

Effects of food availability on sex change in two species of *Crepidula* (Gastropoda: Calyptraeidae)

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ABSTRACT: Sex allocation theory predicts that growth rates should influence the optimal size at sex change, with sex change occurring at larger sizes with increasing growth rates. We examined how variation in food availability affects growth rates and therefore influences size at sex change in 2 protandrous calyptraeid gastropods, *Crepidula* cf. *marginalis* and *C. incurva*. The lowest food concentration tested resulted in starvation and was discarded. The medium and high food concentrations resulted in differences in growth rates, and in both species the penis was lost and sex change was initiated at larger sizes with more food. However, food concentration had little effect on the size at which the female stage was attained. Food-induced alterations in the timing of sex change were not in the expected direction. *C. cf. marginalis* raised with less food initiated sex change earlier, but completed sex change later in time than those raised with more food. *C. incurva* both initiated and completed sex change later when raised with less food compared to those individuals raised with more food. In both species the transitional phase was completed more rapidly with more food. These results are consistent with the idea that, as more resources become available, time dedicated to active sexual phases is maximized and the duration of the non-reproductive transitional phase is minimized. Under 'good' conditions this is manifest by a delay in the loss of the male phase, but no change in the attainment of the female phase.

KEY WORDS: Protandry · *Crepidula incurva* · *Crepidula* cf. *marginalis* · Plasticity · Food availability

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INTRODUCTION

Sex allocation

One branch of life-history theory focuses on sex allocation and particularly on sequential hermaphroditism or sex change, a phenomenon that occurs in widely divergent, primarily marine, taxa. Sex allocation theory predicts that sex change can be explained in an adaptive way if male or female reproductive success depends on size or age, and if the shape of the relationship is different for each sex (Ghiselin 1969, Warner 1975, 1988, Charnov 1982). For example, in some harem-forming, protogynous (female

first) species of fishes, male mating success increases rapidly as size increases, whereas egg production increases only moderately with female size. In protandrous (male first) species female reproductive success should increase more rapidly with size than does male reproductive success. Specifically, sex allocation theory predicts that individuals change sex when the potential future reproductive success becomes greater when acting as the second sex than it would be if individuals continued as the first sex. These predictions have been largely supported by empirical studies across the great diversity of sexual strategies and mating systems employed by sex-changing animals (Charnov 1982, Munday et al. 2006). However,

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the reasons why more animals are not sex changers, the costs of sex change, and the cues used to induce sex change remain relatively poorly explored (Munday et al. 2006, Kazancıoğlu & Alonzo 2009).

The predicted optimal size at sex change depends on the size and sex ratio of all the individuals in a population or local mating group (Charnov 1982). These parameters can vary among populations, habitats, or seasons. It is not surprising, therefore, that size at sex change is largely influenced by interactions with conspecifics in a number of species (e.g. fishes: Robertson 1972, Cole & Robertson 1988, Warner & Swearer 1991, Cole & Shapiro 1995, Sakai et al. 2001; molluscs: Hoagland 1978, Wright 1989, Warner et al. 1996, Chen et al. 1998, Collin et al. 2005; polychaetes: Franke 1986, Premoli & Sella 1995). Studies of bluehead wrasse have shown that removing the male from a harem will induce the largest female to begin sex change in only a few days (Warner & Swearer 1991). Likewise, in pair-forming fishes and worms the body condition or sex of one of the pair can induce sex change in its partner (Berglund 1986, Munday et al. 1998, Munday 2002).

Sex change is also predicted to respond to changes in growth or mortality rates. For example, high mortality will reduce the chances of any individual attaining a large size, increasing the sex ratio bias toward the first sex, and therefore increases the benefits of earlier sex change. Higher growth rates act in the opposite direction, with increased growth resulting in sex change at a larger size (Charnov 1982, Warner 1988, Munday et al. 2006), if other demographic parameters remain the same. This pattern has been demonstrated by a number of field studies (reviewed in Munday et al. 2006, Kazancıoğlu & Alonzo 2010), but has also recently been shown, in theory, to depend on the details of the shape of the relationship between reproductive success and size or age (Kazancıoğlu & Alonzo 2010). Because this relationship is difficult to measure, it is often assumed to be roughly linear (see Charnov 1982). In most models age and size are linked so that a prediction of 'later' sex change implies that individuals are both older and larger at sex change. When growth rates differ, however, age and size are uncoupled and the form taken by the 'delay' in sex change depends on the relationship between reproductive success, size and age. For example, at higher growth rates, a larger size at sex change could occur at a younger age, but this is not necessarily the case.

