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Simultaneous hermaphroditism in the marine shrimp *Lysmata wurdemanni* (Caridea: Hippolytidae): an undescribed sexual system in the decapod Crustacea

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Abstract Simultaneous hermaphroditism with out-crossing, a previously unconfirmed sexual system in decapod crustaceans, is documented in the shrimp *Lysmata wurdemanni* (Gibbes), using time-lapse video observations on mating in pairs of “female-phase” (FP) individuals. Copulations between FPs resulted in successful spawning and development of embryos. However, female-phase hermaphrodites maintained in isolation were unable to self-fertilize spawned eggs. All smaller individuals possessed characters typical of caridean males, including male gonopores, appendices masculinae, ejaculatory ducts, and cincinnuli on the first pleopods. However, the gonad of these male-phase (MP) individuals was an ovotestis with an undeveloped ovarian portion. FPs, which spawn eggs and incubate embryos, also had male gonopores and an ovotestis terminating in ejaculatory ducts containing sperm. In FPs, male pleopod characters were absent or reduced, and a female incubatory character, expanded pleopod flanges, showed a greater relative size. Smaller size classes of the population were composed only of MPs while larger size classes consisted of both MPs and FPs. “Transitionals,” individuals with MP characters but an ovotestis containing vitellogenic oocytes, were rare and overlapped completely in size with FP and larger MP individuals. It is suggested that not all MPs transform into FP hermaphrodites. Social interactions may mediate MP change into FP hermaphrodites in *L. wurdemanni*. The social system of *L. wurdemanni* and other

Lysmata spp. may determine the exact form of the sexual system in these species.

Introduction

Although most species of decapod crustaceans have separate sexes, sequential hermaphroditism in the decapod Crustacea has been documented in a number of species, particularly caridean shrimp (Yaldwyn 1966; Policansky 1982; Bauer 1986). The form of hermaphroditism previously described has been protandrous, in which individuals first mature as males and then, with increasing age and size, change sex to females. Several variations of protandry are recorded in carideans. In some species, such as *Pandalus platyceros* and *Pandalopsis dispar* (Butler 1964, 1980), as well as *Lysmata seticaudata* (Spitschakoff 1912; Dohrn 1950), all individuals are reported to transform from males into females. However, in other protandric carideans, a variable proportion of the population mature as “primary” females without a male phase (Pandalidae: Butler 1964, 1980; Charnov et al. 1978; Bergström 1997; Crangonidae: Fréchette et al. 1970; Processidae: Noël 1976; Boddeke et al. 1991). Conversely, in the hippolytid *Thor manningi*, the population is composed of 50% primary males and 50% protandric hermaphrodites (Bauer 1986). Primary males and sex changers compose the population of the alpheid *Athanas kominatoensis* (Nakashima 1987), while a mix of primary females, primary males, and sex changers are found in *A. indicus* (Gherardi and Calloni 1993).

Simultaneous or synchronous hermaphroditism which is functional, i.e. an individual may serve male or female at any given time, has not previously been documented in the order Decapoda, nor in any other malacostracan (Charniaux-Cotton and Payen 1985). Buchanan (1963) reviewed and added to morphological and demographic observations that suggested simultaneous hermaphroditism in the burrowing thalassinid

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Calocaris macandreae. Kagwade (1982) concluded, on similar grounds, that the hippolytid caridean *Exhippolysmata ensirostris* first passed through a male phase and then retained functional testicular tissue and male ducts in the female phase. However, no mating observations have been done to confirm these possible cases of synchronous hermaphroditism.

Spitschakoff (1912) illustrated and described the gonads of the caridean *Lysmata seticaudata*. In this species, the gonads are ovotestes, in which the testicular part is well developed in smaller individuals while the ovarian part becomes prominent in older, larger (female-phase) individuals. The species has been described as a protandrous hermaphrodite by various workers such as Spitschakoff himself, Dohrn (1950), Berreur-Bonnefant and Charniaux-Cotton (1965), Charniaux-Cotton (1975), and Charniaux-Cotton and Payen (1985). Berreur-Bonnefant and Charniaux-Cotton demonstrated with histology that the anterior region of male-phase individuals of *L. seticaudata* was the ovarian portion of an ovotestis while the posterior part of the gonad was testicular. Extensive histological study of the gonads and gametogenesis of *L. seticaudata* by these authors showed not only that male ducts are retained in the female phase but also that spermiogenesis and sperm production continue after the sex change. The possibility of concurrent male and female sexual function was not addressed.

Reports on *Lysmata* spp. in which "females" are described as mating and producing fertile offspring with other "females" have become frequent in the non-refereed literature (Bundy 1983; Riley 1994; Wilkerson 1994). Fletcher et al. (1995) suggested, on the basis of preliminary observations, that *L. debelius* and *L. amboinensis* are simultaneous hermaphrodites. These anecdotal reports and preliminary observations, as well as previous indications of synchronous hermaphroditism in the literature, stimulated the present investigation on the sexual system of *L. wurdemanni* (Gibbes), the "red cleaning" or "peppermint" shrimp from the Gulf of Mexico. The purpose of this study was to determine if female-phase individuals are true simultaneous hermaphrodites, capable of mating and reproducing as either sex during the same reproductive period. The hypothesis that individuals of *L. wurdemanni* are simultaneous hermaphrodites was tested with observations on matings recorded with time-lapse video, other aspects of the reproductive biology of live animals, reproductive morphology, and population structure.

Materials and methods

Populations of *Lysmata wurdemanni* (Gibbes) were sampled with 1 mm-mesh dipnets during nocturnal low tides from pools and under ledges of the rock jetty adjacent to the marine laboratory of the University of Texas, Austin, at Port Aransas, Texas, USA (27°50'N; 97°03'W). Shrimp collected on 19 and 20 July 1994 and 11 July 1995

were preserved in 10 to 15% seawater formalin or Davidson's solution, later washed in water, and stored in 70% ethyl alcohol. From the combined July 1994 sample, some individuals were removed haphazardly for live work before the rest of the sample was preserved. All individuals in the July 1995 sample were preserved. Live specimens used for the mating study were mainly collected on 12 June 1995, with a few specimens taken from the July 1994 sample, along with several reared in the laboratory by one of the authors (GJH) (breeding and rearing techniques given in Riley 1994).

Morphometric observations were done on specimens from the July 1994 sample. Carapace length, a measure of body size, was measured as the distance from the posterior edge of the eye orbit to the middorsal posterior edge of the carapace. Appendix masculina length was measured from its base to its tip, spines excluded. Presence or absence of openings (male gonopores) on the coxae of the fifth pereopods was recorded. An ejaculatory duct was dissected from the body wall/coxa juncture of the fifth pereopod, and its maximum width was measured. The number of coupling hooks (cincinnuli) on the endopod of the first pleopod was counted, as well as the number of spines on the appendix masculina. Width of the flange on the basipod of the second pleopod, narrow in males but expanded in incubating females in carideans (Höglund 1943) was measured. Degree of ovarian development in the gonad was staged from 1 (no vitellogenic oocytes observed) to 4 (maximum filling of ovarian portion with vitellogenic oocytes) as in Bauer (1986); degree of development of incubated embryos was also staged from 1 (just spawned) to 4 (near hatching) as in Bauer (1986). Measures of carapace length, as well as observations on ovarian condition, incubated embryos, presence or absence of appendices masculinae, and Pleopod 1 cincinnuli were done on specimens from the July 1995 collection for construction of length-frequency diagrams of population structure.

