Assessing the Use of Geometric and Feature Cues During Goal-directed Navigation in Whip Spiders (*Phrynus marginemaculatus*): A Pilot Study

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ABSTRACT

Phrynus marginemaculatus, a species of whip spider, regularly relies on multi-sensory integration of various cues to orient in and navigate their natural environment. The aim of the current study was to investigate the visuospatial cue hierarchy that guides spatial learning and memory in *P. marginemaculatus*. First, spiders were trained to discriminate a single goal location (an open shelter) from three alternative locations (closed shelters), defined by its position to both geometric (boundary shape) and feature (colored card) cues, in a rectangular arena. Probe trials were then intermittently conducted to isolate the use of each cue or to set the two cues in conflict. The data revealed above chance performance when using either the geometric or feature cue alone, but random performance when the two cues were set in conflict. While this pilot data is encouraging, we must incorporate another probe to directly assess learning of the goal location (currently underway).

BACKGROUND

- Homing behavior (i.e., the ability to return to a shelter/nesting site following a period of exploration) is displayed by a variety of animals (Papi, 2012).
- Homing insects have been shown to use a variety of cues and strategies to orient themselves and navigate through space (Mandal, 2018). For example:
  - Ants (*Gigantiops destructor;* Wystrach & Beugnon, 2009) and bumblebees (*Bombus terrestris;* Sovrano et al., 2012) can use the shape an enclosure (i.e., geometric information) for orientation.
  - Honeybees (*Apis mellifera*) can learn visual features associated with a goal location and/or nearby objects (Cheng, 2000).
- Homing whip spiders (*Amblypygids*) have been found capable of using tactile, olfactory, and visual cues to navigate (Ortega-Escobar, 2020).
- The current study set out to…
  - test the ability of whip spiders to use geometric information to learn about a goal location.
  - assess the visuospatial cue hierarchy for spatial learning in whip spiders.

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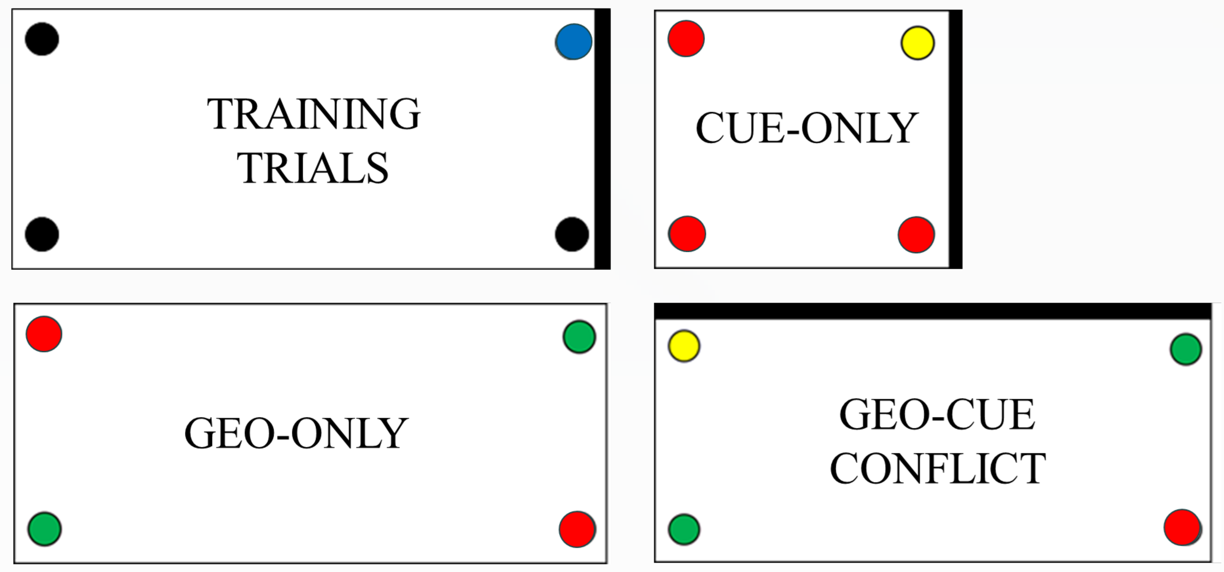
METHODS

- *Subjects* consisted of 4 wild-caught (Florida) whip spiders (*Phrynus marginemaculatus*).
- *Training* trials were conducted in a rectangular 60 x 30 x 20 cm arena made of white acrylic (Figure 1) and surrounded by a dark curtain.
  - One 120w light bulb was situated centrally overhead.
  - Only one shelter location was open during training trials.
  - The short wall adjacent to the correct location was made green with a thin piece of cardboard (visual cue).
- *Probe trials* in which the arena was manipulated were employed to assess cue use (Figure 1).
- Each animal completed 6 trials each day, over 15 days.
  - Days 1-3: exclusively training trials.
  - Days 4-15: the first and last trials were the same of one probe type; four middle trials were training trials.
  - Probe type was pseudorandomized across days (4 days of each probe type).
- Each trial began with the animal placed at center of the arena.
  - Given 10 minutes to find the correct shelter (training) or choose a shelter (probe).
  - During training trials, the animal was given a two-minute reward time in the shelter; there was no reward time on probe trials.
  - If no choice is made, then the trial is marked "no choice" and the spider is guided to the correct shelter.
  - Between trials, animals were disoriented for one minute

