Tests of Hypotheses on the Adaptive Value of an Extended Male Phase in the Hermaphroditic Shrimp *Lysmata wurdemanni* (Caridea: Hippolytidae)

RAYMOND T. BAUER

Department of Biology, University of Louisiana at Lafayette, Lafayette, Louisiana 70504-2451

Abstract. Hypotheses on delayed sex change in the protandric simultaneous hermaphrodite *Lysmata wurdemanni* were tested with observations from population samples, mating experiments, and experiments on sex change under optimal and suboptimal breeding conditions. Male-phase individuals (MPs) much larger than the minimum size of sex change were most frequent in a natural population from fall through early spring. The hypothesis was tested that some MPs delay sex change to the simultaneous hermaphrodite female-phase (FP) because MPs are more competitive in obtaining copulations with parturial FPs than are FPs mating as males (MFPs). In different experiments, parturial FPs were maintained with two potential male mating partners (large MP and MFP, small MP and MFP, large MP and small MP) through the parturial molt and spawning; activities were recorded with time-lapse video. MFPs gained the single copulation with the parturial FP as frequently as MPs, large or small, but large MPs copulated with more FPs than small MPs. The hypothesis of FP reversion to large MP was tested experimentally and rejected. Rate of change of MP to FP was much lower in large MPs maintained under suboptimal (fall/winter) than optimal (spring/summer) breeding conditions. The results presented here suggest that the occurrence of large MPs from the fall to early spring is better explained by abiotic proximate factors related to breeding than by socially mediated sex change in different demographic environments.

Received 16 April 2002; accepted 6 September 2002.
E-mail: rtbauer@louisiana.edu

Abbreviations: AG, androgenic gland; CL, carapace length; ESD, environmental sex determination; FP, female-phase individual (simultaneous hermaphrodite in *Lysmata wurdemanni*); MP, male-phase individual; MFP, a FP acting as a male mating partner; PFP, parturial FP; PSH, protandric simultaneous hermaphroditism; TR, transitional individual.

Introduction

Many organisms, both plant and animal, partition reproductive output into male and female components. Such sex allocation may occur before fertilization or during embryonic development. In hymenopteran insects (ants, wasps, bees), haplodiploid sex determination allows females to produce male and female offspring in variable proportions (Wilson, 1976; Thornhill and Alcock, 1983). In turtles and some other reptiles such as alligators, sex determination is temperature dependent, so that females may control the sex ratio of offspring by the seasonal timing of egg laying or the location of nests (Bull, 1980; Ferguson and Joanen, 1982). Higher male fetal mortality may occur in mammalian females under stressful conditions, resulting in female-biased litters (Wilson, 1976; see also Charnov, 1982). In other organisms, sex allocation may occur after birth or hatching in the sexually mature organism. Simultaneous hermaphrodites may vary resources to male and female gamete production (Charnov, 1982). In sequential hermaphrodites, sexual function is partitioned ontogenetically because an individual first reproduces as one sex and then the other (sex changers; Ghiselin, 1969; Warner, 1975; Charnov, 1982; Policansky, 1982). Simultaneous and sequential hermaphroditism is found in many plant taxa, in most invertebrate groups (except insects), and in many fishes (Chan and Yeung, 1983; Ross, 1990). The direction of sex change (female to male; protogyny; male to female; protandry) is fairly well explained by the size-advantage model (Ghiselin, 1969), especially in fishes and crustaceans, two groups in which sequential hermaphroditism is relatively common (Warner, 1975). What is not often well explained, especially in decapod shrimps, is variation in the timing of sex change, *i.e.*, the amount of time spent as male and female.

Protandric hermaphroditism occurs in several genera and
families of caridean shrimps (Bauer, 2000). In the simplest form of caridean protandry, all individuals develop first as males and then later change to breeding females. The sexual system may be more complex, with a mixture in the population of primary females, which develop only as females, and protandric individuals, as in Pandalus jordani, P. borealis, P. eous, P. montagui, and P. danae (Butler, 1980; Bergström, 2000). In other species, the population may consist of protandric individuals and primary males, individuals that never change to females, as in Thor munningi (Bauer, 1986) and Athanas kuminatoens (Nakashima, 1987).

Throughout the last century, Lysmata seticaudata was described as a completely protandric species (Spitschakoff, 1912; Dohrn, 1950; Charniaux-Cotton and Payen, 1985). Smaller individuals have external male characteristics, but the gonads are ovotestes with immature oocytes in the ovarian region. With increasing size, individuals lose external male characters and change to breeding females that incubate embryos. Unlike other protandric carideans, the female-phase individuals (FPs) retain the male ducts and testicular portion of the ovotestes, and sperm production continues (Berreur-Bonnenfant and Charniaux-Cotton, 1965; Charniaux-Cotton, 1975). The ability of L. seticaudata FPs to function as males was not addressed in these studies. However, FPs of Lysmata wurdemanni (Bauer and Holt, 1998) and L. amboinensis (Fiedler, 1998), with sexual changes during ontogeny similar to L. seticaudata, mate as males and fertilize other FPs, although they are unable to fertilize themselves. They are functional, outcrossing simultaneous hermaphrodites. Bauer (2000) termed this sexual system “protandric simultaneous hermaphroditism” (PSH), in which male-phase individuals (MPs) function only as males, but FPs are functional simultaneous hermaphroditoids. Morphological and behavioral observations on these and other Lysmata spp. indicate that PSH is widespread in the genus (Bauer, 2000, 2001).

