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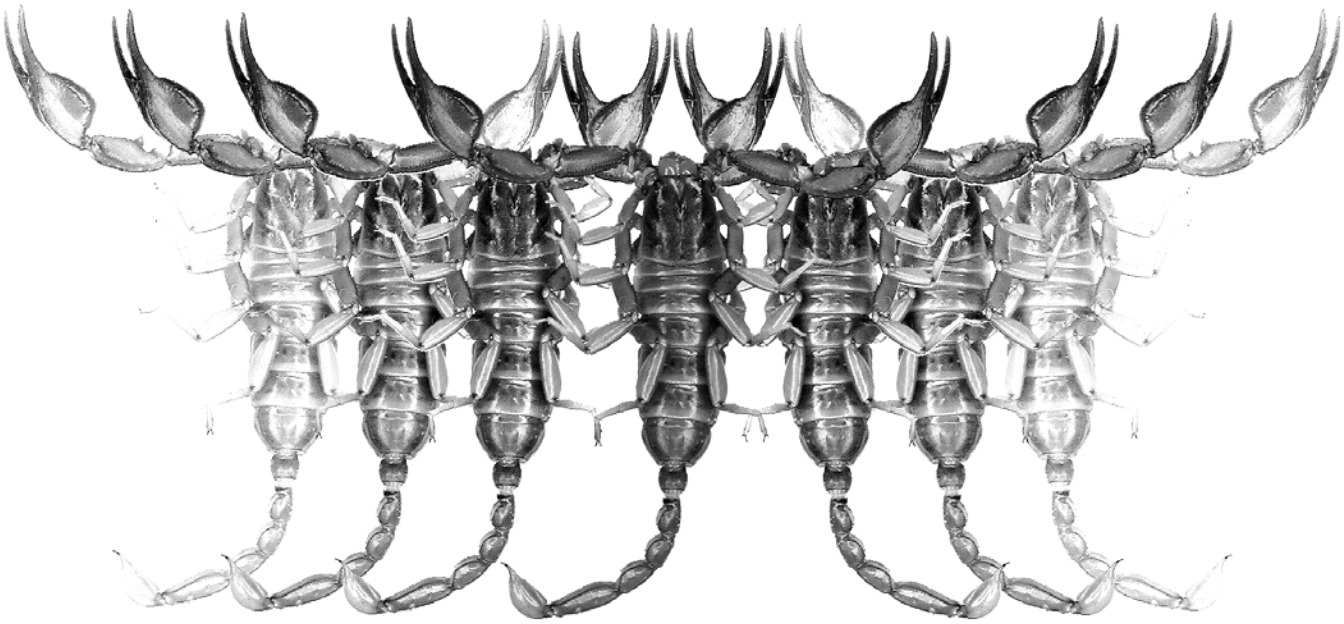

Observations on Prey-Capture Behavior of *Androctonus crassicauda* (Olivier, 1807) (Scorpiones: Buthidae) in Northern Iraq

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Summary

Modern studies of scorpion prey-capture behavior have included several genera from a variety of habitats and have demonstrated that scorpions have a limited, yet similar, repertoire of reactions towards their prey. These experiments, however, by necessity have dealt with scorpions under the ecologically artificial conditions of an indoor laboratory. The experimental design presented here included both indoor and outdoor laboratory experiments to study the prey capture in *Androctonus crassicauda* (Olivier, 1807). Thirty indoor and twenty outdoor experiments recorded scorpion activities from initial prey recognition to prey ingestion. By experimenting with this indigenous species in its harsh environment (an outdoor laboratory, which was 7° C hotter and 11% drier than the indoor laboratory), there was an 11 minute reduction in total prey-capture time and a 40% reduction in scorpion inactivity during outside prey-capture sequences. This increase in prey-capture efficiency is probably related to a negative response due increasing metabolism and desiccation stress when on/near the surface; thereby, ensuring a quicker return to the more equable burrow.

