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Colony defense and behavioral differentiation in the eusocial shrimp *Synalpheus regalis*

Received: 8 October 2001 / Revised: 30 November 2001 / Accepted: 3 January 2002 / Published online: 14 March 2002
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Abstract The sponge-dwelling shrimp *Synalpheus regalis* and several congeners exhibit monogynous, eusocial colony structures, and field data suggest that these social species may have a competitive advantage in crowded environments. We explored mechanisms of colony defense, a likely contributor to such a social advantage, by measuring responses of resident shrimp to conspecific and heterospecific intruders in experimental nests. To test for behavioral differentiation among classes of individuals, we compared responses of juvenile, large male, and breeding female (queen) *S. regalis* to each intruder type. Colony residents discriminated nestmates from both foreign conspecific and heterospecific intruders, making more aggressive snaps and fewer contacts with such intruders than with controls from the same colony. *S. regalis* colonies also exhibited behavioral differentiation among classes of individuals: most snaps against intruders were by large males. Thus, large males defended the colony, allowing juveniles and the queen to feed and grow unmolested. On a per-individual basis, intruders more frequently contacted the queen than either large males or juveniles, possibly indicating that signals from the queen provide information about colony status as they do in many social insects. Sampling of unmanipulated field colonies showed that colony reproductive output increased linearly, by a factor of 177 times, throughout the range in sampled colony sizes (2–356 individuals), whereas average per capita reproductive output decreased by only 61% over the same range. These data suggest that non-breeding colony members may gain inclusive fitness benefits by remaining in and helping defend the natal sponge. Differentiation among individuals in reproductive and defensive behavior may enhance the

efficiency and productivity of *S. regalis* colonies and contribute to their apparent competitive advantage over less social congeners.

Keywords Division of labor · Eusociality · Snapping shrimp · *Synalpheus regalis*

Introduction

Understanding the evolution of cooperative breeding systems, including the extreme of eusociality, requires understanding the selective pressures that favored the transition(s) from solitary life to group living, and to helping behavior by non-breeders. In many social vertebrates, the evolution of helping appears related to limited opportunities for individuals to breed independently, stemming from both environmental constraints (Emlen 1982a; see also Koenig et al. 1992) and life history features that together result in low turnover of suitable breeding opportunities (Hatchwell and Komdeur 2000). When opportunities for independent breeding are severely limited, selection favors delayed dispersal and the retention of adult offspring in the parental territory (Emlen 1982a). Helping behavior by the non-breeding offspring can then be favored by kin selection, often in combination with some form of behavioral manipulation or incentive by the breeders (Emlen 1982b). There is considerable support for the importance of ecological constraints at the intraspecific level in cooperatively breeding vertebrates (Emlen 1984, 1991) and somewhat more indirect support for social insects (Lin and Michener 1972; Evans 1977; Strassmann and Queller 1989; Brockmann 1997).

Colonies of the social snapping shrimp *Synalpheus regalis* (Alpheidae) share several features with those of primitively eusocial insects and cooperatively breeding vertebrates (Duffy 1996a). *S. regalis* inhabits the internal canals of tropical sponges, living in colonies of up to a few hundred individuals. Colonies consist of close kin groups containing adults of at least two generations,

Communicated by J. Heinze

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which cooperatively defend the host sponge using their large and distinctive snapping claws, and in which invariably only a single female breeds (Duffy 1996a). *S. regalis* also shares several life history characters that appear to predispose bird lineages to cooperative breeding (Arnold and Owens 1998 ; Hatchwell and Komdeur 2000), namely low dispersal rate, probable low adult mortality within its protected environment in sponges, sedentary lifestyle, and restriction to the relatively constant environment of tropical reefs. Several lines of evidence are consistent with the Ecological Constraints model, and more specifically the “habitat saturation” hypothesis (Selander 1964), for the origin and maintenance of this social organization in *S. regalis* and its several social congeners. First, suitable unoccupied habitat appears nearly non-existent: fewer than 5% of individual sponges of the two species used by *S. regalis* in Belize are unoccupied in the field (Duffy 1996b), suggesting that there may be strong competition for suitable “nest” sites and that shrimp attempting to disperse and breed on their own would have low success. Sympatric species of social *Synalpheus* overlap little in host species used and, more importantly, rarely co-occur in individual sponges where host range does overlap (Duffy 1996b, c), suggesting that shrimp also exclude heterospecific competitors. Moreover, because of their specialized host requirements (Duffy 1992, 1996b, c), there is little or no “marginal” habitat available to dispersing non-breeders. In over a decade of field work, we have rarely if ever found a *S. regalis* that was clearly outside a sponge at the time of collection. Theory suggests that such habitat saturation should favor philopatry and set the stage for helping (Emlen 1982a; Koenig et al. 1992). Both genetic data and colony structure confirm that many offspring indeed remain in the natal sponge through adulthood in *S. regalis*, where relatedness among colony members averages 0.50 (Duffy 1996a), as well as in some social congeners (Duffy 1998 ; Duffy and Macdonald 1999).

As the data on host occupation patterns support the premise of the habitat saturation hypothesis, comparative data are consistent with its (often implicit) proposed result, that cooperative groups should have a competitive advantage over less organized groups or individuals (Oster and Wilson 1978). To our knowledge this hypothesis has not been tested experimentally, although it is consistent with much indirect evidence from distribution and abundance of social insects (Wilson 1990). For sponge-dwelling shrimps, Duffy et al. (2000) found indirect support for a connection between sociality and competition in a comparative analysis, showing that highly social species of *Synalpheus* were significantly more likely than less social species to dominate their host sponges in terms of density of individuals.

