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RESEARCH PAPER

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Ontogenetic approach reveals how cognitive capability and motivation shape prey-searching behavior in *Pholcus phalangioides* cellar spiders

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Abstract

An important part of understanding the evolution of behavior is understanding how and why behavior develops and changes throughout ontogeny. Patterns of behavior are shaped by an animal's capabilities as well as its motivations, both of which are subject to selection. We ran an experiment to see how spiders' efforts to recover lost prey change with age and to determine the relative contributions of shifts in capability and motivation. We found that as spiders mature, they spend less time searching to recover lost prey, and they discriminate less between prey of different sizes. We also found that even the youngest, least experienced spiders are cognitively equipped to search for lost prey. Thus, predatory behavior in spiders fluctuated primarily with each age group's motivations to capture and consume prey, and did not seem to be hindered by behavioral or cognitive limitations at young ages.

KEYWORDS

behavioral limitations, behavioral motivations, cognition, cognitive phenotypes, development, prey-searching

1 | INTRODUCTION

Behavior sits at the intersection of capability and motivation. An animal can only perform actions that its anatomy and cognition allow for, and it should only perform actions that are necessary or rewarding. Throughout an animal's life, these two aspects change. Capability tends to increase with maturity (up to a point), while motivation fluctuates based on context—it can change over months and years (e.g., during sexual maturation) or over seconds (e.g., after the sudden appearance of a predator).

Developing capabilities and varying motivations influence the ontogeny of animal behavior, and exploring this interplay is one of the major goals of the evolutionary study of behavior (Tinbergen, 1963). A key challenge in this enterprise is to elucidate the precise mechanisms that are involved in ontogenetic change in behavior (Ryan & Wilczynski, 2011). This is a difficult task, for two main reasons. On the one hand, behavior is the consequence of neural processes that constitute a special kind of trait—cognitive phenotypes (Mendelson et al., 2016). These traits determine behavior, decision-making, and how animals engage with their surroundings, yet they are ephemeral and difficult to study objectively. On the other hand, the ontogeny of a given behavior can be affected by the development of various other capabilities. In other words, not only must a capability be present, but the ability to exhibit the capability must also be present—for example, experimental tests for object permanence generally require that the animal be capable of either searching for an object or showing surprise (Baillargeon, Spelke, & Wasserman, 1985; Dore & Dumas, 1987; Hoffmann, Rüttler, & Nieder, 2011; Singer & Henderson, 2015). Understanding the ontogeny of behavior thus requires untangling webs of interrelated developing capabilities that are influenced in their expression by changing motivations.

In this study, we analyze the ontogeny of memory in a web spider. Memory is a basic cognitive capability that allows animals to gather and use information in order to improve their behavioral responses to a range of contexts (Shettleworth, 2010). Working with a web spider affords a particularly clear analysis in terms of changes in capability and motivation.

Our assay of the contents of memory is based on the searching behavior that many web spiders perform when they lose prey that they have captured (Rodríguez & Gamboa, 2000; Rodríguez & Gloudeman, 2011; Rodríguez, Briceño, Briceño-Aguilar, & Höbel, 2015; Rodríguez, Kolodziej, & Höbel, 2013). Searching for lost prey can help spiders reduce rates of prey loss caused by wind disturbances, or prey escaping, or by kleptoparasites that steal captured prey (Rodríguez & Gamboa, 2000; Rodríguez & Gloudeman, 2011).

When a web spider searches for lost prey, it moves about the web and tugs or plucks the threads, sending out vibrations that help it locate objects in its web. The effort put into recovering the lost prey is a function of the spider's memory of the features of the prey and its preference for those features (e.g., searching longer for more valuable or preferred prey items) (Rodríguez & Gamboa, 2000; Rodríguez & Gloudeman, 2011; Rodríguez et al., 2015, 2013). Therefore, spider prey-searching behavior is an illuminating measurement of a cognitive phenotype; specifically, it allows us to analyze the contents of spiders' memories of prey features. We used this assay of memory of captured prey to test for variation between individuals across life stages.

We studied the ontogeny of the formation of memory of captured prey and its use in prey recovery efforts in long-bodied cellar spiders, *Pholcus phalangioides* (Fuesslin, 1775). We tested two hypotheses to analyze change in searching behavior in terms of potential changes in capability and motivation. The two hypotheses are not mutually exclusive. Our goal was to determine which factor is the principal driver of behavioral change for these spiders. The hypotheses are as follows:

1.1 | Changing capability hypothesis

This hypothesis states that young spiders are limited in their ability to evaluate prey and form memories, and that ontogenetic changes in behavior are driven by improvements in capability as spiders grow older. Development in many species involves orders-of-magnitude increases in brain size (Quesada et al., 2011), as well as substantial changes to patterns of neuronal connectivity (Supekar, Musen, & Menon, 2009), both of which are likely to have direct impacts on cognition (Chittka & Niven, 2009; Eberhard, 2007; Eberhard & Wcislo, 2011). The changing-capabilities hypothesis makes the following predictions: (a) Young spiders will search very little upon losing prey they have captured, and searching efforts will increase with the age of the spider; (b) young spiders will discriminate less strongly between prey of different sizes than older spiders. A lack of prey size discrimination in young spiders could indicate that they have trouble either evaluating or remembering the size of prey.