The size (age) of sex change in protandric species may be variable (Charnov et al., 1978; Charnov, 1981; Charnov and Anderson, 1989; Bergström, 1997, 2000). The size of sex change may be a phenotypically flexible trait governed by the demographic environment in which an individual occurs (environmental sex determination, or ESD; Charnov et al., 1978; Charnov, 1981; Charnov and Anderson, 1989). In Pandalus spp., for example, a male in a population with abundant females might gain more inseminations by delaying change to female because males can potentially inseminate several females during the breeding season. A Pandalus female, in contrast, produces only one brood per year. When females are not abundant in the population, a male might benefit by early change to female because insemination of a female’s brood of eggs is assured. As a male, the individual would have to compete with numerous other males to inseminate one of the scarcer females. On the other hand, a male changing to female later in life would begin female production with larger broods, since female fecundity is correlated with size in carideans including Pandalus spp. (Bauer, 1991; Bergström, 2000). However, a later change to female might cause the loss of female breeding for an entire season, given only one brood per year, thus countering the fecundity advantage of late change to female. The relative proportions of males and females in the population, perceived by an individual through social interactions, would serve as the environmental factor stimulating or inhibiting male change to female.

However, the hypothesis of ESD in Pandalus danae was rejected by Marliave et al. (1993) after they conducted extensive experiments involving the rearing of a large number of young males in different demographic environments. They concluded that the sexual composition of the population has no effect on sex determination (change) in this protandric species. Other mating and rearing experiments led Marliave et al. (1993) to conclude that the timing of sex change was under genetic control. They proposed that any adaptive change in the proportions of sexual morphs would occur by between-generation selection on sexual genotypes rather than a within-generation, socially determined response as proposed by Charnov et al. (1978) and Charnov (1981). Bergström (1997), on the basis of results of a demographic analysis, concluded that the variable type and size of sex change in Pandalus borealis was best explained by frequency-dependent selection on genotypes for sex change at different ages (sizes). According to this hypothesis, different combinations of mortality, recruitment, and growth favor different sex-change genotypes. Conover and Munch (2002) showed experimentally in the fish Menidia menidia (Atherinidae) that size-dependent selection, like that acting on commercially fished populations, lowers the age of sexual maturity. Charnov et al. (1978) and Charnov (1981) reported a decrease in the size (age) of sex change in fished pandalids. This might result from size-related mortality (selection) (Conover and Munch, 2002) rather than from an adaptive response (decision) by individuals to demographic changes in the population. On the other hand, Zupo (2001) clearly showed that seasonal variations in diet of newly recruited shrimps determine whether or not sex change takes place in the protandric shrimp Hippolyte inermis.

In Lysmata wurdemanni, individuals from a natural population vary in size from 3–14 mm carapace length (CL) (Bauer and Holt, 1998; Bauer, 2002). Individuals enter the population as MPs and can change to FPs at a size as small as 6 mm CL; the majority change by 7.5 mm CL. However, there is frequently considerable overlap between MP and FP size-frequency distributions. Many MPs do not change sex until a much larger size, with some MPs becoming as large as the larger FPs (Bauer and Holt, 1998; Bauer, 2002). Some MPs might never change to FP, although all are
morphologically capable of doing so (Bauer and Holt, 1998; Bauer, 2000; Lin and Zhang, 2001). Bauer and Holt (1998) and Bauer (2000, 2001) suggested that delayed change to the reproductively advantageous FP, a simultaneous hermaphrodite, might be selected for in certain demographic situations if large MPs were competitively superior to FPs (mating as males) and small MPs in inseminating FPs near spawning. Lin and Zhang (2001) reared MPs of L. wurdemanni singly and in groups of from 2 to 10 individuals, finding that change to FP increased with increasing MP group size. Replicate size was quite small in this experiment, varying from 2 to 9 replicates per treatment. The authors concluded that these results demonstrated social control of sex ratio. Furthermore, they proposed that persistence of MPs in some treatments suggested a mating advantage for large MPs in certain demographic situations.

Other explanations are possible for large MP size in Lysmata wurdemanni. Large MPs might be FPs that have reverted to the male phase. Bidirectional sex change is known in some animals, such as gobid fishes (Munday, 2002) and polychaetes (Berglund, 1986). A secondary change back to male purportedly takes place in some females of the protandric alpheid Athanas indicus (Suzuki, 1970). In L. wurdemanni, abiotic factors related to female breeding might account for the seasonally related occurrence of large MPs. The largest MPs, including transitional MPs changing to FP, are found in winter and early spring (Bauer, 2002). Suboptimal conditions for breeding (lower water temperatures, short daylength) and presumably larval survival (planktonic food supply) occur during the late fall through early spring in this primarily tropical species studied near the northern edge of its range (Bauer and Holt, 1998; Bauer, 2002). Abiotic environmental conditions unfavorable for female breeding might cause a decrease in female hormone levels, resulting in a shift in female/male hormones in favor of the male system. Female-phase individuals, with both male and female reproductive systems, might molt back into a male phase, with male external sex characters. Alternatively, suboptimal breeding conditions might delay change to FP in MPs that reach the size of sex change during the fall, resulting in growth to the very large MPs observed by Bauer (2002) in the late winter and early spring.

The objective of this study was to examine components of hypotheses explaining the presence of large MPs in Lysmata wurdemanni. The male mating abilities of large MPs, small MPs, and FPs were compared to determine the possible male mating advantages of delayed sex change. The presence of large MPs might alternatively be explained by factors not related to the sexual composition of the population and socially determined sex change. The possible effects of seasonal abiotic factors unfavorable to female breeding resulting in either (a) reversion of FPs to large MPs or (b) delayed MP change to FPs were examined by maintaining (a) FPs and (b) MPs capable of sex change under suboptimal (nonbreeding season) and optimal (breeding season) conditions.