The hypothesized competitive advantages of social groups stem in large part from division of labor, which enhances efficiency of foraging, resource processing, and defense (Wilson 1971 ; Oster and Wilson 1978; Karsai and Wenzel 1998). In this paper we demonstrate a similar form of behavioral differentiation among classes

of individuals in the important activity of colony defense by *S. regalis*, and show that colony members discriminate between nestmates and others in this aggressive behavior. We then examine indirect evidence for an advantage of large colony size by estimating, from field data, the pattern of colony reproductive output with colony size.

Methods

Natural history of the system

Synalpheus regalis is one of >20 described species in the *gambarelloides* species-group, a monophyletic clade (Duffy et al. 2000) of common and often abundant inhabitants of sponges in the tropical West Atlantic (Dardeau 1984 ; Duffy 1992, 1996c). These shrimp live their entire lives within the internal canals of the host sponge, feeding on host tissues and perhaps also detritus (Ruetzler 1976 ; Erdman and Blake 1987 ; Duffy 1998 ; Ríos and Duffy 1999). Like several other species within the *gambarelloides* group (Dobkin 1965, 1969), *S. regalis* has direct development. Eggs hatch directly into crawling juveniles, many of which remain in the natal sponge (Duffy 1996a). There is no planktonic stage, and the means of dispersal is unknown. Predatory eunicid polychaetes are occasionally found in some of the sponge species occupied by *Synalpheus*, but otherwise potential predators appear to be rare. *Synalpheus* species are often the most abundant macroscopic animals within Caribbean sponges, and their most likely enemies are conspecific and congeneric competitors for space within sponges. Most alpheids are very aggressive toward both heterospecific and conspecific individuals other than their mates (Hazlett and Winn 1962; Nolan and Salmon 1970). Aggressive contests involve the large and distinctive major chela (claw) borne on one of the first pair of walking legs, which is used in both threat displays and physical attacks (Hazlett and Winn 1962; Nolan and Salmon 1970; Schein 1977; Conover and Miller 1978; Knowlton and Keller 1982; Hughes 1996a, b). When the chela is closed rapidly, a plunger-like tooth on the movable finger fits snugly into an opposing socket, producing both an intense snap and a rapid, focused jet of water which is directed at the opponent. At close range the resulting water jet is capable of stunning or even killing small animals (MacGinitie and MacGinitie 1949; Schultz et al. 1998), although it rarely injures conspecifics. Because evidence from host occupation patterns (Duffy 1992, 1996b, c) and aggressive behavior (Duffy 1996a) indicates that competition among sponge-dwelling *Synalpheus* is strong, our tests for division of labor focused on colony defense against intruders.

The reproductive biology of *Synalpheus* is poorly known. There is no sign of sexual dimorphism among juvenile *Synalpheus*, nor any secondary sexual character that reliably distinguishes mature males from juveniles (Dardeau 1984). This situation probably indicates environmental sex determination, which is known in the alpheid genus *Athanas* (Suzuki 1970 ; Nakashima 1987; Gherardi and Calloni 1993). In contrast, mature females are easily identified by the ovaries, which are visible from a relatively early state through the carapace and dorsal abdomen, and by the modification of the pleopods to form a brood pouch. Thus, large non-ovigerous individuals are either mature males or sexually undifferentiated. Because sex is indistinguishable externally in all individuals other than mature females, we refer to non-ovigerous individuals as either “juveniles” (small) or “males” (large). To our knowledge, mating has not been described for any alpheid shrimp, but in several other carideans it involves the male attaching spermatophores to the female’s abdomen (Bauer 1976, 1979), followed by external fertilization when the female subsequently ovulates. Coutière’s (1899) anatomical study of *Alpheus* illustrated a simple tubular oviduct without seminal receptacle, and Knowlton (1980) reported that *A. armillatus* is unable to store sperm as females produced infertile eggs in the absence of males. The ab-

sence of sperm storage in alpheids suggests that the female must mate each time she ovulates. Eggs are laid in batches rather than continuously, and the clutch is carried in the mother's brood pouch, sometimes with minor attrition, until the embryos hatch. We have collected egg-bearing females at all times of year, and mature females without eggs are extremely rare (personal observation), indicating that breeding is continuous in female *Synalpheus*.

Field sampling of colony structure

We measured colony size (number of shrimp per sponge), and clutch size of the resident queen, in 48 colonies of *S. regalis* occupying the sponges *Xestospongia* sp. and *Hyattella intestinalis*. Data from 17 of these colonies were reported previously (Duffy 1996a). Sponges were collected using SCUBA between 1993 and 1999 from the outer reef ridge at Carrie Bow Cay, Belize (16°48'N, 88°05'W), returned immediately to the laboratory, and dissected to remove all associated animals. Shrimps were preserved in 10% formalin or 95% ethanol. We tabulated the number of individuals cohabiting within the sponge and the number of reproductive females in the colony, as assessed by presence of brooded embryos or visible ovaries. For each female, all embryos were removed and counted (hereafter clutch size).

Following Michener (1964), we used regression analysis to obtain a first-order empirical estimate of the relationship between colony size and reproductive output. Colony reproductive output was estimated as the number of embryos brooded by the queen at the time of collection. Because identifying potential breeders is complicated by absence of secondary sexual characters and the likelihood of sex change (see last section), we calculated average per capita egg production among all colony members regardless of size or sex. Before estimating regressions, we tested for heterogeneity of variances using Cochran's test. Where the test was positive, we transformed the data by $\log(100x+1)$; in all cases this transformation eliminated variance heterogeneity.

Establishment of experimental colonies

We established experimental colonies for observation in chambers resembling "ant farms" (formicaria). A slice of living sponge (*Lissodendoryx* cf. *colombiensis*) 5–10 mm in thickness was placed between two transparent plates of plexiglass and wrapped with a ring of styrofoam to prevent shrimp from escaping. This chamber was supplied with continuously flowing, unfiltered seawater via a narrow tube that pierced the styrofoam ring. The apparatus remained submerged throughout the assembly period to avoid injuring the sponge by exposure to air. The chamber was then clamped to the inside wall of a filled glass aquarium such that the sponge slice could be observed from the side. Shrimp were introduced to the chamber through a temporary hole in the top of the styrofoam ring.