It is not known whether individuals use their own growth rate and/or the size distribution of the population with which they interact as the proximate cue for

altering sex change in response to changes in growth rate (Charnov 1982, Munday et al. 2006). We used 2 common species of calyptraeid gastropods, *Crepidula* cf. *marginalis* and *C. incurva*, to test the following hypotheses about the direct effects of food-mediated differences in growth rates on sex change:

(1) Animals grown at high food concentrations, and therefore with higher growth rates, change sex at a larger size than those grown at low food concentrations. (2) Animals grown at high food concentrations, and therefore with higher growth rates, change sex later in time than those grown at low food concentrations. (3) We also examined the effect food concentration has on the duration of sex change.

Sex change in calyptraeid gastropods

Protandrous marine gastropods in the family Calyptraeidae, especially species in the genus *Crepidula*, were some of the first sex-changing animals to be studied (Orton 1909, Gould 1917, 1919). All calyptraeids are sedentary, protandrous suspension-feeders, and socially mediated sex change has been demonstrated in all species that have been studied (Coe 1938, 1953, Gould 1917, 1919, Hoagland 1978, Warner et al. 1996, Collin et al. 2005). An early study (Coe 1938) showed that mated males delayed sex change relative to solitary males. Results from recent experimental studies that manipulated social environment in the laboratory suggest that the presence of females delays sex change from the default condition, while sex change is not altered from the default in the presence of other males (Warner et al. 1996, Collin et al. 2005). Sex change is not, however, delayed indefinitely in the presence of females, and all animals eventually change sex (Collin et al. 2005).

The significant differences in size at sex change that result from different associations with conspecifics under controlled laboratory conditions are small relative to the total variation in the size at sex change observed in the field. The effect of conspecific association on *Crepidula* cf. *onyx* and *C. incurva* accounts for only 34 and 11%, respectively, of the observed size range of transitional individuals collected in the field (Collin et al. 2005). The wide range in size at sex change in these field samples is typical of other calyptraeids (Warner et al. 1996, Collin 2006). In addition, in *C. incurva*, the size at sex change in the field is considerably smaller than it is in the laboratory, with sex change in the laboratory occurring well above the size range of sex-changing animals collected in the field (Table 1). If the snails alter the timing of sex change in

response to variation in environmental conditions, such as food availability, this could account for the unexplained variation in size at sex change and the unexplained effect of laboratory rearing on *C. incurva*.

MATERIALS AND METHODS

Study species and field collection

Juveniles of *Crepidula cf. marginalis* and *C. incurva* were collected by hand from Playa Chumical (8° 30' N, 79° 40' W) near the town of Veracruz, Panama, during the spring tides of February 11 to 14 and March 11, 2009, and January 30 to 31, 2010. Both species were collected from within an approximately 2 ha area. Animals were gently removed from the substrate and immediately transported submerged in seawater to the laboratory.

At Chumical, *Crepidula incurva* occurs patchily in the intertidal between approximately -0.5 and -1 m, usually attached to shells of large snails, especially cerithiids and muricids, which congregate in shallow tide pools and below large rocks. Sex ratio and size at sex change for *C. incurva* have been reported elsewhere (Collin et al. 2005, Collin 2006; Table 1).

At the same site, *Crepidula cf. marginalis* are abundant, attached directly to rocks, a little higher in the intertidal than *C. incurva*. Size at sex change in this species has not been previously investigated, but our observations show that the sex ratio is significantly male-biased ($\chi^2 = 80.8$, $p < 0.0001$; $N = 285$): 79% of breeding animals were males. The size at sex change for field-collected animals estimated by a logistic regression (L_{50}) is 11.0 mm (CI = 10.3 to 12.2 mm), and males and females both occur between 9.6 and 11.3 mm.

Measurements and sex determination

Shell length was measured with calipers to the nearest 0.1 mm, and sex was determined at the beginning of the experiment and every subsequent week. Males were identified by the presence of a penis behind the right tentacle, and females, by a genital papilla (FGP) and capsule gland on the right side of the mantle cavity. Transitional individuals either have the organs of neither sex or of both sexes. Small specimens without sexual characteristics were considered to be juveniles, which was verified by subsequent observations during the laboratory experiments.

Small (3 to 5 mm) *Crepidula cf. marginalis* and *C. incurva* were allocated haphazardly among the different treatments (see below) within 2 or 3 d of being collected from the field. There were 25 to 30 ind. treatment⁻¹ for each species. Animals were kept alone in 350 ml cups, at approximately 20°C (18 to 21°C), water was replaced with UV-filtered seawater 3 times a week, and they were fed the microalga *Isochrysis galbana* (Strain T-iso). Because we were interested in the direct effects of differences in growth, and not in the indirect effect mediated by cues from other animals, we raised each animal alone, but otherwise followed the protocol of our previous study on the effects of conspecifics on sex change (Collin et al. 2005).

