the introduced extract. At 2% dilution, they reacted and began foraging. This observation indicates that their olfactory limit lies between a ratio of 1:75,609,800 and 1:151,219,600 dilution.

The amount of extract introduced did not appear to influence the reaction time of the salamanders, as reaction times did not show a noticeable pattern (Fig. 18a); however, the length of the subsequent foraging time seemed to be influenced by a lower percentage of diluted extract (Fig. 18b)

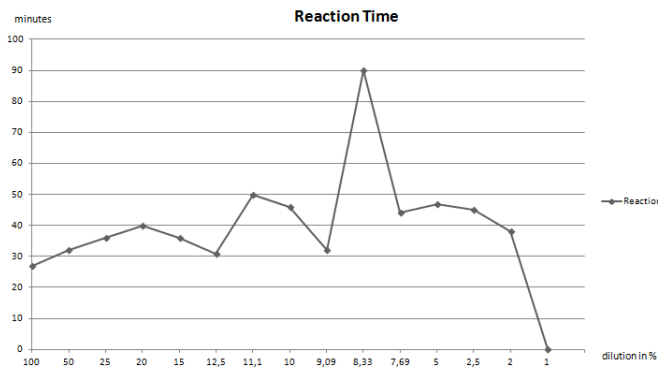


Fig. 18a. The reaction time reflects the time that passed between the introduction of the extract and an observable reaction of the animals. The average reaction time was 42 seconds.

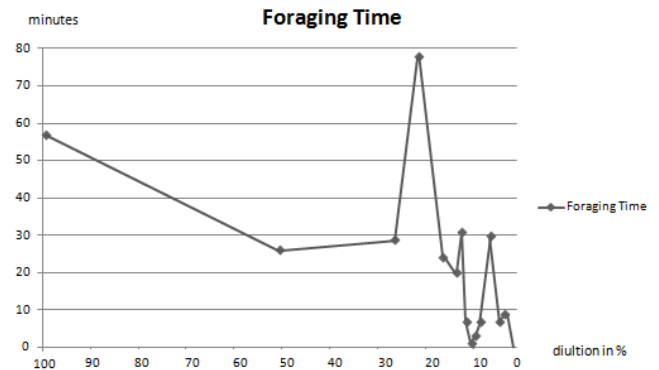


Fig. 18b. The foraging time is the time that the animals were active after the introduction of the extract. The foraging time was considered to be over when the animals went back to a resting state for 3 minutes. The average foraging time was about 23 minutes but decreased with the dilution of relative extract to water ratio

Lower percentages of diluted extract (10 % and below) tended to result in shorter foraging times than the average of about 23 minutes of foraging after the introduction of extract. While animals were foraging, different behaviours and amount of activity was observed and recorded. The activity levels were grouped in 4 activity categories: resting, alert, crawling and swimming (Ethogram: Appendix; II.a). During the recording interval of one minute, it was note whether at least one individual was showing one of the four activity categories.

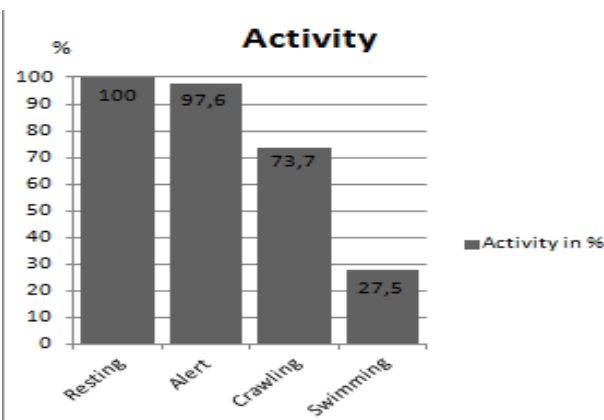


Fig. 19. Activity levels during the foraging time

Because Eastern Hellbenders are nocturnal, it is not surprising that at least one animal was motionless at all times during the observation (Fig. 19). Observations conducted in a dark room would therefore probably have changed the results drastically. Besides that, a lot of minor movements (classified as alert) and crawling could be observed. Swimming was relatively rare, and could be observed only about 27.5% of the observation time. Since there was generally no activity visible during the pre-baseline, it was possible to see the increase in activity that the introduction of the extract procured.

5.1. Eastern Diamondback Rattlesnake - *Crotalus adamanteus* (Beauvois, 1799)

5.1.1. Species description

The Eastern Diamondback Rattlesnake is a venomous pit viper species found in the southeastern United States, inhabiting the Lower Coastal Plain from North Carolina to southern Florida (Fig. 20). They prefer pine flatwoods, sandhill habitats, and low lying palmetto stands (Hammerson, 2007).



Fig. 20. Distribution of the Eastern Diamondback Rattlesnake.



Fig. 21. Eastern Diamondback Rattlesnake. Photograph courtesy of Smithsonian's National Zoo, Meghan Murphy.

It is the most massive venomous snake in the Americas and the largest rattlesnake (Graham, 2001). The Eastern Diamondback is a heavy, thick-bodied snake with large diamond-shaped markings that have a whitish outline and a black, brown or yellowish background (Fig. 21). Their rattle, which gives the rattlesnakes their name, is relatively large compared to those of other rattlesnakes (Gibbons, Green, and Mills, 2009). The rattle mechanism probably evolved as a warning to large grazing mammals, helping both animals avoid a deadly encounter. The muscles that shake the rattle are some of the fastest known, going more than twice as fast as a hummingbird's flight muscles (Graham, 2001). Like all snakes in the Viperidae family, they possess hollow, folding fangs, and like all snakes in the subfamily Crotalinae, Eastern Diamondback Rattlesnakes have heat sensitive facial pit receptors that allow them to sense the body heat of their prey (Graham, 2001).