Colony responses to intruders

To assess the specificity of colony responses to intruding competitors, and potential differences among colony members in mounting these responses, we recorded the reactions of experimental colonies to three types of intruders: a nestmate conspecific (i.e., from the same sponge), a non-nestmate conspecific (i.e., from a different sponge), and a heterospecific (*S. chacei*). In the field, the sponge *Lissodendoryx* sp. is used as a host by both *S. regalis* and *S. chacei*, although rarely if ever simultaneously. Thus, these two shrimp species are potential competitors for space. Each experimental colony consisted of the queen, 15 large males, and 15 small juveniles from a given field-collected colony (sponge). We used individuals from the smallest ("juvenile") and largest ("large male") size classes of non-ovigerous shrimp so that we could keep track of individuals of different sizes, which would have been impossible using the relatively continuous size range of individuals found in natural colonies. The shrimp were introduced to a cham-

ber and allowed to acclimate overnight. Shortly after dawn the following morning, one of the three intruder types was introduced to the surface of the sponge in the chamber. For the next 20 min, we carefully followed the intruder's movements through the sponge and observed (by naked eye) its interactions with the resident colony under natural, dim light conditions. We recorded all physical contacts between residents and intruder, and all snaps by residents, tallying these separately for queen, juveniles, and large males. To avoid excessive disturbance of the colonies that would result from dismantling the colony to remove the first intruder, we introduced the second intruder after this 20-min observation of the first, and the third intruder after these; thus, at the end of the experiment all three intruders were present in the sponge. Order of intruder introduction was alternated in successive replicate colonies, with the possible sequences of treatment ordering roughly equally frequent; resampling analysis confirmed that order of introduction had no effect on total numbers of contacts ($P=0.27$) or snaps ($P=0.51$). Experiments were performed on 14 independent replicate colonies. In one colony, several deaths unrelated to aggressive interactions occurred, so this colony was excluded from the analysis. Thus, 13 replicate colonies were used in the analysis.

The resulting data sets for counts of contacts and snaps had large numbers of zeros, strong variance heterogeneity among treatments, and were clearly not normally distributed, rendering conventional parametric statistics invalid. We therefore tested differences among treatments by repeatedly ($n=10,000$) resampling the data matrix, with replacement, to generate a distribution of resampled treatment means against which our empirical data could be compared. Reported P values indicate the proportion of these bootstrapped pseudoreplicates in which differences between the smallest and largest treatment mean equal or exceed the observed difference. To account for variation in behavior among colonies, we resampled within colonies (within rows in the data matrix), treating colonies in essence as a blocking factor. Resampling was performed using the resampling add-in for Excel 2000 (Blank et al. 1999).

We tested three sets of hypotheses using this resampling procedure. First, to assess whether colonies responded differently to the three types of intruders we tested four a priori hypotheses: the total number of contacts (pooled across all residents of a colony) was no lower with (1) a heterospecific intruder, or (2) a foreign conspecific, than with a control; and the total number of snaps was no greater against (3) a heterospecific intruder, or (4) a foreign conspecific, than against a control. These were directional (one-tailed) hypotheses, based on the expectation (Duffy 1996a) that shrimp aggression should increase in the order colony-mate (control), foreign (non-nestmate) conspecific, heterospecific. The second set of hypotheses tested for differences in behavior among resident "castes" (sensu Oster and Wilson 1978), i.e., juveniles, large males, and queen. As an overall estimate of behavioral differentiation among castes, we pooled data across the three intruder treatments and tested for differences among juveniles, large males, and the queen in number of contacts and snaps against intruders. Finally, the third set of hypotheses also tested differences among castes, but used only data from the heterospecific intruder treatment, which elicited the most snaps in this (see Results) and a previous experiment (Duffy 1996a). The second and third sets of tests were two-tailed, as predictions were less certain a priori.

Results

Colony structure and productivity in the field

Among the 48 colonies of *S. regalis* sampled, colony size (i.e., number of shrimps in the sponge) ranged from 1 to 356, with median 129.5 and no clear mode (Fig. 1). Forty-two of these colonies (88%) contained a single, mature female (queen), all of whom were either brooding embryos or had ripe ovaries and a distended, empty

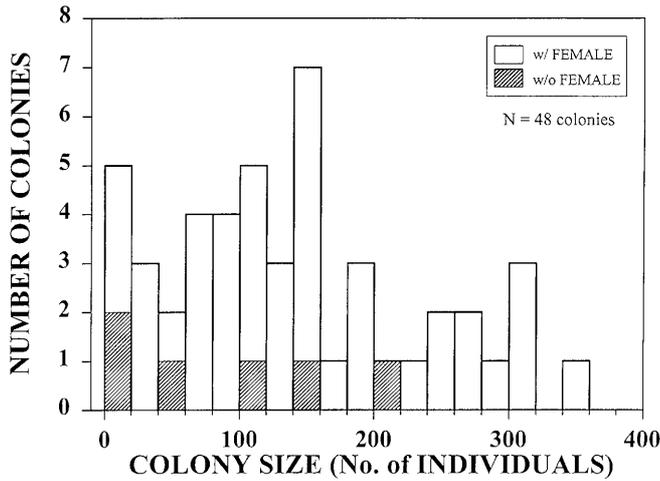


Fig. 1 Frequency distribution of *Synalpheus regalis* colony sizes (number of individuals per sponge) in field samples from Carrie Bow Cay, Belize

brood area indicating that embryos had recently hatched. In six colonies, no queen could be found; these ranged in size from 1 to 207 individuals (median 88). Colonies containing a queen ranged in size from 3 to 356 individuals (median 145).