Introduction

Scorpions are nocturnal predators, which hunt and capture a variety of prey types. Commonly, prey capture occurs because of a sit-and-wait strategy; whereby, a scorpion waits for prey to wander within a few centimeters of its burrow opening before attempting its capture. First scientific publications on this behavior appeared by the end of 19th century (e.g., Pocock, 1893). Pocock's description of feeding behavior was a qualitative study of two scorpion species and their reactions to common cockroaches. Even though anecdotal in nature, Pocock's observations correspond well to later, quantitative analyses of prey-capture (Hadley & Williams, 1968; Alexander, 1972; Bub & Bowerman, 1979; Casper, 1985; Rein, 2003).

The first work on quantitative prey-capture behavior was published by Hadley & Williams (1968) who made laboratory and field-based observations of five scorpion species where common prey-capture behaviors were noted. Later, Bub & Bowerman (1979), using *Hadrurus arizonensis* Ewing (Caraboctonidae), were able to identify and discuss different prey-capture behaviors. These behaviors were compiled into a flow chart (ethogram). Most recently, Rein (2003) conducted a quantitative analysis of prey-capture behavior using two buthid species (*Parabuthus leiosoma* Ehrenberg and *P. pallidus* Pocock). The observed behavioral components were identified and a species-corrected ethogram was presented. All of these studies achieved similar results; however, Rein (2003) opted to modify and add to the behavioral-component descriptions. These modifications were made to suit his study using two buthids and to better analyze their stinging behaviors. To date, prey-capture studies have shown that scorpions, regardless of their systematic position (e.g., *Parabuthus*, *Hadrurus*) have a limited repertoire of reactions when hunting/capturing their prey.

Geographic, temporal, and weather-related restrictions have forced scorpion prey-capture studies to be completed in an indoor laboratory. This indoor laboratory, although accessible and comfortable (to the scientist), may be restrictive to generating true-to-life data. Instead of doing experiments in the indoor laboratory where, as Hadley (1990) stated "test animals are confined to conditions that are often artificial and ecologically meaningless" an outdoor laboratory should be used. There are many aspects of a natural environment that may be missing from indoor studies, such as: fluctuating temperatures, cloudiness, humidity, precipitation,

Figure 1: Location map showing a portion of the Middle East and the approximate location of the study area (red star) in northern Iraq, just north of Samarra.

and wind. In addition to these climatic factors, other factors such as photoperiod and stellar/lunar recognition (e.g., astromenotaxis) may be just as important to behavioral studies. (e.g., McReynolds, 2004). The easiest way to "replicate" these natural environmental conditions is to move the controlled setting outdoors, into the scorpion's habitat. This is exactly how a portion of this study was completed.

In this indoor- and outdoor-laboratory study, environmental conditions were recorded and analyzed along with the prey-capture sequence of one species – *Androctonus crassicauda* Olivier, 1807. Assuming that prey-capture behaviors are similar amongst scorpions, then what differences can be noted by adjusting environmental conditions? Are some aspects of the prey-capture sequence more affected by changed environmental conditions, or will they remain the same?

Methods

Species studied. *Androctonus crassicauda* (Olivier, 1807) (Buthidae) is a medically important species (e.g., Tuncer & Onur, 1996; Gaijre & Dammas, 1999), which inhabits the Palaearctic region, primarily the Middle East (Fet & Lowe, 2000). Adults of this species vary in color from light brown to reddish to black and can reach lengths greater than 10 cm. Described as a generalist desert species (Fet et al., 1998) it has been noted as anthropotolerant (Crucitti & Cicuzza, 2001) and is commonly found "in the ruins of old, neglected buildings... ." (Birula, 1917; quoted from a Nakhichevan native in modern Azerbaijan).