Eastern Diamondback Rattlesnakes prey on small mammals, such as cotton rats, squirrels, and rabbits. They are ambush predators and sometimes remain in one location for several weeks, waiting for prey to come by (Gibbons, Green, and Mills, 2009). Snakes are often considered the least social of all reptile groups (Brattstrom, 1974; Wilson, 1982). Indeed, a common view is that outside of mating, snakes engage in virtually no intraspecific interactions. Because snakes are such solitary animals, it is not surprising that olfactory cues are their main communication channel for intraspecific communication (Blair, 1968). Chemical communication does not require a face to face meeting of two animals, making it the ideal communication channel for snakes. Visual signals, such as rearing above the ground to identify with others of its kind, are also used for intraspecific communication. Because the Eastern Diamondback is a rattlesnake, sound, specifically rattling, also plays an important role

in communication (Blair, 1968).

Rattlesnakes are unique to the Americas, known for their tenacity and potential deadliness, through which they earned respect from the first settlers. The fear of those snakes inspired negative propaganda against them and they were – and still are – hunted and killed in so called “Rattlesnake roundups” (Graham, 2001). Although the IUCN rates the endangerment of the Eastern Diamondback Rattlesnake as *least concern* (Hammerson, 2007), the Eastern Diamondback Rattlesnake is considered to be one of the most endangered snake species in the United States (Gibbons, Green, and Mills, 2009).

5.2.2. Olfaction and trailing behavior

Olfaction plays a major role in the lifestyle of Eastern Diamondback Rattlesnakes. Snakes in general are well known to be guided by the olfactory and vomeronasal system in courtship, reproduction and trailing of prey (Brown and McLean, 1983). Unlike human tongues, their tongues are not involved in gustation (Schwenk, 1994). They possess deeply forked tongues, specialized for picking up scent molecules from the air and ground. The morphology of its tongue is perfectly adapted to the requirements of an olfactory-based lifestyle. The long lingual protrusion may reflect the tongue’s ability to reach the substrates to be sampled. Deep lingual forking serves the snake for scent trailing and its sampling ability (Cooper, 1997). The ability to simultaneously sample two points of a chemical scent trail is the basis for the assessment of the course of the trail. If the scent molecules are denser on one tip of the tongue, the snake knows that the trail goes in that direction. For that reason, forked tongues are associated with a wide searching mode of foraging. The tongue is a delivery system for the chemosensors located inside the snout, the vomeronasal organs. When sampling, the tongue is rapidly protruded, sometimes oscillated, and then retracted into the mouth. The tongue tip often contacts the ground or an object during sampling chemicals from the environment (Schwenk, 1994).

Many laboratory tests have been done concerning the olfaction and trailing behavior of snakes (Smith, Kardong, and Lavín-Murcio, 2000; Smith, Bevelander, and Kardong, 2005; Shine et al. 2005; Ford, 1981). One parameter often used in such scent experiments is the rate of tongue flicks (RTF). This parameter either measures the snake’s relative interest in an object or indicates chemosensory searching.

High RTF’s in a novel environment or situation can also be interpreted as exploratory behavior (Chiszar et al. 1980). Thus, when a new object is introduced into a snake’s enclosure, the relative interest of a snake in that object can be read through its amount of tongue flicking during a certain time.

When rattlesnakes strike, they typically bite, inject venom in prey, and release. The snake usually waits after the strike. The prey moves away but soon dies from the venom. Soon the snake begins to track (indicated by high RTF) the envenomated prey by its olfactory trail; this behavior is labeled as Strike-induced Chemosensory Searching (Chiszar and Radcliffe, 1976; Chiszar, Radcliffe, and Scudder, 1977; Chiszar, Radcliffe, and Smith, 1978).

5.2.3. Scent experiment: Methods and procedures

The experiments conducted tried to investigate whether the level of interest and investigation in an object increases as the object increases in resemblance to a food item (Ethogram and Methodology: Appendix: III.a und III.b). Could these novel objects be considered enrichment?

The Eastern Diamondback Rattlesnake at the National Zoo is kept in an enclosure by itself due to its solitary lifestyle. Its home cage was also used for the experiments. To count the tongue flicks, a counter clicker was used in combination with a stopwatch.

Experiment 1: Introduction of different items into snake enclosure:

Before introducing a new object in the enclosure of the rattlesnake, the animal was observed for 5 minutes. The RTFs recorded during this period were generally low and were considered as the undisturbed baseline RTF. The snake was tested in three different trials, each with a different object introduced. For consistency, the trials were conducted on separate days although at the same time of day. The objects were sealed in an opaque plastic box perforated with small holes. A new, but identical, plastic box was used for every object so that the odor of other objects would not interfere with the results. Each trial lasted 20 minutes after the introduction of the new object. The trial objects were 1) clean mouse bedding, 2) dirty mouse bedding, and 3) a dead rat.

Experiment 2: Envenomated prey

A fourth trial was conducted to determine the difference between exploratory behavior and predatory behavior (i.e., Strike-induced Chemosensory Searching). The experiment was constructed like a regular feeding procedure where a dead rodent was offered to the snake with tongs. As in the first three experiments, the trial was preceded by a 5 minutes baseline observation period, and the trial lasted 20 minutes thereafter. At the beginning of the trial, the snake was allowed to strike a dead prey item (rat) and then the envenomated prey was placed in the enclosure, hidden as much as possible from the snake.

