



## Behavioural syndromes and their fitness consequences in a socially polymorphic spider, *Anelosimus studiosus*

JONATHAN N. PRUITT\*, SUSAN E. RIECHERT\* & THOMAS C. JONES\*†

\*Department of Ecology and Evolutionary Biology, University of Tennessee, Knoxville

†Department of Biological Sciences, East Tennessee State University, Johnson City

(Received 14 December 2007; initial acceptance 3 February 2008;

final acceptance 14 March 2008; published online ■ ■ ■; MS. number: A07-20027)

The temperate comb-footed spider, *Anelosimus studiosus*, shows a social behaviour polymorphism: individuals may defend asocial nests against intrusion by conspecifics or cooperate with them in multifemale nests. A suite of behavioural traits, including response to predators and prey, degree of superfluous killing, exploratory behaviour and general level of activity, was examined in laboratory trials to: (1) establish the extent to which these traits are correlated with social phenotype and (2) explore the potential adaptive value of this trait suite to the respective asocial and social phenotypes. Populations from the two latitudes studied, 26° and 36°, showed similar suites of correlated traits or syndromes. The individuals tested generally fell into two groups in the behavioural trials, although the asocial individuals from the mixed-phenotype population at 36° latitude were intermediate with respect to some of the measures. Individuals scored as 'social' generally showed less aggressive behaviour towards predatory cues, were less responsive to prey encountering their webs, showed little evidence of superfluous killing of prey and showed lower levels of activity than did most individuals scored as 'asocial'. These results suggest linkage or pleiotropy between social behaviour and these traits. The behaviour of individuals in staged, mixed-phenotype feeding pairs indicated that social individuals may suffer fitness consequences in polymorphic populations. These results are discussed with respect to the stability and dynamics of the respective phenotypes in polymorphic populations.

*The Association for the Study of Animal Behaviour. Published by Elsevier Ltd.*

**Keywords:** *Anelosimus studiosus*; behavioural syndrome; correlated traits; polymorphism; social spider; social structure; sociality

Sih et al. (2004) defined behavioural syndromes as suites of behavioural traits that are correlated across contexts and situations. Behavioural syndromes are of interest to evolutionary biologists because they have been shown to have fitness consequences (see Sih et al. 2004 for a recent review). Although suites of correlated traits may be adaptive in some cases (Cheverud 1996), they also have been shown to constrain adaptation (Arnqvist & Henriksson 1997; Johnson & Sih 2005). We examined the extent to which behavioural syndromes might facilitate or limit the shift of species from selfish, individualistic replicators to cooperatively social reproducers in which individual reproductive success is not maximized.

*Correspondence:* J. N. Pruitt, Department of Ecology and Evolutionary Biology, University of Tennessee, Knoxville, TN 37996-1610, U.S.A. (email: jpruitt6@utk.edu).

Recent advances in our understanding of the evolution of sociality have come from systems that have variable social structures (e.g. studies involving the social structure variability observed in halictid bees: Michener 1990; Seger 1991; Crespi 1996; Danforth 2002; Richards et al. 2003). The comb-footed spider, *Anelosimus studiosus* (Araneae, Theridiidae) is similar to some halictid bee species in that it shows a social behaviour polymorphism within populations as well as variable social structures along a latitudinal gradient (Jones et al. 2007; Riechert & Jones, in press). Adult female comb-footed spiders adopt one of two strategies: (1) defence of asocial nests against intrusion by adult conspecifics or (2) cooperation with other adult females in multifemale nests. In southeastern North America, multifemale nests are first observed at 30° latitude and increase in frequency to 14% at 36° (Riechert & Jones, in press). Jones et al. (2007) developed a model that predicts that the social phenotype represents a bet

hedge in colder environments where there is an increasing probability that a mother will die before its brood has reached independence. In the event of a female's death, other resident females will care for its offspring. Empirical and experimental evidence supporting this model is presented in Jones et al. (2007). Because spiders are diploid, they lack the haplodiploid inheritance pattern present in the halictids and other social hymenopterans that facilitates social evolution. All female spiders also retain their reproductive totipotency, similar to cooperative vertebrate taxa. Thus, comb-footed spiders may provide a more general model for social evolution in the animal kingdom than would a social hymenopteran possessing an unusual inheritance pattern and the morphological and behavioural caste systems it favours.

We investigated the behavioural correlates or behavioural syndrome associated with the transition from an asocial to a social existence in comb-footed spiders. Behavioural syndromes have in the past been referred to as suites of correlated traits in the biological literature and as personalities in the psychology literature. Riechert has produced a series of papers on a suite of correlated traits associated with aggressiveness in the American desert spider, *Agelenopsis aperta* (Araneae, Agelenidae). Although all *Ag. aperta* show an asocial territorial social structure, local populations show ecotypic variation in this set of traits: an aggressive arid-land phenotype versus a nonaggressive riparian phenotype (see Riechert et al. 2001 for a recent review). The phenotypically correlated traits include agonistic behaviour, territory size, antipredator behaviour, superfluous killing, latency to attack prey, diet breadth and sexual cannibalism. Riechert & Maynard Smith (1989) suggest that pleiotropy underlies the phenotypic correlations existing among these traits and presented a two-gene complex model that explains trait-value variation between arid-land and riparian phenotypes and different genetic classes produced in breeding experiments. This 'behavioural syndrome' produced maladaptive phenotypes in *Ag. aperta* when population mixing occurred between phenotypes adapted to different competitive and predation environments (Riechert et al. 2001). If a similar syndrome is also present in comb-footed spiders, it is possible that the trait correlations will pose costs to the transition from an asocial to a social structure and perhaps explain the fact that the majority of nests of this species at all latitudes are solitary.

