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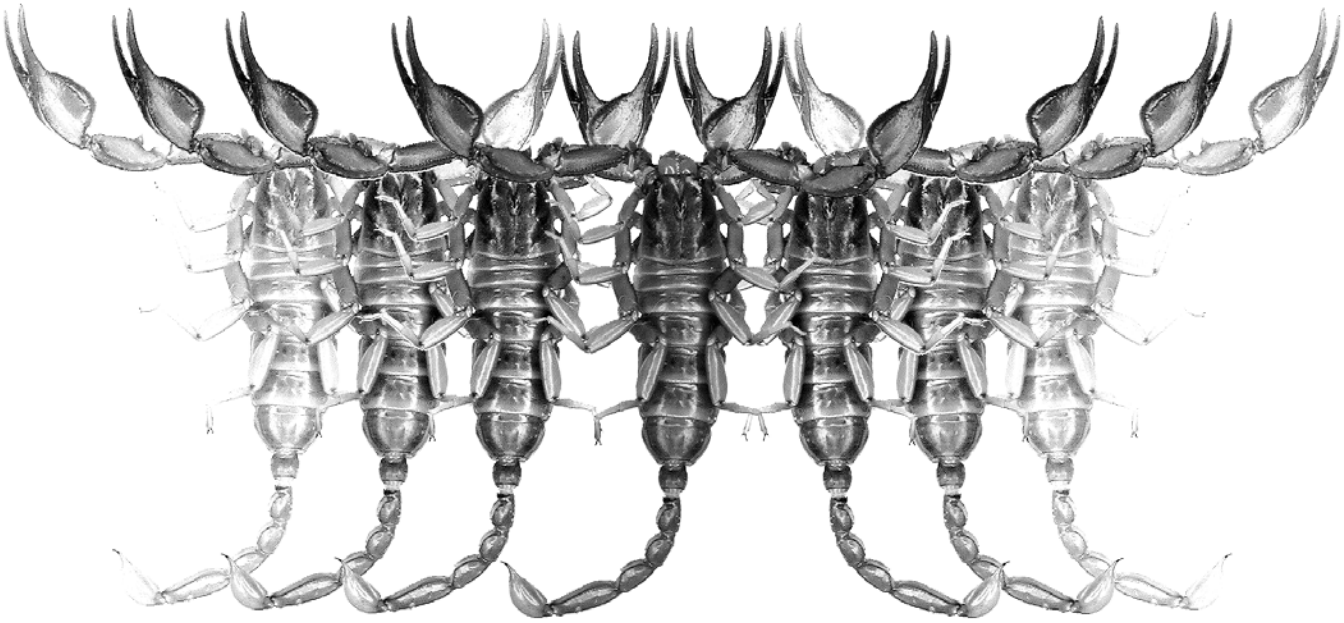
Occasional Publications in Scorpiology



Aerial Insects Avoid Fluorescing Scorpions

Carl T. Kloock

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Aerial insects avoid fluorescing scorpions

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Summary

The ecological function of scorpion fluorescence under ultraviolet light is unknown. In fact, no response of any organism to scorpion fluorescence has been documented. To determine whether or not some potential prey, specifically aerial insects, respond to scorpion fluorescence, I compared the number of aerial insects captured on sticky traps containing fluorescent scorpions to the number captured on traps containing non-fluorescent scorpions during both full and new moons. The results show that aerial insects avoid fluorescing scorpions during the full moon, when fluorescence is at its peak, but not during the new moon when it is weakest. Avoidance of fluorescing scorpions by potential prey is likely to reduce the scorpions' prey capture rate. This apparent cost of fluorescence highlights the likelihood that fluorescence has a positive function which maintains the trait in spite of this cost.