The test of this hypothesis is confounded by the possibility that experience may help shape spider prey-searching behavior. If this is the case, then an increase in search behavior with age could be attributed to spiders learning how to search, which is downstream of the ability to form and use memories. We suspect that prey loss events like those we create in this experiment (where the spider has already subdued and secured its prey before the prey goes missing) are relatively rare in nature, so it is unlikely that the spiders we test have much experience. Nevertheless, we deal with this potential confound in three ways. First, many of our trials involve testing individuals twice. This allows us to test for short-term experience effects—if experience is important, we would expect to see spiders consistently searching differently in Trial 2 compared to Trial 1. Second, a subset of our spiders is laboratory-hatched naïve spiderlings, which have absolutely zero experience capturing or losing prey. Comparing this group to more experienced groups will help shed light on the effects of experience. Third, we consider the effects of long-term experience. Many of the spiders in this experiment were caught in the wild, and so we have no way of knowing their previous experience with capturing and losing prey. However, because this experience is uncommon, it is likely that some of the spiders we tested had multiple experiences losing prey while others had absolutely none. This range of variation in experience across individuals is only likely to increase with age, so if long-term experience has a strong effect on behavior, we would expect to see behavioral variation increase with age.

1.2 | Changing motivation/need hypothesis

This hypothesis states that all individuals are similar in their memory capabilities, but that the needs of the spiders change over ontogeny. This hypothesis makes the following predictions: (a) Young spiders will search the most, as they have greater need of energy for growth and development, whereas mature spiders will search the least, as they have more energy reserves and motivation to engage in sexual behavior rather than in foraging. (Note that it might be argued that adult females have a high need of nutrients for developing eggs (cf. Rickers, Langel, & Scheu, 2006); however, our prediction is based on the fact that females that consume more prey as juveniles grow to larger sizes and reach higher fecundity (Skow & Jakob, 2003). Therefore, females are preparing for reproduction well before maturity). (b) Young spiders will discriminate more strongly between prey of different sizes than older spiders.

Note that as with the previous hypothesis, experience is a potential confound for the age-based tests outlined here. Specifically, spiders could learn that searching for lost prey is not worthwhile. If this is the case, then an decrease in search behavior with age could be attributed to spiders learning not to search, which is similar to, but distinct from, a shift in motivation. The predictions described above that are designed to detect effects of experience apply here as well.

Differences in the life history of males and females in our spiders suggest additional predictions for this hypothesis. Juvenile *P. phalangioides* males grow more quickly than females (Uhl, Schmitt, Schäfer, & Blanckenhorn, 2004), and so are likely to place a higher premium on prey; therefore: (c) Juvenile males will search more than females for preferred prey. Additionally, when males reach sexual maturity, their behavior shifts away from capturing prey toward actively seeking mates (Escalante, 2013; Foelix, 2011; Uhl, 1998); therefore: (d) Mature males will exert particularly little search effort.

2 | METHODS

Pholcus phalangioides is a widespread synanthropic spider (Uhl, 1998). We collected *P. phalangioides* spiders from populations in six houses in Milwaukee, Wisconsin (USA) during the summers of 2015 and 2016. The spiders ranged in age from juveniles to adults. We recorded the mass of each spider on the day of capture, and then we immediately set them up in individual rectangular plastic shoe boxes. Each box measured approximately 30 × 17 × 10 cm (L × W × H) and was lined with a 6-cm-wide strip of poster board along the sides (to give spiders a better surface to climb on and attach silk to). We also placed a thin sheet of plastic wrap under the lid of each box to discourage spiders from attaching silk to the lid of the box.

Among the collected spiders were eight gravid females, which we did not test, but rather set aside and tested their spiderlings after they hatched. We separated spiderlings from their mothers once they reached their second instar (at approximately one week of age), which is when they naturally disperse (Uhl, 1998; Uhl et al., 2004). At this point, we recorded the mass of each spiderling, and we excluded any that weighed considerably more than its siblings (over 20% greater than the family median), as we took this to be a sign of cannibalism, and we wanted spiderlings with no experience of capturing or eating prey of any kind. We housed spiderlings in individual, clean, round plastic takeout containers (approximately 8 cm high and 11 cm in diameter at the top), each with a flat-bottom standard coffee filter pressed flat along the bottoms and sides to give spiderlings a decent surface to climb on and attach silk to. Spiderlings were ready for testing as soon as they built their first web.