Expt 1

In 2009 we assigned animals to 3 different treatments: high food (38×10^6 cells d⁻¹), medium food (9.5×10^6 cells d⁻¹), and low food (2.4×10^6 cells d⁻¹). Food was split into 2 feedings d⁻¹ to avoid rejection of food due to saturation of the feeding apparatus in the

Table 1. *Crepidula cf. marginalis* and *C. incurva*. Summary of size at sex change. L_{50} : the length at which a logistic regression of sex on shell length gives a 50% chance of being male. -: no data

| Treatment | Measure of size at sex change | <i>C. cf. marginalis</i> (mm) | <i>C. incurva</i> (mm) |
|---------------------------------|---|-------------------------------|------------------------|
| Field ^a | L_{50} (size overlap of both sexes) | 11.0 (9.6–11.3) | 8.05 (6.3–9.8) |
| Alone ^a | Average (last male size, first female size) | – | 10.9 |
| Paired with female ^a | Average (last male size, first female size) | – | 12.0 |
| Medium food | Average (last male size, first female size) | 10.17 | 11.48 |
| High food | | | |
| Expt 1 | Average (last male size, first female size) | 10.85 | 12.33 |
| Expt 2 | Average (last male size, first female size) | 11.31 | 12.46 |
| 2× high food | Average (last male size, first female size) | 11.89 | 12.19 |
| 4× high food | Average (last male size, first female size) | 11.80 | 12.5 |

^aData for *C. incurva* from Collin et al. (2005)

high food treatment (Chaparro et al. 2004). The low food treatment was not sufficient to support growth beyond an average of 8 mm in *Crepidula cf. marginalis* and of 5.5 mm in *C. incurva*, well below the normal size at sex change in each species. Therefore, the low food treatment was not considered further here.

Expt 2

We repeated Expt 1 in 2010 using higher food concentrations. The lowest ration was the same as the high food treatment in the first experiment (50×10^3 cell ml^{-1}), and the other treatments were twice (2 \times) and 4 times (4 \times) this amount. To avoid accumulation of uneaten food, water was changed every day, and in the 2 \times and 4 \times treatments food was divided into 2 or 4 feedings d^{-1} each separated by at least 1 h.

Analyses

The effect of each treatment on the growth rate was analyzed using a repeated measures analysis of variance (RM ANOVA) on the weekly measurements of shell length, implemented as a MANOVA (multivariate ANOVA) in the JMP statistical package (SAS Inc.). The effect of each treatment on size at sex change was analyzed with a 1-way ANOVA, on both the size at which each individual was last reported to have a penis and the size at which they were first reported to have an FGP. Differences in the time until sex change in each treatment were examined in a similar way, but the number of weeks from the initiation of the experiment until sex change was log-transformed to conform to the requirements of ANOVA. Differences in the duration of sex change between treatments were examined using a Kruskal-Wallis non-parametric ANOVA on the number of weeks between the last week the animals had a penis and the first week they were reported to have an FGP. When the transitional phase involved the co-occurrence of male and female features this value was <0 .

RESULTS

Crepidula cf. marginalis

Food availability had a significant effect on the growth rates of *C. cf. marginalis* (Fig. 1). In Expt 1 size differed between food treatments by Week 8 (RM ANOVA univariate tests: $p < 0.05$) indicating

that medium concentrations of food became limiting at this time. In Expt 2, which used higher food levels, food limitation became evident in Week 16, when RM ANOVA univariate tests showed significant differences between the high level treatment compared to the 2 \times and 4 \times treatments. This is well after sex change was complete in most of the animals in all 3 treatments.

The size at which the penis is lost increased with food availability, but the size at which animals became female did not differ between food treatments (Fig. 2). In Expt 1, the size at which they lost the penis was smaller in the medium food treatment than in the high food treatment (ANOVA: $F_{1,63} = 21.92$; $p < 0.0001$; Fig. 2) and a similar, almost significant, trend for larger shell length at penis loss in higher food concentrations was evident in Expt 2 ($F_{2,83} = 2.86$; $p = 0.06$). In neither experiment did food availability influence the size at which animals became female (Expt 1: $F_{1,61} = 2.26$; $p > 0.10$; Expt 2: $F_{2,83} = 1.12$; $p > 0.3$).

As food availability increased, the penis was lost later and the FGP developed earlier, resulting in a more rapid sex change. Animals with more food ended the male phase later than those with less food in Expt 1 (ANOVA log [last week male]: $F_{1,63} = 4.42$; $p = 0.03$) and Expt 2 ($F_{2,83} = 4.96$; $p = 0.009$) (Fig. 3). Animals became female earlier with more food than with less food in Expt 1 (ANOVA log [first week female]: $F_{1,61} = 10.2$; $p < 0.003$), but there was no significant effect in Expt 2 ($F_{2,83} = 1.16$; $p = 0.32$).