Introduction

Biologists have long used scorpion fluorescence under ultraviolet light to help them locate and study scorpions in the field (Sissom et al., 1990). Recently, several of the molecules responsible for fluorescence have been isolated (Stachel et al., 1999; Frost et al., 2001). However, there has not been much more than speculation as to the function of this trait. Lourenço & Cloudsley-Thompson (1996) and Fasel et al. (1997) speculated that fluorescence functions as a protection against ultraviolet light. Several potential ecological functions have been proposed, including detection of ultraviolet light (Hjelle, 1990), prey attraction (Lourenço & Cloudsley-Thompson, 1996; Fasel et al., 1997), and intraspecific communication (Hjelle, 1990). It is also possible that fluorescence acts as aposematic coloration. For fluorescence to attract prey, be useful as communication or repel predators, it must be detectable by organisms in the scorpion's environment. To date there is no empirical evidence that any organism responds to scorpion fluorescence under natural illumination. This is obviously a necessary first step in evaluating several of the hypotheses regarding the function of scorpion fluorescence.

Specifically, I wanted to determine whether or not insects were being lured by scorpion fluorescence. Aerial insects make ideal candidates for being susceptible to this type of fatal attraction because of their tendency to be both more mobile and more visually oriented than their ground-dwelling counterparts. Several types of flying insects are preyed upon by scorpions (McCormick & Polis, 1990), and Polis (1979) identified aerial insects as comprising 10 % of the diet of the vaejovid scorpion *Smeringurus mesaensis* (formerly *Paruroctonus mesaensis*). In this study, I set out to determine whether aerial insects respond to scorpion fluorescence under natural conditions.

Methods

The basic design of this experiment is a comparison of the number of aerial insects collected on sticky traps bearing fluorescent scorpions to the numbers on traps bearing non-fluorescent scorpions. This was done during both the full and new moons in order to study the influence of differing illumination on aerial insect responses to fluorescence. Overall nocturnal light intensity is strongly influenced by moon phase (Silberglied, 1979), and using the full and new moons provides maximum contrast in natural illumination levels.

A paired design was adopted to control for spatial, temporal and/or seasonal variation in insect availability and environmental factors such as light intensity, cloud cover, temperature, etc. Each pair consisted of one freeze dried scorpion (*Vaejovis* sp., identified using Williams, 1980) capable of fluorescing paired with a size-matched freeze dried scorpion made incapable of fluorescing by dip coating the scorpion in a clear, UV resistant varnish (McClosky Man O' War marine spar varnish, #6505). A VirTis model 24DX24 Specimen Freeze dryer was used to dry the specimens. Each scorpion treated with varnish was given two coats and observed under a 40 W fluorescent ultraviolet light to ensure that no fluorescence was visible to the human eye under high

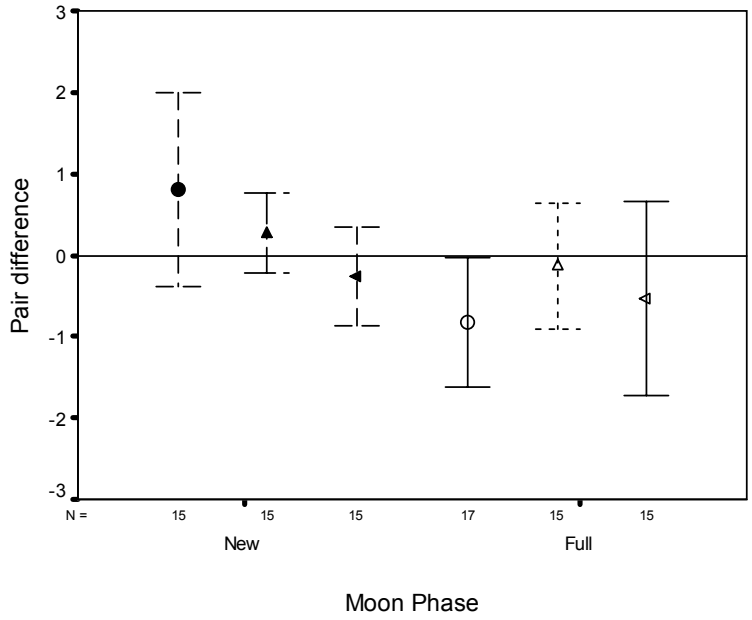

Figure 1: Mean difference (with 95% confidence intervals) between aerial insects captured on sticky traps bearing fluorescing scorpions and sticky traps bearing non-fluorescing scorpions on six nights during the summer of 2004. Values > 0 indicate that more aerial insects were captured on traps bearing fluorescing scorpions than non-fluorescing scorpions, while values <0 indicate the reverse.