In this study, we examined most of the behavioural traits examined by Riechert in *Ag. aperta* in the *An. studiosus* system. We tested individuals from two different latitudes for their social structure phenotype and then examined the extent to which the attraction to conspecifics needed to become social is correlated with an overall loss of aggression in other contexts: foraging behaviour, degree of wasteful/superfluous killing, exploratory behaviour and antipredatory behaviour. Historically the evolution of sociality has been framed in a 'costs versus benefits' framework, which typically considers foraging and antipredatory costs and benefits (Raffa & Berryman 1987; Rypstra & Tiley 1991). We applied this framework to the behavioural syndrome identified for comb-footed spiders and, thus, identified costs to becoming social that are not generally considered.

## METHODS

### Study Species

*Anelosimus studiosus* inhabits understory vegetation in deciduous forests throughout much of eastern North America. This species has an affinity for water and, thus, the densities of nests of both phenotypes are highest on small tree and shrub branches at the edge of bodies of water.

Based on work on comb-footed spiders completed at a subtropical latitude (26°) in the southeastern United States, Brach (1977) classified them as subsocial, in which the mother shows extended care of its offspring but defends its nest against intrusion by other adult females. Avilés (1997) has suggested that such extended offspring care, rare to spiders, is a precursor to the evolution of social behaviour, which is limited to approximately 50 species inhabiting tropical areas. Comb-footed spiders are unique among social spider species, in that individuals showing the cooperative brood care, foraging and web-maintenance characteristic of the cooperatively social spiders live in temperate habitats at higher latitudes (i.e., 30° and above; Furey 1998; Jones et al. 2007). Riechert & Jones (in press) have quantified the shift in frequencies of the asocial (subsocial) and social phenotypes with increasing latitude and in various temperature environments within the same latitude. This is the first study that extends the analysis of the behavioural polymorphism beyond social behaviour to other fitness-linked behavioural traits, as well as examining the nature of the interactions that might occur between asocial and social individuals in mixed-phenotype populations.

### Collection and Lab Maintenance

We collected penultimate spiders from two populations separated by 10° latitude, the Everglades in south Florida (26° latitude) and a Tennessee Valley Authority-managed river system in east Tennessee (36°). We collected spiders along pre-established transects by covering webs with plastic bags and cutting off the stems of the plants used as structural support for the web. This technique ensures that females that drop from the web because of the disturbance are still collected. We housed all spiders individually in clear, plastic containers (59 ml) and provided tangled poultry netting as a support structure on which webs could be built. We housed all individuals in the laboratory between 22 and 24 °C and fed them weekly a combination of 1-week-old crickets, fruit flies (*Drosophila melanogaster*) and termite workers. We used females in test trials when they reached maturity, with all tests completed on an individual within 4 weeks of its maturation. To reduce stress, a minimum of 24 h elapsed between tests for individual spiders. Trials were completed in the order presented here. Except where noted, we ran all tests pertaining to the traits potentially correlated with social phenotype with 30 individuals scored as asocial from 26°, 20 individuals scored as asocial from 36°, and 26 individuals scored as social from 36° latitude.

## Tests

### *Interindividual distance*

We assigned behavioural phenotype using a distance test after Riechert & Jones (in press). We marked two spiders with fluorescent powder and placed them in a clear, square plastic container ( $6.5 \times 6 \times 2.5$  cm). We placed both spiders in the central portion of the container and gave them 24 h to settle and construct webs. We assigned a spider to the asocial versus social phenotype based on the distance between the two spiders at the end of this 24 h period. *Anelosimus studiosus* generally prefers to settle in the corners of these containers. Two spiders were typed as belonging to the social, aggregative phenotype if, after 24 h, they settled in the same corner. Conversely, spiders that placed themselves at a distance from each other in either opposite or adjacent corners were typed as asocial in nature. (Note that in Riechert & Jones, in press, spiders settling in adjacent corners were not assigned to either phenotype.) It is possible in the distance trials that a social individual might score as asocial because it had been repelled by the asocial spider it was paired with. To prevent this error in phenotype assignment, we completed a second trial on all individuals scored as asocial in which each spider was paired with an individual that had previously scored as social. In addition to recording the relative positions of the two interacting individuals with respect to corners of the arena, we measured the distance in millimetres between them using a digital calliper. We used these interindividual distance scores in calculating the correlations between social phenotype and other behavioural trait scores. We used the coarser corner position assignment merely in typing individuals as to social phenotype. By using the interindividual distance scores in the correlation analyses we allowed for the possibility of intermediate phenotypes or even a continuous distribution of asocial–social behaviour.

To confirm whether interindividual distance was a repeatable measure we tested 20 pairs of individuals in four different distance trial runs with a minimum interval of 24 h between trials. In all, we scored 65 individuals for social phenotype from the 26° latitude site and 211 from 36°. Individuals assigned to social phenotype from these trials formed the pool from which we selected test subjects at random for the following behavioural trials.