In Expt 1, animals in the medium food treatment also took significantly longer to change sex (~11 wk) than those in the high food treatment (~1 wk; Kruskal-Wallis: $F_{1,67} = 76.58$; $p < 0.0001$). There was a similar trend in Expt 2, where sex change happened more quickly in the 4 \times food treatment (0.7 wk) than in the 2 \times and high treatments (1 and 1.5 wk, respectively; Kruskal-Wallis non-parametric ANOVA: $F_{2,83} = 7.08$; $p = 0.001$). In all treatments, the median duration of sex change was ≥ 0 , indicating that most animals lost the penis before acquiring visible female structures.

The development of the penis was more sensitive to food concentrations in *Crepidula cf. marginalis* than in *C. incurva*. Four of the *C. cf. marginalis* in the medium food treatment never developed a penis, and some developed a small penis. In the high, 2 \times and 4 \times food treatments, all *C. cf. marginalis* developed a large penis before becoming female. In almost all cases the penis disappeared before the female features developed. In contrast, all *C. incurva* developed a large penis, and the penis was usually retained until after the female structures were well developed.

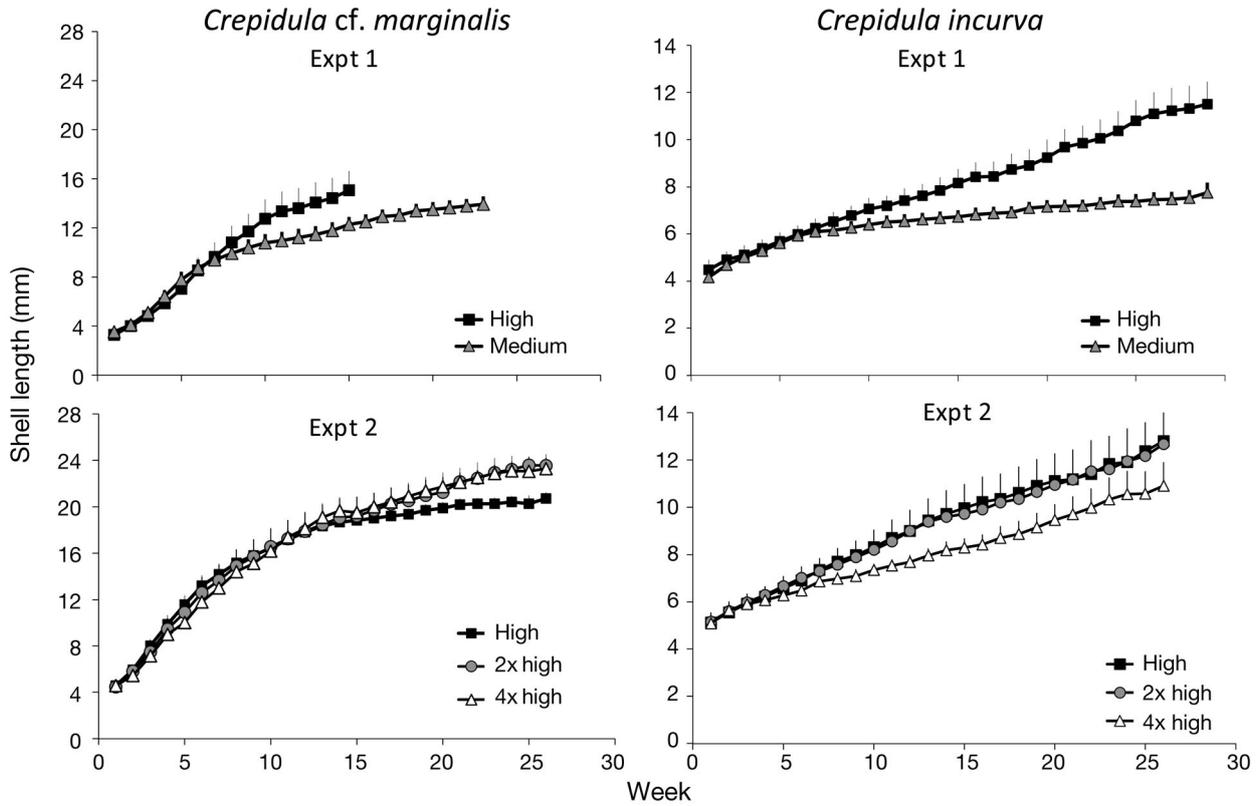


Fig. 1. *Crepidula cf. marginalis* and *C. incurva*. Growth of *C. cf. marginalis* and *C. incurva* exposed to different feeding treatments (high, medium, 2× high, 4× high). Error bars represent 95% confidence intervals. Data in the top left graph end before Week 30 because animals in Expt 1 were sacrificed 3 wk after sex change was complete and the dry body weight was measured