UV illumination. Unfortunately, a non-UV blocking coating that was chemically similar enough to the UV blocking varnish to act as a valid control was not available, so fluorescent scorpions were left uncoated. This also has the advantage of leaving the scorpions at their maximum possible fluorescence.

Scorpion pairs were size matched using a size index derived from a principal components analysis of three size variables: carapace width, telson width, and length of the 5th metasomal segment. The first principal component of this analysis explained 94.5 % of the variation in all preserved scorpions (eigenvalue = 2.834, n=85). Scorpions were ranked according to the first principal component score and paired using this ranking. A paired t-test of the subsequent scores revealed no significant difference in size between fluorescent and non-fluorescent scorpions used in the study (t=0.660, p=0.516). Some preserved scorpions were destroyed by various means over the course of the study; these were replaced with similar-sized scorpions to maintain the original size-match of each paired sticky trap.

Scorpions were collected in the same locality where the field experiment was conducted, approximately 10 km west of the town of Buttonwillow in Kern County, California, USA. This area is typical of the natural vegetation in this part of Kern County, dominated by widely dispersed salt bush with occasional low grasses, but with significant bare areas between plants. Germano et al. (2001) provide a good general description of the region. This site is approximately 25 miles away from Bakersfield, the nearest large city. While there is undoubtedly some small amount of light pollution in the night sky, due to the city glow and light from nearby petroleum industry activities being reflected by the atmosphere, there are no lights near enough (none within at least 2 miles) to the study site to provide any direct illumination of the traps. Because of the distance to these artificial light sources, any effect they may have should be randomly dispersed across the traps, and the paired design of the study controls for any local effects.

Insect traps were made by coating pieces of black construction paper (17.4 cm x 12.3 cm) with Tanglefoot Tangle Trap liquid insect trap coating. The traps were made in this way rather than using commercially available traps to avoid the presence of fluorescent compounds often used in these traps. The traps made in this fashion did not fluoresce to the human eye when exposed to a 40 W UV light that causes scorpions to fluoresce strongly. Two traps were glued into the inside surface of a 17.5 cm x 12.4 cm plastic box: one trap was placed in the top and another in the bottom of each box, yielding a box which could be conveniently opened and closed to reveal and cover the traps as needed. Each trap was placed on the ground, and opened with the traps lying horizontally on the desert floor, facing the sky. Traps were placed far enough from the low bushes so

Source	Numerator df	Denominator df	F	Sig.
Intercept	1	86	0.470	0.495
Moon Phase	1	86	5.176	0.025
Moon Phase (Date)	4	86	1.194	0.319

Table 1: Results of a mixed model analysis of variance on the experimental pairs using trap ID as a repeated subjects variable, Moon phase as the main variable of interest (combined n= 45 on the new moon, 47 on the full moon), and date nested within moon to test for seasonal effects. The dependent variable is the difference between aerial insects captured on sticky traps bearing fluorescent scorpions and sticky traps bearing non-fluorescing scorpions on seven nights during the summer of 2004.

that they would not be shaded from moon or starlight except potentially when the moon was extremely low in the sky. Potential variation in shading between traps was controlled for by the paired design of the study. In one of the traps a non-fluorescent scorpion was placed roughly in the center of the trap surface, while the paired trap received a fluorescent scorpion in the same location. For each trial, the traps were set up in a 5 box x 4 box grid, with boxes being spaced approximately 5 m apart (total grid size: ~20 x 25 m). Fluorescent and non-fluorescent scorpions were oriented in the same direction within each pair, and randomly across pairs with respect to compass direction.