Length-frequency diagrams describing population structure and breeding condition were constructed for the 1994 and 1995 samples. "Male-phase" (MP) individuals were characterized by the occurrence of cincinnuli on the first pleopods (P1), a male character in most carideans (Bauer 1976; Butler 1980), along with a non-vitellogenic gonad. "Female-phase" (FP) individuals were those lacking P1 cincinnuli; "transitional" individuals were those with P1 cincinnuli which displayed stages 2 to 4 ovarian development in the gonad.

Live specimens were maintained in aquaria with seawater at 33 to 36‰, 25 to 28 °C, and a 14 h light:14 h dark cycle. In mating experiments, pairs of female-phase individuals, one in Ovarian Stage 3–4 (near a prespawning molt), were confined by a porous partition into a space 21 long × 24 wide × 15 cm high within a 38 liter aquarium. Individuals chosen for observations on mating and spawning carried embryos or had just hatched a brood, i.e. had reproduced as females. Activities were recorded with an infrared-sensitive surveillance camera equipped with an 8 or 12.5 mm lens, connected to a time-lapse video recorder. Day illumination was by overhead fluorescent lights, that at night by infrared lamps (880 nm). Videotapes were recorded at 24 h speed (5 frames/s) daily until molting and spawning of the prespawning individual. After the molt of the latter, observations were made on possible spawning (gonad condition, presence or absence of embryos under the abdomen). Embryos in spawned broods were checked every 2 to 3 d until eyespots became visible, confirming successful fertilization and embryonic development.

Another group of female-phase individuals, carrying embryos and with obvious vitellogenic gonads, were maintained individually in aquaria. Each was checked daily through hatching of embryos, the posthatching molt, and possible spawning. After molting, observations were continued to determine if successful spawning had occurred (gonadal condition, presence of embryos under the abdomen).

Scanning electron microscopy of spermatophore material and sperms was done with standard SEM techniques as given in Bauer (1996a)

Results

Reproductive morphology and population structure

All individuals of *Lysmata wurdemanni* that were examined had gonopores on the coxae of the fifth pereopods, typical of male caridean shrimp. Dissections showed that all individuals in the July 1994 sample ($n = 184$), including “females” brooding embryos, had an ejaculatory duct (Figs. 1; 2d) characteristic of male carideans. Individuals with well-developed male appendices (“male-phase, transitionals”) occurred in both smaller and larger size classes of the population, while those carrying embryos (“female-phase”) were present only in larger size classes (Fig. 3).

Female-phase morphs (FPs), defined by the absence of cincinnuli on Pleopod 1 (Figs. 1a,b; 5a), varied from 5.5 to 9.1 mm CL (carapace length) in the 1994 sample ($\bar{x} = 7.6 \text{ mm} \pm 0.8 \text{ SD}$, $n = 49$) and from 6.7 to 9.8 mm CL in the July 1995 sample ($\bar{x} = 8.1 \text{ mm} \pm 0.6 \text{ SD}$, $n = 68$) (Fig. 3). Appendices masculinae, a characteristic of male caridean shrimp, could be observed in a reduced condition on the second pleopods of many but not all FPs (Figs. 1a,b; 4a). When appendices could be observed at 50 \times with a stereomicroscope, they were usually short, compared to other sexual morphs, and without spines (Figs. 4a; 5b). In some FPs recorded as lacking an appendix masculina, a possible vestige of this structure could be observed at higher magnification (100 \times) with transmitted light (Fig. 1b).

Male-phase morphs (MPs) were defined by the presence of cincinnuli on the first pleopod (Figs. 1d,e; 5a) and a non-vitellogenic ovotestis (Fig. 1d,e). Carapace length varied in MPs from 3.6 to 8.2 mm ($\bar{x} = 5.6 \text{ mm} \pm 1.2 \text{ SD}$, $n = 108$) and from 3.5 to 7.8 mm ($\bar{x} = 5.8 \text{ mm} \pm 1.0 \text{ SD}$, $n = 116$) in the 1994 and 1995 collections, respectively. MPs had well-developed appendices masculinae, with several spines, on the endopods of the second pleopods (Figs. 1d,e; 4a; 5b). The correlation between appendix masculina length and body size (carapace length) was highly positive and statistically significant in MPs compared to a weakly negative one in FPs (Table 1).

Many FP morphs had large vitellogenic oocytes in the gonad (Figs. 1a; 2c; 3), and constituted that part of the population which incubated embryos (Fig. 3). However, in all FPs examined ($n = 49$ in July 1994 sample; $n = 63$ from mating and spawning observations, see following subsection), the gonad, in addition to bearing oviducts, terminated in a testicular portion with vasa deferentia leading to ejaculatory ducts typical of male carideans (Figs. 1a,b; 2a,b,d; 4b). Ejaculatory ducts of FPs were comparable in relative size to those of MPs (Fig. 4b). Correlations between ejaculatory duct size (width) and body size (carapace length) were positive and significant for both FPs and MPs, but the correlation for FPs was lower, reflecting higher variability in FPs for this character (Fig. 4b; Table 1).

Spermatophore material squeezed from the ejaculatory ducts of live FPs, including those carrying spawned embryos, contained thumbtack-shaped sperm typical of male carideans (Fig. 6). Oviducts as well as vasa deferentia were readily observable on the gonads of larger MPs, but were not apparent in dissections of smaller MPs (Fig. 1d, e). The relative width of the Pleopod 2 flange appears greater in FPs than in MPs (Fig. 4c). The positive correlation between flange width and carapace length is higher in FPs than in MPs (Table 1).

Transitionals, individuals with cincinnuli on the first pleopods, with vitellogenic oocytes in the gonad (Stage 2 to 4 ovarian condition) (Fig. 1c), but without attached embryos, were rare, ranging in size from 5.5 to 8.7 mm CL ($\bar{x} = 6.8 \text{ mm} \pm 1.0 \text{ SD}$, $n = 8$) in July 1994 and 6.4 to 8.3 mm CL ($\bar{x} = 7.5 \text{ mm} \pm 0.6 \text{ SD}$, $n = 11$) in 1995 (Fig. 3). Transitionals resembled MPs in the presence of well-developed appendices masculinae with spines (Figs. 1c; 4a; 5b), although the correlation between appendix masculina length and carapace length was weak (Table 1).

Transitionals were intermediate between MPs and FPs in the relative size of the pleopod flange (Fig. 4c; Table 1). Like both MPs and FPs, transitionals had well-developed ejaculatory ducts (Fig. 1c; 4b); the correlation coefficient of ejaculatory duct width with body size (carapace length) was positive, weaker than that for MPs but stronger than that for FPs (Table 1). However, none of the correlation coefficients calculated for the transitionals, with a small sample size ($n = 8$) were statistically significant.

Mating and spawning in female-phase pairs

The hypothesis that FPs are able to copulate successfully both as male and female during the same reproductive period was tested by observing pairs of FPs ($n = 22$ replicates) in which at least one of the pair was nearing a prespawning molt. Both individuals of these pairs were incubating embryos at the beginning of the observation period; it was thus certain that they had previously reproduced as females. Seventeen of the pairings were recorded with time-lapse video. Recordings were made daily until the initial prespawning female had molted and spawned; in 8 of these 17, observations were continued until both FPs had molted once. Spawned individuals were maintained and followed to determine if embryonic development of spawned eggs had occurred (Fig. 2e,f).