Clutch size of the queen, and thus total colony reproductive output, was strongly and linearly related to colony size, which explained 84% of the variance in reproductive output among those colonies containing queens (Fig. 2, number of eggs = $0.158 \times \text{colony size}$, $P < 0.0001$). Including the six colonies that lacked queens, which had zero reproductive output, had little effect on this relationship, although required log transformation reduced its explanatory power somewhat ($r^2 = 0.65$, $\log [100 \times \text{number of eggs} + 1] = 0.0132 \times \text{colony size}$, $P < 0.0001$). We then made a rough estimate of the average inclusive fitness of non-breeding colony members by dividing the resident queen's egg production by the number of individuals in the colony. This approach assumes that sociality and monogyny are obligate in this species (last paragraph, and Duffy 1996a) and applies only to inclusive fitness of the trait for remaining and helping given that individual reproduction is not an option for such individuals (Queller 1996). The decline in this average non-breeder fitness (per capita reproductive output) with increasing colony size was surprisingly weak (Fig. 2). Indeed, when the six queenless colonies were included in the regression, there was no significant effect of (log-transformed) colony size on per capita output ($r^2 = 0.003$, $P = 0.74$). Excluding the queenless colonies produced a significant relationship ($r^2 = 0.35$, $\log [100 \times \text{number of eggs} + 1] = 1.60 - 0.0018 \times \text{colony size}$, $P = 0.0010$), but the shallow decline in per capita output with colony size remained.

Whereas the mean per capita egg production decreased relatively little with increasing colony size, variance in per capita reproduction declined substantially. The variance in per capita reproductive output was about

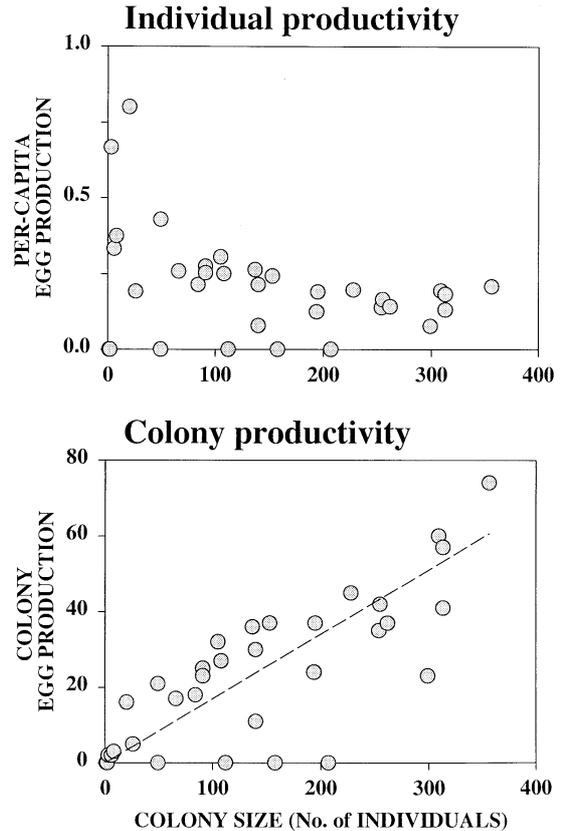


Fig. 2 Egg production of *Synalpheus regalis*, expressed as both average per capita production and total colony production, as a function of colony size. Number of eggs represents the size of the clutch carried by the queen at the time of collection, and average per capita production is this clutch size divided by the number of individuals in the colony. See text for regression analyses

an order of magnitude greater in colonies smaller than the median size compared with larger colonies, a significant difference ($F_{15,17} = 7.80$, $P < 0.001$).

Colony responses to intruders

Experiments began with the intruder placed on top of the sponge in the chamber. Most intruders eventually entered the sponge. When an intruder came into close proximity with a resident, particularly a larger individual, first contact characteristically involved energetic tapping of one another's antennae. Nestmate conspecifics (controls) then were generally accepted into the colony immediately and without incident. In contrast, when a heterospecific intruder was encountered, the initial antennal contact was usually brief and followed immediately by snapping, rapid retreat, or both. Heterospecific encounters sometimes led to extended bouts of snapping between resident(s) and intruder. Responses to non-nestmates were intermediate between these extremes.

The experiment introducing different types of intruders confirmed that heterospecific shrimp were treated very differently than conspecifics (Fig. 3). When re-

Fig. 3 Responses of resident shrimp to three types of intruders in experimental colonies of *Synalpheus regalis*. The upper panels show the total number of events summed across all residents; lower panels show counts standardized to the number of individuals of that caste present in the colony (15 juveniles, 15 males, 1 queen). See Table 1 for statistical analysis. $n=13$ independent replicate colonies