### *Superfluous killing*

The superfluous killing trials follow the protocols developed by Riechert and Maupin to test for aggressiveness towards prey in several different web-building spider species (Riechert & Maupin 1998; Maupin & Riechert 2001). We performed trials after 7 consecutive days without feeding or any other testing of the spiders. We introduced termites, one at a time, at 3-min intervals to each test spider's web. Additional time was provided if the spider required more time to subdue a particular prey item. We recorded the reaction of the test spider to each prey item entering the web: whether it failed to respond to the prey, retreated from it, or attacked it. A trial continued until the test spider failed to attack two consecutive prey

items. At the end of the observation period, we removed all rejected prey items from the web and gave the spider 24 h to feed. At the end of 24 h, we inspected the captured prey items visually and assigned them to one of the following categories: partially consumed, fully consumed, or uneaten. We verified inspections by comparing weight determinations for remaining prey to a regression relationship established from mass loss determinations made on termites ( $N = 35$ ) after 24 h of maintenance in the absence of potential feeding by comb-footed spiders. We classified prey items as fully consumed if they weighed less than 10% of their original body mass, partially consumed if between 10 and 25% and uneaten if greater than 25%. If a test spider was still feeding on prey after 24 h, we checked it again after an additional 24-h period and identified the feeding classification of the prey at that time.

### *Prey attack sequence*

Three days after a maintenance feeding, we offered a spider a single prey item in its web. We placed the prey 2.0 cm distant from the spider and recorded the capture sequence, timing all behaviour using a stopwatch. We recorded (1) the time taken to orient to the prey item or first intention movement shown by the spider ('latency to first response'), (2) the time from first response until contact with the prey ('latency to attack'), (3) the location of the first bite as evidenced by recoil of the prey as contact is made (score: 1, abdomen; 2, thorax; 3, head) and (4) the number of bites issued before the first feeding bout.

### *Antipredator*

This test is modelled after that applied by Riechert & Johns (2003) in scoring the behavioural phenotype of the desert spider *Ag. aperta*. In this test an individual is placed in a 36-cm circular track and the distance to its settling (no movement for a minimum of 30 s) is recorded. At this time, the individual is touched at the rear with the end of a probe and its response recorded as to action taken and distance travelled. After the spider settles, this procedure is repeated for a front prod. Whether the front or the rear prod is issued first alternates between test subjects.

Because of the diversity of possible antipredatory responses, we scored individuals based on their behaviour, from 0 to 1, 1 being highly aggressive towards predators and 0 being completely submissive. We assigned a single score for the most aggressive behaviour shown by an individual from the front and rear cue. We assigned scores as follows: 1, turn attack; 0.9, attack; 0.8, raise front legs; 0.7, lay silk; 0.6, walk 0–5 cm; 0.5, walk 5–15 cm; 0.4, walk 15 cm or greater; 0.3, run 0–5 cm; 0.2, run 5–15 cm; 0.1, run 15 cm or greater; and 0, huddle. Huddling behaviour is characterized by the spider tightly drawing in its legs as in a death feint.

### *Exploration and boldness*

We placed the test spider in an unfamiliar, gridded container ( $6.5 \times 6 \times 2.5$  cm). When we closed the lid, 100% of the individuals initially went into a huddle posture. We observed the subsequent behaviour of the test

subject over a 5-min period. We recorded the following measures: (1) time lapsed between the initial positioning in a huddle on release and the first subsequent movement as measured by stopwatch, (2) number of times the spider moved in a 5-min period following first movement out of the huddle (activity), and (3) total distance the spider traversed in this 5-min interval (exploration). For the activity portion of this trial, we scored movements as independent if they were separated by 10 or more seconds of quiescence.

### Group feeding

We performed group-feeding trials to determine the fitness consequences of possessing the asocial or social phenotype in competitive interactions. Because no individuals from 26° showed the social phenotype, only individuals from 36° represented the social phenotype in the group-feeding trials. After a week of starvation, we marked one asocial and one social spider individually with fast-drying enamel paint and placed them in a web made by a third spider. We had removed the spider that made the web 1 week earlier to reduce chemical signatures on the web ( $N = 25$ ). After 15 s, we placed a termite equidistant between the two spiders being tested and recorded the behaviour of each individual in the order in which it occurred until the prey item was consumed. We also recorded (1) which spider reacted to the prey item first, (2) which spider reached the prey item first, (3) which spider ultimately captured and subdued the prey and (4) a time course of individual feeding on the prey from the start of feeding to abandonment of the prey item by both individuals. Individuals that had scored as social were paired in the control runs of this experiment ( $N = 25$ ).

## RESULTS

### Interindividual Distance Measure of Phenotype

The position results of the interindividual distance trials identified all individuals from the 26° latitude site ( $N = 65$ ) as asocial. The population from the site at 36° was polymorphic: 81% of the individuals scored as asocial and 19% as social ( $N = 211$ ). From the between-individual distances measured we learned that social behaviour may be a quantitative trait as some individuals scored as intermediate between social (in same corner) and asocial (in different corners). Repeated tests run on a subset of the pairs of individuals scored indicated that social phenotype shows a high repeatability ( $r = 0.64$ ,  $N = 20$ ).

### Tests for Correlated Traits

Except where noted, we used Statistical Analysis Software 9.1 (SAS Institute 1998) in all tests on these data. Nonparametric correlations (Spearman) with Bonferroni correction completed within the two populations identified a number of behavioural measures related to exploration, foraging and antipredatory contexts that showed

significant correlations with interindividual distance scores (Table 1).