Materials and Methods

Population samples and live specimens were collected from a population of Lysmata wurdemanni inhabiting the rock jetty at Mustang Island, Port Aransas, Texas (27° 50' N, 97° 03' W). Collection was during low tides at night (Bauer, 2002); when tides are lower than −0.5 ft mean sea level, the rock ledge and tidepool habitats of these shrimps are exposed, and the shrimps are active at night, moving out into pools and the perimeter of the jetty rocks where they can be taken with dipnets. Hand dipnets were used to take the shrimps from tidepools; long-handled dipnets were more effective in sweeping deep under rock ledges. All nets had a 1-mm mesh and were effective in collecting the smallest (= newly recruited) individuals of the population (Bauer, 2002). In any particular location, groups of individuals showed no apparent segregation by sexual morph or size, and the samples are assumed to represent a random sample of the population. Population samples were taken monthly from June 1999 through June 2000, and then bimonthly from August 2000 through June 2001 (Bauer, 2002). Shrimps collected in population samples were first preserved in 10% seawater formalin and later transferred to 70% ethanol for permanent storage. Live specimens for experiments were collected separately from population samples and transported in sealed collecting bags containing seawater oxygenated just before the 8–10-h trip from the collecting site to the home institution.

Sexual morphs were identified and classified as in Bauer and Holt (1998). Male-phase individuals (MPs) have cincinnuli (coupling hooks) on pleopod 1 endopods and appendices masculinae on pleopod 2 endopods. Transitionals (TRs) are MPs in which the ovarian portion of the ovotestes is filling with vitellogenic oocytes, which is easily observed through the carapace in living and preserved specimens. At the next molt, TRs change to FPs and spawn for the first time. Female-phase individuals (FPs) lack pleopod 1 cincinnuli and appendices masculinae. They are typical caridean females in external morphology, and they brood embryos. However, they can mate as males (simultaneous hermaphrodites) (Bauer and Holt, 1998). As in females of other carideans, FPs of L. wurdemanni near spawning (ovarian portion of gonad full of vitellogenic oocytes) become receptive to mating immediately after a prespawning (parturial) molt (Bauer and Holt, 1998; Bauer and Abdalla, 2001). As in other carideans, the male deposits an external spermatophore on the underside of the female, and there is no sperm storage (Bauer, 1976; Bauer and Holt, 1998). In this report, FPs very near or just completing the parturial molt are termed parturial FPs (PFPs).
Mating experiments tested the hypothesis of no difference in male mating ability among FPs (mating as males = MFPs), small MPs, and large MPs. In three experiments, competition for inseminating the PFP was between a large MP and a MFP, a small MP and a MFP, and a large MP and a small MP. Parturial FPs near the prespawning molt were identified by gonad condition (vitellogenic oocytes filling the cephalothoracic space behind the eyes and above the cardiac stomach) and developmental state of brooded embryos (near hatching, with large eyes and little or no yolk). MFPs carried embryos but, unlike PFPs, were not close to a parturial molt. Small and large MPs were defined as MP < 6.5 mm CL and > 6.5 mm CL, respectively. The size of 6.5 mm CL was chosen to define “large” MP because MP can change to FPs above this size (Bauer and Holt, 1998; Bauer, 2002).

In each replicate, a premolt PFP was placed with two potential mating partners. The median duration of replicates, from placement of individuals into the experimental chamber until spawning by the PFP, was 2 days (1–9 d, n = 90). The MP or MFP that copulated with and inseminated the PFP was identified from time-lapse video recordings. Replicates in which the PFP copulated with more than one male mating partner (4 of 94 replicates) were not included in the data analyses. In all other replicates, there was only a single copulation, except for one replicate in which a MFP copulated twice with the PFP. Spawned PFPs were maintained alive for 5 days to determine if the attached brood of eggs had been fertilized and contained developing embryos (Bauer and Holt, 1998).

Replicates of mating experiments were conducted in aquaria partitioned into a space 20 cm wide × 25 cm long × 15 cm high to allow for sufficient magnification using the CCD surveillance video camera, which was equipped with an 8-mm lens sensitive to both low-intensity white and infrared light. Activities were recorded with a time-lapse video at a speed of 5 pictures per second (24-h recording mode). Fluorescent and infrared (880 nm) lamps provided day and night illumination, respectively. In replicates with a large and a small MP, individuals had to be marked to distinguish between them in videotapes, especially at night when most mating occurred. Small pieces of reflective tape were fixed with cyanoacrylate glue on the carapace of one MP and the posterior abdomen of the other, with position assigned at random. No marking was needed in replicates with a MP and a MFP because the latter could be identified by its brood of embryos. After hatching of embryos prior to the mating molt, the PFP was identified by the absence of embryos and the presence of vitellogenic oocytes. Reflective tape, embryo broods, and vitellogenic gonads show clearly on videotapes recorded with infrared (nighttime) illumination. As in Bauer and Holt (1998), shrimps were maintained prior to and during experiments at water temperatures of 25–28 °C, salinities of 33–36 ppt, and a photoperiod of 14-h day: 10-h night. Shrimps were fed shrimp pellets daily.

The hypothesis that FPs may revert to large MPs was tested by an experiment in which FPs were maintained under favorable and less favorable breeding conditions for 9 weeks. In the “optimal” treatment (2 September through 2 November, 2000), 50 FPs with embryos were maintained in groups of 10 individuals occupying 38-l aquaria at a water temperature, controlled by aquarium heaters, of 26 °C and a photoperiod of 14-h light: 10-h dark. In the “suboptimal” treatment (22 September to 24 November, 2000), another 50 FPs with embryos were similarly maintained except without heaters so that water temperatures would be cooler than those in the optimal treatment. The ambient temperature of the aquatic laboratory during this period was 19 °C; the photoperiod was set at 10-h light: 14-h dark. Prior to the experiment, individuals used in both treatments were maintained at a salinity of 33–36 ppt and an optimal temperature (26 °C) and photoperiod (14-h light: 10-h dark). They were fed daily, and the aquarium water was partially changed weekly. To reduce cannibalism of newly molted shrimps, which was an observed source of mortality during this experiment, small concrete block shelters were placed in each aquarium for this thigmotactic species. Molts (exuviae) readily visible in the aquaria were removed and recorded daily. Absolute counts of molts were not feasible because (a) the shrimps may pick apart and eat some exuviae before they are observed, and (b) other exuviae are hidden by shelters and aquarium tubing and could not be observed without major disturbance of the aquarium environment. At the end of the experiment, all individuals were preserved and checked for sexual condition.