Materials. Specimens were collected and studied during the summer and fall of 2004, approximately 50 kilometers north of Samarra, Iraq while the author was a soldier in the midst of Operation Iraqi Freedom II (Figure 1). The specimens were located by a LPD LLC 5-LED shortwave, ultraviolet light (385nm, 4.0mW) during evening Nautical and Astronomical Twilights (i.e., when the sun was greater than 12 degrees below the horizon) in and around derelict buildings. Specimens were found roaming within one meter of outside walls, sitting in crevices or pre-made "burrows" at the wall-substratum interface, or residing, vertically, on the wall face (no more than 0.5 meters up). Pre-made "burrows" appeared to be modified crevices or the opportunistic

Figure 2: *Androctonus crassicauda* (Olivier, 1807) in active position. Scale in cm.

burrow use of removed animals. The substratum was densely packed silts and sands with areas adjacent walls being broken and fissured (generating some pre-made "burrows").

Studied specimens (e.g., Figure 2), of undetermined age, varying from 25 to 40 mms in length (pro and mesosoma; mean=30mms), were photographed, then housed individually, at differing times, in both an indoor terrarium (Hagen, Flat Faunarium (HG0458), 46x30x17 cm) (Figure 3) and an outdoor terrarium (Self-made, glass, open-cell-sponge base, ~100x30x30 cm) (Figure 4). The area and volume of the outdoor terraria were different from the indoor terraria because they were handmade from scavenged/cannibalized glass from destroyed buildings. An area-difference correction was attempted by placing a cardboard divider into the outdoor terraria prior to feeding. Both indoor and outdoor terraria were equipped with approximately 15 cms of substratum comprising locally derived silt and sand, premade "burrows," a flat cobble, a sponge for water (applied weekly) and three red-alcohol thermometers (-40 to +120^{o} F). Both indoor and outdoor laboratory environmental conditions were recorded by an Oregon Scientific wireless weather station (model WMR968) at the beginning of each feeding. The indoor laboratory environmental conditions were "controlled" by the room's insulating nature, with temperature and humidity all contingent on ambient room conditions (see Table 1) and illumination was controlled by an approximately 0.5-meter^2 northeast-facing window to the outside. Outdoor environmental conditions were ambient with the local environment (see Table 2).

The prey item used in the experiments was the American cockroach (*Periplaneta americana* Linnaeus). Cockroaches were acquired every few nights from similar locations as the scorpions. Their abundance and proximity to observed scorpions suggests they are probably a common source of food. Cockroaches used for feeding ranged from 15 to 40 mm (mean, 31 mm) and were the same length as the scorpions (see Table 1).

Experiment

This prey-capture experiment was conducted every seven days using 10 specimens, at differing times, in both indoor and outdoor laboratories. Immediately prior to prey-capture observations, environmental data was recorded (see Table 2). In order to reduce stress on the scorpion, scorpions were not translocated to an "observation terrarium" (Rein, 2003; Bub & Bowerman, 1979). Instead, direct prey-capture observations were made using alert and/or ambulatory scorpions in their home terrarium. To initiate the prey-capture data collection, one cockroach was dropped into the center of the terrarium. Data acquisition started at first recognition of prey item by the scorpion and finished upon ingestion, which was noted by cyclical movements of coxae of the first

Specimen and Prey Size	Inside (n=30)	Outside (n=20)	Δ
Specimen	30 ± 0.7	30 ± 1	0
Prey	31 ± 1	29 ± 1	-2
Specimen:Prey (length ratio)	1 to 1	1 to 1	0

Table 1: Mean sizes of scorpions and prey (mm) with standard error. Delta shows difference between the inside and outside.

Environmental Data	Inside (n=30)	Outside (n=20)	Δ
Ambient Air Temperature (^{o}C)	29 ± 0.3	36 ± 0.5	+7
Relative Humidity (%)	23 ± 0.8	12 ± 0.7	-11
Substratum Temperature (^{o}C)	28 ± 0.3	36 ± 0.3	+8

Table 2: Environmental data taken at the beginning of prey-capture observations for both indoor and outdoor study areas with standard error. Delta shows difference between inside and outside data.

legs. Observations were made under low-intensity red light with one feeding being captured with a Sony camcorder in infrared, Nightshot® mode; neither light source seemed to affect the scorpion's behavior (Machan, 1968; Blass & Gaffin, 2005).