Within 1 to 4 d after hatching previously attached embryos, females with a prespawning ovotestis molted [median (md) < 1 d, $n = 25$] (Figs. 7a, b, c; 8a, b, c). In 20 of the 23 prespawning molts observed on video, the molted FP was approached by the other FP (Figs. 7d; 8d) within a median time of 28 s [95% confidence limits (cl), 5, 80 s] and copulated with it a single time (Figs. 7e; 8d). In 5 of these 20 cases, the individual acting as a male seized the molting FP before it had completely emerged from the exuviae (molt skin). In all cases, the non-molting FP

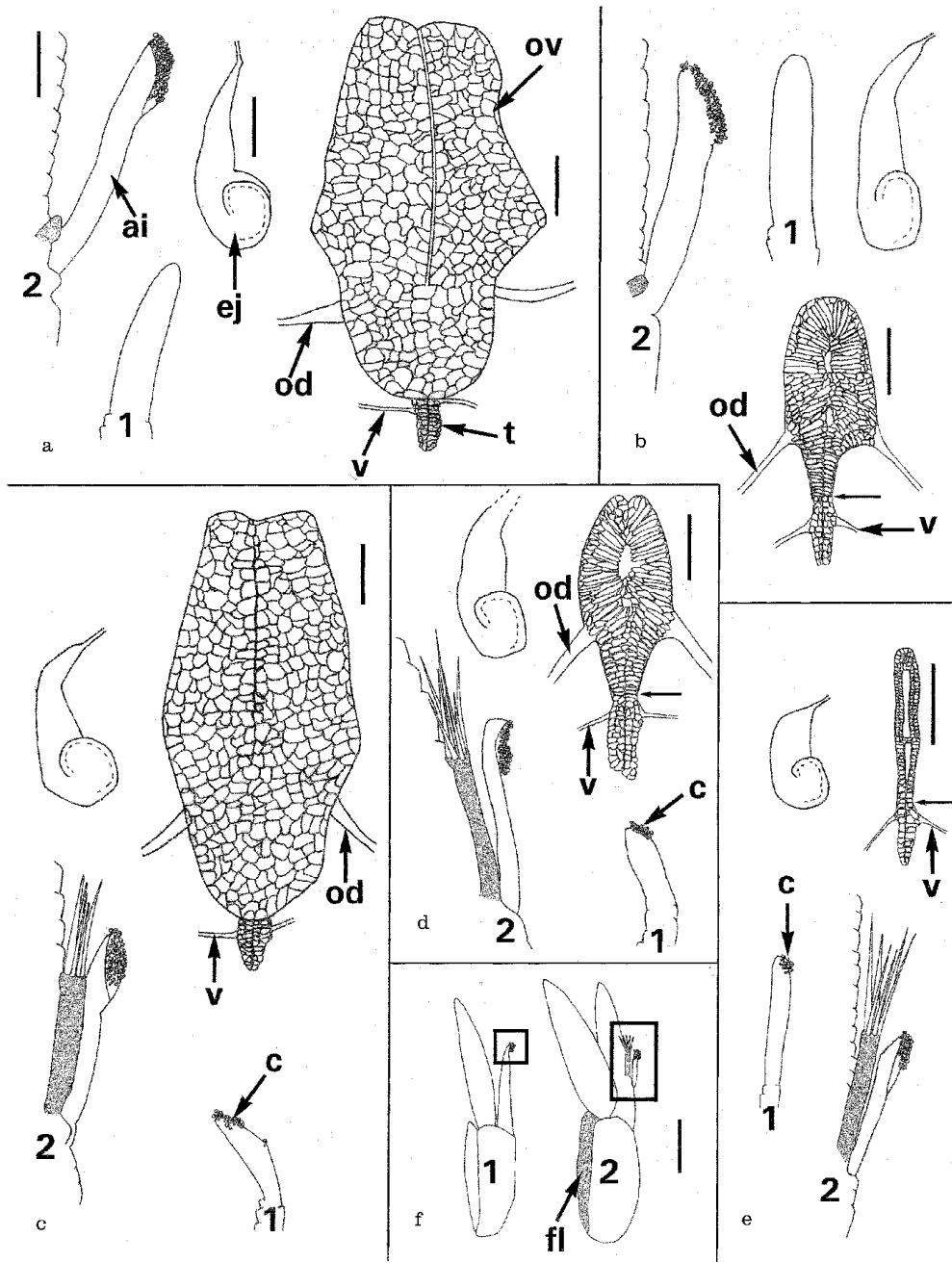


Fig. 1 *Lysmata wurdemanni*. Reproductive morphology of sexual morphs: gonads, ejaculatory ducts, sexual characters of Pleopods 1 and 2. **a** Female-phase (FP) individual, 8.1 mm CL (carapace length), that carried embryos near hatching, with ovarian portion of gonad (ov) distended by vitellogenic oocytes; **b** FP, 7.9 mm CL, that bore recently-spawned embryos, with ovarian portion of gonad in undeveloped (spawned out) condition; **c** transitional individual, 7.5 mm CL, with ovarian portion distended by vitellogenic oocytes; **d** large male-phase (MP) individual, 7.8 mm CL; **e** small MP, 4.6 mm CL; **f** diagram of left Pleopods 1 and 2, anterior view (boxes areas of endopods amplified in **a** to **e**) [Unmarked arrows to right of gonads in **b**, **d** to **e** indicate point at which distinct change occurred in staining with acid fuchsin between anterior (oviduct-bearing) and posterior (vas-bearing) parts of gonad; in **a** and **c**, large anterior part was filled with

vitellogenic oocytes, distinct from small testicular portion; no oviducts could be observed in gonad of small MP in **e**; ai appendix interna of endopod of Pleopod 2 (labeled only in **a**); c cincinnuli (coupling hooks) on tip of endopod of Pleopod 1, a male character; fl basipod flange (shaded) of pleopod; od oviduct; t testicular portion of gonad (labeled only in **a**); v proximal vas deferens; 1,2, in **a-e** magnified portions of endopods of Pleopods 1 and 2; shaded portions of endopods of Pleopods 2 are appendices masculinae fully developed with spines in **c** to **e**, reduced to spineless nubs in **a** and **b**) Scale bars (= 1 mm) are given to right side of each gonad in **a** to **e**; ejaculatory ducts in **a** to **e** are to same scale [scale bar to right of ejaculatory duct (ej) in **a**], as are all endopod areas in **a** to **e** (scale bar to left of Endopod 2 in **a** 0.25 mm); scale bar in **f** = 1 mm