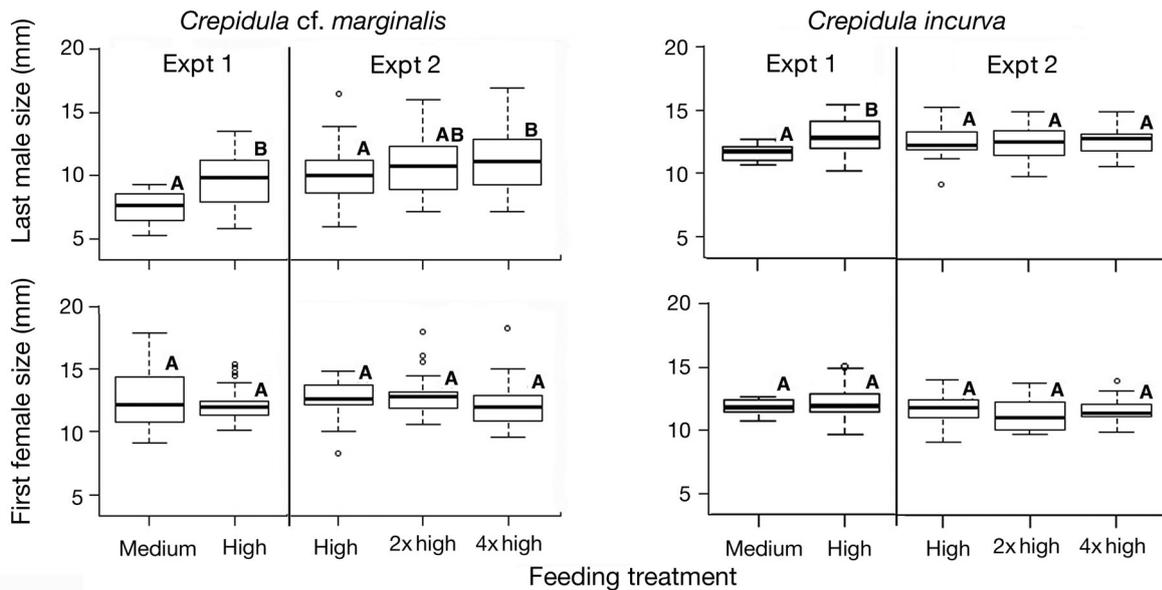


Fig. 2. *Crepidula cf. marginalis* and *C. incurva*. Box plots showing the size at sex change as indicated by the last male size and first female size for *C. cf. marginalis* and *C. incurva* in different feeding treatments (high, medium, 2× high, 4× high). The box is drawn between the quartiles, with a thick line at the median value. Whiskers indicate the range of data, with outliers shown as circles. Groups marked with the same letter within the same experiment are not significantly different according to Tukey's honestly significant differences post hoc test

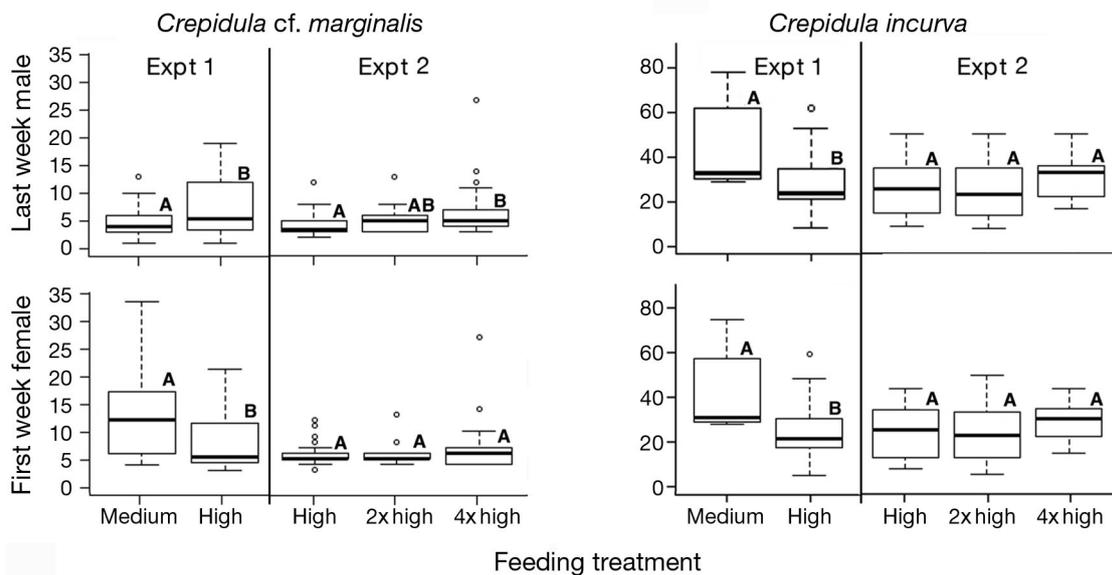


Fig. 3. *Crepidula cf. marginalis* and *C. incurva*. As in Fig. 2 except showing the time until sex change as indicated by the week the animals were last reported as male or first reported as female