We performed a nonmetric multidimensional scalar ordination (MDS) with Euclidian distance measure (Primer-E software; Primer-E 2001) to explore spider behaviour in latitude and social phenotype space. The analysis produced a global  $r$  of 0.31 significant at  $P < 0.001$  and a low stress statistic of 0.01, suggesting an adequate fit. The results of this analysis indicate that both latitude and social phenotype significantly affect the nature of individual behaviour, as all pairwise Analysis of Similarity (ANOSIM) comparisons among the three categories of individuals (asocial 26°, asocial 36° and social 36°) differed significantly at an  $\alpha$  of 0.05.

To evaluate further where the differences in trait values lie between social and asocial individuals, we partitioned the sample scores from each behavioural measure that showed a significant correlation ( $\alpha = 0.05$ ) into three groups: asocial individuals from 26° latitude, asocial individuals from 36° latitude and social individuals from 36° latitude. We performed a MANOVA including all significant characters from the analysis above for all three groups (MANOVA: Wilk's  $\lambda = 0.06$ ,  $F_{18} = 22.34$ ,  $P < 0.001$ ). We then completed a series of univariate ANOVAs with Tukey tests for each trait independently to determine which classes of individual vary from one another for the particular trait measure.

The results of the ANOVAs for each trait are summarized in Table 2 and the Tukey groupings are noted in Fig. 1. Inspection of Fig. 1 indicates that there is a significant distinction between the behaviour of asocial individuals from 26° latitude and that of social individuals from 36° latitude with respect to all of the behavioural trait scores.

**Table 1.** Correlations between behavioural assay scores indicated and interindividual distance scores

Test and measurement	26° ( $df=28$ )	36° ( $df=44$ )
<b>Antipredator</b>		
Response to front prod	0.32	0.47*
Response to rear prod	0.50*	0.48*
<b>Prey attack sequence</b>		
Latency of first response to prey	-0.57*	-0.63*
Latency to attack prey	-0.80*	-0.70*
Number of bites to prey during attack	0.10	0.32
Location of bites to prey item	0.22	0.31
<b>Exploration</b>		
Latency of return to activity following huddle	-0.56*	-0.47*
Activity level (number of times moved)	0.47*	0.44*
Total distance moved	-0.01	0.05
<b>Superfluous killing</b>		
Number of prey killed	0.69*	0.67*
Number of prey killed and left uneaten	0.63*	0.65*

Spearman correlation with Bonferroni adjustment for populations at 26° ( $N = 30$ ) and 36° latitude ( $N = 46$ ).

\*Significant at  $\alpha = 0.05$ .



**Table 2.** Summary of univariate ANOVAs performed for each trait

Test and measure	$F_{2,74}$	$P$
Interindividual distance score		
Distance score	72.41	<0.001
Antipredator		
Response to front prod	16.41	<0.001
Response to rear prod	12.91	<0.001
Latency to return to activity following huddle	33.4	<0.001
Superfluous killing		
Number of prey killed and left uneaten	14.22	<0.001
Number of prey killed	13.28	<0.001
Prey attack sequence		
Latency of first response to prey	12.62	<0.001
Latency of attack towards prey	22.09	<0.001
Exploration		
Activity level (number of times moved)	4.58	<0.01

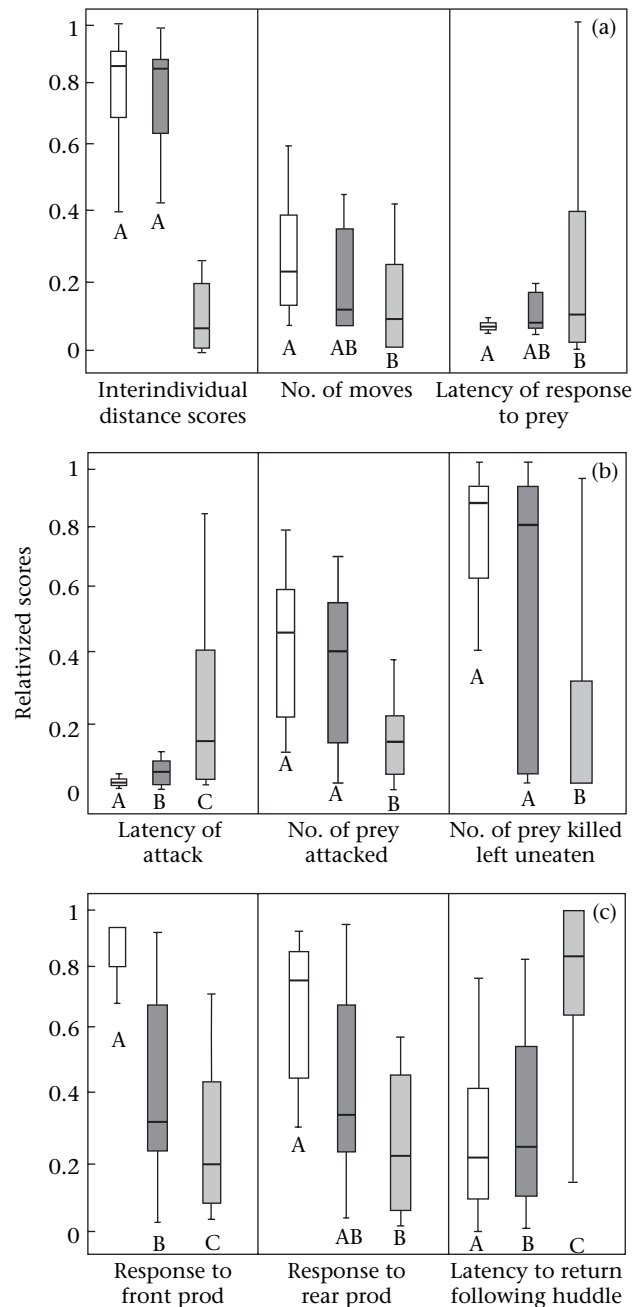
Results for each behaviour are summarized for three groups: 26° asocial ( $N = 30$ ), 26° asocial ( $N = 20$ ) and 36° social ( $N = 26$ ).