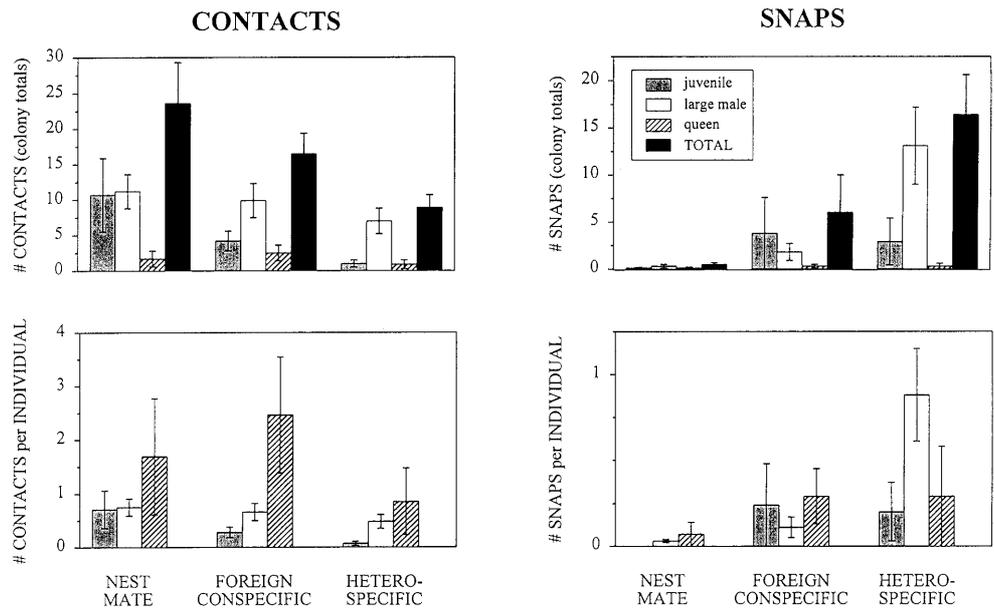


Table 1 Results of resampling tests of selected hypotheses from the intruder experiment (data in Fig. 3). $n=10,000$ re-sampled replicates, with replacement, for each hypothesis

Null hypothesis	Nominal P
Among intruder types (data pooled across castes within colonies), one-tailed tests:	
No fewer contacts with heterospecific than with control	<0.0001
No more snaps against heterospecific than against control	<0.0001
No fewer contacts with conspecific non-nestmate than with control	0.0136
No more snaps against conspecific non-nestmate than against control	0.0074
Among castes (data from heterospecific intruder treatment only), 2-tailed tests:	
No difference in raw number of contacts (colony totals)	0.0002
No difference in raw number of snaps (colony totals)	0.0010
No difference in per-individual number of contacts	0.1154
No difference in per-individual number of snaps	0.0454

sponses of all resident individuals in a colony (juveniles, large males, and queen) were pooled, intruding *S. chacei* elicited fewer than half the contacts, and about an order of magnitude more snaps, from residents than did conspecific nestmates from the same colony (Fig. 3); both differences were significant (Table 1). Non-nestmate conspecific intruders elicited intermediate responses, with somewhat fewer contacts and more snaps compared with controls (Fig. 3); these responses are significantly different than responses to nestmate controls even when α is adjusted to 0.025 to account for the two comparisons involving the control (Table 1).

Responses of residents to intruders indicated several differences in behavior among castes (juveniles, large males, and queens), which were generally more pronounced for heterospecific intruders. Large males were most active and aggressive, and were responsible for more contacts and snaps with intruders overall than were juveniles or the queen (Fig. 3). Heterospecific intruders, in particular, were contacted and snapped at almost exclusively by large males (Fig. 3). Even at the individual level, large males were significantly more likely to snap at heterospecific intruders than were juveniles or queens

($P=0.045$, Table 1, Fig. 3), indicating a behavioral difference among the three castes. The relative paucity of interactions between queen and intruders was due in part to the larger number of males and juveniles present (30) in the colony compared with the single queen. Indeed, when behaviors are expressed on a per-individual basis, the queen was significantly more likely to contact intruders (pooled across all three intruder types) than were males or juveniles ($P=0.0044$, Fig. 3).

Discussion

Nestmate recognition and colony cohesion

Social species of *Synalpheus* such as *S. regalis* are unusual among alpheid in their exceptional tolerance of conspecifics. The generally peaceful interactions within colonies are presumably attributable in part to close genetic relatedness among colony-mates. Allozyme data revealed that relatedness within colonies of *S. regalis* is high, averaging 0.50, and together with demographic evidence of natal philopatry, indicate that colonies in this

species represent close kin groups (Duffy 1996a). The existence of such groups is an important prerequisite of explanations of social evolution based on kin selection (Hamilton 1964), and some ability to distinguish kin from non-kin broadens the conditions under which altruism can evolve (Perrin and Lehmann 2001). Our intruder experiment provides evidence for discrimination between nestmates and other conspecifics. When faced with a foreign conspecific, resident shrimp contacted it less and snapped more frequently than they did when faced with a nestmate (Fig. 3, Table 1). As nestmates are generally close kin in *S. regalis*, this discrimination may reflect kin recognition and should in any case help maintain the integrity of kin-structured social colonies.

Colony residents also responded quite differently to heterospecific than to conspecific intruders entering the host sponge, a pattern demonstrated previously in artificial habitats (Duffy 1996a). Resident *S. regalis* were much more wary and aggressive toward the heterospecific intruder *S. chacei*, contacting them less and snapping at them far more frequently than they do toward conspecifics. Greater aggression toward heterospecifics than toward conspecifics also appears to be common among social insects. For example, experiments with a range of termite taxa have shown that aggression is often noticeably greater against other termite species than against conspecifics (Thorne and Haverty 1991; Leponce et al. 1996; Polizzi and Forschler 1999). In social shrimp this may reflect an advantage of accepting foreign conspecifics into the nest occasionally for the purpose of outbreeding.

Nestmate discrimination likely involves both waterborne and contact chemical signals, both of which have been shown to mediate sex recognition in other alpheids (Schein 1975; Jeng 1994; Hughes 1996b). Water-borne cues presumably explain our finding that resident *S. regalis* contacted foreign conspecifics and heterospecifics less frequently than controls (Fig. 3). But the mutual antennulation that initiates nearly all contacts in alpheids (Nolan and Salmon 1970; Knowlton and Keller 1982; Jeng 1994; authors' personal observation) suggests that contact chemical signals are of central importance in mediating recognition in these shrimp, as they are in many social insects (Hölldobler and Wilson 1990; Blum 1996). In this context, the distinctly higher frequency of intruder contacts with the queen than with other residents, when expressed per individual (Fig. 3), is intriguing. This may merely reflect a general attraction of intruding males to a fertile female. However, in many social insects, normal colony function is facilitated by exchange of pheromones that are produced by the queen and distributed among other colony members by frequent touching of her body (Hölldobler and Wilson 1990; Winston and Slessor 1992). Indeed, experiments with myrmecine ants showed that workers were attracted to both conspecific and heterospecific queens (Vienne et al. 1998), which the authors interpreted as evidence for a non-species-specific, volatile pheromone produced by queens. Perhaps a similar mechanism explains the

high frequency of intruder contacts with the queen in *S. regalis*.

