

Supplemental Materials

Pilot Studies

Initial tests consisted of a rectangular homing arena and a circular testing arena. The homing arena had a sand mound in the southwest corner of the rectangle and scorpions were given unlimited time to burrow. We observed that most scorpions would burrow within four days of placing them in the homing arena. We also determined that mounds needed to be moistened to provide additional structural support and that added illumination from overhead drop lights induced more burrowing. We played with displacing the animals from the rectangular homing arena to the testing arena using various containers including clear beakers, bottle caps, and cups placed on top of the animal. We also tried to use playing cards, sieves, and perforated cards to slide underneath the animal. We found that a sieve and an opaque cap was the most effective mechanism for transferring the scorpion. We felt it important to allow the home arena sand to sift from below the captured animal to remove residual home arena cues that could affect the animals' behavior in the testing arena. We also determined that one minute under the cap when placed in the testing arena was sufficient to calm the animal before release. We did not use displacement after prey capture because scorpions are not consistently motivated by food as they only eat approximately every 5 days [Polis 1990]. We felt displacement should instigate homing for the animals in our tests. We completed a series of runs with several animals and recorded the testing arena with IR cameras processed through a MATLAB script. While this study helped us refine our protocol, there were problems. One being that the rectangle to circle conversion made it difficult to determine the home position relative to the scorpion. It also required that the radius of our circular arena needed to be larger than the diagonal of the rectangular arena to allow the animal sufficient room to reveal a directional choice before contacting the testing arena wall. At this point, we adjusted our protocol for two reasons: 1) we did not have ready access to sufficiently large circular arenas and 2) larger arenas would make it difficult to accurately track the scorpions via overhead IR cameras.

The next series of pilot studies consisted of four circular arenas: two smaller homing arenas and two larger testing arenas like in our final experiment. We completed a series of trials with methods like those currently described in our main experiment, but without the added plastic pizza saver inside the mound (see section 2.2). Eight of the 10 animals used in these pilot studies burrowed in the sand mounds within four days. Of these eight animals, three completed at least one usable run with an average of 1.66 runs per animal and a total of six usable runs. We plotted these runs as independent trials in three differently rendered plots (see section 2.4) and determined the resultant vector angle that the scorpions crossed the decision line (Figure S1A). We used these vectors to calculate the composite vectors and determine the significance of the runs using Raleigh's Z test (Figure S1B). We found there was significant directionality in the animals' raw paths biased toward the southwest corner of the experimental room, which was in the direction of a large mirror that hung on the wall. We removed the mirror because we felt it may have been reflecting extraneous light entering the room that may have affected the scorpions' movements. We also minimized extraneous light sources by drilling a hole through the wall and running the camera wires through a light blocking grommet to exclude light from the adjacent storage room where the computers were stationed. We also put the adjacent room on the same reverse night:day cycle as the experimental room to further restrict any extraneous light. In addition, a light blocking curtain was strung between the homing arenas to prevent variations in light levels within the homing arenas due to light cast from the neighboring arena's lamps (the homing arena lamps were never on during trials in the testing arenas). All these actions helped ensure that there was uniformity in the experimental room to avoid any potential visual bias.