Thus, asocial individuals from 26° latitude made more moves and travelled greater total distances in the exploration trials than did social individuals; they were more aggressive with respect to all the foraging measures, from first response to prey to number of prey captured and left uneaten in a feeding bout, and they showed more aggressive responses to predatory cues and returned more quickly to a normal stance after huddling.

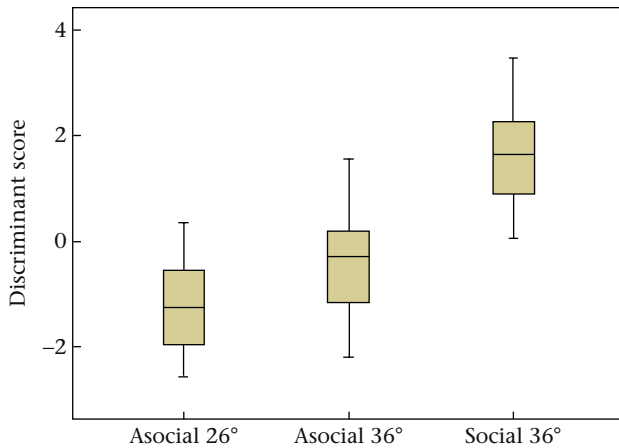
The distinction between asocial individuals from 36° latitude and the social phenotype from the same latitude was not as consistent across traits as between asocial spiders from 26° latitude and social individuals from 36° (Fig. 1). Asocial individuals from 36° latitude, however, differed significantly from social individuals from the same latitude in their exhibition of shorter latencies to attack prey, greater levels of superfluous killing, more aggressive antipredator responses towards front prods and shorter latencies to return to activity after positioning in a huddle.

We performed a discriminate analysis with the goal of selecting traits that show low within-phenotype variation but high between-phenotype variation to understand better the relative behavioural trait positioning of the three classes of comb-footed spiders (asocial 26° and 36°, social 36°). We pooled data from both populations in this analysis. The discriminant function shown in Fig. 2 was significant (Wilk's  $\lambda = 0.48$ , canonical correlation = 0.74,  $P < 0.001$ ) and successfully classified the social phenotype of a female in 78% of cases. Note also from Fig. 2 that the asocial individuals from 36° latitude are classified as intermediate between the social individuals at the same latitude and the asocial individuals from 26° latitude. Although intermediate, the distribution of this class is biased towards the 26° latitude asocial class.

The weighted values of behavioural measures that contributed to the distribution of the three classes are presented in Table 3. The high numbers of prey killed and those killed but left uneaten in the superfluous killing trials were the strongest discriminating characteristics of the asocial



**Figure 1.** The distribution of the behavioural scores for three classes of temperate comb-footed spider, asocial phenotype from 26° latitude (white bars), asocial phenotype from 36° latitude (dark grey bars) and social phenotype from 36° latitude (light grey bars), determined by performance in interindividual distance trials. Values presented for each behavioural measure were relativized by dividing all scores within the trial type by the highest score achieved. Filled vertical bars represent 25% quartiles, medians are demarcated by the horizontal lines within bars and vertical lines reflect the full range of scores for the measure indicated. Different letters indicate there were significant differences between classes at  $P < 0.05$  using Tukey tests. (a) Distance, exploration and prey attack sequence test results. (b) Antipredator behaviour test results. (c) Latency to attack a prey item from prey sequence trials and voracity and the number of prey killed and left uneaten from the superfluous killing trials.



**Figure 2.** The distribution of individual behaviour in discriminate space (y axis) of the asocial phenotype from both latitudes and the social phenotype from 36°. See Table 3 for the relationships of various behavioural measurements to the discriminant score distribution shown here.

phenotype, whereas the long latencies to return to activity following a predatory cue and less aggressive response to a front prod from the antipredator trials were the major contributors to discrimination of the social phenotype.

### Phenotype Performance in Paired-Feeding Contexts

Following Bonferroni correction, the asocial female was statistically the first individual to respond to the prey item ( $\chi^2_1 = 18.7$ ,  $P < 0.01$ ), make contact with it ( $\chi^2_1 = 13.4$ ,  $P < 0.01$ ), and subsequently capture it ( $\chi^2_1 = 21.7$ ,

**Table 3.** Behavioural measures contributing to discriminant scores assigned to individuals showing asocial versus social phenotypes (see Fig. 1)

Test and measure	Standardized discriminant function 1 coefficients
Antipredator: response to front prod	-0.57
Superfluous killing	
Number of prey killed and left uneaten	-0.22
Number of prey killed (voracity)	-0.15
Prey attack sequence	
Location of bites to prey item	-0.03
Latency of first response to prey	0.08
Exploration: activity level (number of times moved)	0.11
Prey attack sequence: latency of attack towards prey	0.27
Exploration: total distance moved	0.37
Prey attack sequence: number of bites to prey during attack	0.40
Antipredator	
Response to rear prod	0.65
Latency to return to activity after huddle	0.99

Coefficients are listed from lowest to highest.