For the rest of the spiders, we standardized hunger prior to running trials. First, we withheld food from spiders for at least four days after capturing them, then we fed them a single cricket whose mass was one quarter of the spider's mass at capture, and then we waited an additional two days before testing them. This controlled feeding, combined with the standardized periods without food, ensured that spiders started their trials at similar levels of satiation.

We kept all spiders (including spiderlings) in an environmental chamber (Percival Scientific, Inc., Perry, Iowa, USA), where we controlled the light:dark cycle (14:10 hr) and the temperature, which ranged from 21.4 to 27.6°C. In preliminary versions of the statistical models presented below, we included temperature as a term and found that it had no effect, so we removed it from our final analyses. The interior dimensions of the chamber were 2.54 × 2.39 × 2.1 m (L × W × H).

2.1 | Overview of trials

All trials involved us giving a cricket to a spider as prey, then stealing the cricket and recording the spider's behavior as it searched for the lost prey (described in detail below). We tested spiderlings only once to observe their behavior in the complete absence of prey-capture experience. We tested all other spiders twice, two days apart, once with a relatively small prey and once with a relatively large prey (prey size details below) in random sequence. In our preliminary analyses, we included trial sequence in our statistical models to look at short-term effects of experience. It had no effect, so we removed it from the final analyses presented below.

Each trial began with us attaching a cricket to a miniature crane that we used to lower prey onto the horizontal webs of our spiders (Figure 1). If the spider did not promptly respond to the arrival of the cricket, we used an electric toothbrush (Colgate 360° Total Advanced) to vibrate the cricket and simulate struggling in order to attract the attention of the spider. (We used the toothbrush in 84% of trials. In preliminary versions of our models, we included whether or not the toothbrush had been used. We found that it had no statistical effect, so we removed it from our final analyses).

We allowed the spider two minutes to wrap up its prey before we scared it away with several sharp puffs of air from a 2-ml plastic pipette. The spider would retreat, leaving the tethered prey behind, at which point we used the crane to raise the cricket off from the web, and we used a hot soldering iron to gently cut any strands of silk that attached it to the web.

After stealing each spider's prey, we took note of behaviors indicative of searching (e.g., tugging, defined in Table 1; see Supporting Information Video S1), as well as nonsearching behaviors (e.g., attaching threads or grooming, defined in Table 1). We recorded video of

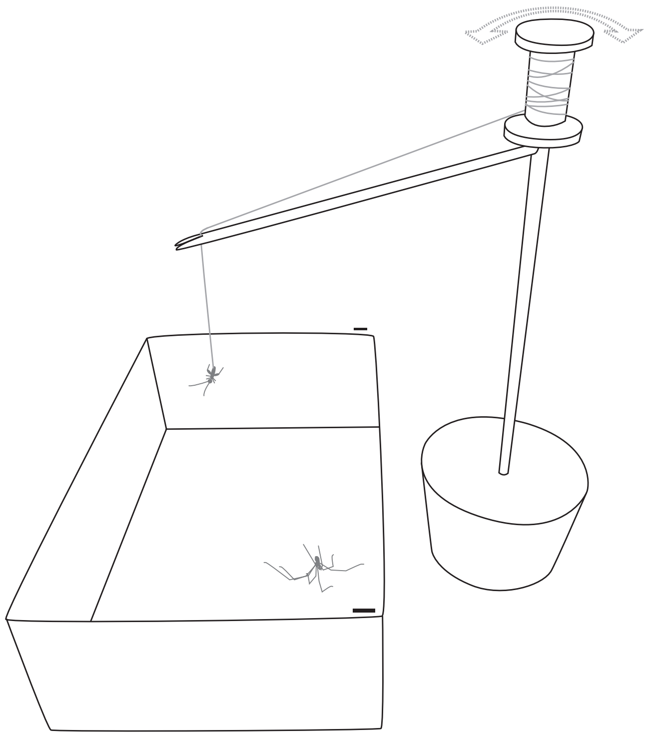

FIGURE 1 Experimental setup, including a plastic shoe box holding one of the spiders hanging from its horizontal web (web not shown), and the crane we used to lower crickets onto the spider's web. Scale bars along the lip of the box indicate 1 cm. The crane is made of bamboo garden stakes with a spool of thread mounted on top. Rotating the spool clockwise or counterclockwise (indicated by arrows) raises and lowers the cricket, which is attached to the end of the thread by a tiny hook

TABLE 1 Descriptions of spider behaviors observed during trials. See Figure 2. Not every behavior was used in the final analysis of search behavior—for that, we primarily looked at tugs and descents—but all together, they helped expand our general view of how each spider spent time during trials