Crepidula incurva

Food availability had a significant effect on the growth rates of *Crepidula incurva* (Fig. 1). In all treatments the juveniles grew quickly during the first 2 to 3 wk. In Expt 1, medium and high food treatments differed by Week 8 (RM ANOVA univariate tests: $p < 0.05$). In Expt 2, animals with the highest (4 \times) food ration had the lowest growth rates, and the high and 2 \times treatments did not differ (Fig. 1). Univariate tests in the RM ANOVA showed that the treatments differed between Weeks 7 and 19 ($p < 0.05$), but no significant difference was detected after that.

In *Crepidula incurva* food ration had a significant positive effect on the last size at which animals were male in Expt 1 ($F_{1,30} = 6.30$; $p < 0.02$), but not in Expt 2 (ANOVA $F_{2,60} = 0.10$; $p = 0.90$; Fig. 2). Food treatment did not significantly affect the first size they were observed to be female (ANOVA—Expt 1: $F_{1,30} = 0.70$; $p > 0.4$; Expt 2: $F_{2,60} = 0.79$; $p > 0.4$; Fig. 2).

The timing of sex change did not differ at the high food concentrations in Expt 2 (last date with penis: ANOVA log [last week male]: $F_{2,60} = 0.94$; $p > 0.3$; first date with FGP: ANOVA log [first week female]: $F_{2,60} = 1.07$; $p > 0.3$; Fig. 3), but, in Expt 1, animals in the medium food treatment lost male features later and became females later than the animals in the high food treatment (ANOVA log [last week male]: $F_{1,30} = 7.02$; $p < 0.02$; ANOVA log [first week female]: $F_{1,30} = 9.29$; $p < 0.01$). There was no significant difference between the treatments in the duration of sex

change (Kruskal-Wallis—Expt 1: $F_{1,30} = 3.70$; $p = 0.06$; Expt 2: $F_{2,60} = 0.38$; $p > 0.6$). In Expt 1, the trend was for sex change to occur more slowly with less food. Animals in the medium food treatment generally lost the penis in the same week as the FGP appeared, while those in the high food treatment grew the FGP 3 wk before the final loss of the penis.

DISCUSSION

Socially mediated sex change, where the optimal size to change sex is determined as a function of the other individuals in a population, is almost ubiquitous among sex changing animals (Munday et al. 2006). Similarly, it is predicted that growth rate and mortality rate affect the size at sex change, but the influence of these factors, independent of the social effects mediated by population differences, has not been demonstrated. The cues by which the animals detect differences in growth or mortality could be either direct (individual assessment of their own growth rate or mortality risk) or indirect (based on an assessment of the immediate social environment, which reflects growth and mortality in the area). We endeavored to remove any possible social effects by raising animals individually while we looked at the effects of differences in growth rates induced by different food rations.

The significant differences we found are generally consistent with our initial hypotheses with respect to

size and the conflicting results with respect to timing.

(1) Both *Crepidula cf. marginalis* and *C. incurva* initiate sex change at smaller sizes when less food was available, but showed little difference in the size at which they complete sex change.

(2) *Crepidula cf. marginalis* raised with more food initiated sex change later in time than those raised with the least food, but *C. incurva* raised with more food initiated sex change earlier in time than those raised with the least food. Both species showed later completion of sex change in the treatment with the least amount of food.

(3) In addition animals of both species raised under low food conditions took longer to complete sex change than did those raised with higher food levels.

Food availability, growth rates, and sex change

Crepidula cf. marginalis raised at medium and high food concentrations differed in growth rate, but those raised at the 3 highest rations (38×10^6 , 76×10^6 , and 15×10^7 cells d^{-1}) did not differ until they were well past the average size at sex change. They did, however, show a slight difference in the maximum size attained. In *C. incurva* the lowest (medium) food concentration and the highest (4 \times) concentration both showed slower growth rates than the other treatments. Slower growth at the highest concentration may have resulted from a concentration of microalgae that exceeded the animals' ability to collect and process food (which results in rejection of the food; see Chaparro et al. 2002, 2004), but this treatment did not have any effect on the size or timing of sex change.