$P < 0.01$ ) in feeding trials involving an asocial and a social individual (Table 4). The asocial member of the pair also significantly monopolized feeding on the captured prey in 97% of the trials, significantly more than did the social individual ( $\chi^2_3 = 63.0$ ,  $P < 0.0001$ ). The behaviour of the two social spiders, designated as individuals a and b in Table 4, was not significant for any measure (e.g. for monopolizing time:  $\chi^2_3 = 2.38$ ,  $P < 0.50$ ). In fact, individuals shared feeding on the single termite in 70% of the trials compared to 0% in the mixed trials involving an asocial individual paired with a social one.

## DISCUSSION

### Social Polymorphism and Latitude

All individuals collected at 26° latitude tested as asocial, whereas the population from 36° latitude was polymorphic in nature, with 19% of the spiders testing as social and 81% as asocial. This percentage of social individuals is slightly higher than that reported in a previous study: Riechert & Jones (in press) obtained a high estimate of 14% social individuals. In that study, 50 nests from each of two large local populations were collected at random along a 500-m transect for testing. We applied the interindividual test to a much larger sample of individuals in this study and chose our test subjects from known local polymorphic populations. Phenotype assessment for a large number of populations in east Tennessee is planned to determine the degree to which the representation of the two phenotypes varies among populations and over time as well as to examine the factors that might underlie such variation.

### Behavioural Syndrome

Social tendency in comb-footed spiders is phenotypically correlated with a number of behaviour patterns associated with functionally different contexts, including antipredator behaviour, foraging behaviour and general level of activity. Social individuals were generally less aggressive than asocial individuals, in that they were slow to respond to and attack prey, showed a lower percentage of superfluous killing, were less active, were less aggressive in their response to predator cues and showed a longer latency to return to activity after a huddle response.

Although the asocial individuals from 36° scored as aggressive as the asocial individuals from 26° in the interindividual distance trials and for traits associated with superfluous killing (i.e. number of prey killed and number of prey captured but left uneaten), MDS and discriminant analysis results identified this class as being intermediate in behaviour. The conservative Tukey univariate test results further identified the 36° asocial class as not significantly different from the 26° asocial nor from the 36° social class with respect to several traits and as significantly different (being intermediate) from both 26° asocial and 36° social classes in others. Possible explanations for the observed performance of asocial 36° latitude individuals include any one or a combination of the following possibilities: (1) a larger sample of individuals showing different

**Table 4.** Comparison of individual behaviour in paired-feeding trials involving mixed phenotype versus same phenotype in temperate comb-footed spiders

Behaviour	Treatment	df	Individuals
First response to prey item	Mixed	1	Asocial 0.86 versus social 0.14*
	Control	1	Social <sup>a</sup> 0.40 versus social <sup>b</sup> 0.60
First touch prey item	Mixed	1	Asocial 0.81 versus social 0.19*
	Control	1	Social <sup>a</sup> 0.32 versus social <sup>b</sup> 0.68
Capture prey item	Mixed	1	Asocial 0.89 versus social 0.11*
	Control	1	Social <sup>a</sup> 0.36 versus social <sup>b</sup> 0.64
Monopolize feeding	Mixed	1	Asocial 0.97 versus social 0.03*
	Control	1	Social <sup>a</sup> 0.16 versus social <sup>b</sup> 0.0

Values represent proportion of trials. Superscript a and b designate the two control social spiders.

\*Significant frequency difference at  $\alpha = 0.05$  after Bonferroni correction from chi-square test.

interindividual scores is needed; (2) the individual traits measured vary in the degree to which they are correlated with social phenotype because of different underlying genetic mechanisms; (3) if assortative mating with respect to social phenotype exists at all, it is incomplete; (4) the selection regime that favours a decrease in aggressiveness in 36° females has facilitated the evolution of the social phenotype. We are currently engaged in a breeding study with the cross-fostering of clutches, which should provide insight into the results we obtained here.

A behavioural syndrome similar to the one described in comb-footed spiders, *An. studiosus*, was described for the desert spider, *Ag. aperta*, and attributed to a hormone regulatory system (Maynard Smith & Riechert 1984; Riechert & Maynard Smith 1989). Although different behavioural contexts may have unique context-specific genes that influence them, hormones can affect an organism's general tendency to be nonaggressive or aggressive on some aggression continuum, thereby influencing trait values of a multitude of behaviour patterns. Maynard Smith & Riechert (1984) first developed a model based on conflicting hormones, one controlling levels of aggression or tendency to attack and the other fear or tendency to flee, to explain the contest behaviour of this spider and the size of territories it demanded. In a series of breeding experiments Riechert & Maynard Smith (1989) further showed that the fear component of an individual spider's aggressiveness is a quantitative autosomal trait, and the aggression component is sex-linked with only a single gene or a small number of genes involved. Ultimately, this model adequately explained between-population variation in a suite of traits including agonistic, foraging, antipredator and mating contexts (see, in addition to references cited above, Hedrick & Riechert 1989; Riechert & Hedrick 1993; Riechert et al. 2001). The influence of hormones on correlated behaviour extends beyond spiders. For instance, the hormone testosterone has been implicated in generating trade-offs in levels of aggression in male vertebrates (Hau 2007).