Results and Discussion

Prey-capture sequence

A total of 50 feedings from 10 specimens were recorded. Thirty of these observations (mean = 3 per specimen) were completed in an indoor laboratory, while the remaining were completed in an outdoor laboratory. Atmospheric conditions were significantly different between observation locations. Mean atmospheric data compiled at beginning of feeding is listed in Table 2. The significant variables were the temperature and relative humidity, where the outdoor observations were, on average, 7^{o}C hotter and relative humidity was 11% lower (see Table 2).

The total prey-capture time is the temporal sum of the steps in the flow chart (Figure 5 and Table 3) used by the scorpion. For example, the quicker capture sequences involved orienting toward the prey, grasping and stinging successfully, then manipulating the prey (cephalon first) and locomotion to a burrow for ingestion. In opposite, the slower capture sequences involved employing the above phases multiple times with an addition of inactive periods (sometimes greater than ten minutes). Table 4 shows the more affected phases in the experiments by comparing the mean phase use between inside and outside experiments. In order to test whether the phases (e.g., sting or manipulation) were similar or dissimilar, each phase's use was compared between indoor and outdoor experiments using the t-test of paired means (indoor/outdoor) assuming equal variance (Hollander & Wolfe, 1973; Donnelly, 2004). The hypothesis statement for these tests is: H_o:$\mu_1=\mu_2$ and H_1:$\mu_1\neq\mu_2$; where μ_1 = indoor phase and μ_2 = outdoor phase. Of all the paired tests of the same indoor and outdoor phase, only one was significantly different – the Inactive Phase.

The null hypothesis of the Inactive Phase ($\rho=<0.05$) was rejected. During each feeding in an indoor laboratory, specimens tend to enter this phase 1.8 times (mean); during outdoor feedings they only use it 1.1 times (mean). This divergence suggests that while feeding under real (outdoor) environmental conditions, the scorpion enters this phase 61% as often, compared with the indoor setting. No other phases were significantly different between the indoor and outdoor-type laboratory experiments. Marginal, but not significant phases were Sting, Active, and Locomotion (see Table 4). Interestingly, however, both the Passive Phase and the Cleaning Phase (used by Rein, 2003; "sand thrust" in Bub & Bowerman, 1979) were not observed to be a part of this species' feeding strategy. The lack of Passive Phase was a result of experimental design; feeding scorpions only when active on the surface or in a sit-and-wait posture at their burrow entrances in their home terrarium. The