In addition, a small number of traps were constructed to test for the effects of the varnish used. These controls consisted of traps constructed just as above, but instead of placing scorpions on the trap, a 2.5 cm x 4.3 cm rectangle of the black construction paper was either left uncoated or coated with the same varnish used to remove fluorescence from the scorpions. No tangletrap covered this rectangle, so it was not an insect capturing part of the trap, just as the preserved scorpions were not insect capturing surfaces in the main traps. As with the main traps, these control traps did not visibly fluoresce when exposed to 40 W UV lights.

Six trials were carried out during the summer of 2004: three during the full moons (maximum UV availability) on July 2, July 31, and August 31, and three during the new moons (minimum UV availability) on July 17, August 15, and September 14. Weather conditions at the site on all nights sampled were generally clear, with some scattered clouds, which obscured the moon for brief intervals during the trapping period. Traps were set up at 20:00 and collected at 06:00 the following morning. Traps were laid out and collected during the twilight hours in order to avoid the necessity of using artificial lights during these activities. At this site, artificial lights attract large numbers of flying insects very rapidly. Although this extended the hours of collection into periods of time when sunlight reflected from the sky could enter the traps, it was felt that any effect of this would be randomly distributed across the traps, and would be far less than the potential effect of shining artificial lights into the traps during placement or collection. It also allows collection of insects during those twilight periods when many scorpions tend to most active (Warburg & Polis, 1990).

Slight variations between traps in total exposure time and microenvironmental conditions (cloud cover, temperature etc.) are controlled for by the paired design, which ensured that both traps in each pair, which were side-by-side, experienced as similar a micro-environment as possible.

The first sample (July 2nd) contained 17 paired scorpions and three control traps. Two of the scorpion pairs were damaged during analysis and removed from subsequent trials. The remaining five trials consisted of 15 pairs of scorpions plus five control trap pairs. This provided a total sample size of 47 pairs of scorpions during full moons and 45 pairs of scorpions during new moons. One of the control traps during a new moon was destroyed and removed from analysis, yielding a final control sample size of 13 and 14 control pairs on full and new moons, respectively.

Traps were collected by placing a piece of aluminum foil or waxed paper between the traps to prevent the possibility of insects struggling free from one side being trapped on the other, then closing the box to prevent further captures. Insects captured on each trap were counted and identified to order (following Borer & White, 1970). Due to the design of the traps, insects could only enter the trap from the air. Flightless arthropods were therefore assumed to have been blown in randomly and were excluded from analysis.

Relative success was measured as the number of aerial insects trapped in the side with the fluorescent scorpion minus the number of aerial insects trapped on the side with the non-fluorescent scorpion. For convenience, this measure will be referred to as the pair difference. A positive pair difference indicates that the fluorescent trap caught more than the non-fluorescent, and a negative pair difference indicates the opposite. Freeze-dried scorpions were re-used in order to maintain sample size across trials, so the differences were analyzed using a mixed model ANOVA with pair number as a subject