It was expected that the RTF as an investigatory behavior would vary between items. Presumably that the olfactory enrichment conditions, i.e., scented items in box, would result in higher RTFs than in the baseline condition, and it was predicted that the RTF would increase from each experimental introduction from clean mouse bedding in a box to the actual food item (dead rat).

5.2.4. Data analysis and resultsExperiment I: Introduction of different items into snake enclosure:

According to the RTFs measured in the Experiment I, it was apparent that the box with the dead rat (\bar{O} 26.15 RTF) produced the highest search activity of the Eastern Diamondback Rattlesnake (e.g. Fig. 22), followed by the dirty mouse bedding (\bar{O} 12.35 RTF). The box filled with clean mouse bedding (\bar{O} 3.75) did not cause a major reaction and was disregarded by the snake almost completely (Analysis: Appendix: III.c).

Experiment 2: Envenomated prey

It was expected that the RTF of this trial would be the highest rate of all four experiments, but with \bar{O} 10.7 RTF (the time taken out of the calculations for when the snake devoured its prey), the average RTF was relatively low compared to the other trials. The first two minutes after striking the prey, the tongue flicks were extremely low (Fig. 23). In the third minute though, the RTF rocketed upwards right before the snake found its prey (\bar{O} 52/min). That was one of the highest RTFs observed in any of the intervals (Compare: Appendix: III.c).

In the first experiment, the enrichment items closest to a prey item raised the interest of the Eastern Diamondback Rattlesnake the most. In comparison to a regular feeding (experiment II: envenomated prey), the RTF was noticeably higher for the trial with the rat in a box and the trial with dirty mouse bedding in a box.

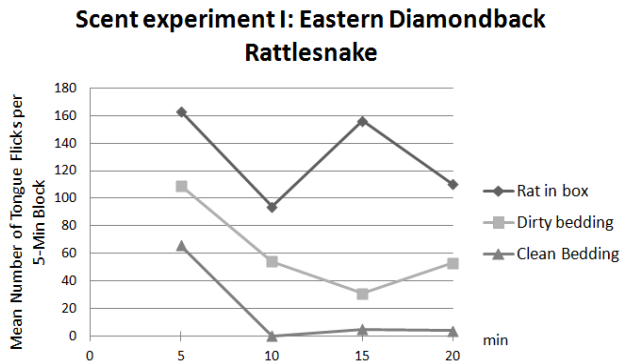


Fig. 22. Mean rate of tongue flicking (RTF) over five intervals of 5 min duration. The first block represents the baseline levels. The other four blocks represent the 20 min trials divided into four 5 minute phases.

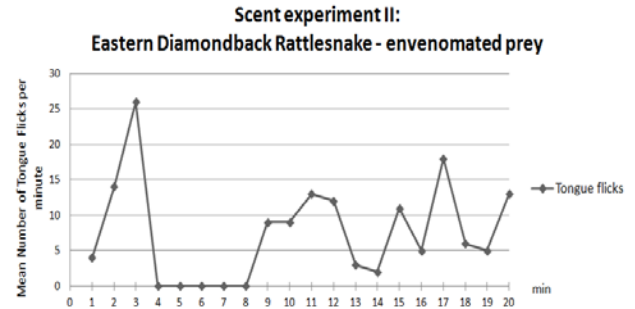


Fig. 23. Mean rate of tongue flicking (RTF) over 21 intervals of 1 min duration. The snake was devouring the prey in min 2:30- 8:29.

5.2.5. Discussion

From these results it can be concluded that the enrichment items closest to a prey stimulated the highest level of exploratory behavior. Trials with prey odor generated the highest interest and for an extended period of time. The envenomated prey trial kept the snake busy only for a short amount of time. One explanation is that because it was constructed like a regular feeding, the snake behaved in the same manner as in normal feeding and did not remain stimulated long.

The experiments showed that providing novel items (here called enrichment) can have an invigorating effect compared to a regular feeding procedure. The snake was actively exploring the boxed, prey scented items for an extended period of time. A regular feeding only invoked a short arousal and little exploratory behavior as well. When providing olfactory enrichment for Eastern Diamondback Rattlesnakes, it is therefore recommendable to use prey scented items over other items.

5.3. Komodo Dragon - *Varanus komodoensis* (Ouwens 1912)

5.3.1. Species description

As its name indicates, Komodo Dragons lives on the Indonesian island of Komodo, and they occur also on the neighboring islands Rinca, Padar, Gili, Motang, Gili Dasami and Flores (Fig. 24) (Sprackland, 2010). The Komodo Dragon lives in savannas, dry or moist tropical deciduous monsoon forests, and semi-evergreen forests, from sea level to about 800m (Sastrawan and Ciofi, 2002).