Behavior	Description
Tug	The spider quickly and sharply contracts all of its legs in and then immediately returns them to a resting state. This is a typical single tug. When a spider performs multiple tugs over a short period of time, it is indicative of search behavior. We defined a bout of searching as a series of tugs in which each tug was no more than 20 s apart from the next
Build	The spider works on constructing or maintaining its web. We recorded every time a spider attached a new thread to its web and every time it cut old strands with its chelicerae
Descend	The spider drops down, tethered by a silk dragline, from its web to the floor of its enclosure. Often when the spider is on the floor, it waves its front legs around in an apparent exploration of its surroundings. The spider usually returns up its silk line back to its web after several seconds. Sometimes, the spider attaches a thread to the floor of the box before returning to the web. If it does, we consider this and the entire trip down as web-building behavior. On the other hand, when a spider descends and only explores, we count this as searching behavior
Move	The spider moves around its web, usually hanging from the underside of the web, with its ventral side up
Handle debris	The spider encounters a piece of debris in the web and spends time investigating it and removing it from the web
Groom	The spider stops moving around and cleans its legs. It brings a leg up to its mouthparts and pulls the tarsus through its chelicerae, presumably to remove tiny bits of dirt or silk
Rest	Periods of inactivity

each trial (using a Canon VIXIA HF R600 camcorder). We also entered behavior data in real-time into a custom computer program that we created for this purpose. To enter data quickly and efficiently, we programmed keyboard hotkeys to correspond to different behaviors, and we used a one-handed keyboard (Twiddler 3 by Tek Gear) for our input. This allowed us to record behavior while keeping one hand and both eyes free for the trial. With this setup, we created behavior logs with precision down to a fraction of a second that were digitized and ready for analysis as soon as the trial was complete, and we had video recordings that we double-checked afterward to resolve any inconsistencies or unusual sequences in our behavior logs.

We ended each trial when the spider had finished searching. Our criterion was when it had gone five minutes without tugging on its web. Our observations ranged in duration from a little over 5 min up to 52 min. In the event that a spider did not search at all (which happened in 16 of 144 trials), we waited a full 15 min after stealing the prey to declare the trial over. After running a spider through its final trial, we euthanized it, recorded its mass, and preserved it in 75% EtOH. We deposited voucher specimens at the personal collection of M. Draney at the University of Wisconsin-Green Bay.

2.2 | Prey used in trials

The prey used for the trials were commercially acquired crickets (*Gryllodes sigillatus*). For each trial, we selected a cricket based on its size relative to the spider. After euthanizing spiders, we were able to take more precise measurements of relative prey size, using spider sternum width as a proxy for spider body size (Lee, Somers, & Chown, 2011; Suter & Stratton, 2011). A spider's sternum is a single sclerotized plate, and unlike other measurements, like mass or body length, sternum width does not change based on how recently or how much a spider has eaten. Additionally, it is an external structure with clear landmarks (measurement procedures described below). Our measure of cricket size was the length of its body from the front of its head to the end of its abdomen. Finally, our measure of relative prey size was the ratio of cricket body length over spider sternum width. The relative prey size values in all of our trials varied across a range from about 4 to 8.

2.3 | Spider size measurements

After preserving the spiders (including the spiderlings), we measured their sternums using an Olympus SZ61 microscope (Olympus Corporation, Tokyo, Japan) with a Moticam 2500 digital camera (Motic, Causeway Bay, Hong Kong) attached that was hooked up to a computer running Motic Images Plus 2.0.10. We photographed the sternum of each spider and a calibration slide for scale, and then we used ImageJ 1.51j8 (National Institutes of Health, USA) to measure the widths of the sternums.

2.4 | Data analysis

We used custom scripts written in Python 3.5.2 (https://www.python.org, RRID:SCR_008394) and R 3.4.2 (R Core Team, 2017; RRID:SCR_001905) to process the behavior logs from the trials and calculate the total amount of time spiders spent actively searching for prey (e.g., Figure 2, Table 1). To do this, the scripts looked for two main things: periods of frequent tugging and time