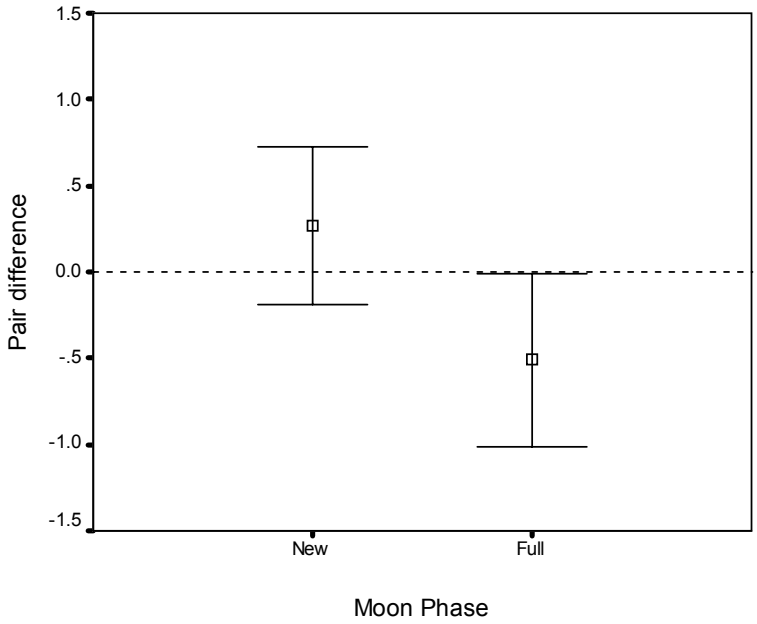

Figure 2: Mean difference (with 95% confidence intervals) between aerial insects captured on sticky traps bearing fluorescent scorpions and sticky traps bearing non-fluorescing scorpions, using data pooled from new moon nights (total n=45) and full moon nights (total n=47).

variable (using SPSS for Windows, Version 11.0.1, SPSS, 2001).

Results

Main effects: On each night of the full moon, the pair difference was negative; traps bearing fluorescent scorpions caught fewer insects than paired traps bearing non-fluorescent scorpions. In contrast, on two out of three new moon nights, the pair difference was positive (Fig. 1). The difference between new and full moons was significant (F=5.176, p=0.025; Table 1).

To further explore the difference between full and new moons, data were pooled by moon phase (Fig. 2), and a one-sample t-test against a hypothesized mean pair difference of zero was carried out independently for each moon phase. This analysis shows that during the new moon, the pair difference was not statistically significantly different from zero (t=1.182, p=0.244, n=45): i.e., prey capture is random with respect to fluorescence. During the full moon, the pair difference was significantly less than zero (t=-2.053, p=0.046, n=47), indicating that traps bearing fluorescent scorpions captured fewer insect than traps bearing non-fluorescent scorpions. During full moons, traps with fluorescent scorpions averaged 0.5 fewer insects per night than traps with non-fluorescent scorpions.

Control pairs: Control pairs were analyzed using the same procedure as the main traps. This analysis showed no significant effect of moon phase (F=0.035, p=0.853) or date nested within moon phase (F=0.241, p=0.912) on pair difference, indicating that there was no effect of varnish in the experiment. The observed difference between pair differences of new and full moon was 0.01. By contrast, this difference in the main experiment was 0.78. However, the small sample size of control traps argues for caution in interpreting these non-significant results (Peterman, 1990). For this reason, a power analysis was conducted, using the observed pair differences in the main experiment. In effect, this analysis asks how powerful the control sample size (27 control pairs) would be in detecting an effect of the size observed in the main experiment. The freeware program Gpower was used with the estimated effect size from the main experiment (0.78, yielding a partial η^2 of 0.062) and a combined sample size of 27 for a standard ANOVA. This yielded an estimated β of 0.25, or about a 75 % chance of detecting a difference of the size seen in the main experiment.

Taxonomic distribution of captured insects: Lepidoptera dominated the sample, comprising 59.0 % of insects captured. Diptera (18.9 %) and Hymenoptera (6.6 %) were also well represented. Nine other orders were captured only occasionally: none comprised >4% of the sample. Combined, these nine orders plus uniden-

Order	Total	Full Moon		New Moon	
		Fluorescent	Non-fluorescent	Fluorescent	Non-fluorescent
Lepidoptera	134	31	40	36	27
Diptera	43	12	20	6	5
Hymenoptera	15	1	8	3	3
Neuroptera	9	5	2	1	1
Homoptera	7	3	2	1	1
Psocoptera	6	0	0	2	4
Thysanoptera	4	2	2	0	0
Coleoptera	2	1	1	0	0
Isoptera	2	0	1	1	0
Orthoptera	2	2	0	0	0
Odonata	1	0	0	1	0
Trichoptera	1	0	0	0	1
Unidentifiable	1	0	1	0	0
Totals	227	57	77	51	42

Table 2: Taxonomic distribution of insects captured on sticky traps. Numbers are the total number over the entire study, broken down by moon phase and trap type.

tifiable insects make up only 15.4 % of the total (Table 2).