Behavioral differences among colony members

Most hypotheses to explain cooperative breeding or eusociality entail some form of adaptive division of labor, which results in enhanced reproductive output of the breeders and inclusive fitness benefits for the non-breeding helpers (Wilson 1975 ; Emlen 1991). Our experiment aimed to characterize differences in behavior among classes of colony members with respect to colony defense, an activity with probable strong consequences for colony fitness in the space-limited environment (Duffy 1996b ; Duffy et al. 2000) of *S. regalis*. Whereas small individuals tended to be rather sedentary, often congregating in groups to feed, large males frequently moved around the sponge with an appearance of boldness, cocking the chela upon contacting another individual. The preponderance of large male interactions with intruders in our experiment reflects significant behavioral differences among castes. On a per-individual basis, large males were more aggressive than other castes when facing heterospecific intruders, and at the level of the colony were responsible for most contacts and snaps against all intruders (Fig. 3, Table 1). Such size- or age-related polyethism is a common component of labor specialization among social insects (Wilson 1971; Oster and Wilson 1978; Robinson 1992) and vertebrates (Emlen 1991) as well.

At the level of the colony, the queen participated in a small fraction of the total contacts and snaps against intruders (Fig. 3). This largely reflects the fact that she is greatly outnumbered – each experimental colony contained 15 juveniles and 15 large males, but only a single queen. Thus, the number of interactions expected from the queen is 1/15th that of either juveniles or large males if all behave identically. Nevertheless, because natural colonies of *S. regalis* invariably contain only a single queen (Duffy 1996a), these results reflect a significant phenomenon in which the queen is partially insulated from interaction with intruders, in the sense that an intruder is much more likely to encounter a large male than any other type of individual. Indeed, in natural colonies, the queen is even better protected since the median size of *S. regalis* colonies we measured in Belize was 129.5 individuals, ~fourfold larger than our experimental colonies. Thus, our observations and experiment show that large males shoulder the burden of defense, leaving small juveniles free to feed and grow, and the queen free to feed and reproduce. This shielding of the queen and juveniles from contact with danger is likely to translate into important fitness advantages as aggressive encounters between alpheid shrimp can cause serious injuries or even death (Knowlton and Keller 1992 ; Duffy 1996a).

Behavioral division of labor, particularly defense, among colony members in *S. regalis* could potentially provide a mechanism that contributes to the greater dom-

inance of host sponges by social shrimp species compared with less social taxa (Duffy et al. 2000). But the important question remains whether non-breeding defenders in *S. regalis* indeed enhance productivity of the breeder. Answering this question definitively will require experiments. Indirect evidence from our field data is consistent with such enhancement, however, in that reproductive output of the lone queen increased steadily with colony size. Clearly such correlational field data cannot prove that group size affects reproductive success (Emlen 1991). Group size might be a consequence, rather than a cause, of breeder reproductive success, for example through high territory quality or breeder experience. In *S. regalis*, queen body size varies directly with colony size (Duffy 1996a) so larger queen size in large colonies could result from any combination of aid provided by helpers, better territory quality, or simply from greater queen age. Thus, the mechanism for this relationship remains an open question. Nonetheless, as competition for sponges (Duffy 1996a, b) likely places a fitness burden on individuals and small groups, and aggressive activities of large males demonstrably reduce the queen's need to defend against such competitors (Fig. 3), there is a plausible mechanism for a link between such helping and colony reproductive output. Interestingly, the characteristic loss of the major chela in queens of the closely related social *Synalpheus filidigitus* (Duffy and Macdonald 1999) is also consistent with protection by other colony members in that species.

Why should a shrimp remain in, or join, a group as a non-breeder rather than attempting to breed independently? One likely benefit is enhanced survival. Pairs and small colonies have higher probabilities of death or disappearance than larger colonies in social wasps, bees, and spiders (Strassmann and Queller 1989; Itô 1993; Avilés and Tufiño 1998). Indeed, Strassmann and Queller (1989) concluded that the risk of reproductive failure in individuals and small groups has been a primary factor selecting for eusociality. More generally, enhanced demographic stability, and specifically reduced risk of total reproductive failure, in larger colonies may also favor colonial life (Wenzel and Pickering 1991). The hypothesis of greater survivorship of large colonies cannot be tested rigorously with data on static colony structure such as ours (Fig. 2) because such collections do not include data on failed colonies. Nevertheless, our data offer preliminary support for the pattern of smaller groups having higher risk of reproductive failure. Variance in per capita reproductive output was significantly greater in smaller colonies, and four of the six colonies in our sample that lacked females, and thus were at risk of reproductive failure, were below the median colony size. These patterns suggest that reproductive failure in social *Synalpheus* is likely more frequent in smaller colonies, as it is in many other social animals, and that selection may favor remaining in (or joining) a social group to minimize this risk. Finally, under the assumption that monogynous sociality is obligate in *S. regalis*, as suggested by our field data (also Duffy 1996a), the in-

clusive fitness of the conditional helping strategy should increase to the extent that the helper enhances the breeders' productivity (Queller 1996). If the enhanced productivity of larger colonies (Fig. 2) does result in part from helping by non-breeders, it will also enhance their inclusive fitness for the helping trait.

Previous research showed that *S. regalis* lives in colonies with strong similarities to those of many social insects and social vertebrates (Duffy 1996a). The similarity in colony structure mirrors a common environmental challenge to many of these animals, that is, a low turnover rate of breeding opportunities. The data presented here demonstrate both nestmate (probably kin) recognition and a form of division of labor in colonies of *S. regalis* with respect to a behavior that appears closely related to fitness, namely colony defense against competitors. By shouldering most of the burden of this potentially dangerous task large defenders may allow the queen to forage and reproduce, and her offspring to survive and grow, at higher rates, enhancing colony productivity and stability in a competitive environment. Experiments will be required to test this hypothesis definitively.