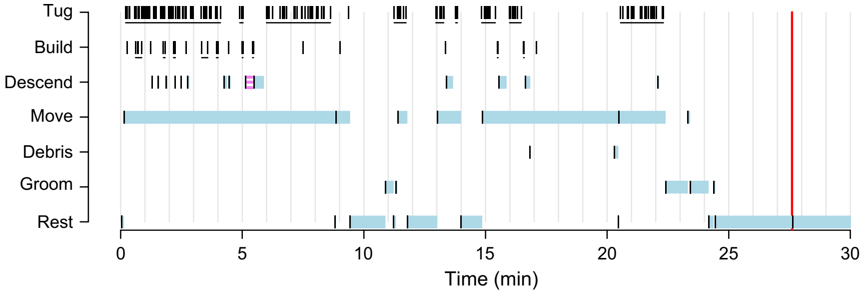

FIGURE 2 Example visualization of a spider's behavioral data from a trial. Categories of behaviors (defined in Table 1) are labeled along the *y*-axis, with markers showing instances of those behaviors through time, starting from when the spider's prey was stolen. Horizontal lines under the markers for *tug* and *build* indicate sequences broken by no more than 20 s (for *tug*, this was our major criterion for search behavior). For the behaviors that continue over time (*descend*, *move*, *debris*, *groom*, and *rest*), markers indicate the start of the behavior, and light trailing bars indicate the duration. Note that for the *descend* category, we make the distinction between descents in which spiders attached threads (striped) and those in which they did not (solid) (see Table 1). The thick vertical line near the end indicates that five minutes have passed since the last tug—our usual criterion for ending observation. [Colour figure can be viewed at wileyonlinelibrary.com]

spiders spent exploring the floors of their enclosures. Tugging is a conspicuous component of searching behavior, but spiders tug on their webs in other contexts as well, for example, when testing the tension of their threads or when orienting themselves in their web. The main difference is that during searching, a spider tugs frequently over a period of time, as opposed to performing infrequent and isolated tugs. Therefore, we only counted tugs that occurred within 20 s of other tugs. Each sequence of tugs that was not broken by a gap of 20 s or more was considered to be a bout of searching behavior (see Supporting Information Video S1 for a video of tugging behavior). In addition to tugging, spiders sometimes descend from their webs and search the ground for fallen prey (Table 1). We counted this as searching behavior as well, as long as the spider did not attach any threads to the floor before returning to its web (we did not want to mistakenly count web building as prey-searching).

For our analyses, we divided spiders into four groups based on age: (a) spiderlings, which were hatched and raised in the laboratory and were tested during their second instar posthatching; (b) early juveniles, which were older than spiderlings, but were still too small for us to distinguish their sexes; (c) advanced juveniles, which were old enough that we could tell them apart by sex, but were not yet fully mature; and (d) adults, who were sexually mature (see Figure 3 for sample size and mass).

2.5 | Statistical analysis

We used linear mixed models implemented in JMP Pro 13.0.0 (https://www.jmp.com, RRID:SCR_014242). To test the first prediction of each hypothesis, we tested for variation in overall searching behavior with spider age. We used a model with search time as the dependent variable, and the following fixed independent terms: age, relative prey size, and the age x relative prey size interaction. The model also included a random term for replicate: this represented family identity for spiderlings and individual identity for all other spiders.

To test the second prediction of each hypothesis, we tested for variation in how strongly spiders of different ages discriminated between prey of different relative size. Our measure of discrimination was the slope of the line representing change in search time over change in relative prey size. A flat slope indicates low importance of prey size, while a steep slope (either positive or negative) indicates high importance. We analyzed differences in the absolute value of slopes across three age groups: early juveniles, advanced juveniles, and adults (we omitted spiderlings from the formal analysis because their slopes came from family-level regressions, not the individual-level). We did this by running a one-way ANOVA with the absolute values of slopes as the dependent variable and age group as the independent variable. We used Tukey HSD post hoc analysis to determine which groups were statistically different from the others.

The changing motivation hypothesis has two additional predictions that deal with differences in search behavior between males and females, so we ran a model that explicitly tested for the effects of sex. As sex is indistinguishable in spiderlings and early juveniles, this model only included advanced juveniles and adults. The dependent variable was search time. The fixed independent variables were age, relative prey size, sex, and their interactions. We also included a random term for spider identity, as each spider was tested twice. We do not know for sure whether any of the adult females we tested were gravid, because we caught them as adults from the wild. We also do not know what effects being gravid would have on search behavior. All of our adult females decreased in mass from the beginning of the experiment to the end, but two in particular decreased proportionately less than the others. In case this was a sign of them being gravid, we ran our models with these individuals excluded. These additional models were qualitatively identical to the models that included these females (data not shown).

One test for the effects of experience involves looking at changes in variance of search behavior as a function of age. We ran a Brown–Forsythe test for unequal variances across the four groups. We also ran a mean-corrected test, looking at the correlation between the coefficient of variation (CV) and age group.

Finally, we were curious about family-level and individual-level sources of variation in search behavior, as this would help give us a broad sense for the variation available for natural selection to potentially act on. To measure family-level variation, we ran a model that only included spiderlings. It used relative prey size as a main term, family as a random term, and it included a family × prey size interaction. Our exploration of individual-level variation comes from the random terms in the model described above.

3 | RESULTS

In evaluating the age-related predictions from the two hypotheses, we found that spider maturity had a significant effect on search time, with younger spiders spending more time searching than older spiders (Table 2, Figure 3). We found a marginally significant effect of relative prey size on search time, with spiders overall tending to search longer for relatively smaller prey (Table 2, Figure 3). There was no significant interaction between maturity and prey size (Table 2).