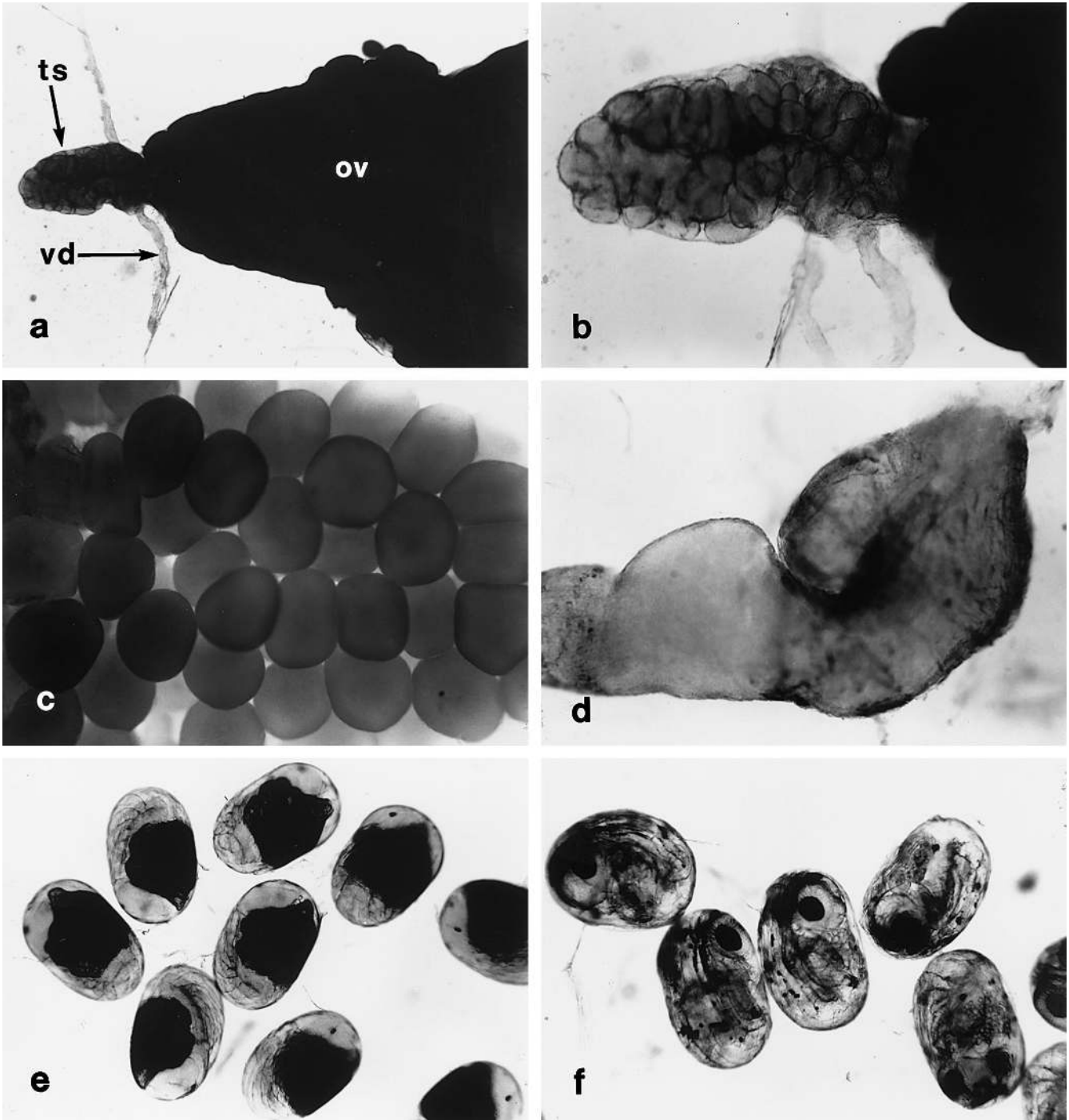


Fig. 2 *Lysmata wurdemanni*. Reproductive morphology and embryos. **a** Gonad (ovotestis), posterior half, dorsal view from female-phase (FP) individual incubating embryos, showing ovarian (*ov*) and testicular (*ts*) portion with vas deferens (*vd*); **b** higher magnification of posterior end of ovotestis, showing testes and right vas deferens at end of ovarian portion; **c** mature oocytes from gonad shown in **a** and **b**; **d** ejaculatory duct at end of vas deferens from same gonad as in **a** to **c**; **e** developing embryos at eyespot stage, dark mass in embryo is unused yolk; **f** embryos near hatching, spawned and incubated by FP individual which copulated with another FP. Scale bars = 1.1 mm in **a**, 370 μ m in **b** to **f**

copulated from below (Figs. 7e; 8e), a position common in caridean males. The copulatory position was maintained for several seconds ($md = 7$ s; 95% $cl = 5,9$ s).

In two cases, both FPs of each pair were near a spawn and molted within hours of each other on the same night. In these pairs, each FP copulated as a male when the other member of the pair molted and then as a female when it molted itself. All four FPs involved in these reciprocal matings then spawned a successful brood of embryos. In six other pairs, maintained long enough for both FPs to undergo a prespawning molt, six FPs

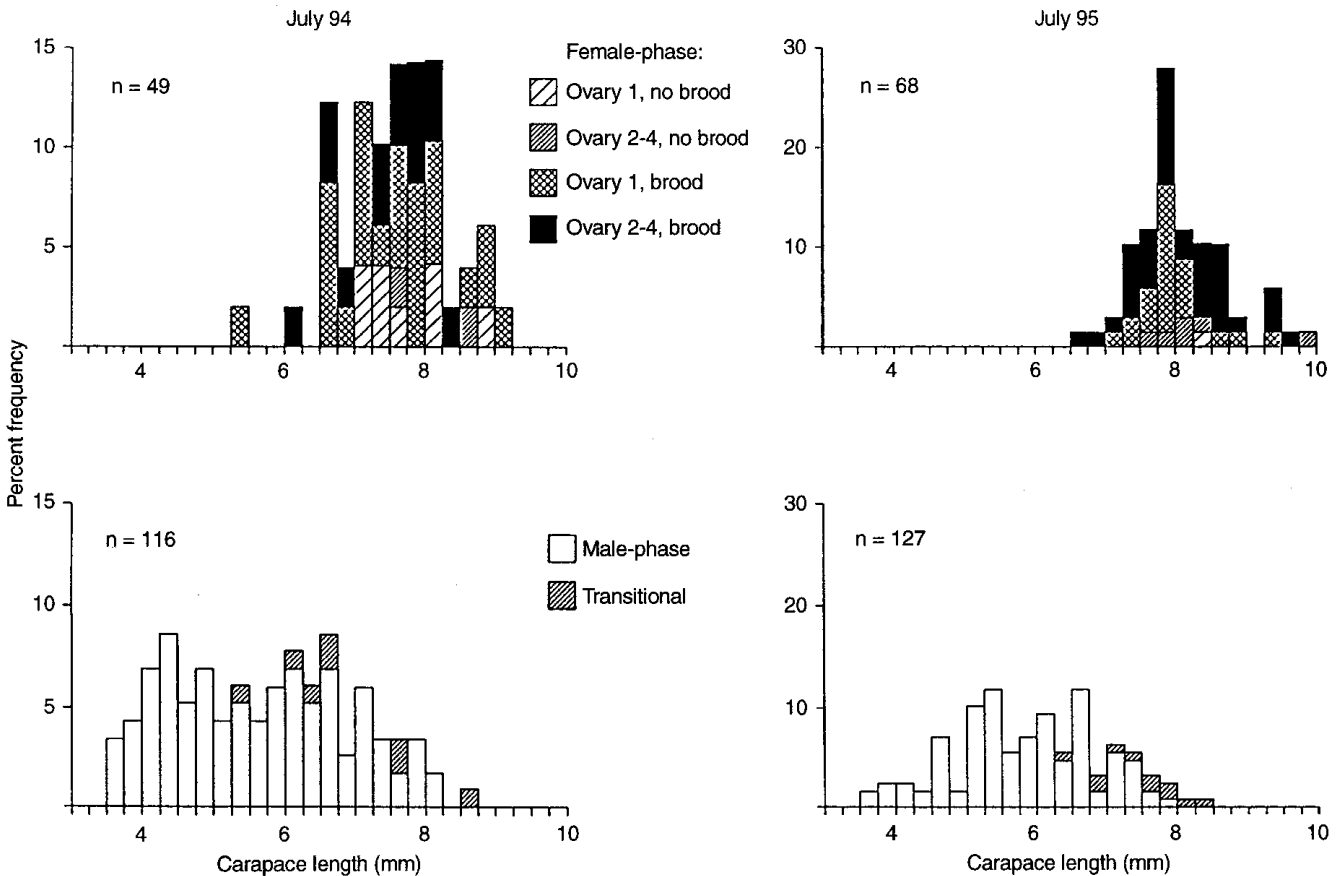


Fig. 3 *Lysmata wurdemanni*. Population structure and reproductive condition of sexual morphs from collections made in July 1994 and July 1995 (*Ovary Stages 1 to 4* after Bauer, 1986: see "Materials and methods")

copulated as both male and female, two as female only, two as male only, and in one pair there was no copulation after the molt of neither FP.