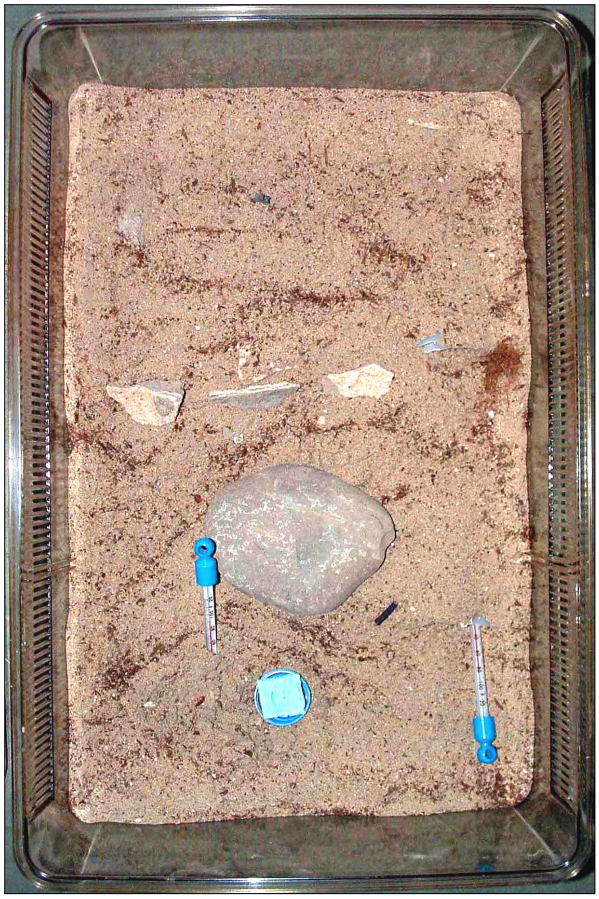

Figure 3: Example of inside terrarium showing typical set-up such as the thermometers, water dish and cobble; approximate dimensions are 50 x 25 cm.

Cleaning Phase has been observed by Rein (pers. comm.), in more than 50% of his prey-capture observations, involving over 30 species and 100's of feedings. This study, however, did not recognize a Cleaning Phase, and may be a result of a misinterpretation of scorpion activity during prey-capture observations or a combination of venom toxicity (killing/subduing the prey quickly) and lack of body fluid eruptions during capture (Rein, pers. comm.).

Another t-test of paired means was applied to the total prey-capture times for indoor against outdoor data (see Table 4); where the difference between feeding locations was statistically significant (ρ=<<0.05). Because of environmental differences, the hotter, dryer outdoor-type laboratory produced an 11 minute reduction in the prey-capture sequence.

Environmental effects

The two significant differences between the indoor and outdoor data sets are the overall prey-capture sequence timing and the use of the Inactive Phase, both of which seem to derive from one facet of the scorpion's life cycle – metabolism. The metabolic activity of scorpions is the lowest for all arthropods, except ticks (Brownell & Polis, 2001), and is largely a function of substratum and ambient air temperatures and humidity (Brownell, 2001). In response to these physiological and metabolic stresses imposed by their harsh environments scorpions have a suite of autoecological traits, which are particularly suited to cope with them (Polis, 2001). With mean ambient air temperatures for the outdoor laboratory, during feeding, being 36^{o}C and relative humidity being 12%, scorpions may undergo both a vapor-pressure deficit and desiccation stress if on the surface or at burrow entrance for an extended period of time. Together, these stresses may alter the scorpion's physiology. Riddle (1979) and Lighton et al. (2001) have shown changes in metabolic activity with relation to experimental thermal changes. For example, using the mean ambient air temperature for outdoor feedings and assuming relevance to scorpions in general, the data of Lighton et al. (2001) would suggest a two- to three-fold increase in metabolic rate (from approximately 300μW to 800μW; from their fig. 2, p. 610) if the scorpion allowed itself to equilibrate with this stressful environment. A sensation of increasing metabolism and desiccation stresses, associated with an increase in observed temperature and decrease in humidity, may help explain the differences in prey-capture behavior for *Androctonus crassicauda*.

For example, the Inactive Phase is the only prey-capture phase whereby it is hypothesized that the scorpion is able to recuperate from the energy expenditure of the Sting, Locomotion and/or Grasping phases. The reduction in this phase while in the outdoor laboratory, probably based on the sensation of an increasing metabolism from its burrow-based metabolism, has a multi-fold benefit to the scorpion. One of the primary aspects of scorpion lifestyle is its low metabolism and its ability to preserve this low metabolic rate for survival. Decreasing the inactivity levels on the surface, during feeding, allows the scorpion to return to its burrow where environmental stressors are attenuated (e.g., Hadley, 1990). This environmentally pleasant burrow, moreover, adds another benefit to the scorpion—protection from predation. Scorpions live most their lives hidden away from the elements and predators only to come out to feed and mate (e.g., Lighton et al., 2001). If a scorpion can lessen its time above ground, it will increase its survivability. By making observations in the indoor laboratory, environmental effects are attenuated, with surface and burrow temperatures in equilibrium; thereby, making prey-capture more time consuming and hazardous. Primarily, indoor laboratories eliminate this increasing-metabolism signal (resultant of increased environmental stressors) and permit the scorpion to spend more time on the surface during prey capture. Without these environmental

Figure 4: Outdoor laboratory showing the four outside terraria; approximate dimensions are 100 x 100 cm, each holding pen approximately 100 x 25 cm.

stressors, scorpion prey-capture behavior studies are not accurate and do not reflect a scorpion's "reaction" to its environment.