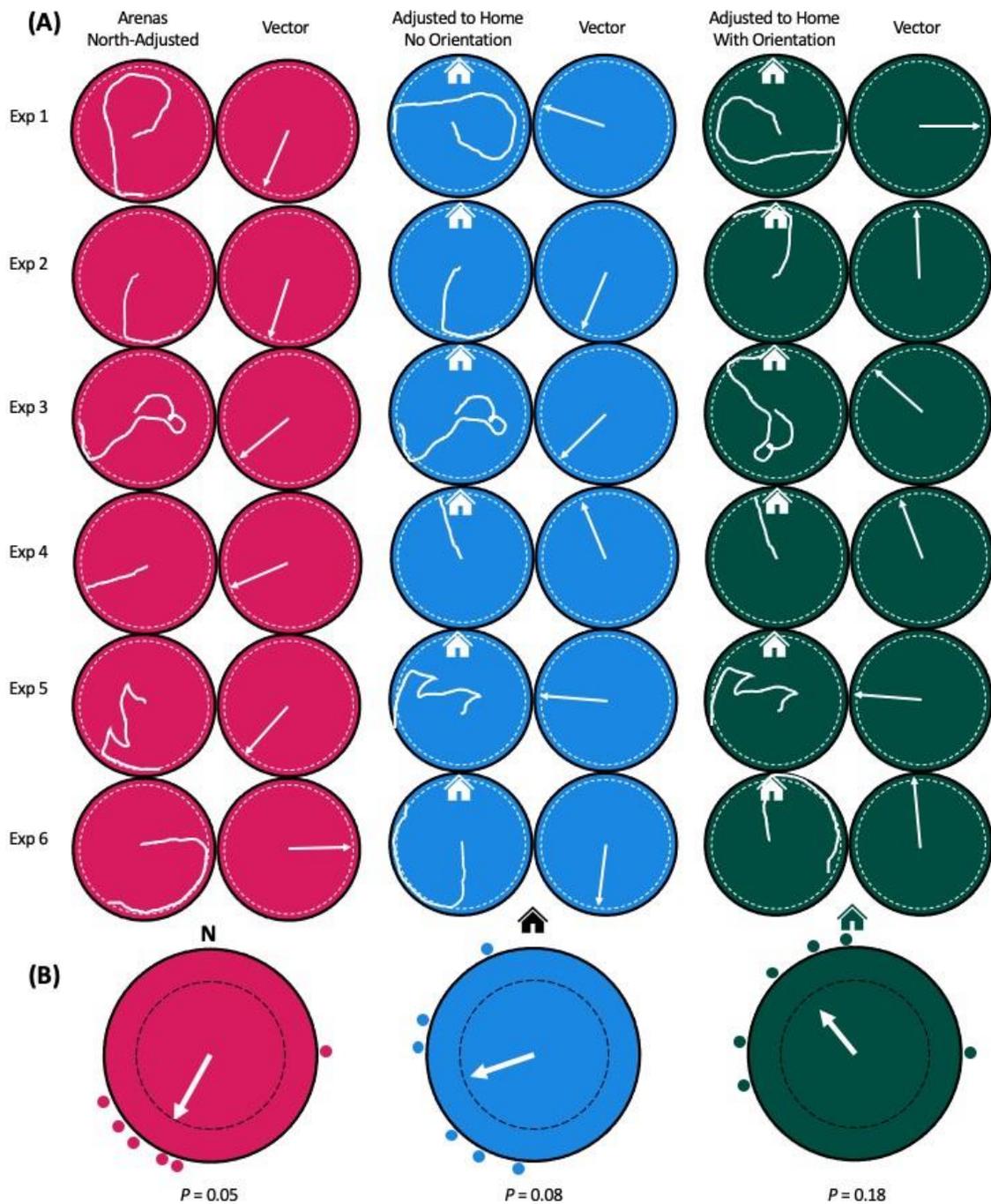


Figure S1. Pilot study movements and patterns. **(A)** The movements of all animals in our pilot study are shown based on the adjustments described in the main text. The first column in each group shows the animal's path and the second column the angle where the animal crossed the decision line. **(B)** The mean resultant vectors for each of the groups of adjusted trials are shown along with the individual angles as dots on the periphery. The dashed black lines represent $P = 0.05$ significance level for the resultant vectors. The P -values at bottom indicate statistical significance based on the Rayleigh test for uniformity.