Of 20 prespawning, postmolt FPs observed on video copulating with another FP, all spawned a brood of eggs within a few hours of mating (md = 2.7 h; 95% ci = 2.5, 2.8 h) (Figs. 7f; 8f). In 15 of these 20 FPs, embryos were fertilized and underwent embryonic development (Fig. 1e, f). In the remaining 5 cases, the presumably unfertilized eggs were detached from the abdomen within a few hours to days. In the five replicates not recorded with video, all molting FPs produced a successful spawn of embryos.

All FPs used in mating experiments were later examined and found to have fully formed ejaculatory ducts. Gonadal condition of FPs which had served as male mating partners was examined; all stages of ovarian development were observed. FPs incubating embryos had a similar rate of success in copulating as males and fertilizing other FPs (75%, $n = 16$) as did FPs which, due to hatching, had no attached embryos when copulating as males (83%, $n = 12$).

Spawning in solitary female-phase individuals

The possibility of self-fertilization or fertilization from stored sperm was tested by individually isolating 17 incubating FPs which had gonads full of mature oocytes.

These FPs were observed daily up to and after the posthatching, prespawning molt. There was no successful spawning by any of these FPs. In ten cases, eggs were attached for 1 to 2 d after spawning but were discarded before development could be observed. In four cases, spawned eggs were never observed, but mature oocytes in the gonad had disappeared 1 to 2 d after molting, indicating unsuccessful spawning. In three cases, spawning apparently never took place, since abundant mature oocytes remained in the gonad; with the passage of days and weeks, these oocytes gradually became smaller and appeared necrotic. All 17 FPs were later examined and all had typical ejaculatory ducts.

Exuviae recovered from the prespawning molts of these FPs, as well as many of those used in the mating observations, were examined ($n = 39$) for presence of sperm storage structures, e.g. cuticular spermathecae. No stored sperm nor sperm-storing structures were observed.

Discussion

Functional simultaneous hermaphroditism is known in crustaceans, occurring in remipedes (Yager 1991), cephalocarids and thoracican barnacles (Charniaux-

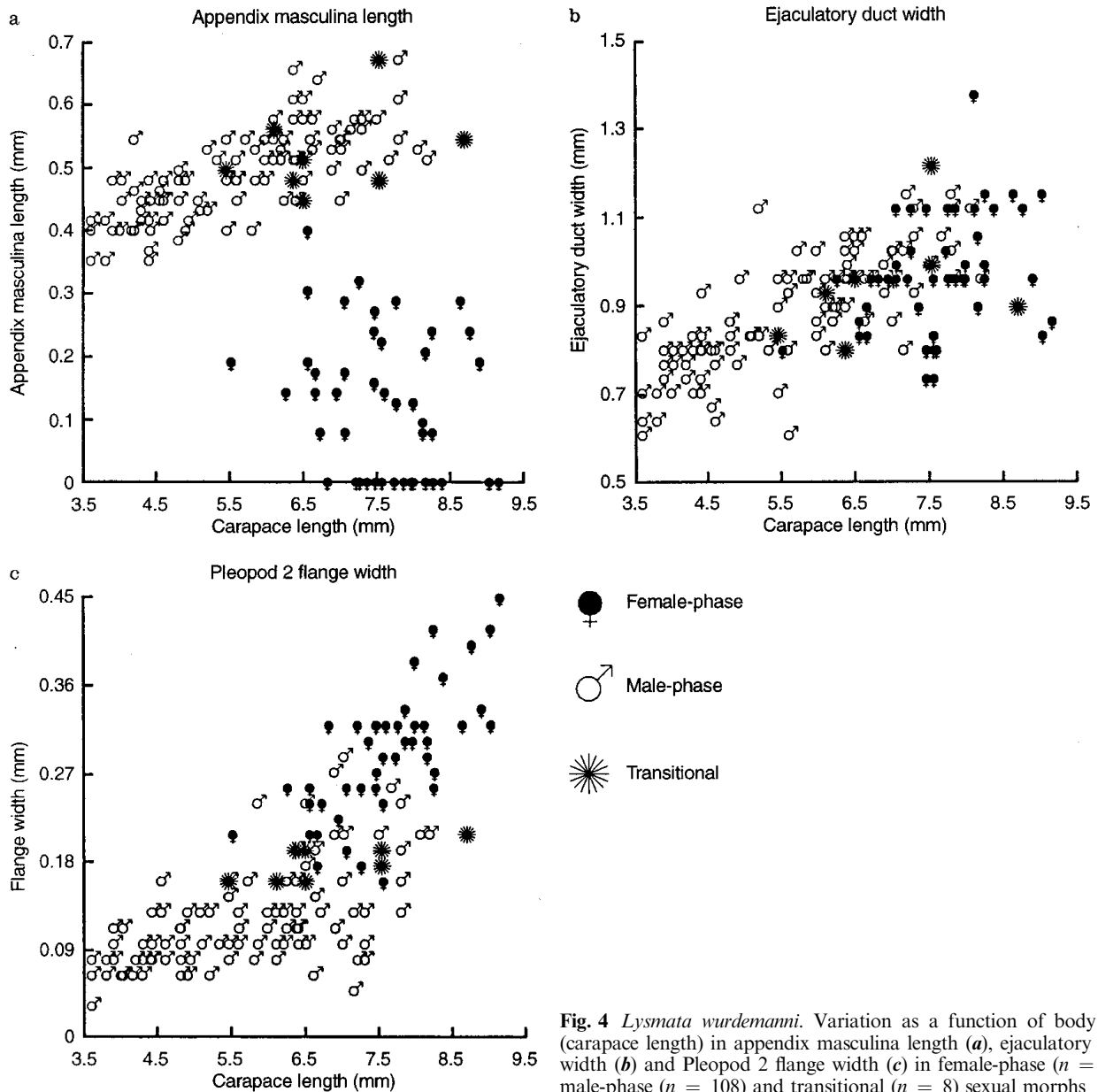


Fig. 4 *Lysmata wurdemanni*. Variation as a function of body size (carapace length) in appendix masculina length (a), ejaculatory duct width (b) and Pleopod 2 flange width (c) in female-phase ($n = 49$), male-phase ($n = 108$) and transitional ($n = 8$) sexual morphs

Cotton and Payen 1985), notostracans (Ghiselin 1969), and in conchostracans, (in which selfing may occur: Sassaman and Weeks 1993). The sexual system described in the present report for *Lysmata wurdemanni*, documented with mating studies, is the first confirmed case of synchronous hermaphroditism in decapods or any malacostracan. Sex change, however, is relatively common in malacostracans. Protandry, in which an individual first matures as a male and then reproduces as a female later in life, is the form of sequential hermaphroditism found in caridean shrimps (Yaldwyn 1966; Bauer 1986). Thus it seems probable that the sexual system of *L. wurdemanni* has evolved from a protandric ancestor. Apparently in this species, smaller (younger) individuals can only reproduce as males; only larger, (older)

individuals are functional hermaphrodites, serving as the breeding females of the population. Instead of losing male capabilities when entering the female phase, as in other sex-changing carideans, female-phase individuals retain male ducts, sperm, and the ability to copulate successfully as males. The gonads resemble those of *L. seticaudata*, in which Berreur-Bonnefant and Charniaux-Cotton (1965) confirmed, with histological studies, Spitschakoff's (1912) conclusion that these structures were ovotestes in all individuals.