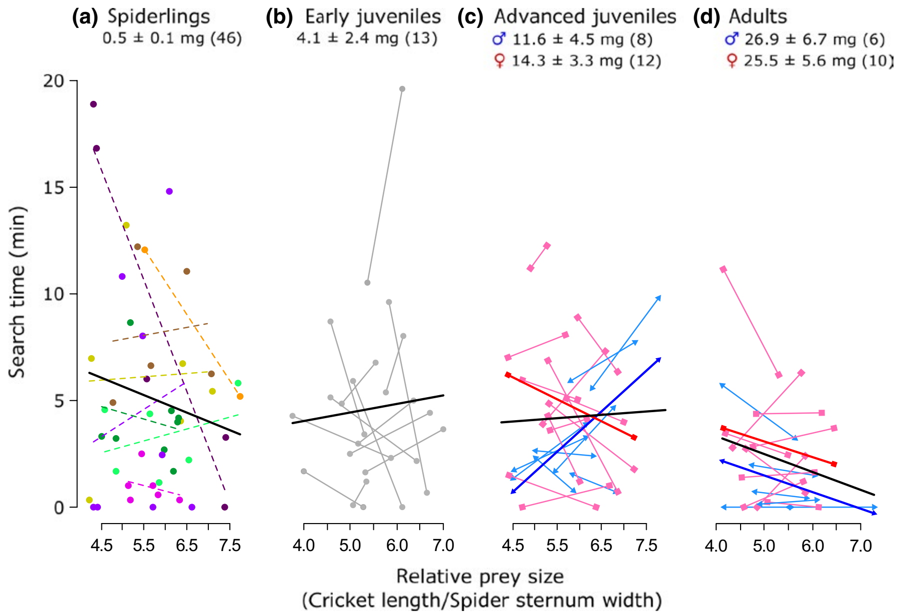

FIGURE 3 Time that spiders (*Pholcus phalangioides*) spent searching for lost prey of varying relative sizes, plotted by spider age category. Each panel displays the average mass ± *SD* for the corresponding categories, followed by sample size in parentheses. (a) Spiderlings were naïve second-instar spiders that were raised in the laboratory. They are grouped by family (eight families total, distinguished by color). Dashed lines show simple linear regressions for each family, and a solid black line shows the regression across all spiderlings. (b) Early juveniles were young spiders caught in the wild that had not yet developed sexual characteristics. Each juvenile was tested twice, and each thin gray line connects data points for an individual's two trials. A thick black line shows the simple linear regression through all points. (c) Advanced juveniles were young spiders caught in the wild that were not yet mature, but old enough for us to be able to distinguish the sexes. Thin lines connect data points from two trials for males (triangles) and females (squares). Thick colored lines represent the linear regression for the sexes, and a thick black line represents the linear regression through all the points. (d) Like panel (c), but for fully mature spiders. [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 2 Statistical results from a linear mixed model looking at the effects of maturity and prey size on search time in *Pholcus phalangioides* spiders. Maturity is broken into four categories (spiderling, early juvenile, advanced juvenile, and adult, as described in the text), and relative prey size is a continuous variable calculated from linear measurements of the prey and the spider. We included an interaction between these two variables to test for differences in prey size preferences between age groups, and we included replicate as a random variable. Replicate codes for family in spiderlings (which were all tested once) and for individual in all other spiders (which were tested multiple times). Significant and marginally significant terms indicated with bold text. Data visualized in Figure 3

Fixed effects	**DF num, DF den**	***F*-ratio**	***p*-Value**	
Spider maturity	3, 49.80	**3.11**	**0.035**	
Relative prey size	1, 93.95	**3.72**	**0.057**	
Maturity × prey size	3, 116.0	0.75	0.522	
Random effect	**Var. comp.**	**95% CI**	**% of total var.**	**Wald *p*-value**
Replicate	15,049	2,354–27,744	28.1	**0.020**