When behavioural characters are linked in some way, selection must operate on the multiple traits simultaneously and balance the influences from each, thereby limiting the optimization of behaviour. As such, an aggression syndrome might impose potential costs to the evolution of sociality and may explain why sociality

is so rare in the Araneae. For instance, if decreased aggressiveness towards conspecifics is a necessary condition to cooperative living, and agonistic behaviour is correlated with foraging behaviour and antipredatory behaviour, prey attack and feeding rates will be lower for social individuals as well as potential nest defence. These would be fitness costs that would need to be offset by the benefits of cooperative living.

Spiders by their nature are asocial aggressive animals, and sociality in spiders is extremely rare. If aggression levels are under hormonal control, as was indicated for *Ag. aperta* (Riechert & Hedrick 1993), the fishing spider *Dolomedes triton* (Pisauridae) (Johnson & Sih 2005) and now the theridiid spider *An. studiosus*, then selection for increased aggressiveness in a functional context such as foraging may well prevent a species from evolving the necessary tolerance towards conspecifics to become social. Spiders show a number of traits that facilitate a feast and famine existence, including a sit and wait foraging strategy, external digestion and an extensible abdomen for taking large meals, and polyphagy. High levels of aggressiveness towards prey and competitiveness towards conspecifics are traits that also fit this spider niche (see review in Riechert & Harp 1987).

A role for behavioural syndromes in the evolution of sociality is not limited to spiders. For example, in an artificial selection experiment on cyprinodontid fish, Ruzante & Doyle (1991, 1993) found that increased shoaling tendency was correlated with a decrease in aggression during foraging. These findings suggest that aggression syndromes linking grouping behaviour with other aspects of aggression might be influential in many taxa. Socially polymorphic systems make for an ideal starting place for such investigations.

### Group-Feeding Trials

The behavioural syndrome results described for comb-footed spiders in this study link tolerance/attraction to conspecifics with a slower reaction time to prey, a longer latency of attack and a lower general level of activity than that shown by the asocial phenotype of the species. As expected from the results of the correlated trait tests, asocial individuals dominated mixed-phenotype feeding

trials. Asocial individuals were more sensitive to movement in the web, were quicker to react to that movement and dominated feeding rights. No individual dominated feeding in the control trials involving two social individuals. Thus, when the two phenotypes are in competition for prey in the natal nests and in coalescent webs, the asocial phenotype may deprive social individuals of feeding opportunities and even attack and injure them in the latter case.

These results suggest that the persistence of the asocial phenotype in a mixed population poses a serious threat to the success and stability of social groups, especially when the social phenotype is at low concentrations. Even in large nests composed of mostly social individuals, the presence of asocial individuals could result in a disproportionate distribution of resources skewed in favour of the asocial phenotype. These dynamics resemble social systems in social parasites (Johnson & Herbers 2006). They differ in that the damage to the social spider group is not limited to siphoned resources; asocial females occasionally prey on social females in multifemale webs maintained in the laboratory (Pruitt, personal observations).

We have shown that the tolerance of conspecifics needed to become social is phenotypically correlated with a suite of other functionally dissimilar behaviours in comb-footed spiders and that these linked traits have costs (maladaptive spillover) when social individuals compete for resources with nonsocial individuals. Social individuals are less active, more fearful, and less aggressive than asocial individuals. This predisposes them to be exploited and even cannibalized by nonsocial individuals in mixed-phenotype clutches in the home nest.

Riechert (1985) noted similar pleiotropic effects in the African social spider, *Agelena consociata*, though this spider lacks phenotypic variation in social behaviour and all interactions were with other social individuals. Riechert suggested that correlated traits such as these would hinder the evolution of sociality in spiders and that high levels of inbreeding might be needed to overcome it. The comb-footed spider system is an important test system, because social structure polymorphism is a within-population phenomenon and even mature social individuals are likely to encounter asocial individuals, because social individuals tend to extend silk pathways from their nests to those of other comb-footed spiders (personal observations). Together, correlated behaviour and intraspecific competition with the asocial phenotype present strong, and unexplored, obstacles to the evolution and stability of nascent sociality, something that can be studied in this spider system.

### Acknowledgments

We thank Sarah Duncan for her assistance in data collection, Ben Fitzpatrick, Jim Fordyce and especially Jen Schweitzer for her patience and guidance while assisting with the MDS procedure. Finally, we thank two anonymous referees for their helpful comments, which greatly improved the quality of this paper. This research

was supported by Grant 0235311 from the Animal Behaviour Program of the National Science Foundation.