Our mating observations showed that FPs of *Lysmata wurdemanni* are true outcrossing hermaphrodites, with the remarkable capability of copulating and inseminating as males on the very night in which they themselves undergo a spawning molt and copulate, as

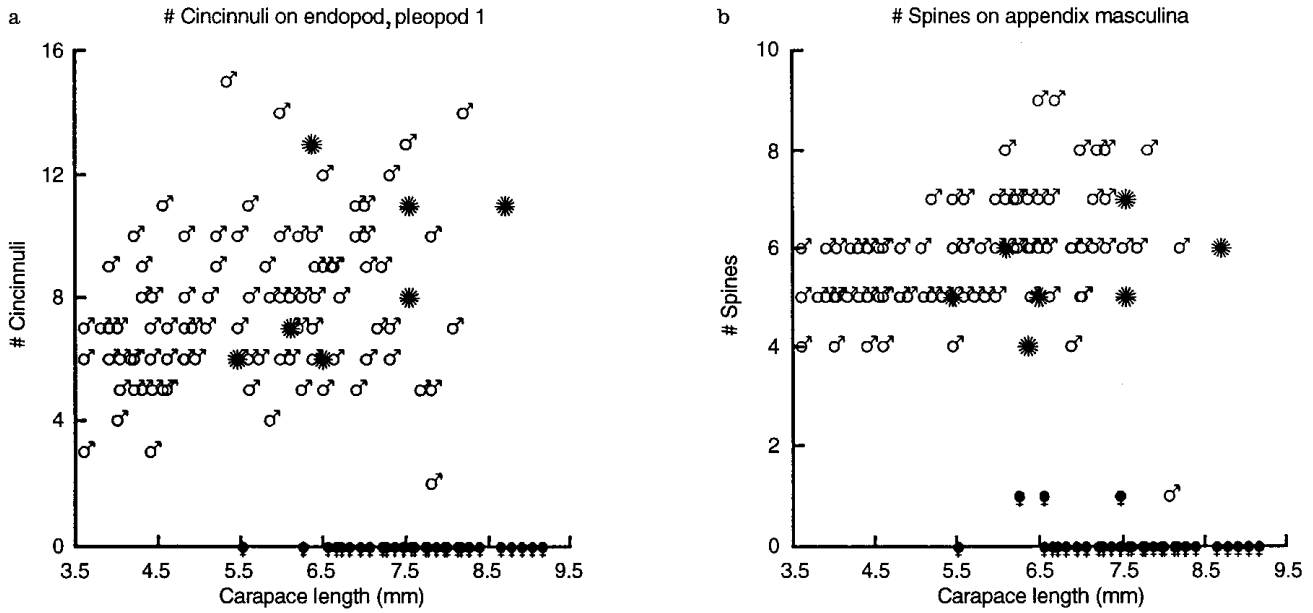


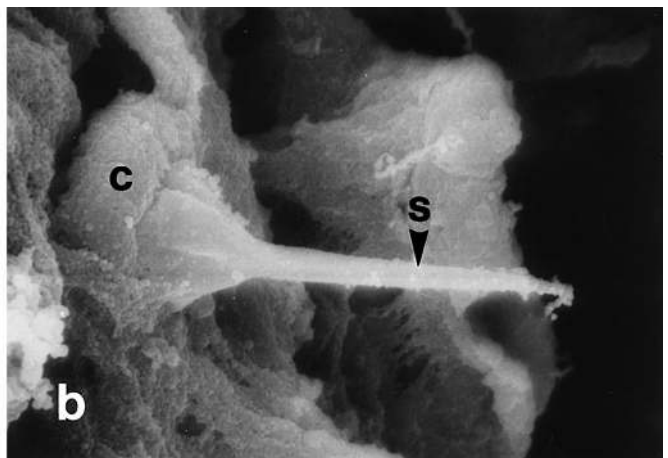
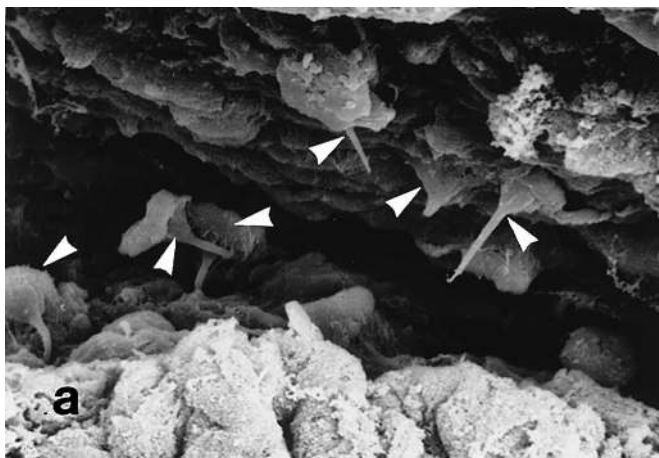
Fig. 5 *Lysmata wurdemanni*. Variation as a function of body size (carapace length) in number of cincinnuli on endopod of first pleopod (a) and spines on appendix masculina (b) in female-phase ($n = 49$), male-phase ($n = 108$) and transitional ($n = 8$) sexual morphs (Symbols as in Fig. 4)

females, with another FP. FPs mated with other FPs produced fertile spawns of embryos which developed in an apparently “normal” manner, i.e. went through the stages of embryonic development resembling those described for other caridean species (Bauer 1991). When development was followed to completion, embryos hatched as swimming larvae. However, isolated FPs were incapable of producing fertile spawns. As in most other caridean species, there is no sperm storage in female or female-phase individuals (see Boddeke et al.

Fig. 6 *Lysmata wurdemanni*. Sperm from spermatophore material ejaculated from female-phase individual incubating brood of embryos. a Several sperm (at tips of arrows) embedded in spermatophore material; b single sperm cell composed of typical caridean cap (= main body) (c) and spike (s). Scale bars = 8 μ m in a, 2 μ m in b

Table 1 *Lysmata wurdemanni*. Correlations of measures of various sexual characters with body size (carapace length) in sexual morphs (Fig. 4) [FP female-phase morph; MP male-phase morph; T transitional morph; r_s Spearman’s rank correlation coefficient; p = probability of null hypothesis that $r_s = 0$ with significance value (α) = 0.050; N number of observations]

Character, sexual morph	r_s	p	(N)
Length of appendix masculina			
FP	-0.284	0.051	(48)
MP	0.776	< 0.002	(108)
T	0.164	> 0.050	(8)
Width of ejaculatory duct			
FP	0.388	0.007	(49)
MP	0.793	< 0.002	(108)
T	0.553	> 0.050	(8)
Width of flange, Pleopod 2			
FP	0.715	< 0.002	(48)
MP	0.631	< 0.002	(108)
T	0.684	> 0.050	(8)