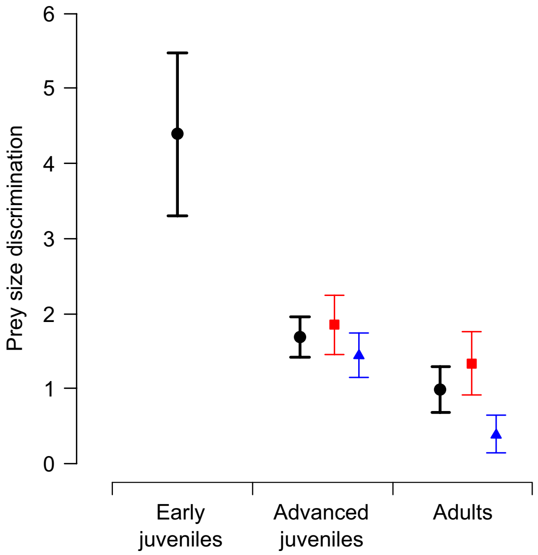

FIGURE 4 Prey size discrimination, measured as the steepness of search time ~ prey size relationships, for spiders of different ages and sexes. Data are calculated from the absolute values of the slopes in Figure 3. Black points represent the overall mean for the group. Squares and triangles represent the means for females and males, respectively, in those groups where the spiders are old enough to be told apart by sex. Error bars indicate standard error. Spiderlings were omitted from the analysis, because they were tested only once each, and so their slopes (mean ± *SE* = 1.53 ± 0.65) come from family-level regressions, not individual-level behavior, like the groups plotted here. A one-way ANOVA, with age group as the independent variable, followed by a Tukey HSD post hoc analysis, showed early juveniles to be significantly higher than the advanced juvenile and adult age groups ($F_{2,46} = 9.14$, $p < 0.05$). [Colour figure can be viewed at wileyonlinelibrary.com]

The analyses for the discrimination-related predictions yielded similar results. The search time ~ prey size slopes revealed that early juveniles had much steeper differences in their search behavior between large and small prey (Figure 4). These slopes were significantly steeper than those of the advanced juveniles and the adults ($F_{2,46} = 9.14$, $p < 0.005$).

When we looked at only those life stages for which we could tell the sexes apart (to evaluate additional predictions of the motivation hypothesis), we continued to see the significant effects of maturity (Table 3). We also found that males and females searched differently for prey of different sizes (significant prey size × sex interaction), with juvenile males preferring larger prey and juvenile females preferring smaller prey (Figure 3c; Table 3). Furthermore, these differences between the sexes varied across maturity levels (significant maturity × prey size × sex interaction), with males sharply decreasing their search behavior once reaching full maturity (Figure 3c, d; Table 3).

Adult males were the group that searched the least, and the nature of their searching was distinctly different from any other group—we observed tugging search behavior in only one male trial of 12. Search behavior in males almost exclusively manifested as exploring the bottoms of their enclosures. Compare this with mature females, who used tugs in their search behavior in 16 of 20 trials—similar to spiders in the other age groups.

Our sample size of males is relatively low and is lower than our sample of females (Figure 3), so there is a risk that we undersampled variation in males. However, based on the F-ratios in our statistical models (Table 2), we believe that the patterns we report are real. Additionally, we should address the possibility that two of the mature females we tested could have been gravid. This is based on the fact that their final masses were a bit higher (relative to their mass at capture) compared to the other mature females. However, their search behavior was no different from other females, and their exclusion from statistical models did not change any patterns. Therefore, one of two things is true: either (a) being gravid has little effect on search behavior or (b) these individuals were not gravid.

In our tests for the effects of experience, we looked at how variance in search time changed with age. We found a significant difference in variance across the four age groups, with higher variance in the younger spiders and lower variance in older spiders (Supporting Information Figure S1a). When correcting for changes

TABLE 3 Statistical results from a linear mixed model looking at the effects of maturity, prey size, and sex (and their interactions) on search time in advanced juvenile and adult *Pholcus phalangioides*. Spider identity was included as a random term because each spider was tested twice. Significant and marginally significant terms indicated with bold text. Data visualized in Figure 3

Fixed effects	DF num, DF den	*F*-ratio	*p*-Value	
Spider maturity	1, 33.18	**5.19**	**0.029**	
Relative prey size	1, 39.49	0.69	0.410	
Spider sex	1, 33.24	2.76	0.106	
Maturity × prey size	1, 37.92	1.25	0.270	
Maturity × sex	1, 33.18	0.01	0.934	
Prey size × sex	1, 39.49	**9.96**	**0.003**	
Maturity × prey size × sex	1, 37.92	**5.83**	**0.021**	
Random effect	**Var. comp.**	**95% CI**	**% of total var.**	**Wald *p*-value**
Spider identity	18,224	5,748–6,959	68.7	**0.002**

TABLE 4 A closer look at the random effect from the model in Table 2. We ran a similar model looking only at spiderlings to see whether there were family-level differences in search behavior. We used relative prey size as a fixed effect, family ID as a random term, and an interaction between the two

Fixed effects	DF num, DF den	*F*-ratio	*p*-Value	
Relative prey size	1, 8.94	0.55	0.479	
Random effects	**Var. comp.**	**95% CI**	**% of total var.**	**Wald *p*-value**
Family ID	22,187	−14,320–58,694	25.6	0.234
Family × prey size	12,254	−8,697–33,207	14.2	0.252

in the mean, we found no correlation between age and CV of search time (Supporting Information Figure S1b).