Discussion

These results clearly show that the response of aerial insects to fluorescing and non-fluorescing scorpions depends upon moon phase. Insects detect and avoid fluorescing scorpions during the full moon, when fluorescence is at maximum intensity, but not during the new moon, when fluorescence is at minimum intensity. The paired design of the study eliminates the possibility that variation in insect availability, scorpion size, or local environmental variations (moonlight exposure, cloud cover, etc.) caused this difference. Although caution is warranted because of the low power of the control experiment, it revealed no significant differences. Although there is no generally accepted criteria for levels of β (Peterman, 1990), given a β of 0.25 and that the difference observed in the controls is about 1 % the magnitude of the difference seen in the main experiment, the conclusion that varnish had no effect on the outcome of the experiment appears reasonable. However, a stronger control with a larger sample size would be desirable. This is the first experimental demonstration that scorpion fluorescence is detectable by other organisms under natural conditions.

The most common insects trapped in this study are generally strong flyers: Lepidoptera, Diptera, and Hymenoptera. These three orders drove the results of the study. Lepidoptera and Hymenoptera are both cited as prey items of several scorpion species (McCormick and Polis, 1990). The aerial prey cataloged in Polis (1979) included Lepidoptera, Diptera, Hymenoptera, and Neuroptera. Thus, the insects captured on these sticky traps appear to be representative of actual scorpion prey.

Although there is not much information on the visual spectra of nocturnal insects, Kelber et al. (2003) show that nocturnal hawkmoths exhibit visual sensitivity peaks in the UV, blue, and green, and that they can utilize color vision in starlight conditions. Other moths and nocturnal insects may possess similar abilities. Since scorpions fluoresce in the green portion of the spectrum, visual detection of fluorescence by nocturnal insects with these receptors is possible. It must also be remembered that scorpions absorb UV in order to fluoresce. Thus it is possible that they can be detected not by the green fluorescence, but by their absorption of UV (Silberglied, 1979). Since there are sensitivity peaks in both the green and the UV, it is entirely possible that scorpion detection involves both of these receptors.

This experiment was performed against a black background, which provides for maximum contrast of the fluorescent scorpion. Against a more natural background, detection of fluorescence by these insects may be more difficult, or the effect could even be reversed. This experiment shows that aerial insects are capable of detecting fluorescence under natural illumination levels; now it is a matter of determining how easily detectable scorpions are to these organisms under a variety of natural backgrounds.

Earlier studies have shown that scorpions increase their activity levels on moonless nights (Skutelsky, 1996; Warburg & Polis, 1990). Skutelsky (1996) attributed this pattern to predator avoidance. Here, I suggest that insect response to fluorescence may also be an important factor in setting this activity pattern. If potential prey—and possibly predators—can more easily detect scorpions on moonlit nights, then moonlight foraging trips would bear a double penalty: increased predation risk coupled with reduced foraging success.

These results imply a cost to fluorescence in scorpions: reduced predatory efficiency. This raises an interesting puzzle. While the possibility that fluorescence has no function must of course be considered (Gould & Lewontin, 1979), the existence of a cost associated with fluorescence makes it sensible to look for potentially offsetting benefits of fluorescence.

Potential benefits of fluorescence include ultraviolet protection (Lourenço & Cloudsley-Thompson, 1996; Fasel et al., 1997; Frost et al., 2001), vision enhancement (Hjelle, 1990), and intraspecific communication (Hjelle, 1990). In addition, the possibility that fluorescence functions as an aposematic warning should be considered. Any of these proposed benefits to fluorescence could potentially offset the cost in terms of prey avoidance and account for the maintenance of scorpion fluorescence.