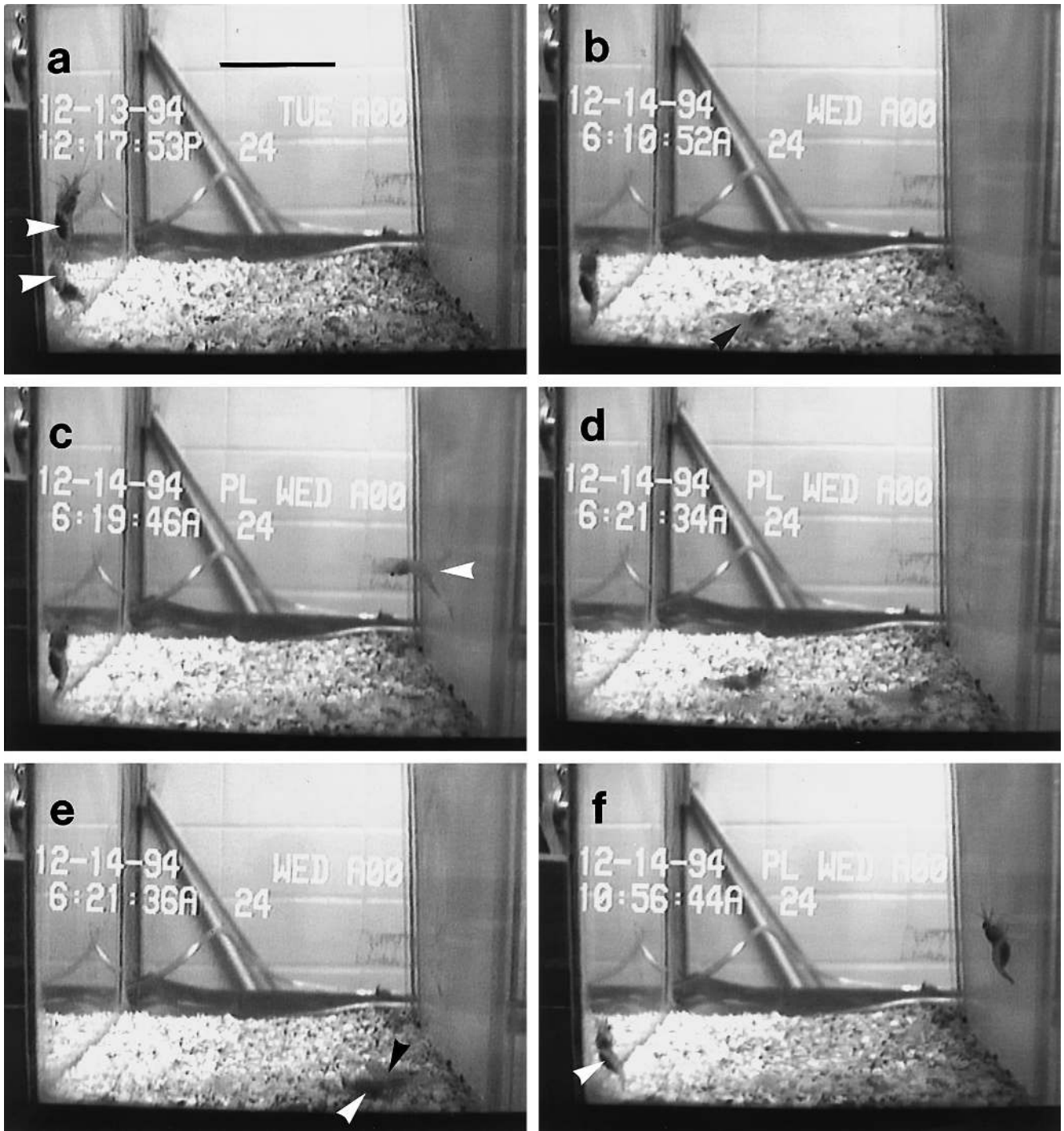
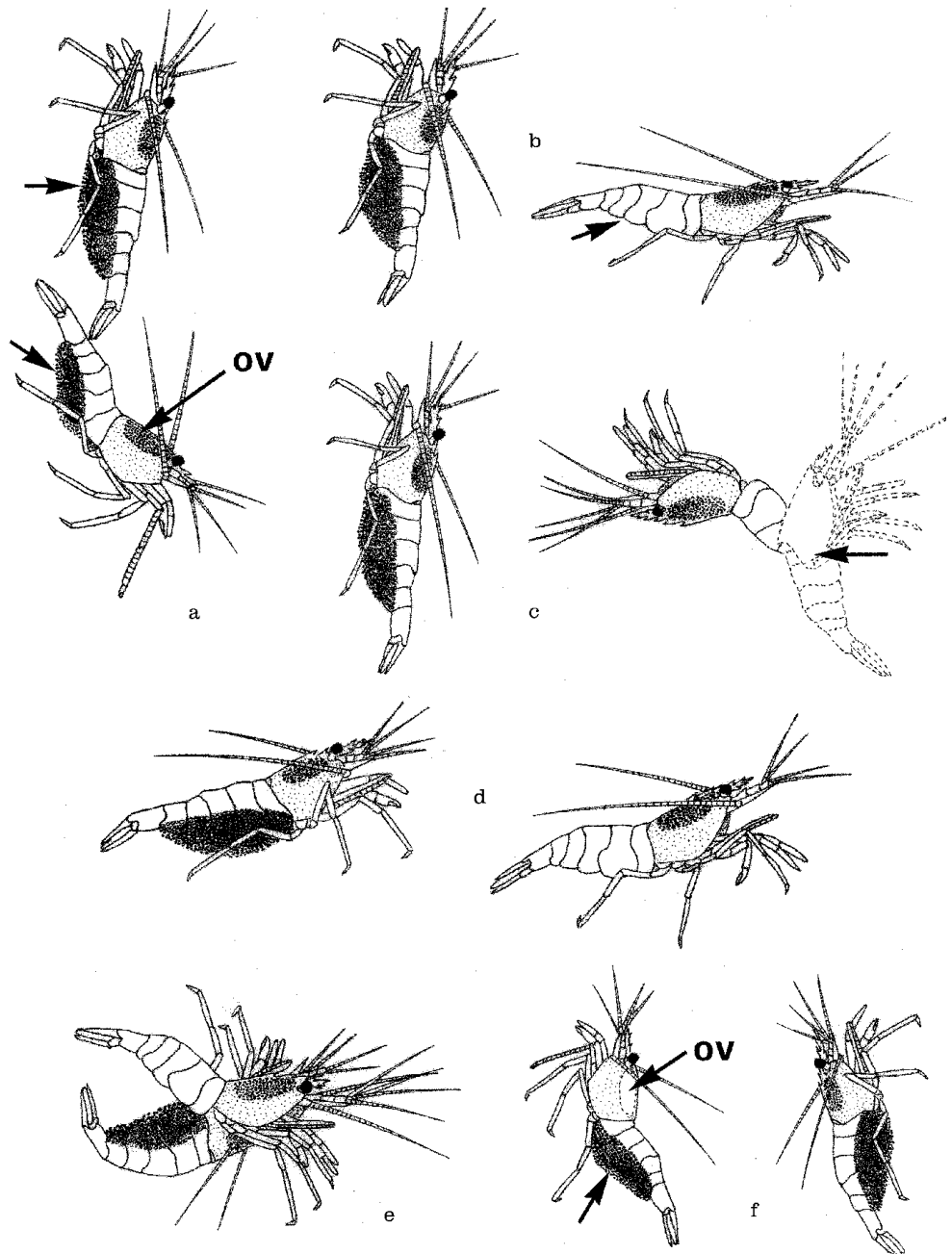


Fig. 7 *Lysmata wurdemanni*. Video sequence of molting, mating, and spawning in female-phase (FP) individual paired with another FP, 13-14 December 1994. **a** Both FPs with brood of embryos (dark masses at tips of arrows) below abdomen (late afternoon 13 December); prespawning FP perched below other FP; ovarian portion of gonad filled with vitellogenic oocytes (*ov* in Fig. 8a). **b** Following afternoon, embryos of prespawning FP have hatched (arrow indicates embryo mass absent); hatching occurred between 22:30 and 23:00 hrs. **c** Ecdysis by prespawning female, seen emerging from exuviae