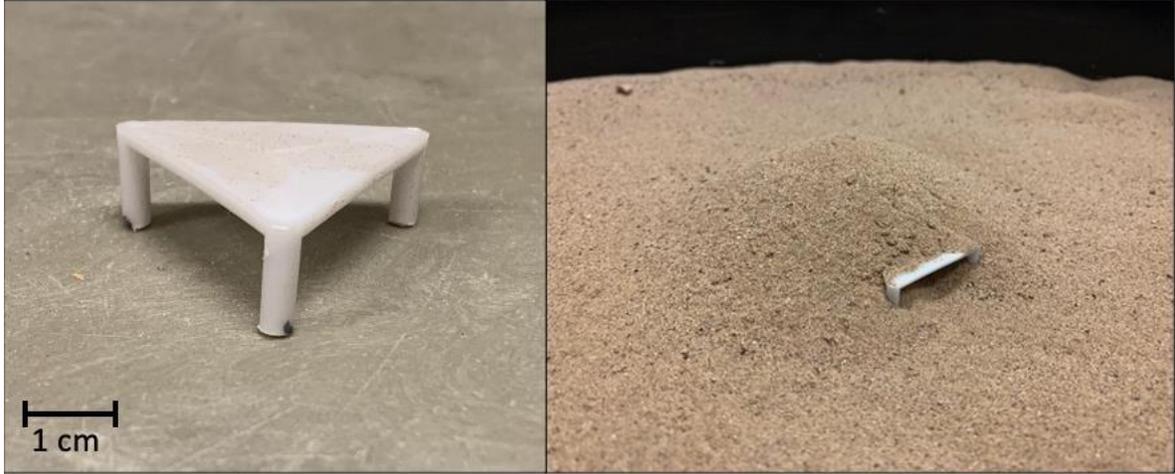


Figure S2. Pizza saver aid in burrowing. A triangular pizza saver was placed in the center of each homing arena and used as a structural support under the 90 ml mound of sand used by scorpions to create a home burrow (Right). The pizza saver on its own (Left) is 1 cm tall and 2.5 cm wide with three legs.

Animal burrowing and movement information

Animal	Burrow	Number of Runs	Time Before Moving	Time to Cross Decision Line	Trial Number
1	No	0	--	--	--
2	No	0	--	--	--
3	No	0	--	--	--
4	No	0	--	--	--
5	Yes	4	12.3	9.5	1
6	No	0	--	--	--
7	Yes	4	45.0	75.8	2
8	No	0	--	--	--
9	No	0	--	--	--
10	No	0	--	--	--
11	Yes	1	25.0	18.0	3
12	Yes	1	101.0	49.0	4
13	Yes	1	23.0	34.0	5
14	Yes	1	13.0	36.0	6
15	Yes	3	1.7	13.0	7
16	Yes	2	13.5	14.5	8
17	Yes	4	243.8	54.0	9
18	Yes	4	153.3	38.9	10
Avg	55% burrowed	2.5	63.2	34.3	
STD	--	1.4	75.5	19.3	
SE	--	0.4	23.9	6.1	

Movement durations

Run	Cap to 1st move	1st move to cross	Cap to Cross
Exp01-01	8	15	23
Exp01-02	1	7	8
Exp01-03	35	11	46
Exp01-04	5	5	10
AVG	12.25	9.50	21.75
Exp02-01	31	85	116
Exp02-02	63	65	128
Exp02-03	80	22	102
Exp02-04	6	131	137
AVG	45.00	75.75	120.75
Exp03-01	25	18	43
Exp03-02			
Exp03-03			
Exp03-04			
AVG	25	18	43
Exp04-01	101	49	150
Exp04-02			
Exp04-03			
Exp04-04			
AVG	101	49	150
Exp05-01	23	34	57
Exp05-02			
Exp05-03			
Exp05-04			
AVG	23	34	57
Run	Cap to 1st move	1st move to cross	Cap to Cross
Exp06-01	13	36	49
Exp06-02			
Exp06-03			
Exp06-04			
AVG	13	36	49
Exp07-01	1	16	17
Exp07-02	1	15	16
Exp07-03	3	8	11
Exp07-04			
AVG	1.67	13.00	14.67
Exp08-01	22	9	31
Exp08-02	5	20	25
Exp08-03			
Exp08-04			
AVG	13.50	14.50	28.00
Exp09-01	796	49	845
Exp09-02	96	140	236
Exp09-03	54	16	70
Exp09-04	29	11	40
AVG	243.75	54.00	297.75
Exp10-01	9	12	21
Exp10-02	370	17	387
Exp10-03	225	40	265
Exp10-04	9	86	95
AVG	153.25	38.75	192.00
SUMMARY			
Avg	63.14	34.25	97.39
STD	79.52	21.27	92.33
SE	25.15	6.73	29.20

Figure S3. Movement and burrowing data for all animals used in the study.