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References

BORER, D. J. & R. E. WHITE. 1970. *Insects. Peterson Field Guides.* Houghton Mifflin, Boston, 440 pp.

FAZEL, A., P. A. MULLER, P. SUPPAN & E. VAUTLEY. 1997. Photoluminescence of the African scorpion *Pandinus imperator*. *Journal of Photochemistry and Photobiology B: Biology*, 9: 96–98.

FROST, L. M., D. R. BUTLER, B. O'DELL & V. FET. 2001. A coumarin as a fluorescent compound in scorpion cuticle. Pp. 365–368 *in* Fet, V. & P. A. Selden (eds.). *Scorpions 2001. In Memoriam Gary A. Polis*. British Arachnological Society, Burnham Beeches, Bucks.

GERMANO, D. J., RATHBUN, G. B. & SASLAW, L. R. 2001. Managing exotic grasses and conserving declining species. *Wildlife Society Bulletin*, 29: 551–559.

GOULD, S. J. & R. G. LEWONTIN. 1979. The spandrels of San Marco and the Panglossian paradigm: a critique of the adaptationist programme. *Proceedings of the Royal Society of London, Series B*, 205: 581–598.

HJELLE, J. T. 1990. Anatomy and morphology. Pp. 9–63 *in* Polis, G. A. (ed.). *Biology of Scorpions*. Stanford University Press, Stanford, CA.

KELBER, A, A. BALKENIUS & E. J. WARRANT. 2003. Colour vision in diurnal and nocturnal hawkmoths. *Integrative and Comparative Biology*, 43: 571–579

LOURENÇO, W. R. & J. L. CLOUDSLEY-THOMPSON. 1996. The evolutionary significance of colour, colour patterns and fluorescence in scorpions. *Revue suisse de Zoologie, Vol. hors série*, 449–458.

MCCORMICK, S. J. & G. A. POLIS, 1990. Prey, predators, and parasites. Pp. 294–320 *in* Polis, G. A. (ed.). *Biology of Scorpions*. Stanford University Press, Stanford, CA.

PETERMAN, R. M. 1990. The importance of reporting statistical power: the forest decline and acidic deposition example. *Ecology*, 71: 2024–2027.

POLIS, G. A. 1979. Prey and feeding phenology of the desert sand scorpion *Paruroctonus mesaensis* (Scorpionida: Vaejovidae). *Journal of Zoology (London)*, 188: 333–346.

SILBERGLIED, R. E. 1979. Communication in the ultraviolet. *Annual Review of Ecology and Systematics*, 10: 373–398.

SISSOM, W. D., G. A. POLIS & D. D. WATT. 1990. Field and laboratory methods. Pp. 445–461 *in* Polis, G. A. (ed.). *Biology of Scorpions*. Stanford University Press, Stanford, CA.

SKUTELSKY, O. 1996. Predation risk and state-dependent foraging in scorpions: effects of moonlight on foraging in the scorpion *Buthus occitanus*. *Animal Behaviour*, 52: 49–57.

SPSS. 2001. *SPSS for Windows, Version 11.0.1*. SPSS Inc.

STACHEL, S. J., S. A. STOCKWELL & D. L. VAN VRANKEN. 1999. The fluorescence of scorpions and cataractogenesis. *Chemistry and Biology*, 6: 531–539.

WARBURG, M. R. & POLIS, G. A. 1990. Behavioral responses, rhythms, and activity patterns. Pp. 224–246 *in* Polis, G. A. (ed.). *Biology of Scorpions*. Stanford University Press, Stanford, CA.

WILLIAMS, S. C. 1980. Scorpions of Baja California, Mexico, and adjacent islands. *Occasional Papers of the California Academy of Sciences*, 135: 1–127.