The hypothesis that sex change of large MPs to FPs is greater under optimal (spring/summer) than suboptimal (fall/winter) breeding conditions was tested with an experiment using a setup similar to that just described for the previous experiment. The MP used in both treatments (all nontransitional) were maintained at optimal temperature and photoperiod prior to the beginning of the experiment. In the optimal treatment (n = 46 MPs), water temperature was maintained with heaters at 26 °C, and photoperiod was 14-h light: 10-h dark. In the suboptimal treatment (n = 45 MPs), cooler water temperatures were obtained by not using water heaters, with water temperature following the ambient air temperature of the laboratory, which was not under control of the investigator. The experiment was begun when a decrease in ambient laboratory temperature was predicted from outside weather conditions. Water temperature in the suboptimal treatment was 23 °C at the beginning of the experiment and decreased to 18–19 °C (weekly median temperature = 20 °C); photoperiod was 10-h light: 14-h dark. After 14 weeks (20 October 2001 to 23 January 2002), the laboratory air temperature and thus the water temperature of the suboptimal treatment began to rise. Heaters were
then placed in aquaria of the suboptimal treatment to increase water temperature to that of the optimal treatment, and photoperiod was changed to 14-h light: 10-h dark, simulating a change from fall/winter to spring/summer conditions. The experiment then continued for another 8 weeks.

The shrimps were maintained and checked for molts as in the previous experiment. Male-phase individuals that changed to FP (brood of embryos observed) were removed from the experiment and preserved. When only one MP remained in an aquarium, it was removed when it reached the late transitional stage (mature vitellogenic ovotestes), with the assumption that it would have transformed to FP at the next molt. This was done because it could not have produced a brood of embryos (visible evidence of sex change) upon molting to FP without a mating partner.

Results

Occurrence of large MPs

Large MPs, defined as MPs > 6.5 mm CL, composed 17.6% to 100% of the MP population (Fig. 1), with higher proportions of large MPs in the fall through early spring (Fig. 1). The proportions of large MPs ≥ 8.5 mm CL (“very large MPs”) followed a similar pattern. Some (28.7%; n = 150) of the very large MPs were transitionals that would change to the female phase at the next molt. The highest frequency of these very large transitionals occurred from mid-winter through late spring (Fig. 2).

Mating experiments

Precopulatory and copulatory behaviors were qualitatively similar to those described in Bauer and Holt (1998), in which a single FP served as the male mating partner, except for the following observations. In this study, MPs and MFPs began to approach, follow, and remain near the PFP as the latter neared the parturial molt. These behaviors became obvious about 1 h before the PFP molt. In addition, another behavior was observed (“perching”) in about 20% of the replicates, in which a MP or a MFP perched on the PFP from above and remained in that position as long as several seconds. Although individuals of Lysmata wurdemanni can be aggressive—for example, fighting over food particles using the third maxillipeds and anterior pereopods—no obvious aggressive behavior between the potential male mating partners over a parturial female was observed in the videotapes.

The hypothesis of no difference among sexual morphs in their ability to copulate with and inseminate parturial females (PFPs) was tested with mating experiments. In the experiment with large MPs and small MPs as potential male mating partners (Table 1), large MPs gained the single PFP copulation in more replicates than small FPs (Fig. 3A). The hypothesis of no difference from a 50:50 distribution of copulations between large MPs and small MPs was tested with chi-square and is rejected ($\chi^2 = 6.53; P = 0.01; n =$...
Table 1

Size data for potential male mating partners placed with newly molted parturial females in mating experiments

| Experiment         | SMP | LMP | MFP |
|--------------------|-----|-----|-----|
| SMP vs. LMP        | 5.4 (4.4, 6.4) | 7.4 (6.6, 9.4) |
| SMP vs. MFP        | 5.6 (4.5, 6.5) | 8.8 (7.2, 12.3) |

Size given is median carapace length (mm), with minimum and maximum values in parentheses (n = 30 for all categories). LMP, large male-phase; MFP, non-parturial female-phase (simultaneous hermaphrodite); SMP, small male-phase.

In another experiment, large MPs and MFPs (Table 1) were equally successful in obtaining the PFP copulation (Fig. 3B) ($\chi^2 = 0; P = 1; n = 30$). In the 19 replicates in which the large MP was smaller than the MFP, the MP and MFP gained the PFP copulation equally ($n = 9, 10$ respectively). Similarly, when the large MP was larger than the MFP ($n = 8$ replicates), each gained the single PFP copulation in an equal number of replicates. In the small MP vs. MFP experiment (Table 1), MFPs obtained the PFP copulation in more replicates (Fig. 3C), but the difference is not statistically significant ($\chi^2 = 1.2; P = 0.27; n = 30$). The copulated PFP produced a fertile brood of embryos in 87 of the 90 replicates of the three experiments.

Correlations of large MP and FP relative abundance

The effect of demographic influence (relative abundance of FPs) on the proportion of large MPs was investigated. From population samples, Spearman rank correlation coefficients ($r_s$) were calculated for possible correlations between % large MP and % FP, and % very large MP and % FP (Fig. 1). The null hypothesis of no correlation ($r_s = 0$) between % large MP and % FP and between very large MP and % FP was accepted ($r_s = -0.32, P > 0.20$) respectively; $n = 17$ months in both tests).