Acknowledgements We are grateful to the National Science Foundation (DEB 98-15785), the Smithsonian Institution's Caribbean Coral Reef Ecosystem (CCRE) program, and the BBC Natural History Unit for support of this research. We thank Richard Kirby and Martha Holmes for making video observations possible; Klaus Ruetzler, Mike Carpenter, Brian Kensley, and the staff of the Pelican Beach resort for facilitating work in Belize; and Christoph Schubart, Jürgen Heinz and one anonymous reviewer for comments that improved the MS. This is contribution 2452 from the Virginia Institute of Marine Science and contribution 636 from the CCRE.

References

- Arnold KE, Owens IPF (1998) Cooperative breeding in birds: a comparative test of the life history hypothesis. *Proc R Soc Lond B* 265:739-745
- Avilés L, Tufiño P (1998) Colony size and individual fitness in the social spider *Anelosimus eximius*. *Am Nat* 152:403-418
- Bauer RT (1976) Mating behaviour and spermatophore transfer in the shrimp *Heptacarpus pictus* (Stimpson) (Decapoda: Caridea: Hippolytidae). *J Nat Hist* 10:415-40
- Bauer RT (1979) Sex attraction and recognition in the Caridean shrimp *Heptacarpus paludicola* Holmes (Decapoda: Hippolytidae). *Mar Behav Physiol* 6:157-174
- Blank S, Seiter C, Bruce P (1999) Resampling stats in Excel. Version 1.1. Resampling Stats, Arlington, Va.
- Blum MS (1996) Semiochemical parsimony in the Arthropoda. *Annu Rev Entomol* 41:353-374
- Brockmann HJ (1997) Cooperative breeding in wasps and vertebrates: the role of ecological constraints. In: Choe JC, Crespi BJ (eds) *The evolution of social behavior in insects and arachnids*. Cambridge University Press, Cambridge, pp 347-371
- Conover MR, Miller DE (1978) The importance of the large chela in the territorial and pairing behaviour of the snapping shrimp *Alpheus heterochaelis*. *Mar Behav Physiol* 5:185-192
- Coutière H (1899) Les Alpheidae. Morphologie externe et interne, formes larvaires, bionomie. *Ann Sci Nat*, 8 sér, Zool 9:1-560
- Dardeau MR (1984) *Synalpheus* shrimps (Crustacea: Decapoda: Alpheidae). I. The Gambarelloides group, with a description of a new species. *Mem Hourglass Cruises* 7, 2:1-125
- Dobkin SR (1965) The first post-embryonic stage of *Synalpheus brooksi* Coutière. *Bull Mar Sci* 15:450-462