### References

- Arnqvist, G. & Henriksson, S. 1997. Sexual cannibalism in the fishing spider and a model for the evolution of sexual cannibalism based on genetic constraints. *Evolutionary Ecology*, **11**, 255–273.
- Avilés, J. 1997. Causes and consequences of cooperation and permanent-sociality in spiders. In: *Evolution of Social Behaviour in Insects and Arachnids* (Ed. by J. Choe & B. Crespi), pp. 476–498. Cambridge, U.K.: Cambridge University Press.
- Brach, V. 1977. *Anelosimus studiosus* (Araneae: Theridiidae) and the evolution of quasisociality in theridiid spiders. *Evolution*, **31**, 154–161.
- Cheverud, J. M. 1996. Developmental integration and the evolution of pleiotropy. *American Zoologist*, **36**, 44–50.
- Crespi, B. J. 1996. In: *Phylogenies and the Comparative Method in Animal Behavior* (Ed. by E. P. Martins), pp. 253–287. New York: Oxford University Press.
- Danforth, B. N. 2002. Evolution of sociality in a primitively eusocial lineage of bees. *Proceedings of the National Academy of Sciences*, **99**, 286–290.
- Furey, R. E. 1998. Two cooperatively social populations of the theridiid spider *Anelosimus studiosus* in a temperate region. *Animal Behaviour*, **55**, 727–735.
- Hau, M. 2007. Regulation of male traits by testosterone: implications for the evolution of vertebrate life histories. *Bioessays*, **29**, 133–144.
- Hedrick, A. V. & Riechert, S. E. 1989. Population variation in the foraging behavior of a spider: the role of genetics. *Ecologia*, **80**, 533–539.
- Johnson, C. A. & Herbers, H. M. 2006. Impact of parasite sympatry on the geographic mosaic of coevolution. *Ecology*, **87**, 382–394.
- Johnson, J. C. & Sih, A. 2005. Precopulatory sexual cannibalism in fishing spiders (*Dolomedes triton*): a role for behavioral syndromes. *Behavioral Ecology and Sociobiology*, **58**, 390–396.
- Jones, T. C., Riechert, S. E., Dalrymple, S. E. & Parker, P. G. 2007. Fostering model explains variation in levels of sociality in a spider system. *Animal Behaviour*, **73**, 195–204.
- Maupin, J. L. & Riechert, S. E. 2001. Superfluous killing in spiders: a consequence of adaptation to food-limited environments? *Behavioral Ecology*, **12**, 569–576.
- Maynard Smith, J. & Riechert, S. E. 1984. A conflicting-tendency model of spider agonistic behavior—hybrid-pure population line comparisons. *Animal Behaviour*, **32**, 564–578.
- Michener, C. D. 1990. In: *Social Insects: an Evolutionary Approach to Castes and Reproduction* (Ed. by W. Engels), pp. 77–121. New York: Springer.
- Primer-E. 2001. *PRIMER Version 5.2.8*. Plymouth, U.K.: Primer-E Ltd.
- Raffa, K. F. & Berryman, A. A. 1987. Interacting selective pressures in conifer-bark beetle systems—a basis for reciprocal adaptations. *American Naturalist*, **129**, 234–262.
- Richards, M. H., von Wettberg, E. J. & Rutgers, A. C. 2003. A novel social polymorphism in a primitively eusocial bee. *Proceedings of the National Academy of Sciences*, **100**, 7175–7180.
- Riechert, S. E. 1985. Why do some spiders cooperate? *Agelena consociata*, a case study. *Behavioral Ecology Symposium Florida Entomology Society*, **68**, 106–116.
- Riechert, S. E. & Harp, J. 1987. Nutritional ecology of spiders. In: *Arthropod Nutrition* (Ed. by F. Slansky & J. G. Rodriguez), pp. 318–328. New York: Academic Press.
- Riechert, S. E. & Hedrick, A. V. 1993. A test for correlations among fitness linked behavioural traits in the spider *Agelenopsis aperta* (Araneae, Agelenidae). *Animal Behaviour*, **46**, 669–675.



- Riechert, S. E. & Johns, P. 2003. Do female spiders select heavier males for the genes or behavioral aggressiveness they offer their offspring? *Evolution*, **57**, 1367–1373.
- Riechert, S. E. & Johns, T. C. 2008. Phenotypic variation in the social behaviour of the spider *Anelosimus studiosus* along a latitudinal gradient. *Animal Behaviour*.
- Riechert, S. E. & Maupin, J. L. 1998. Spider effects on prey: tests for superfluous killing in five web-builders. In: *Proceedings of the 17th European Colloquium of Arachnology, Edinburgh 1997* (Ed. by P. A. Selden), pp. 203–210. Burnham Beeches, Buckinghamshire: British Arachnological Society.
- Riechert, S. E. & Maynard Smith, J. 1989. Genetic analyses of two behavioural traits linked to individual fitness in the desert spider, *Agelenopsis aperta*. *Animal Behaviour*, **37**, 624–637.
- Riechert, S. E., Singer, F. D. & Jones, T. C. 2001. High gene flow levels lead to gamete wastage in a desert spider system. *Genetica*, **112/113**, 297–319.
- Ruzzante, D. E. & Doyle, R. W. 1991. Rapid behavioral changes in medaka (*Oryzias latipes*) caused by selection for competitive and noncompetitive growth. *Evolution*, **45**, 1936–1946.
- Ruzzante, D. E. & Doyle, R. W. 1993. Evolution of social behaviour in resource-rich structured environments: selection experiments with medaka (*Oryzias latipes*). *Evolution*, **47**, 456–470.
- Rypstra, A. L. & Tirey, R. S. 1991. Prey size, prey perishability and group foraging in a social spider. *Oecologia*, **86**, 25–30.
- SAS Institute. 1998. *Statview 5.0*. Cary, North Carolina: SAS Institute.
- Seger, J. 1991. Behavioural Ecology: an Evolutionary Approach. In: . 3rd edn (Ed. by J. R. Krebs & N. B. Davies), pp. 338–373. Oxford: Blackwell Scientific.
- Sih, A., Bell, A. M. & Johnson, J. C. 2004. Behavioral syndromes: an ecological and evolutionary overview. *Trends in Ecology & Evolution*, **19**, 372–378.