Komodo Dragons are the largest living species of lizards. They are also known as the “Komodo Monitor” and, in their native Indonesia, as the “Ora” (Connors, 2001). Adult males can grow up to 3m in total length and weigh up to 90 kg. The females of the species are usually smaller, measuring less than 2m and weighing less than 50kg (Walsh, Visser, and Lewis, 2003). The largest verified dragon was 3.13 meters and was claimed to weigh 166 kg (Ciofi, 1999a). The reason for their abnormally enormous body size has been attributed to island gigantism, since there are no other carnivorous animals on their home islands (Burness, Diamond, and Flannery, 2001). Less competition for food and territory allowed the species to develop into the colossal lizards they are today. This competitive release may therefore explain the gigantic body size of the island dwelling reptiles (Jessop, et al., 2006). Only little is known of the life expectancy of free ranging Komodo Dragons, but captive lizards can live over 20 years (Auffenberg, 1981). The longest known life span for a captive Komodo Dragon was 24 years at the Taronga Zoo in Sidney, Australia (Bennett, 1995). The body color of adults

is uniform colored and is generally earthen red to slate gray or black (Fig. 25), but it can vary in different populations (Walsh, Visser, and Lewis, 2003).

The Komodo Dragons' diet is influenced by its body size. Smaller, juvenile feed primarily on insects and small lizards (Auffenberg, 1981). They spend most of their time above ground in trees to escape larger dragons and other predators (Burghardt, et al., 2002). Cannibalism of smaller conspecifics by larger animals is not unusual (Auffenberg, 1981; Ciofi, 1999a). As they grow larger, dragons start feeding on rodents and birds, which are mainly captured on the ground. Large adult dragons are too large to climb. They mainly feed on deer, other large vertebrates, or carrion. They, ambush large prey items by waiting for them to come close enough to strike (Auffenberg, 1981), but like most other monitors, also forage widely for smaller prey items. They can track prey from as much as four kilometers away (Ciofi, 1999a) and under favorable conditions they will be able to smell a carcass from 11 km (Burghardt, et al., 2002). Their amazing sense of smell is enhanced by their long, yellow forked tongues and their wide searching mode of foraging (Schwenk, 1994). Not only do they have forked tongues like snakes, and recent studies indicate that they are also venomous (Fry, et al., 2009). Further, there are at least seven kinds of highly septic bacteria in their saliva (Ciofi, 1999a).

Komodo Dragons belong to the family Varanidae, lizards that use both chemical and tactile signals for communication. Since all varanids are solitary hunters, communication between Komodo Dragons is rare. The only times they socialize with other conspecifics is while congregating around carrion (King, Pianka, and Green, 2002), or during the initial stages of courtship. At this time, male Komodo Dragons tongue flick females at various positions along the body (Zug, Vitt, and Caldwell, 2001; Ciofi, 1999a; Pianka and Vitt, 2003), thereby communicating sexual motivation toward the females (Burghardt, et al., 2002). Their sense of hearing and vision is less developed than their olfactory sense.

They hear only in a restricted range, probably between about 400 and 2,000 hertz (humans hear frequencies between 20 and 20,000 hertz). Monitors like the Komodo Dragon can see up to 300 meters, so vision does play a role in their lifestyle, but they have poor vision in dim light (Ciofi, 1999a). Monitor lizards are considered the most intelligent lizards and some Komodo dragons in captivity are known to be eager for social play, similar to domestic dogs and cats (Burghardt et al., 2002).

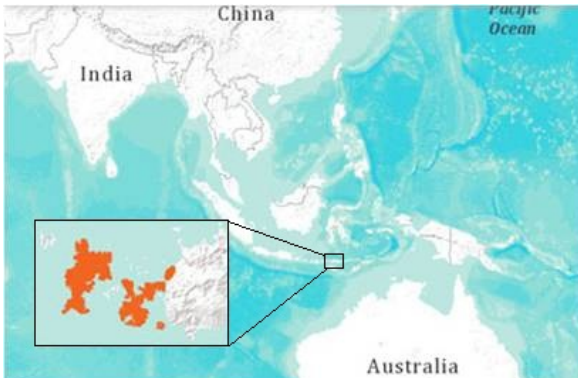


Fig. 24. Distribution of the Komodo Dragon.



Fig. 25. Komodo Dragon at the National Zoo, Washington D.C. Photograph courtesy of Smithsonian's National Zoo, Meghan Murphy

The Komodo Dragon is classified as *vulnerable* by the IUCN (World Conservation Monitoring Centre, 1996). The populations are threatened through habitat loss and the poaching of deer, their main prey. These factors have substantially reduced Komodo Dragon distribution on the islands in the last few decades. It is estimated that fewer than 3500 dragons live within their home range (Ciofi, 1999a). Much attention has focused in the past decade on the role that genetics plays in the formation of management strategies in conservation. In small populations, such as in the Komodo Dragon populations, it is important to avoid inbreeding (Ciofi et al. 1999b) by using unrelated individuals. An international studbook is being maintained to aid the proper genetic management of the captive population. The studbook contains data from 64 zoos maintaining this taxon in order to ensure a diverse gene pool (Conners, 2001).

5.3.2. Olfaction and trailing behavior

Monitor lizards and snakes have common ancestry and are all members of the order Squamata, hence share many evolutionary features. Monitor lizards such as the Komodo Dragon have forked tongues like snakes. Their tongues are used as a chemosensory edge detector to follow pheromone trails of potential mates and prey (Schwenk, 1995). The mechanism monitor lizards use for chemosensory detection is the same that snakes use. Through tongue flicking, odor molecules are picked up from the air and delivered to the vomeronasal organ or Jacobson's organ. A higher concentration of molecules present on one tongue tip rather than on the other tells the lizard to approach its prey or mate from that side (Ciofi, 1999a). This way the giant lizards are able to track others over long distances. Similar to snakes that stop and swing their head from side to side when they lose a pheromone trail within two tongue flicks (Schwenk, 1994), Komodo Dragons walk in an undulating manner, swinging their head from side to side (Ciofi, 1999a). This behavior ensures that an individual stay on a scent trail without periodic stop.