Conclusions

Fifty prey-capture experiments, both indoors and outdoors, were conducted using *Androctonus crassicauda* in northern Iraq. By following the scorpion's prey-capture sequence three main conclusions were made:

1) Overall prey-capture times are about 50% shorter outdoors than indoors, probably because of increased environmental stresses such as increased metabolism due to higher temperatures and increased desiccation stress due it being hotter and dryer outdoors.
2) After a successful capture, scorpions, when observed outdoors, became inactive about 40% less than when observed indoors.
3) Plausible reasons for increase efficiency in the outdoor environment are the dangers of increased metabolism and desiccation stresses; thus, leading to quicker prey capture and return to an equable and safer burrow.

In order to better understand this problem, an equal number of experiments should be conducted both inside and outside, which will allow better statistical control on the experiment. Additionally, this experimental design of indoor versus outdoor can be applied to any laboratory animal where their home environmental conditions are not replicated in the indoor laboratory. For maximum reality, field studies with minimal constraints would be optimal, although they are more difficult. These more true-to-life studies will likely show significant difference between observed behaviors.

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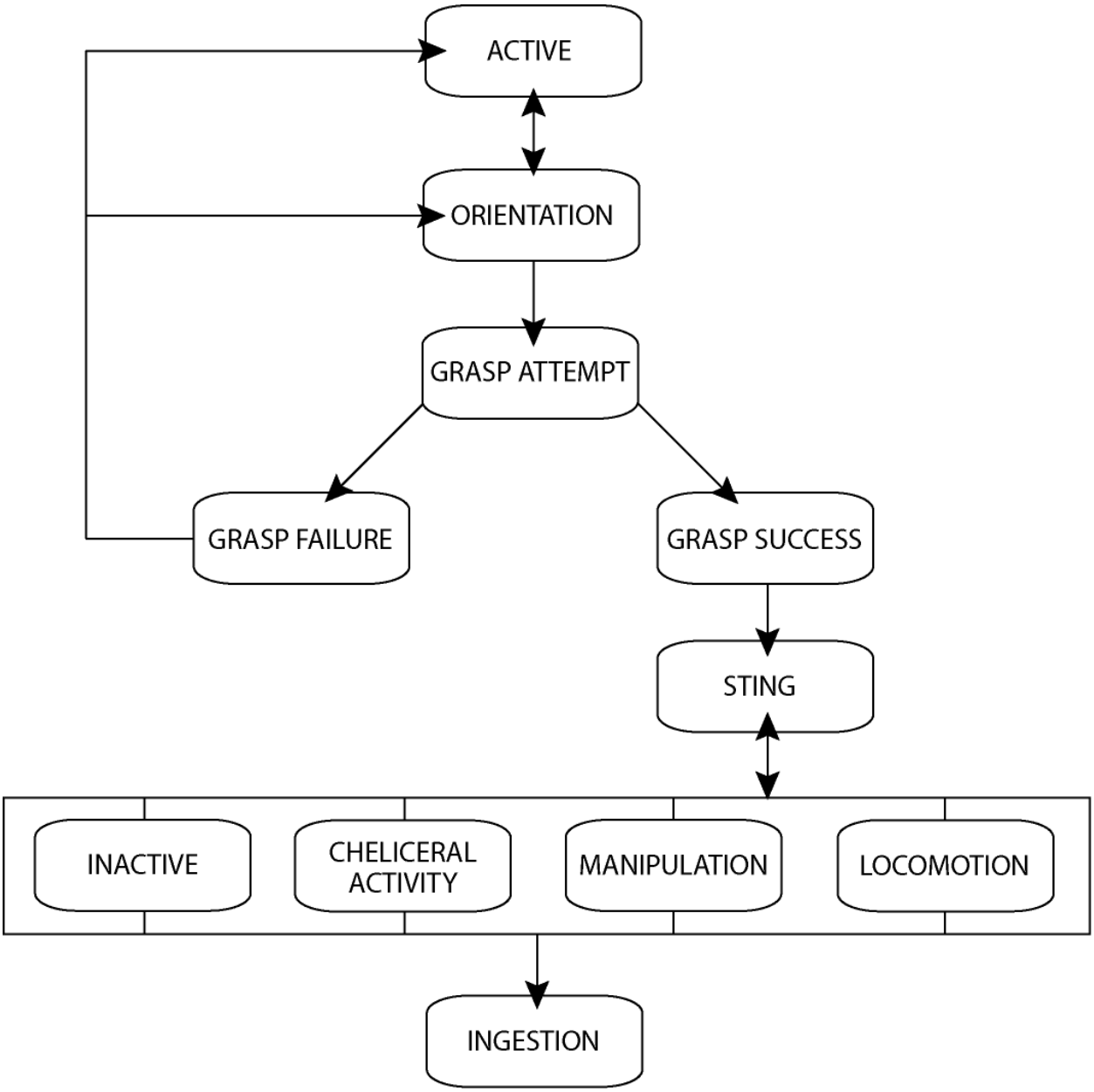