Figure 1

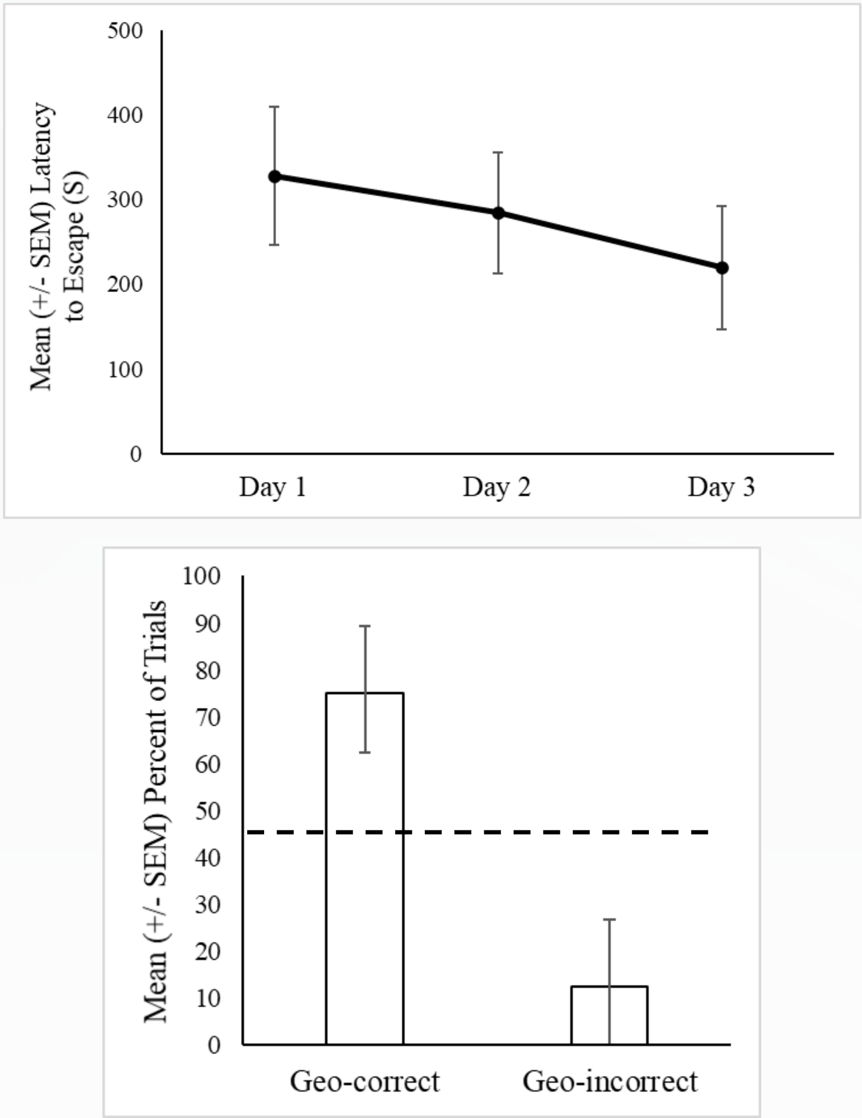


Overhead view of training and probe configurations. In training configuration, blue circle indicates open shelter while black circles indicate closed shelters. In probe trials (all shelters open), green circles indicate a geo-correct, yellow a cue-correct, and red an incorrect choice. Thick wall indicates the green cue wall in all configurations (save GEO-ONLY).

RESULTS

- Dependent measures:
  - Training trials (first 3 days only): average latency to escape.
  - Probe trials: percentage of geo-correct, cue-correct, and incorrect.
    - First and second probe on each day were separately analyzed.
    - Only the last two days of each probe were used in the analyses.
- A repeated-measures ANOVA revealed a non-significant decrease in the latency to escape over the three initial training days, $f(2, 6) = 0.54$, $p = .61$ (Figure 2).
- One sample t-tests failed to find significant differences in the percentage of geo-correct, cue-correct, or incorrect choices compared to chance performance (50% for geo, 25% for cue) during any of the probe trials.
- However, an interesting trend was found regarding the 2nd probe trials of GEO-Only probes (Figure 2):
  - While the percentage of geo-correct choices (M = 75.00%, SD = 28.90%) was not significantly greater than chance (50%), $t(3) = 1.73$, $p = .18$, the percentage of geo-incorrect choices (M = 12.50%, SD = 25.00%) was nearly significantly less than chance, $t(3) = 3.00$, $p = .06$.

Figure 2



(*Top*) mean (+/- SEM) latency to escape (in seconds) across the first 3 days (18 trials) of training (no probe trials). (*Bottom*) mean (+/- SEM) percent of probe trial 2 (i.e. trial 6) choices in that were geo-correct and geo-incorrect (last two GEO-ONLY probes). Dashed line indicates chance performance (50%).

DISCUSSION

- *Conclusions*
  - The current study failed to find evidence of learning to locate the goal location as measured by the latency to escape over the first three days (18 trials) of training.
  - However, whip spiders showed promise in learning of geometric information by the end of training each day, though these analyses failed to reach statistical significance.
- In sum, the data suggests that whip spiders, like ants (Wystrach & Beugnon, 2009) and bees (Sovrano et al., 2012), *might* be cable of using geometric information during spatial learning, and that perhaps such information precludes the use of visual features (considering the lack learning about the visual cue in the current study).
- *Limitations* include a small sample size (n = 4), habituation to training (decreased motivation in later trials), and lack of a probe trial designed to test learning of the trained shelter location.
- We are currently testing more animals following the same procedures described here, but with the addition of a TRAINING probe (i.e., with the arena in the training configuration, but with all shelters open).
- *Future studies* might simplify the procedures to focus solely on geometry use and consider shortening the amount of time animals are trained.

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