5.3.3. Scent trail experiment: Methods and procedures

The experiment conducted tried to investigate whether the RTF of the Komodo Dragon would increase in the immediate proximity to a hidden prey item (Ethogram and Methodology: Appendix: IV.a und IV.b). The Komodo Dragon at the National Zoo is kept in an enclosure with a dirt substrate; the enclosure can be separated into two parts with a guillotine door controlled from outside. While the Komodo Dragon was kept in one room of his enclosure, a dead rat was buried in the other room behind the closed guillotine door. A scent trail was created that led from the door to the prey. The monitor was then released into the enclosure.

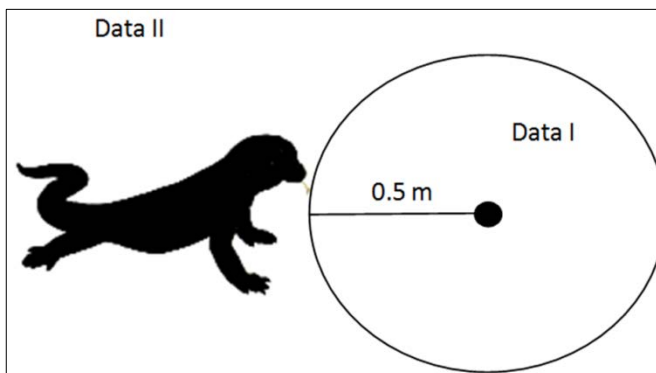


Fig. 26. Division of the results into two different sets of data. Data I: represents all the data points that were noted while the Komodo Dragon was in a range of 0.5 meters of the prey. Data II: represents all the other data points when the animal was further away than 0.5 meters

Because several trials were conducted consecutively, it was decided to forgo a baseline observation. The trials lasted until the animal found the hidden prey and the RTF was counted for every 10 second interval. There were a total of 15 trials. A counter clicker was used in combination with a stopwatch to count the tongue flicks, and a second person recorded the results.

It was expected that the tongue flicks would increase as the lizard approached the buried prey. It was assumed that more scent molecules would be present in close proximity to the prey (in a radius of about 0.5 meters), and their presence would initiate an increase in the tongue flicking rate.

5.3.4. Data analysis and results

The data were divided into two blocks. The first block, Data I, was recorded when the Komodo Dragon was in close proximity to the prey (within a radius of about 0.5 meters from the prey to the Komodo Dragon's snout). The second block, Data II, was recorded when the dragon was outside of that range (e.g. see Fig. 26). The 10 second interval in which the rat was found was excluded from the calculation.

When comparing the average RTFs of Data I (Ø 14.37 tongue flicks per interval) with Data II (Ø 8.92 tongue flicks per interval), it is obvious that close proximity to the prey item noticeably increased the tongue flick rate (Analysis: Appendix: IV.c). An observation not reflected in the data is that the extrusion of the tongue also decreased while approaching the hidden prey.

5.3.5. Discussion

The scent trail experiment clearly shows this Komodo Dragon's RTF increased greatly as it approached the prey and was in immediate proximity to it. The experiment gives insight in how the trailing behaviour is manifested. Shorter and more tongue flicks also indicate intensified investigatory behaviour. Knowing that Komodo Dragons have the ability to follow prey over several kilometres can be the basis for an olfactory-based enrichment regimen. One approach could be to construct an artificial scent trail that leads to a buried food item, similar to the one in the experiment conducted. This approach stimulates the animals to be active. Since many Komodo Dragons in captivity are overweight (J. Gerrits, personal communication 15. Feb. 2012), this feeding approach might increase an individual's physical exercise and be a mental stimulus.

5.4. Green Tree Monitor - *Varanus prasinus* (Schlegel, 1839)

5.4.1. Species description

The Green Tree Monitor, also known as the Emerald Tree Monitor, is a monitor species from New Guinea and Cape York, Australia (Fig. 27) (Pianka and Vitt, 2003; De Lisle, 1996; Losos and Greene, 1988). These arboreal lizards thrive in lowland environments up to 1500 feet above sea level including tropical evergreen forests, palm swamps and cocoa plantations (Cogger, 2000; De Lisle, 1996; Sprackland, 1991). The Green Tree Monitor (Fig. 28) is known for its unusual coloration, ranging from dark jade to lime green (De Lisle, 1996) and dark banding (Sprackland, 1989), serving as a camouflage for an arboreal lizard. It is a medium sized lizard, reaching about 75-100 cm total length with a prehensile tail (Wilson and Burnie, 2001; Cogger and Zweifel, 1992; Greene, 1986).

It is a carnivorous lizard, eating mostly insects, but sometimes also preying on small vertebrates (Pianka and Vitt, 2003; Pattiselanno, Rahuyu, and Wanaggai, 2007; Losos and Greene, 1988). In captivity they will also accept fruit like bananas, melons and berries (Sprackland, 1991). It is widely believed amongst herpetologists that they are deaf, however they have excellent eyesight for stalking their prey (Sprackland, 1991). Like the Komodo Dragon, the Green Tree Monitor relies primarily on its excellent sense of smell when foraging. Their long tongue operates just like a snake's tongue and is for vomeronasal olfaction.



Fig. 27. Green Tree Monitors. Photograph by Sarah Kuppert.



Fig. 28. Distribution of the Green Tree Monitor.