- Dobkin SR (1969) Abbreviated larval development in caridean shrimps and its significance in the artificial culture of these animals. *FAO Fish Rep* 57:935–946
- Duffy JE (1992) Host use patterns and demography in a guild of tropical sponge-dwelling shrimps. *Mar Ecol Prog Ser* 90:127–138
- Duffy JE (1996a) Eusociality in a coral-reef shrimp. *Nature* 381:512–514
- Duffy JE (1996b) *Synalpheus regalis*, new species, a sponge-dwelling shrimp from the Belize Barrier Reef, with comments on host specificity in *Synalpheus*. *J Crustac Biol* 16:564–573
- Duffy JE (1996c) Specialization, species boundaries, and the radiation of sponge-dwelling alpheid shrimp. *Biol J Linn Soc* 58:307–324
- Duffy JE (1998) On the frequency of eusociality in snapping shrimps (Decapoda: Alpheidae), with description of a second eusocial species. *Bull Mar Sci* 63:387–400
- Duffy JE, Macdonald KS (1999) Colony structure of the social snapping shrimp, *Synalpheus filidigitus*, in Belize. *J Crustac Biol* 19:283–292
- Duffy JE, Morrison CL, Ríos R (2000) Multiple origins of eusociality among sponge-dwelling shrimps (*Synalpheus*). *Evolution* 54:503–516
- Emlen ST (1982a) The evolution of helping. I. An ecological constraints model. *Am Nat* 119:29–39
- Emlen ST (1982b) The evolution of helping. II. The role of behavioral conflict. *Am Nat* 119:40–53
- Emlen ST (1984) Cooperative breeding in birds and mammals. In: Krebs JR, Davies NB (eds) *Behavioural ecology. An evolutionary approach*, 2nd edn. Blackwell, Oxford, pp 305–339
- Emlen ST (1991) Evolution of cooperative breeding in birds and mammals. In: Krebs JR, Davies NB (eds) *Behavioural ecology. An evolutionary approach*, 3rd edn. Blackwell, Oxford, pp 301–337
- Erdman RB, Blake NJ (1987) Population dynamics of the sponge-dwelling alpheid *Synalpheus longicarpus*, with observations on *S. brooksi* and *S. pectiniger*, in shallow-water assemblages of the eastern Gulf of Mexico. *J Crustac Biol* 7:328–337
- Evans HE (1977) Extrinsic versus intrinsic factors in the evolution of insect eusociality. *BioScience* 27:613–617
- Gherardi F, Calloni C (1993) Protandrous hermaphroditism in the tropical shrimp *Athanas indicus* (Decapoda: Caridea), a symbiont of sea urchins. *J Crustac Biol* 13:675–689
- Hamilton WD (1964) The genetical evolution of social behavior I, II. *J Theor Biol* 7:1–52
- Hatchwell BJ, Komdeur J (2000) Ecological constraints, life history traits and the evolution of cooperative breeding. *Anim Behav* 59:1079–1086
- Hazlett BA, Winn HE (1962) Sound production and associated behavior of Bermuda crustaceans (*Panulirus*, *Gonodactylus*, *Alpheus*, and *Synalpheus*). *Crustaceana* 4:25–38
- Hölldobler B, Wilson EO (1990) *The ants*. Belknap, Cambridge, Mass.
- Hughes M (1996a) Size assessment via a visual signal in snapping shrimp. *Behav Ecol Sociobiol* 38:51–57
- Hughes M (1996b) The function of concurrent signals: visual and chemical communication in snapping shrimp. *Anim Behav* 52:247–257
- Itô Y (1993) *Behaviour and social evolution of wasps. The communal aggregation hypothesis*. Oxford University Press, Oxford, UK
- Jeng M-S (1994) Effect of antennular and antennal ablation on pairing behavior of snapping shrimp *Alpheus edwardsii* (Audouin). *J Exp Mar Biol Ecol* 179:171–178
- Karsai I, Wenzel JW (1998) Productivity, individual-level and colony-level flexibility, and organization of work as consequences of colony size. *Proc Nat Acad Sci USA* 95:8665–8669
- Knowlton N (1980) Sexual selection and dimorphism in two demes of a symbiotic, pair-bonding snapping shrimp. *Evolution* 34:161–173
- Knowlton N, Keller BD (1982) Symmetric fights as a measure of escalation potential in a symbiotic, territorial snapping shrimp. *Behav Ecol Sociobiol* 10:289–292
- Koenig WD, Pitelka FA, Carmen WJ, Mumme RL, Stanback MT (1992) The evolution of delayed dispersal in cooperative breeders. *Q Rev Biol* 67:111–150
- Leponce M, Roisin Y, Pasteels JM (1996) Intraspecific interactions in a community of arboreal nesting termites (Isoptera: Termitidae). *J Insect Behav* 9:799–817
- Lin N, Michener CD (1972) Evolution of sociality in insects. *Q Rev Biol* 47:131–159
- MacGinitie GE, MacGinitie N (1949) *Natural history of marine animals*. McGraw Hill, New York
- Michener CD (1964) Reproductive efficiency in relation to colony size in hymenopterous societies. *Insectes Soc* 4:317–342
- Nakashima Y (1987) Reproductive strategies in a partially protandrous shrimp, *Athanas kominatoensis* (Decapoda: Alpheidae): sex change as the best of a bad lot. *J Ethol* 5:145–159
- Nolan BA, Salmon M (1970) The behavior and ecology of snapping shrimp (Crustacea: *Alpheus heterochelis* and *Alpheus normanii*). *Forma Functio* 2:289–335
- Oster GF, Wilson EO (1978) *Caste and ecology in the social insects*. Monographs in Population Biology 12. Princeton University Press, Princeton
- Perrin N, Lehmann L (2001) Is sociality driven by the costs of dispersal or the benefits of philopatry? A role for kin discrimination-mechanisms. *Am Nat* 158:471–483
- Polizzi JM, Forschler BT (1999) Factors that affect aggression among the worker caste of *Reticulitermes* spp. subterranean termites (Isoptera: Rhinotermitidae). *J Insect Behav* 12:133–146
- Queller DC (1996) The measurement and meaning of inclusive fitness. *Anim Behav* 51:229–232
- Ríos R, Duffy JE (1999) Description of *Synalpheus williamsi*, a new species of sponge-dwelling shrimp (Crustacea: Decapoda: Alpheidae), with remarks on its first larval stage. *Proc Biol Soc Wash* 112:541–552
- Robinson GE (1992) Regulation of division of labor in insect societies. *Annu Rev Entomol* 37:637–665
- Ruetzler K (1976) Ecology of Tunisian commercial sponges. *Tethys* 7:249–264.
- Schein H (1975) Aspects of the aggressive and sexual behaviour of *Alpheus heterochaelis* Say. *Mar Behav Physiol* 3:83–96
- Schein H (1977) The role of snapping in *Alpheus heterochaelis* Say, 1818, the big-clawed snapping shrimp. *Crustaceana* 33:182–188
- Schultz S, Wuppermann K, Schmitz B (1998) Behavioural interactions of snapping shrimp (*Alpheus heterochaelis*) with conspecifics and sympatric crabs (*Eurypanopeus depressus*). *Zool Anal Complex Syst [Suppl I]* 101:85
- Selander RK (1964) Speciation in wrens of the genus *Campylorhynchus*. *Univ Calif Pub Zool* 74:1–224
- Strassmann JE, Queller DC (1989) Ecological determinants of social evolution. In: Breed MD, Page RE (eds) *The genetics of social evolution*. Westview, Boulder, Colo. pp 81–101
- Suzuki H (1970) Taxonomic review of four alpheid shrimp belonging to the genus *Athanas*, with reference to their sexual phenomena. *Sci Rep Yokohama Natl Univ Section II. Biol Geol Sci* 17:1–38
- Thorne BL, Haverty MI (1991) A review of intracolony, intraspecific, and interspecific agonism in termites. *Sociobiology* 19:115–146
- Vienne C, Errard C, Lenoir A (1998) Influence of the queen on worker behaviour and queen recognition behaviour in ants. *Ethology* 104:431–446
- Wenzel JW, Pickering J (1991) Cooperative foraging, productivity, and the central limit theorem. *Proc Nat Acad Sci USA* 88:36–38
- Wilson EO (1971) *The insect societies*. Belknap, Cambridge, Mass.
- Wilson EO (1975) *Sociobiology*. Belknap, Cambridge, Mass.
- Wilson EO (1990) *Success and dominance in ecosystems: the case of the social insects*. Ecology Institute, Oldendorf/Luhe, Germany
- Winston ML, Slessor KN (1992) The essence of royalty: honey bee queen pheromone. *Am Sci* 80:374–385