FP to MP sex change experiment

This experiment tested the hypothesis that FPs may change back to large MPs. In the treatment with optimal breeding conditions, all surviving individuals (36 of 50) were FPs. Of these 36 FPs, 33 (91.6%) showed female reproductive activity, either incubating embryos (21) or, if without embryos, showing mature vitellogenic ovotestes (12), indicating spawning at their next molt. In the suboptimal treatment, the 39 survivors out of 50 were all FPs. Of these 39, 33 (84%) were reproductively active as females; 26 incubated embryos, and 7 without embryos had mature vitellogenic ovotestes. During the 9 weeks of the experiment, 93 exuviae were collected in the optimal treatment vs. 43 in the suboptimal treatment. The molting rate, adjusted for number of survivors per treatment, was 2.4 times greater in the optimal treatment.

Large MP to FP sex change experiment

During first 14 weeks, 42 of 46 individuals in the optimal treatment changed to FP (including 2 solitary late-stage transitionals), 2 died, and 2 remained MP; in the suboptimal treatment only 23 of 45 changed (5 deaths). The rate of change was relatively constant in the optimal treatment compared to that of the suboptimal treatment (Fig. 4A), in which the rate of sex change slowed with decreasing ambient temperatures (Fig. 4C). Molting rates were always higher in the optimal treatment (Fig. 4B). The mean frequency of individuals molting per week was 31.2% in the optimal and 13.0% in the suboptimal treatment. Given that the period between molts may be estimated by the inverse of
this time, the two MPs of the original optimal treatment had not changed sex (verified by examination of pleopods after preservation). In the suboptimal treatment, after 6 weeks without any sex change and 3 weeks after the switch to optimal conditions, MPs began to change and continued to do so until the experiment was terminated (Fig. 5). In the last 8 weeks, 10 of 17 had changed to FP, while 2 others were late-stage transitionals (close to FP change) when removed from the experiment. Three of the original MPs in the suboptimal experiment remained, including one mid-stage transitional. Two others died during the last 8 weeks of the experiment.

At the beginning of the experiment, the mean size of MPs in the suboptimal (n = 45) and optimal (n = 46) treatments was equal (7.5 mm CL; $t$ test, $P = 0.80$) (Fig. 6). The size of FPs (sex-changed individuals) was compared between the suboptimal (9.2 mm CL; n = 33) and optimal treatments (9.1 mm CL; n = 40) (Fig. 6), and the hypothesis of no difference is accepted ($t$ test, $P = 0.58$). The size of FPs from the suboptimal treatment that transformed in weeks 14–22 (mean = 9.4 mm CL, n = 10) was slightly larger than that of FPs from the optimal treatment (Fig. 6), but the means of these two groups are not significantly different ($t$ test, $P = 0.14$).

**Discussion**

This study addressed components of different hypotheses about the timing and degree of sex change in *Lysmata wurdemanni* that might explain the presence of large MPs (male-phase individuals) in this species. *Lysmata* spp. have a unique protandric sexual system in which sex-changed "females" (FPs) retain the ability to mate as males—that is, are simultaneous hermaphrodites (Bauer, 2000). The size (age) of change from MP to FP might be socially determined, resulting in considerable variation in sex change in...
different demographic environments, as hypothesized for protandric pandalid shrimps (termed environmental sex determination, or ESD; Charnov et al., 1978; Charnov, 1981; Charnov and Anderson, 1989). In ESD, the frequency of sexual morphs in the population (demographic environment), presumably perceived by contacts among individuals (socially mediated), influences the size and age at which males change to females. Delayed sex change might be advantageous to males in populations in which females are abundant. A sexually successful male might inseminate several females during the same period in which a female can produce only one brood of eggs, although those eggs are assured of fertilization by males.

However, “females” (FPs) of *L. wurdemanni* are simultaneous hermaphrodites that can mate as males (Bauer and Holt, 1998). The delayed sex change of MPs would only be adaptive, under an ESD hypothesis, if large MPs had a male mating advantage over the hermaphroditic FPs (Bauer and Holt, 1998; Bauer, 2000). In this study, I tested the relative male mating success of MPs and FPs by presenting a parturial FP simultaneously to a pair of potential male mating partners. Surprisingly, MFPs (FPs mating as males) were just as competitive as MPs, both large and small, in obtaining copulations from parturial FPs. Although these experiments demonstrated no male mating advantage of large MPs over FPs, large MPs were more successful than small MPs in obtaining the copulation of parturial FPs in a competitive situation. If FPs were not simultaneous hermaphroditic and thus could not mate as males, the superiority of large MPs over small MPs in inseminating parturial FPs would agree with an ESD explanation of delayed MP change to FP. However, there is no obvious benefit (selection pressure) favoring delayed change of a MP to a FP (a) whose male mating ability is equal to that of MPs, large and small, and (b) whose reproduction as a female is not only assured but, given spawning every 2 weeks, would be more productive if change to FP occurred as early in life as morphologically and physiologically possible. In a population with few FPs, large MPs would have the mating advantage over small MPs, but there would be few FPs to exercise that advantage with. In a population with abundant FPs, large MPs would have more parturial FPs to mate with, but also more FPs, mating as males, to compete with for the same parturial FPs. Not surprisingly, the percentage of large MPs in the population was not significantly correlated with either low or high percentage of FPs. These results are not concordant with a hypothesis of socially determined sex change in *L. wurdemanni*. However, only large-scale experiments in which sex change in different demographic environments is measured can give a definite answer about whether or not socially mediated ESD occurs in *L. wurdemanni* or other *Lysmata* spp. Based on such large-scale experiments and parentage studies, Marliave et al. (1993) rejected the hypothesis of socially determined sex change in the protandric hermaphroditic shrimp *Pandalus danae*.