The Green Tree Monitor's conservation status is not assessed by the IUCN, but the lizards are protected in their native Indonesia (Iskandar and Erdelen, 2006). The species is also listed in the Convention on International Trade in Endangered Species (CITES) trade database in APPENDIX II. "Appendix II lists species that are not necessarily now threatened with extinction but that may become so unless trade is closely controlled" (CITES, 2012).

Between 1975 and 2005 over 7,000 Green Tree Monitors were exported and sold in international trade as pets. Intensive collection of wild specimens likely will significantly impact the future of Green Tree Monitor populations (Pernetta, 2009).

5.4.2. Puzzles as enrichment

Puzzles are creative ways of presenting food to captive animals (Hernandez-Divers, 2006). These brainteasers are ideal for groups. Monitor lizards, *Varanus*, are known to possess considerable intelligence in comparison to other reptile Mendyk and Horn noted that "extractive foraging, the location and retrieval of food items from embedded matrices is rarely performed by non-avian reptiles, limited mostly to monitor lizards" (p. 345). Their cleverness combined with their fine motor coordination and skilled forelimb movements make Green Tree Monitors predestined for solving complex puzzles.

5.4.3. Olfactory enrichment: Skilled forelimb extraction

One way to construct a puzzle for enrichment purposes for this species is to drill holes in a thick branch. The openings should be small and long enough so that the monitors cannot stick their snouts into an opening. But they must be large enough for them to reach inside with their forelimbs to extract food. It is important that the holes are drilled neatly, so that the lizards cannot injure themselves. The lizards will be able to smell the food, but not necessarily see it. Once the branch is stocked with food, such as neonatal mice, the animals is allowed to forage.

Observation:

Once the prey item was detected by smell or by sight, both lizard tongue-flicked around the opening to investigate its content, then stuck their snouts in the opening, only to discovered that the food was out of reach. After the unsuccessful attempt with their snouts, they reached into the hole with a forelimb while maintaining eye contact with the food. The prey was hooked with the claws,

extracted, and consumed. Extraction of food items out of a small opening requires highly coordinated movements of the wrist and the digits.

5.4.4. Discussion

To solve complex puzzles, the Green Tree Monitors used their learning skills combined with natural instincts to secure food. Since these lizards had used the same technique before, they also demonstrated their ability to learn and memorize patterns. When presenting food in the form of puzzles, the animals have to actively forage, using their senses. They should devise diverse problem solving strategies to get to their meals. This kind of enrichment is certainly not appropriate for every species because it requires the target species to possess certain physical and intellectual abilities.

6. Conclusion

The massive decline in the biodiversity of our planet has forced people to protect the unique multiplicity of the planet's species. Zoos are nowadays major players in the field of conservation biology and they contribute to the preservation of numerous species. While keeping wild animals in captivity can be challenging because each species is unique and has unique requirements, many approaches have been developed in recent years to improve the handling and the physical and mental welfare of zoo animals. While the enclosures and diets must be selected in consideration of the needs of captive animals, their mental and physical demands must not be neglected.

Ectothermic reptiles and amphibians are a very diverse group of animals, and much unlike endothermic birds and mammals. Though reptiles and amphibians differ from endotherms in lifestyle, they also profit from mental and physical stimulation, such as *environmental enrichment*. Few generalizations can be made of what type of enrichment is suitable for animals. Therefore, to find appropriate stimuli for a target species it is vital to study its natural history and it is especially vital to study its communication channels.

Observations and experiments can improve the understanding of how certain species use communication channels. The meaning and medium of communication between animals is not always clear. By observing the animals closely, humans can make sense of, for example, the postural sequences of Veiled Chameleons.

However, knowing how one species uses a communication method through a certain communication channel cannot replace the close study of related species. Green Crested Basilisks and Grand Cayman Iguanas, for example, both use head bob displays to communicate. The bobs look much different for the two species. In combination with other displays a head bob might have a completely different meaning from one species to the next, reaffirming the necessity of closely studying communication channels before even considering enrichment options.

Experiments and observations can also offer great insight into how a communication channel works and details of how it is used. The experiments conducted with Eastern Hellbenders in the context of this thesis gave an idea of the keenness of their olfactory abilities. This insight may help determine the ideal amount of aromatic for potential scent related enrichment regimens.

The more details known about the communication channels used by a target species, the more the enrichment efforts can be adapted to species specific needs and abilities. Knowledge about the olfactory dependence of monitor lizards will help identifying suitable enrichment regimes for these animals. The long-distance foraging abilities of Komodo Dragons may serve as the basis for an enrichment regimen involving a scent trail. Not only will that kind of enrichment stimulate the animals mentally, but provides physical exercise, which can help prevent weight problems. For the Green Tree Monitors, it is possible to offer scent-based puzzles. The message here is clear, being aware of their

skilled forelimb movements, enrichment options focusing on that behavioral skill can be developed. Every species has different skills and abilities that can be incorporated in the enrichment regimens.

Not only can knowledge about communication channels help identify suitable enrichment options, but it can also help to choose the ideal items and substances for enrichment purposes. By comparing different enrichment options through scent experiments, preferences of one enrichment item over another can be measured. Measuring an Eastern Diamondback Rattlesnake's rate of tongue flicks when it is provided different options of scent related enrichment items can help identify the snake's preferences, and consequently the most effective enrichment item.