To test the effects of different growth rates on size at sex change, we could only use the medium and high food ration treatments, which produced differences in growth rates prior to sex change. Our results do support the prediction that increased growth rates increase the size at sex change: in both species the size they initiated sex change was smaller in the medium than in the high food treatment. This supports the hypothesis that sex change is delayed under 'good' conditions. However, other results are not consistent with this hypothesis. There was no significant effect of growth rate on the first female size, and this stage was reached more quickly at high than at medium food treatments. In addition, the size that sex change was initiated increased with food concentration in the absence of significant differences in growth rates across the higher food concentrations in *Crepidula cf. marginalis*. Finally, sex change occurred

more quickly at higher food concentrations. Viewed together these results are consistent with the idea that, as more resources become available, time dedicated to active male and female phases is maximized and the duration of the non-reproductive transitional phase is minimized, thereby possibly increasing potential lifetime reproductive output. When conditions are bad, the duration of the transitional phase increases. Since in protandry female fitness curves increase more steeply with size than do male fitness curves, it is possible that less potential fitness is lost by extending the transition period into the male phase than an extension of the same duration into the female phase. However, it is not possible to determine if this is the case without knowing the details of the shapes of both the fitness curves and without taking into account potential sex-specific differences in growth rates.

Assumptions of sex change theory

Optimal sex allocation theory commonly makes a number of simplifying assumptions in order to generate hypotheses that can be tested with data available from real organisms. The 2 most relevant to the results of the present study are that (1) sex change is instantaneous and (2) the size-specific growth rates of males and females are the same (Charnov 1982).

Our results showed that sex change is not an instantaneous phenomenon and that food availability affects the duration of sex change. In calyptraeids it is not possible to determine the sexual status of the gonad without sacrificing the animal. Therefore, we determined sex using features that could be distinguished from live examination; the female genital papilla to indicate female function and the penis to indicate male function. The use of these characteristics is common in studies of calyptraeid sex change (Chen & Soong 2000, Collin et al. 2005), and they generally reflect the status of the gonad. Because the complete loss of the penis can occur after an individual has become fully functional as a female and the FGP can be completed before the gonad is ready to produce eggs, the reported durations of sex change are minimum estimates. In both species examined in the present study the higher food treatments showed a shorter duration of what we considered to be the transitional stage. In *Crepidula cf. marginalis*, transitional individuals did not usually show any co-occurrence of the penis and FGP. Since animals cannot be functional males without a penis or functional females without the FGP, the 11 wk average duration

of sex change in the medium food treatment represents the minimum time that these animals are incapable of reproduction. Since females can produce a brood every 2 to 3 wk (R. Collin unpubl. data), this represents a considerable potential loss of fitness via female function. In *C. incurva*, however, transitional individuals generally exhibited a co-occurrence of the penis and FGP. There were still significant differences between the treatments in the duration of this transition, but it is difficult to determine the amount of time when these animals would be unable to reproduce.

The second major simplifying assumption of sex allocation theory, that the size-specific growth rates of males and females are the same, is difficult to test. This requires measuring the growth rates of males and females over the size range at which both sexes occur. This is also the size range during which sex change occurs. Because we cannot be sure of the exact duration of sex change in either species and because the duration of sex change varies among treatments and among individuals, it is difficult to make a clear comparison of the size-specific growth rates for males and females.

CONCLUSIONS

Charnov (1982) stated that despite how well animals seem to employ optimal life-history strategies and change sex at optimal times or sizes: 'We can only wonder at what cues the animals may be using' (p. 158) to assess the optimal size at sex change. Associations with conspecifics provide one such suite of cues. Some progress has been made towards understanding the endocrine mechanisms responsible for triggering sex change in fishes (Perry & Grober 2003), but these mechanisms cannot be easily generalized to invertebrates. Few studies have attempted to dissect which aspects of social interactions or external environmental factors trigger sex change in invertebrates. Here we have demonstrated that food ration may affect the size at sex change directly (not mediated via social interactions) and in the direction predicted by theory. However, the amount of variation in size at sex change induced by changes in food concentration relative to the variation observed in the field differs between the 2 species. For *Crepidula* cf. *marginalis* food-induced variation in size at sex change spans a size range similar to the range in size overlap of males and females observed in the field (Table 1). Unfortunately, the effects of conspecifics on sex change have not been assayed for this species.

C. incurva, in contrast, shows a considerably larger size at sex change in our experiment than is observed in the field (Table 1). Experiments on associations with conspecifics in *C. incurva* also showed a larger size at sex change than is observed in the field (Table 1; Collin et al. 2005), suggesting that some, so far unknown, difference between laboratory and field conditions exerts a considerable effect on sex change in this species. Identification of this factor would provide a useful tool for future studies of the cues and mechanisms that induce sex change in these snails.