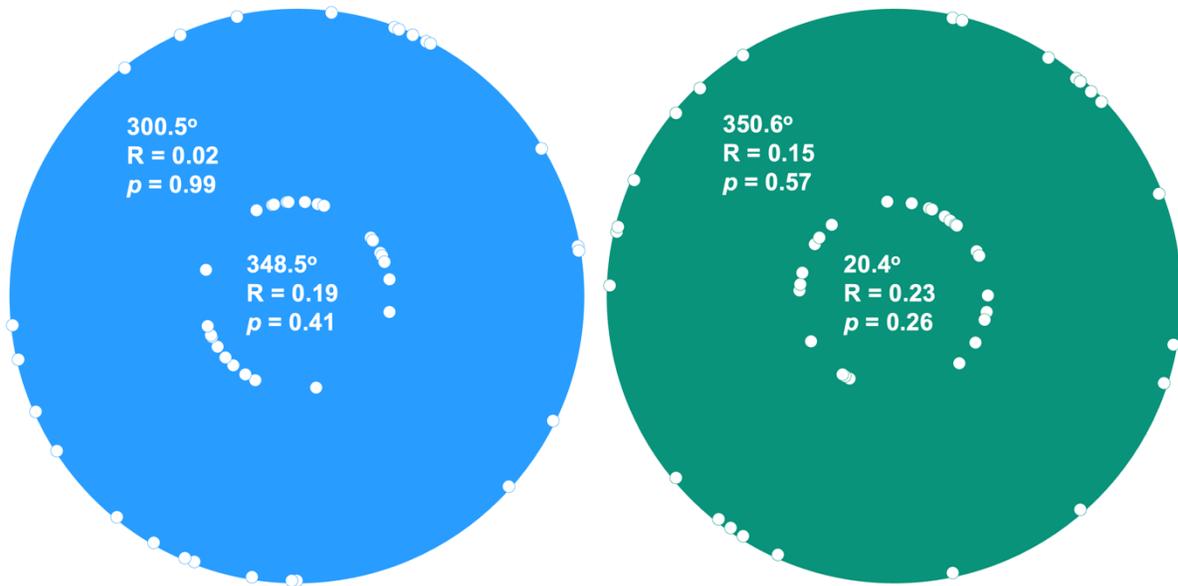
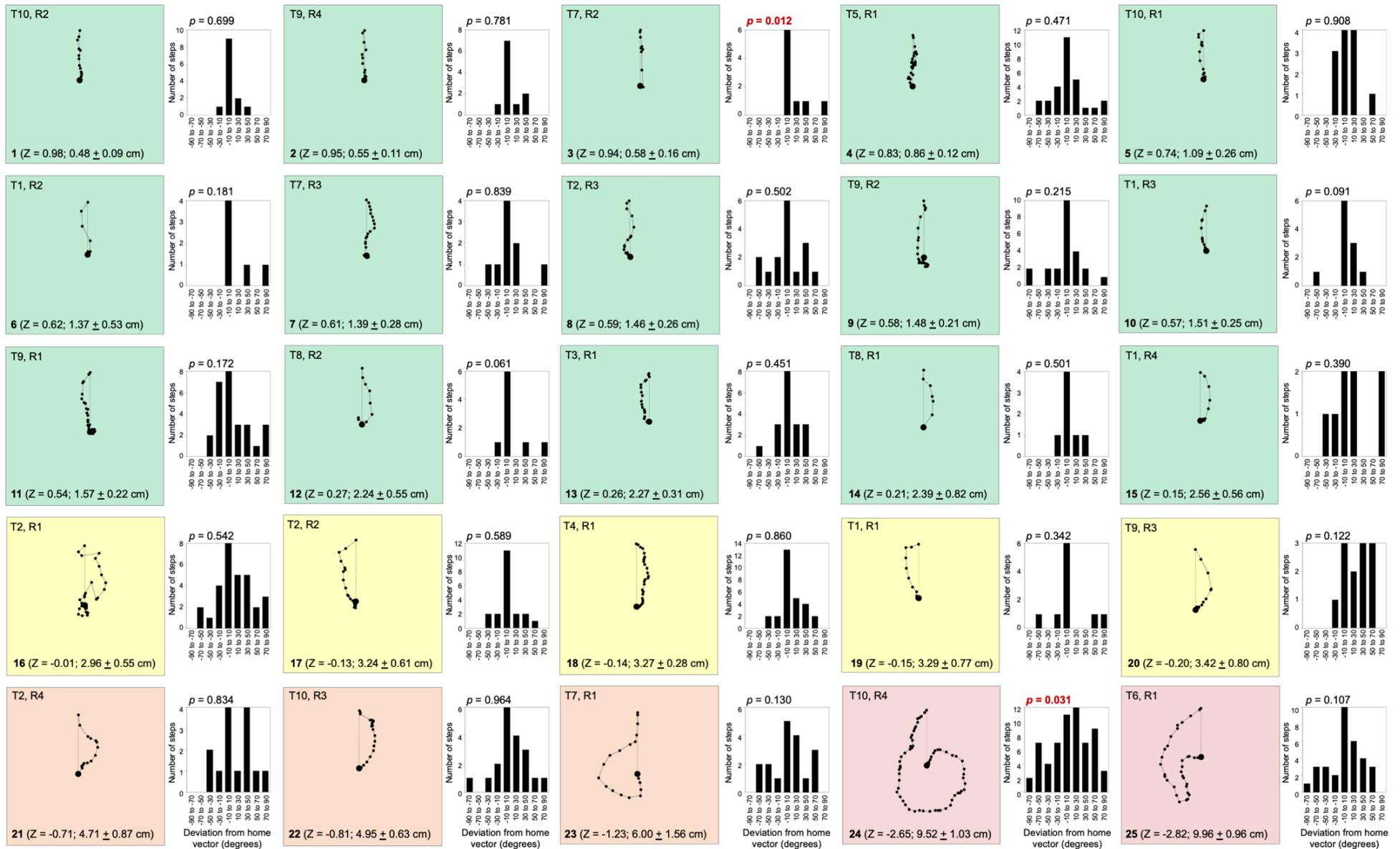