It is especially important to know an animal's way of communicating when using training as a method of enrichment. The animal keeper has to be able to communicate his or her intentions to the targeted species. For example, knowing that crocodylians use visual signals in interspecific communication is paramount in choosing the appropriate visual communication channel for use in training Cuban Crocodiles. Training as a form of enrichment does not only offer mental and physical activity for the animals, but it also simplifies daily contact with these potentially dangerous animals and reduces stress during medical checkups.

The importance of detailed knowledge about an animal's communication channels for enrichment purposes is indisputable. This knowledge can certainly help develop and improve enrichment regimens by focusing on the inherent abilities and skills of different species of reptiles and amphibians. It is vital to make conscious use of this knowledge about communication channels and to further research their details. By raising the welfare of captive animals through successfully implementing enrichment programs, zoos make an important contribution to conservation biology. Furthermore, being concerned with the specifics of non-human communication, the research of animal communication is certainly a valuable contribution to the field of linguistics. This kind of interdisciplinary research will equally help develop the field of linguistics as well as the field of conservation biology. The collaboration of two different fields of study is advantageous for the integral examination of a topic, combining knowledge and approaches of two different backgrounds.

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Appendices

I. Veiled Chameleon

a. Ethogram

(After Kelso & Verell, 2002)

Male Behavior Patterns: 'Non-Courtship' behavior patterns

1. Brighten: The male displays more intense coloration, including increased contrast between light and dark markings.
2. Lateral compression: The male compresses his body laterally while extending his gular (throat) area and tightly curling his tail (this position is held for 5 s).
3. Approach: Walks in the direction of the female.

Female Behavior Patterns: Behavior patterns of non-receptive females

1. Darken: The female displays dark background coloration with bright orange and blue markings.
2. Threaten: The female exhibits lateral compression raises her occipital flaps (lateral margins of the head) and gapes her mouth towards the male.
3. Rock: The female sways from side to side. Also referred to as 'crazy dance' by Cuadrado (2000).
4. Bite: The female attempts to bite the male.

II. Eastern Hellbender

a. Ethogram

Behaviors:

- Resting: (No to minor activity) At least one individual shows no movement. When animals are adjusting inside a cave, it is not counted towards any type of movement.
- Alert: (Little activity) At least individual is moving one or more body parts, but generally stays in the same spot.
- Crawling: (Active) At least one individual is moving in a forward motion, stepping one leg in front of the other. Climbing up the sides of the tanks is also considered as crawling as long as there is no wiggling of the tail visible.
- Swimming: (Very active) At least one individual has all 4 limbs elevated from the ground and the walls of the aquarium using tail as main drive, wiggling it from side to side.

Other behaviors (can occur simultaneously with other behaviors):

- Engulfing: (E) Individual is manipulating food in its mouth.
- Yawning: (Y) Individual is opening mouth wide to a yawn.
- Shedding: (S) Individual is shedding its skin and actively trying to remove it.

b. Methodology

- Research question: At what dilution will the animals stop reacting to the introduced extract?
Sampling method: Focal sampling: Observe one animal for a specified amount of time and record all instances of behavior.

- Recording method:

One-zero: The described behaviors occur or not during each one minute interval.

- Recording interval:

1 minute intervals: Noting all behaviors seen in a 1 minute period.

- Enrichment Phases:

Baseline: No new alterations before observation

Olfactory: Earth worm extract

Additional Comments:

- 1 drop of extract or extract diluted with water will be put in the sump. The time from when the drop touches the water to when a first reaction is seen in the test tank, Tank 1, is recorded.
- If no reaction is visible, it is noted as "no reaction". The assumption for that case is that the dilution is too high and the hellbenders cannot smell it.
- As long as at least one animal in the group is moving, it will be considered as foraging behavior.
- When all animals returned to a resting state for 3 minutes in a row, the foraging time is considered to be over and the post-baseline will be started.

- If an animal is active during the pre-base line, it will be automatically excluded from the observation, since it is not possible to discriminate between actual foraging behavior and general activity.
- If animals show no reaction after 5 minutes into the experiment, the assumption is that the dilution is too high for the animals to smell the extract.

c. Analysis

Table II-I. Extract dilutions and calculation of reaction time and foraging time.

Extract dilution	Dilution ratios Total=extract + water	Reaction Time (minutes)	Foraging g (hours)	Date
1. 100%	Only extract	0:27	0:57	05.12.2011
2. 50% (1:1)	0.2 ml = 0.1 + 0.1	0:32	0:26	07.12.2011
3. 25% (1:3)	0.2 ml = 0.05 + 0.15	0:36	0:29	09.12.2011
4. 20% (1:4)	1 ml = 0.05 + 0.20	0:40	1:18	21.12.2011
5. 1% (1:99)	5ml = 0.05 + 4.95	No reaction	-	30.12.2011
6. 10% (1:9)	0.5ml = 0.05 + 0.45	No reaction	-	03.01.2011
7. 15% (3:17)	0.5ml = 0.75 + 0.425	0:36	0:24	04.01.2012
8. 12.5% (1:7)	0.8ml = 0.1 + 0.7	0:31	0:20	06.01.2012
9. 11.1% (1:8)	0.9 ml = 0.1 + 0.8	0:50	0:31	09.01.2012
10. 10% (1:9) retest	0.5ml = 0.05 + 0.45	0:46	0:08	11.01.2012
11. 9.09% (1:10)	0.55ml = 0.05 + 0.5	0:32	0:01	13.01.2012
12. 8.33% (1:11)	0.6 ml = 0.05 + 0.55	1:30	0:03	18.01.2012
13. 7.69% (1:12)	0.65ml = 0.05 + 0.6	0:44	0:07	20.01.2012
14. 5% (1:19)	1 ml = 0.05 + 0.95	0:47	0:30	23.01.2012
15. 2.5% (1:39)	1 ml = 0.025 + 0.975	0:45	0:07	25.01.2012
16. 2% (1:49)	1 ml = 0.02 + 0.98	0:38	0:09	30.01.2012
17. 1% (1:99) retest	5ml = 0.05 + 4.95	No reaction	-	01.02.2012
18. 2% (1:49) retest	2.5ml = 0.05 + 2.45	0:36	0:09	03.02.2012
AVERAGE		0:42	22.6	

Table II-II. Calculation of activity profiles for each trial.