The hypothesis that FPs may change back to MPs in *Lysmata* under certain conditions, accounting for large MPs, was suggested by the anecdotal observations of colleagues rearing *L. wurdemanni* and by work on *Athenas indicus* (Suzuki, 1970) in which reversion of protandric FPs to MP was reported. Control of sexual differentiation by the androgenic glands (AG), located on the distal vas deferens (ejaculatory duct), has been demonstrated in various malacostracan crustaceans (Charniaux-Cotton and Payen, 1985; Hasegawa et al., 1993). Both males and females have primordia of the AG, but its development in genetic males causes growth of male external characters and testes. However, it is still not established that AG hormone is the only factor controlling sex differentiation in Crustacea (Sagi et al., 1997). Study of AGs in caridean shrimps is limited. Disappearance of the AGs after change to the female phase was reported, on the basis of stereomicroscopical and histological observations, in *L. seticarata* (Berreur-Bonnetfant and Charniaux-Cotton, 1965). In *L. amboinensis*, Fiedler (1998) reported an absence of AGs in FPs, but neither similar histological examination nor other observations on MP androgenic glands was given for comparison. However, as in *L. wurdemanni*, male ducts with sperm production are retained in FPs of both species. Fiedler (1998) showed that FPs of *L. amboinensis*, like those of *L. wurdemanni*, mate as males and inseminate prespawning FPs. A small number of *L. wurdemanni* FPs show external male characters (reduced appendices masculinae) (Bauer and Holt, 1998). Obviously, male function is maintained. Hormones maintaining male characters in FPs might still be produced by remnants of AGs retained from the MP stage but overlooked in histological studies. There must be a balance of male and female hormones in the FPs that allows simultaneous male and
female function. Therefore, the possibility of change from FPs back to large MPs was tested in this study.

In the FP to MP sex change experiment, one group of FPs was maintained under optimal breeding conditions of high water temperature and long day length (Bauer and Holt, 1998; Lin and Zhang, 2001) approximating those found in late spring and summer at the collecting site (Bauer, 2002). Given the hypothesis of socially mediated ESD proposed for pandalid shrimps (Charnov, 1981), reversion to MP, if physiologically possible, might be expected in an all-FP group under optimal breeding conditions. However, mating experiments in this study showed no strong selective advantage for large MPs in a population with abundant FPs, and no reversion to the male phase was observed in the treatment with optimal breeding conditions.

The suboptimal treatment tested the possibility that FPs exposed to less favorable breeding conditions, such as lower water temperatures and short day length, might revert to large MPs. In many caridean species, females molt out of “breeding dress” (characters related to spawning and incubation) outside of the breeding season. In the simultaneous hermaphroditic FP of L. wurdemanni, it was hypothesized that suboptimal breeding conditions might tip the balance of male/female hormones away from the female phase back to the male phase. The FPs in this treatment were exposed to water temperatures and a photoperiod approximating late fall through early winter conditions in the natural habitat of the population sampled (Bauer, 2002). The less favorable breeding conditions of the suboptimal treatment are apparent from a comparison of the reproductive condition and molting rates between the suboptimal and optimal treatments. The percentage of FPs brooding embryos or preparing for spawning was only slightly higher in the optimal than in the suboptimal treatment. However, the molting rate of the optimal treatment, adjusted for number of FPs surviving, was more than twice that of the suboptimal treatment. In L. wurdemanni FPs, spawning occurs at every molt (Bauer and Holt, 1998), so that the higher molting of the optimal treatment indicates that more broods were produced. At the temperature and photoperiod of the optimal treatment, molting and spawning may occur every 10–14 days (Lin and Zhang, 2001; Bauer, pers. obs.). Using a figure of 12 days between spawning molts, a FP in the optimal treatment could have molted and spawned five times during the 9 weeks of the experiment. Molting in the suboptimal treatment was less than half of that in the optimal treatment, so that the number of FP molts in this treatment was about two, with a maximum of two spawns possible. Although breeding conditions were poorer in the suboptimal treatment, no reversion of FPs to MPs occurred. Given that change from MP to FP takes place within a single molt cycle, reversion of FPs back to a male phase could have taken place within a single molt cycle as well. In the suboptimal treatment, FPs passed through two molt cycles, sufficient opportunity for reversion to the male phase, which did not, however, occur.

The high relative abundance of large MPs from fall through early spring might be best explained by suppression of sex change to FPs because of suboptimal breeding conditions. In the fall, MPs that have grown to and beyond the minimum size of sex change may not transform as they enter a period of the year not favorable for embryo production and larval survival. The proximate factors that retard sex change may be seasonally related environmental conditions such as lower temperature or shorter day length. The change of large MPs to FPs was much higher in conditions simulating spring/summer than in those simulating fall/winter conditions. Nearly all large MPs changed to FPs in the optimal treatment during the first 14 weeks of observation. There was much less change to FP during the same time period (14 weeks) when MPs were maintained under suboptimal (fall/winter) conditions. Molting rates were lower, but MPs of sufficient size to change to FP require only one molt cycle to do so (Bauer and Holt, 1998). The average MP in this treatment molted twice during the first 14 weeks.

After 14 weeks, the conditions of the suboptimal treatment were changed to those of the optimal treatment to determine if the slower rate of sex change could be increased in the surviving MPs. After 3 weeks (6 weeks without any MP to FP change), MPs began again to change to FP, and a majority did change or were changing to FP by the end of the experiment. During the same period, neither of the two MP treatments remaining in the original optimal treatment changed. At the end of 22 weeks two MPs from the original suboptimal treatment had neither changed nor showed signs of change to FP in spite of a switch from suboptimal to optimal conditions during the last 8 weeks of observation. Lin and Zhang (2001) found that all MPs reared individually under optimal conditions became FPs; but when MPs were reared in groups of 10, as in this study, only 67% transformed to FP, much less than that observed in the present study. Lin and Zhang began with postlarvae (MPs), and the total period of observation was not specified. In the present study, the experiment began with MPs at or above the minimum size of change to FP. Although Lin and Zhang suggested that a rather high percentage of individuals never change sex (primary males), my results suggest that most, if not all, will change to FP sometime during their lifetime if they live out the maximum natural lifespan (about 1.5 years; A. W. Baldwin and R. T. Bauer, unpubl.). However, in both experimental groups in my study, a low percentage of individuals (4%) never changed under laboratory conditions.