(arrow indicates molt skin; dotted outlines in Fig. 8c). **d** Approach of FP with embryos (lower center) just before copulating with molted FP (lower right). **e** Copulation: FP with embryos (white arrow) upside down in mating position below (black arrow) molted FP. **f** Molted and mated FP (on left) has spawned and carries new brood of embryos below abdomen (arrowed); note clear space (*ov* in Fig. 8f) in upper cephalothorax, behind eyes, which was filled with vitellogenic oocytes prior to spawning (cf. with a). Scale bar in a = 5 cm

Fig. 8 *Lysmata wurdemanni*. Diagrammatic representation of individuals shown in video sequence in Fig. 7 (details as in legend to Fig. 7)



1991 for a possible exception in *Crangon crangon*). Self-fertilization did not occur, although it seems morphologically possible. FPs possess ejaculatory ducts with sperm when they spawn as females; spermatophore material could conceivably be self-deposited in the path of spawned eggs. However, this did not take place. The high density of individuals in the population studied, all morphologically capable of mating as males, would make selfing in FPs unnecessary, especially given the negative genetic consequences of self-fertilization.

Not all male-phase (MP) individuals may develop into FPs. An examination of the population structure shows that there is considerable overlap in size between MP and FP individuals. When an individual changes

from MP to FP, the appendices masculinae become much smaller and lose their spines. The pleopod flanges of MPs which overlapped in body size with FPs followed the growth pattern of smaller MPs. In FPs, the flanges, a character associated with embryo incubation (Höglund 1943), had a somewhat greater relative size. Ejaculatory ducts of the testicular part of the gonad follow a similar growth pattern in all individuals, but with a higher variability in FPs. These observations indicate that the change from MP to FP occurs suddenly, perhaps in any MP above a threshold size.

Transitional individuals grouped with MPs in appendix masculina characteristics, were intermediate between MPs and FPs in flange width, and were similar to

both in ejaculatory duct width. Transitionals, by definition, were MPs in which the ovarian portion of the gonad was filling with vitellogenic oocytes but which were not carrying embryos. Loss or reduction of male characters such as size and spination of the appendix masculina and cincinnuli on Pleopod 1, with development of the female incubatory flanges, all appear to take place at the molt which transforms the transitional to FP. This may be one reason which so few transitionals were found in population samples. Another reason may be that many MPs never make the change to the FP phase.

Although many MPs may not change to FP hermaphrodites, all appear capable of doing so. All MPs dissected, both large and small, had a gonad similar in morphology to that of FPs, with an undeveloped (small, nonvitellogenic) ovarian portion. The larger MPs of *Lysmata wurdemanni* which do not develop into the female phase do not appear to be primary males incapable of developing into a female phase, but rather potential simultaneous hermaphrodites.

What are the proximate and ultimate factors mediating sex change in this species? Two hypotheses proposed for the evolution of hermaphroditism by Ghiselin (1969) are relevant to *Lysmata wurdemanni*. The first, the "size advantage hypothesis" as elaborated for fishes and shrimps by Warner (1975) and Charnov (1979), attempts to explain the evolution of sequential hermaphroditism. In protandry, male reproductive function is most advantageous when the individual is small, but female reproduction is more advantageous at a larger size. In many gonochoristic carideans, males are smaller than females (Bauer 1996b). Males can produce sufficient numbers of the energetically inexpensive sperm at small body sizes to fertilize the spawn of the largest females. Where studied, males of such species do not guard nor defend reproducing females, and thus large body size is not selected for (Bauer 1996b). In fact, small males are more cryptic than larger ones, an advantage for escaping predation. On the other hand, with increasing body size, females can produce larger broods of the energetically expensive, large eggs of carideans (Bauer 1991). While the size-advantage model explains protandry fairly well in a number of species, it does not explain why more gonochoristic species with small males and larger females have not evolved protandry. Nor does it explain why more protandric carideans have not evolved a system similar to *L. wurdemanni*, in which male function is retained with no apparent loss of female function. Also unexplained is why, in *L. wurdemanni*, some MPs apparently do not enter the female phase.

It will be essential, in order to understand the evolution and maintenance of simultaneous hermaphroditism in *Lysmata wurdemanni*, to know what proportion of inseminations are accomplished by MPs compared to those made by FPs acting as male mating-partners. Presumably, FPs acting as males can compete with MPs in nature for matings with receptive, prespawning FPs, thus enhancing their lifetime reproductive output over that of simple protandry. If not, it would be difficult to

comprehend how the simultaneous hermaphroditism of *L. wurdemanni* FPs could evolve and be maintained, since there must be some cost, as yet unknown, of maintaining a male system during the female phase. Knowledge of the relative male reproductive success of individuals during MP and FP phases, along with the demography of a population, will be vital in understanding if and when an MP changes to FP. If a population is low in FPs, and FPs acting as males can successfully compete with MPs for copulations with receptive females, then it would be advantageous for an MP to change to FP, i.e. to have both male and female reproductive functions. Conversely, if a population has an abundance of FPs, the change of an MP to an FP might not be adaptive, given the yet unknown costs of sex change and maintenance of a male system during the female phase.

Simultaneous hermaphroditism can be an advantage when population densities are low, since the probability of meeting another individual of the opposite sex would be slight ("low density model": Ghiselin 1969). Some species of *Lysmata*, such as *L. grabhami*, occur in pairs at low abundance (Limbaugh et al. 1961; Criales 1979). In *L. grabhami*, female-phase hermaphrodites appear to comprise such pairs (personal observation by RTB on specimens donated by P. Wirtz; Wirtz 1997). However, densities in the *L. wurdemanni* population sampled in the present study were qualitatively quite high; several individuals could be collected with a single sweep of the net or could be observed in tidepools at the same time.

Factors related to social organization and mating system, such as dominance relationships among and sex ratios of female-phase and male-phase individuals, have been shown to be important determinants of sex change in sequentially hermaphroditic fish species (Chan and Yeung 1983; Ross 1990), including protandric species (Fricke and Fricke 1977; Godwin 1994). In protandric caridean shrimps, environmental control of sex change through social interactions has been suggested for *Athanas* spp. (Alpheidae) (Nakashima 1987; Gherardi and Calloni 1993), and is indicated by data on pandalid shrimps (Charnov et al. 1978, but see Bergström 1997). In *Lysmata wurdemanni*, in which individuals occur in groups of several to many individuals, a change from MP to FP, based on rank relationships, if they exist, is conceivable. Alternately, some threshold sex ratio (Shapiro and Lubbock 1980) may regulate proportions among FPs and MPs in a group, with the type and frequency of interactions of an individual shrimp with other sexual morphs as the proximate factor triggering change in sexual type.

In *Lysmata grabhami*, in which FPs occur in isolated, female-female pairs (Wirtz 1997), similar social interactions might be proximate factors which trigger change from an MP to a hermaphroditic FP condition. In this case, the "low density" hypothesis might serve as the ultimate (evolutionary) factor accounting for a fully hermaphroditic "female phase," with the "size advantage" hypothesis explaining the sequence of MP to FP

rather than the reverse. All the above is speculative and must be tested with empirical observations on the sexual systems, social structure, and ecology of *Lysmata* spp., since sexual systems may vary even in related, ecologically similar species (e.g. *Thor* spp.: Bauer and VanHoy 1996). Furthermore, phylogenetic relationships of *Lysmata* spp. must be analyzed in order to comprehend the evolution of species of such different social structure (high-density groups vs low-density pairs) but with, apparently, a very similar hermaphroditic system.

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