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LITERATURE CITED

- Berglund A (1986) Sex change by a polychaete worm: effects of social and reproductive costs. *Ecology* 67:836–845
- Chaparro OR, Thompson RJ, Pereda SV (2002) Feeding mechanisms in the gastropod *Crepidula fecunda*. *Mar Ecol Prog Ser* 234:171–181
- Chaparro OR, Thompson RJ, Segura J, Navarro JM (2004) The effect of food supply on feeding strategy in sessile female gastropods *Crepidula fecunda*. *Mar Biol* 144: 79–87
- Charnov EL (1982) The theory of sex allocation. Princeton University Press, Princeton, NJ
- Chen MH, Soong K (2000) Sex change in the hat snail, *Calyptrea morbidia* (Reeve) (Gastropoda: Calyptreaeidae): an analysis of substratum, size, and reproductive characteristics. *Veliger* 43:210–217
- Chen MH, Yang YW, Soong K (1998) Preliminary observations on change of sex by the coral-inhabiting snails *Coralliophila violacea* (Lamarck) (Gastropoda: Coralliophilidae). *J Exp Mar Biol Ecol* 230:207–212
- Coe WR (1938) Influence of association on the sexual phases of gastropods having protandric consecutive sexuality. *Biol Bull* 75:274–285
- Coe WR (1953) Influences of association, isolation and nutrition on the sexuality of snails in the genus *Crepidula*. *J Exp Zool* 122:5–19
- Cole KS, Robertson DR (1988) Protogyny in the Caribbean reef goby, *Coryphopterus personatus*: gonad ontogeny and social influences on sex-change. *Bull Mar Sci* 42: 317–333
- Cole KS, Shapiro DY (1995) Social facilitation and sensory mediation of adult sex change in a cryptic, benthic marine goby. *J Exp Mar Biol Ecol* 186:65–75
- Collin R (2006) Sex ratio, life history invariants and the patterns of sex change in a family of protandrous gastropods. *Evolution* 60:735–745

- Collin R, McLellan M, Gruber K, Bailey-Jourdain C (2005) Effects of conspecific associations on size at sex change in three species of calyptraeid gastropods. *Mar Ecol Prog Ser* 293:89–97
- Franke HD (1986) Sex ratio and sex change in wild and laboratory populations of *Typosyllis prolifera* (Polychaeta). *Mar Biol* 90:197–208
- Ghiselin MT (1969) The evolution of hermaphroditism among animals. *Q Rev Biol* 44:189–208
- Gould HN (1917) Studies on sex in the hermaphrodite mollusk *Crepidula plana*. 2. Influence of the environment on sex. *J Exp Zool* 23:225–250
- Gould HN (1919) Studies on sex in the hermaphrodite mollusk *Crepidula plana*. 3. Transference of the male producing stimulus through sea water. *J Exp Zool* 29:113–120
- Hoagland KE (1978) Protandry and the evolution of environmentally mediated sex change: a study of the Mollusca. *Malacologia* 17:365–391
- Kazancıoğlu E, Alonzo SH (2009) Costs of changing sex do not explain why sequential hermaphroditism is rare. *Am Nat* 173:327–336
- Kazancıoğlu E, Alonzo SH (2010) Classic predictions about sex change do not hold under all types of size advantage. *J Evol Biol* 23:2432–2441
- Munday PL, Caley MJ, Jones GP (1998) Bi-directional sex change in a coral-dwelling goby. *Behav Ecol Sociobiol* 43:371–377
- Munday PL (2002) Bi-directional sex change: testing the growth-rate advantage model. *Behav Ecol Sociobiol* 52:247–254
- Munday PL, Buston PM, Warner RR (2006) Diversity and flexibility of sex-change strategies in animals. *Trends Ecol Evol* 21:89–95
- Perry AN, Grober MS (2003) A model for social control of sex change: interactions of behavior, neuropeptides, glucocorticoids, and sex steroids. *Horm Behav* 43:31–38
- Premoli MC, Sella G (1995) Sex economy in benthic polychaetes. *Ethol Ecol Evol* 7:27–48
- Robertson DR (1972) Social control of sex reversal in a coral reef fish. *Science* 177:1007–1009
- Sakai Y, Kohda M, Kuwamura T (2001) Effect of changing harem on timing of sex change in female cleaner fish *Labroides dimidiatus*. *Anim Behav* 62:251–257
- Warner RR (1975) The adaptive significance of sequential hermaphroditism in animals. *Am Nat* 109:61–82
- Warner RR (1988) Sex change and the size-advantage model. *Trends Ecol Evol* 3:133–136
- Warner RR, Swearer S (1991) Social control of sex change in the bluehead wrasse, *Thalassoma bifasciatum* (Pisces: Labridae). *Biol Bull* 181:199–201
- Warner RR, Fitch DL, Standish JD (1996) Social control of sex change in the shelf limpet, *Crepidula norrisiarum*: size-specific responses to local group composition. *J Exp Mar Biol Ecol* 204:155–167
- Wright WG (1989) Intraspecific density mediates sex-change in the territorial patellacean limpet *Lottia gigantea*. *Mar Biol* 100:353–364

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