The adaptive advantage of not changing to FP during the non-breeding season may be that reproductive effort will be wasted on embryo production during a period unfavorable to larvae. This energy might be channeled into maintenance
and growth so that when spring arrives, the MP will change into a larger FP, with the benefit of a larger brood produced under conditions favorable to larval growth and survival. Brood size in *L. wurdemanni* is highly correlated with body size (Bauer, unpubl.), as in other carideans (Bauer, 1991). Suboptimal conditions did slow and then stop sex change in this study, explaining the presence of large and very large MPs during the fall through early spring in field samples. However, large MPs did not change to FP at a significantly larger size than large MPs reared under optimal (spring/summer) conditions.

In conclusion, mating experiments showed that FPs are competitive with MPs of any size in obtaining copulations with parturial FPs. There seems to be no mating advantage for an individual to remain a large MP, and under optimal breeding conditions, almost all change to FP. The proportion of large and very large MPs in the natural population studied is greatest from fall through early spring, and in the laboratory, change from MP to FP is slowed under the suboptimal breeding conditions of these seasons. Seasonal changes in abiotic proximate factors that regulate breeding, such as temperature and photoperiod, seem a more likely explanation for the occurrence of large MPs in *Lysmata wurdemanni* than does socially mediated ESD in different demographic situations. In *L. wurdemanni*, there may also be genetic variation in the size of sex change, although adaptive advantages to change at a larger size are not apparent from this and previous studies.

The proportion of FPs was often low in the *Lysmata wurdemanni* population sampled (Fig. 1). This is partly because all smaller (younger) individuals of this sex-changing species are male phase. In addition, as shown in this report and elsewhere (Bauer and Holt, 1998; Bauer, 2000), many large MPs capable of changing to FP do not do so for an extended period, so that the population often has a low proportion of FPs (Fig. 1). Therefore, the frequent occurrence of an extended male phase results in a lower number of FPs in the population (Fig. 1). This may affect the total output of larvae and subsequent recruitment potential of the population.

Acknowledgments

This study was funded by the National Science Foundation (Biological Oceanography Program, grant OCE-99822466) and the Neptune Foundation. I thank student assistants Aaron Baldwin and Sara LaPorte for their careful work on the project. This is Contribution No. 91 of the Laboratory for Crustacean Research, University of Louisiana at Lafayette.

**Literature Cited**

Bauer, R. T. 1976. Mating behaviour and spermatophore transfer in the shrimp *Heptacarpus pictus* (Stimpson) (Decapoda: Caridea: Hippolytidae). *J. Nat. Hist.* 10: 415–440.

Bauer, R. T. 1986. Sex change and life history pattern in the shrimp *Thor amboinensis* (Decapoda: Caridea): a novel case of partial protandric hermaphroditism. *Biol. Bull.* 170: 11–31.

Bauer, R. T. 1991. Analysis of embryo production in a caridean shrimp guild from a tropical seagrass meadow. *Pp. 181–191 in Crustacean Egg Production*, A. Wenner and A. Kuris, eds. A. A. Balkema, Rotterdam.

Bauer, R. T. 2000. Simultaneous hermaphroditism in caridean shrimps: a unique and puzzling sexual system in the Decapoda. *J. Crustac. Biol.* 20 (Special Number 2): 116–128.

Bauer, R. T. 2001. Hermaphroditism en camarones: el sistema sexual y su relacion con atributos socioecologicos. *Interciencia* 26: 2–7.

Bauer, R. T. 2002. Reproductive ecology of a protandric simultaneous hermaphrodite, the shrimp *Lysmata wurdemanni* (Decapoda: Caridea: Hippolytidae). *J. Crustac. Biol.* 22: 742–749.

Bauer, R. T., and J. H. Abdalla. 2001. Male mating tactics in the shrimp *Palaeonemus pugio* (Decapoda: Caridea): precopulatory mate guarding vs. pure searching. *Ethology* 107: 185–200.

Bauer, R. T., and G. J. Holt. 1998. Simultaneous hermaphroditism in the marine shrimp *Lysmata wurdemanni* (Caridea: Hippolytidae): an undescribed sexual system in the decapod Crustacea. *Mar. Biol.* 132: 223–235.

Berglund, A. 1986. Sex change by a polychaete: effects of social and reproductive costs. *Ecology* 67: 837–845.

Bergström, B. I. 1997. Do protandric pandalid shrimps have environmentally sex determination? *Mar. Biol.* 128: 397–407.

Bergström, B. I. 2000. The biology of *Pandalus*. *Adv. Mar. Biol.* 38: 1–245.

Berreur-Bonnenfant, J., and H. Charniaux-Cotton. 1965. Hermaphroditisme protéandrique et fonctionnement de la zone germinative chez la crevette *Pandalus borealis* Kroyer. *Bull. Soc. Zool. Fr.* 90: 240–259.

Bull, J. J. 1980. Sex determination in reptiles. *Q. Rev. Biol.* 55: 3–21.

Butler, T. H. 1980. Shrimps of the Pacific coast of Canada. *Can. Bull. Fish. Aquat. Sci.* 202: 1–280.