Figure S4. Analysis of all runs and runs restricted to a closer decision line. These plots show the result of plotting all runs in relation to both the full arena decision line (outside white dots) and a decision line that was only one third the distance from the released animal to the full arena line (inner circle white dots). The left plots (blue) indicate paths adjusted so home is at 0° while the right plots (green) indicate paths adjusted so home is at 0° with change in scorpion orientation factored.



Supplemental Figure S5. Analysis of scorpion path trajectories in testing arenas. The mean distance from the path vector (PV) and the angle of departure were analyzed for all 25 runs. The PV was defined as the straight path from the animal's starting point in the testing arena to its point of crossing the decision line (3.5 cm inside the arena wall). All paths were adjusted so that the PV faced 0°. In each pair of images, the left panel shows the scorpion's path plotted at 1s intervals; the right panel shows the distribution of angle departures from the PV (see Prévost & Stemme 2020 for analysis details [5]). The runs have been rank-ordered based on mean departure distances, which are shown in the bottom of each left-side panel (\pm SE), along with the Z-score for each run. The plots have been color coded based on Z-score (green >0 , yellow 0 to -0.20 , orange -0.20 to 2.00 , red <2.00). The p -values based on the Shapiro–Wilk test of normality for the angle distributions are at the top left of each column graph. Two of the distributions failed to meet normality and are indicated with red text (T7, R2 and T10, R4).