Lastly, we evaluated sources of variation in search behavior. There was a high degree of individual variation (as seen in the identity term in Table 3 as well as Figure 3b–d), whereas family was not a significant source of variation (Table 4).

4 | DISCUSSION

We tested hypotheses about the relative roles of capability and motivation in ontogenetic change in memory of captured prey in a web spider. The first hypothesis was that spiderlings would be limited in their ability to evaluate and remember lost prey, and so would spend less time searching for it compared to older spiders. Our results refute this, as spiderlings searched just as long or longer than older spiders. This hypothesis also predicted that younger spiders would be less discriminating between prey of different sizes. This was also refuted by the fact that the early juveniles had the steepest search time vs. prey size slopes. Thus, early developmental stage was neither a limiting factor in the formation of memories of captured prey nor a limiting factor in the use of those memories in the regulation of searching efforts. This is rather remarkable, considering that the brains of the adults are over 5.5 times the size of the brains of spiderlings (based on the average masses that we measured and the spider brain-volume ~ body-mass relationship described by Quesada et al. (2011). The results from this study also indicate that previous experience with prey is not necessary for a spider to search for lost prey (as none of the spiderlings had ever caught prey before). The lack of trial sequence effects on the spiders' behavior suggests that short-term experience was not a confounding variable in our experiment. Additionally, both the decrease in search time variance with age and the relatively flat CV across age groups suggest that long-term experience does not have a strong effect on search time, relative to the other effects that we tested.

The second hypothesis was that as spiders approached adulthood and their growth slowed down, their motivation to recover prey would decrease. This was supported by our results, as fully mature spiders spent less time searching than spiders in other groups. The motivation hypothesis also predicted the greatest amount of prey size discrimination among young spiders and the least among adults. This too was supported by our results.

The motivation hypothesis made additional predictions about the effects of sex on search behavior. Males grow more quickly than females, and so it is predicted that they would place a premium on larger prey. This prediction was supported by our data, specifically by the significant interaction between prey size and sex in advanced juveniles (Figure 3). It is unclear precisely when this difference first appears. It could be present in the early juveniles—certainly other sex-based differences in behavior, like activity level or prey-capture rate, are known to arise early on for this species (Hoefler, Keiser, & Rypstra, 2010). If so, it might help explain the high variation in slopes across individuals in this group (Figure 3b).

Finally, this hypothesis predicted a decrease in search behavior for mature males, whose priorities shift heavily toward seeking a mate after reaching maturity. The lower search time that we observed in mature males, plus their qualitative change in search behavior (the reduced use of tugging), support this prediction. Males still readily attacked prey, and there is nothing about their morphology that would prevent them from being able to tug, so it seems that they simply have a reduced interest in recovering prey.

Looking at individual differences in search behavior, we find that nearly half of the spiders in our study actually preferred prey from the lower end of the range of sizes we tested (i.e., had negative slopes). The high amount of individual variation in search time, including in inexperienced spiderlings, suggests a diversity of prey size preferences across individuals on which selection may act to shape prey recover efforts.

In short, when it comes to prey-searching in spiders, it is motivation, not capability, that drives ontogenetic change in behavior. Without a doubt, there is some point in a spider's development when it is incapable of searching, but as soon as spiderlings disperse from their mother's web, they essentially fill the same ecological niche as adults, and they perform many of the same actions—they build webs, capture prey, and seek to recapture it when lost. Analyzing the causes of ontogenetic variation in behavior is greatly facilitated with species like this, where direct comparison of similar behavior and tasks is possible. For example, many species of fish allow ontogenetic studies across stages that are ecologically equivalent in many respects, with newborns being miniature adults (Agrillo & Bisazza, 2017). Even in such cases, interesting cognitive limitations may be revealed in younger animals. Guppies, for instance, are safest when swimming in large shoals, but young guppies are not as good as adults at evaluating shoal size; consequently, they face the greatest risk of making wrong shoaling decisions at the most vulnerable stage of their lives (Petrazzini, Agrillo, Piffer, & Bisazza, 2014). Other animals may reach the state of adult-equivalence at older ages and yet still face the risk of performing suboptimal behaviors due to both cognitive limitations and lack of experience. For example, in ravens, caching behavior is affected both by their sense of object permanence and their experience of conspecifics stealing their prey (Bugnyar, Stöwe, & Heinrich, 2007).

In conclusion, the interplay of capability and motivation along ontogeny varies in multiple dimensions between species, and this variation requires explanation. We suggest that progress can be made using behavioral assays to test hypotheses about the evolution and expression of cognitive phenotypes (Mendelson et al., 2016).

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ETHICAL APPROVAL

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

DATA AVAILABILITY

Data analyzed for this study are included in the supplemental material for this article (Supporting Information Data S1).

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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