Chan, S. T. H., and W. S. B. Yeung. 1983. Sex control and sex reversal in fish under natural conditions. *Pp. 171–221 in Fish Physiology*, W. S. Hoar, D. J. Randall, and E. M. Donaldson, eds. Academic Press, New York.

Charniaux-Cotton, H. 1975. Hermaphroditism and gynandromorphism in malacostracan Crustacea. *Pp. 91–105 in Intersexuality in the Animal Kingdom*, R. Reinboth, ed. Springer Verlag, New York.

Charniaux-Cotton, H., and G. Payen. 1985. Sexual differentiation. *Pp. 217–299 in The Biology of Crustacea, Vol. 9, Integuments, Pigments, and Hormonal Processes*, D. E. Bliss and L. H. Mantel, eds. Academic Press, New York.

Charnov, E. L. 1981. Sex reversal in *Pandalus borealis*: effect of a fishery? *Mar. Biol. Lett.* 2: 53–57.

Charnov, E. L. 1982. *The Theory of Sex Allocation*. Princeton University Press, Princeton, NJ. 355 pp.

Charnov, E. L., and P. J. Anderson. 1989. Sex change and population fluctuations in pandalid shrimp. *Am. Nat.* 134: 824–827.

Charnov, E. L., D. W. Gotshall, and J. G. Robinson. 1978. Sex ratio: adaptive response to population fluctuations in pandalid shrimp. *Science* 200: 204–206.

Conover, O., and S. B. Munch. 2002. Sustaining fisheries yields over evolutionary time scales. *Science* 297: 94–96.

Dohrn, P. F. R. 1950. *Studi sulla Lysmata seticaitdata* Risso (Hippolytidae). I. Le condizioni normali della sessualita in natura. *Pubbli. Stn. Zool. Napoli* 22: 257–272.

Ferguson, M. W. J., and T. Joanen. 1982. Temperature of egg incubation determines sex in *Alligator mississippiensis*. *Nature* 296: 850–853.

Fiedler, G. C. 1998. Functional, simultaneous hermaphroditism in female-phase *Lysmata amboinensis* (Decapoda: Caridea: Hippolytidae). *Pac. Sci.* 52: 161–169.
Ghiselin, M. T. 1969. The evolution of hermaphroditism among animals. *Q. Rev. Biol.* **44**: 189–208.

Hasegawa, Y., E. Hirose, and Y. Katakura. 1993. Hormonal control of sexual differentiation and reproduction in Crustacea. *Am. Zool.* **33**: 403–411.

Lin, J., and D. Zhang. 2001. Reproduction in a simultaneous hermaphroditic shrimp, *Lysmata wurdemanni*: any two will do? *Mar. Biol.* **139**: 919–922.

Marliave, J. B., W. F. Gergits, and S. Aota. 1993. *Furcilla* pandalid shrimp: sex determination; DNA and dopamine as indicators of domestication; and outcrossing for wild pigment patterns. *Zoo Biol.* **12**: 435–451.

Munday, P. L. 2002. Bi-directional sex change: testing the growth-rate advantage model. *Behav. Ecol. Sociobiol.* **52**: 247–254.

Nakashima, Y. 1987. Reproductive strategies in a partially protandric shrimp, *Athanas kominatoensis* (Decapoda: Alpheidae): sex change as the best of a bad situation for subordinates. *J. Ethol.* **5**: 145–159.

Policansky, D. 1982. Sex change in plants and animals. *Annu. Rev. Ecol. Syst.* **13**: 471–495.

Ross, R. M. 1990. The evolution of sex-change mechanisms in fishes. *Environ. Biol. Fishes.* **29**: 81–93.

Sagi, A., E. Snir, and I. Khalaila. 1997. Sexual differentiation in decapod crustaceans: role of the androgenic gland. *Invertebr. Reprod. Dev.* **31**: 55–61.

Spitschakoff, T. 1912. *Lysmata seticaudata* Risso, als Beispiel eines echten Hermaphroditismus bei en Decapoden. Z. Wiss. Zool. **100**: 190–209.

Suzuki, H. 1970. Taxonomic review of four alpheid shrimps belonging to the genus *Athanas* with reference to their sexual phenomenon. *Sci. Rep. Yokohama Natl. Univ. Sect. II Biol. Geol. Sci.* **17**: 1–37.

Thornhill, R., and J. Alcock. 1983. *The Evolution of Insect Mating Systems*. Harvard University Press, Cambridge, MA. 547 pp.

Warner, R. R. 1975. The adaptive significance of sequential hermaphroditism in animals. *Am. Nat.* **109**: 61–82.

Wilson, E. O. 1976. *Sociobiology*. Harvard University Press, Cambridge, MA. 697 pp.

Zupo, V. 2001. Influence of diet on sex determination of *Hippolyte inermis* Leach (Decapoda: Natantia) in the field. *Hydrobiologia* **449**: 131–140.
Bauer, Raymond T. 2002. "Tests of Hypotheses on the Adaptive Value of an Extended Male Phase in the Hermaphroditic Shrimp Lysmata wurdemanni (Caridea: Hippolytidae)." The Biological bulletin 203, 347–357. https://doi.org/10.2307/1543577.

View This Item Online: https://www.biodiversitylibrary.org/item/17344
DOI: https://doi.org/10.2307/1543577
Permalink: https://www.biodiversitylibrary.org/partpdf/36032

Holding Institution
MBLWHOI Library

Sponsored by
MBLWHOI Library

Copyright & Reuse
Copyright Status: In copyright. Digitized with the permission of the rights holder.
License: http://creativecommons.org/licenses/by-nc-sa/3.0/
Rights: https://biodiversitylibrary.org/permissions

This document was created from content at the Biodiversity Heritage Library, the world's largest open access digital library for biodiversity literature and archives. Visit BHL at https://www.biodiversitylibrary.org.

This file was generated 16 April 2022 at 15:44 UTC