Figure 5: Flow chart (ethogram) showing the prey-capture behavior for *Androctonus crassicauda*. Prey-capture phases are explained in the Table 3. Arrows indicate the temporal flow of prey-capture phases. Encapsulation of the Inactive, Cheliceral activity, Manipulation, and Locomotion phases shows the completion of these phases subsequent to others and occurring in no particular order. Ethogram modified from Bub & Bowerman (1979) and Rein (2003).

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Prey-Capture Phase	Description
Active	Ambulation prior to feeding or a motionless state with body raised above substratum with tarsi and pectines touching substratum
Orientation	Movement of the anterior part of the scorpion towards detected prey
Grasp Attempt	Attempt to capture prey in one or both pedipalps
Grasp Failure	Unsuccessful attempt at capturing prey
Grasp Success	Successful capture of prey with one or both pedipalps
Sting	Forward movement of metasoma and telson where the aculeus probes and penetrates soft parts (lateroventral/ventral) of prey.
Inactive	Period subsequent to a successful Grasp Attempt or Sting where the scorpion rests motionless
Manipulation	Reorientation of prey by the pedipalps and/or first set of legs, sometimes assisted by chelicerae
Cheliceral Activity	The protraction (abduction) and retraction (adduction) of the cheliceral appendages
Locomotion	Ambulation of scorpion and prey throughout terrarium; usually with prey atop the scorpion in a "piggy-back" position
Ingestion	Intake of pre-digested fluidized prey, as indicated by cyclical movements of coxae of the first legs

Table 3: Prey-capture phases and their descriptions as observed during experiments (modified from Bub & Bowerman (1979) and Rein (2003)).

Significant Prey-Capture Phases	Inside (n=30)	Outside (n=20)	Δ	ρ^*
Total time (minutes)	24 ± 2	14 ± 2	-11	<<0.05
Inactive	1.8 ± 0.2	1.1 ± 0.2	-0.7	<0.05
Active	1.4 ± 0.1	1.6 ± 0.2	0.2	>0.05
Sting	1.7 ± 0.2	1.5 ± 0.1	0.2	>0.05
Travel	2.2 ± 0.2	2.6 ± 0.3	0.4	>0.05

Table 4: Mean number of times the most significant Prey-capture phases were entered during prey-capture observations. Data showing differences (delta) between inside and outside data. *t-test: Two-Sample, Assuming Equal Variances

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