Extract dilution	Resting - % of total	Alert - % of total	Crawling - % of total	Swimming - % of total	Total foraging (hrs - min)
1. 100%	57 - 100	57 - 100	47 - 82.5	15 - 3.8	0:57
2. 50% (1:1)	26 - 100	26 - 100	25 - 96.2	3 - 11.5	0:26
3. 25% (1:3)	29 - 100	25 - 86.2	21 - 72.4	9 - 31	0:29
4. 20% (1:4)	78 - 100	76 - 97.4	70 - 89.7	32 - 41	1:18 - 78
5. 1% (1:99)	-	-	-	-	-
6. 10% (1:9)	-	-	-	-	-
7. 15% (3:17)	24 - 100	24 - 100	22 - 91.7	9 - 37.5	0:24
8. 12.5% (1:7)	20 - 100	19 - 95	17 - 85	11 - 55	0:20
9. 11.1% (1:8)	31 - 100	30 - 96.8	29 - 93.5	16 - 51.6	0:31
10. 10% (1:9) retest	8 - 100	8 - 100	7 - 87.5	-	0:08
11. 9.09% (1:10)	1 - 100	1 - 100	-	-	0:01
12. 8.33% (1:11)	3 - 100	3 - 100	2 - 6.7	-	0:03
13. 7.69% (1:12)	7 - 100	7 - 100	7 - 100	-	0:07
14. 5% (1:19)	30 - 100	30 - 100	23 - 76.7	2 - 6.7	0:30
15. 2.5% (1:39)	7 - 100	7 - 100	5 - 17.1	1 - 14.3	0:07
16. 2% (1:49)	9 - 100	8 - 88.9	6 - 66.7	2 - 22.2	0:09
17. 1% (1:99) retest	-	-	-	-	-
18. 2% (1:49) retest	9 - 100	9 - 100	6 - 66.7	-	0:09
AVERAGE	100 %	97.6 %	73,7 %	27.5 %	22.6 min

III. Eastern Diamondback Rattlesnake

a) Ethogram

Behaviors:

- Tongue flicking: The tongue is rapidly protruded, sometimes oscillated, and then retracted into the mouth.

b) Methodology

- Research question: Does the level of interest and investigation in an object increase as the object increases in resemblance to a food item?
- Sampling method: Focal sampling: Observe one animal for a specified amount of time and record all instances of behavior.
- Recording method: Continuous: The aim is to provide an exact measure of behavior, measuring frequencies.
- Recording interval: 1 minute intervals: Noting all tongue flicks seen in 1 minute intervals during a 20 minute period.
- Enrichment phases:

Baseline – No new alterations before observation (5 minutes)

Olfactory: Experiment I and II

Experiment I

- Rat in Tupperware
- Dirty mouse bedding in Tupperware
- Clean Mouse bedding in Tupperware

Experiment II

- Snake strikes rat, releases and has to search for it in enclosure.

c. Analysis

Table III.I. Experiment I & II: Total of Tongue flicks and mean RTFs.

Date	Item	Σ tongue flicks	$\bar{\emptyset}$ RTF
16.03.2012	Rat in box	523	26.15
19.03.2012	Dirty bedding in box	247	12.35
20.03.2012	Clean bedding in box	75	3.75
21.03.2012	Rat	160*	10.7

* Snake ate for 5 minutes -> the $\bar{\emptyset}$ was calculated excluding this time

Table III.II. Experiment I: Calculation of mean RTFs for four 5-minute intervals.

Item	Interval 1 (0-5 min)	Interval 2 (5-10 min)	Interval 3 (10-15 min)	Interval 4 (15-20 min)
Rat in box	163	94	156	110
Dirty bedding in box	109	54	31	53
Clean bedding in box	66	0	5	4

IV. Komodo Dragon

a. Ethogram

Behaviors:

- Tongue flicking: The tongue is rapidly protruded, sometimes oscillated, and then retracted into the mouth.

b. Methodology

- Research question: Does frequency of tongue flicking increase, as the animal approaches the hidden rat?
- Sampling method: Focal sampling: Observe one animal for a specified amount of time and record all instances of behavior.
- Recording method: Continuous: The aim is to provide an exact measure of behavior, measuring frequencies.
- Recording interval: 10 second intervals: Noting all tongue flicks seen in 10 second intervals, until the Komodo Dragon finds the hidden rat.

- Enrichment phases:

Olfactory: Rat hidden and partially buried in enclosure

c. Analysis

Table. IV.I. Total of tongue flicks and mean RTFs for Data I (outside of a 0.5 meter radius of the prey) and Data II (within a 0.5 meter radius of the prey).

	Σ Tongue flicks	$\bar{\emptyset}$ RTF
Data I	1151	8.92
Data II (0.5m radius)	776	14.37

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