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MORPHOTYPIC DIFFERENTIATION OF MALES OF THE FRESH-WATER PRAWN *MACROBRACHIUM ROSENBERGII*: CHANGES IN THE MIDGUT GLANDS AND THE REPRODUCTIVE SYSTEM

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ABSTRACT

Adult males of the fresh-water prawn *Macrobrachium rosenbergii* in a single-age population can be categorized into three morphotypes. Each morphotype represents a different stage in the development of the adult males from small males (SM) through orange-claw (OC) males to blue-claw (BC) males (Cohen *et al.,* 1981; Ra'anan, 1982). All males are capable of developing through all of the above stages, but individual males largely differ in the rate in which they transform from one morphotype to another. It was observed that SM and BC males are more sexually active than OC males, while the OC males grow more rapidly than SM and BC males (Ra'anan and Sagi, 1985).

To examine further this behavioral observation, two internal organs in each of the morphotypes were measured and compared. The development of the reproductive system (testes, sperm ducts, and ampullae), as an indication of sexual activity, was compared with the development of the midgut gland (hepatopancreas), an organ which is suggested to be in correlation with somatic growth. The hepatopancreas is significantly larger and the reproductive system is relatively smaller, in relation to body size, in OC males. The opposite is true of SM and BC males, in which the reproductive system is significantly larger while the hepatopancreas is much smaller in relation to body size. Thus, the relative sizes of the two organs of an individual prawn are closely associated with its position in the male developmental pathway from the SM through OC to the BC morphotype. Further, the changes in specific ratio between the two organs reflect the inverse relationship between reproduction and growth.

Three male morphotypes comprise a single-age adult population of the fresh-water prawn *Macrobrachium rosenbergii* (Smith *et al.,* 1978; Brody *et al.,* 1980; Cohen *et al.,* 1981; Ra'anan, 1982). These different male morphotypes were defined on the basis of color and spination of the chela, behavior, and growth characteristics (Sagi, 1984; Telecky, 1984; Ra'anan and Cohen, 1985). Kuris *et al.* (1987) have recently summarized operational allometric criteria to recognize the morphotypes and describe the transition between morphotypes.

The three distinguishable male morphotypes include: (1) the large, dominant blue-clawed male (BC), which is territorial, sexually active, and represents the final morphotype in the male developmental pathway; growth of this morphotype is reduced and molting is infrequent, (2) the subdominant orange-clawed male (OC), which is not territorial and is sexually incompetent, and (3) small males with transparent claws (SM), which are not territorial and are sexually competent, using a sneak mating strategy (Sagi, 1984; Ra'anan and Sagi, 1985). Ra'anan (1982) described the transformation of OC to BC and Kuris *et al.* (1987) distinguished a transitional stage from SM to OC. This transitional form was termed the weak-OC (WOC), the fully differentiated OC males being termed the strong-OC (SOC). The OC displaying the first signs of transformation into BC are termed pretransforming SOC (t-SOC) here (see below). Both the BC and the SM are sexually active. Ra'anan and Sagi (1985) concluded that the BC males and SM males represent two alternative mating strategies. The former directly court and hold the females, and the latter engage in sneak mating. The OC males are an intermediate, fast-growing stage. It was of interest, therefore, to examine whether the external morphotypic criteria and behavioral differences are reflected in the relative size of internal organs participating in reproduction, energy storage, and somatic growth.

Here we compare the relative size of the reproductive system and the midgut gland (hepatopancreas) of the various male morphotypes and the transition stages. The relative weight of the reproductive system represents part of the reproductive effort (Hirschfield and Tinkle, 1975), while the relative weight of the hepatopancreas, which plays a role in food assimilation (Yonge, 1924; Dall and Moriarty, 1983) and mobilization of energy during molting (Travis, 1955, 1957; Skinner, 1985) and pigmentation (Ghidalia, 1985), probably reflects on energy spent on somatic growth, morphological changes, and general metabolic activity.

We have found that the relative size of these organs in an individual is highly correlated with its morphotypic stage of development and its relative energy expenditure in growth versus sexual activity.

Materials and Methods

Male prawns were selected from a single-age population, following 150 days of growth in earthen ponds at the Dor Agriculture Research Station, Israel.

Prawns were divided into 6 categories, according to coloration, second pereiopods (claws) spination, propodus and carapace length, and wet weight (Table 1) (Ra'anan, 1982; Ra'anan and Cohen, 1985; Kuris *et al.,* 1987). Wet weight was measured to 0.01 g, using an electronic Sartorius balance. Carapace and propodus length were measured with vernier calipers to 0.1 mm. Carapace length was the distance from the posterior margin of the right eye orbit to the posterior margin of the carapace at the midline. The propodus, with the joint flexed, was measured along the lateral face from the proximal lateral condyle to the distal tip.

Ten prawns of each category were selected for study. Small males WOC, SOC, and BC were selected according to the methods described by Kuris *et al.* (1987). Pretransforming SOC were large OC males which had spiny and relatively larger claws than did other OC males. We hypothesize that these OC males would have transformed to the BC morphotype at the next molt. These prawns combined the SOC morphology with the first appearance of BC coloration and spination. Males of the BC morphotype, but of relatively small body size (carapace length and body weight), were termed old BC (OBC). These individuals were covered with algae and were assumed to be the earliest BC males to have metamorphosed in the process of the development of the social hierarchy. Although at that stage they represent the largest males, eventually, their relative size ranking within the male population decreased, since once becoming a BC male, further growth was largely inhibited while the other males kept growing (Ra'anan and Cohen, 1985).

Each prawn was dissected; the midgut glands and the reproductive system were isolated, blotted, and weighed to 0.01 g. The sperm duct was then stretched and measured with vernier calipers to 0.1 mm.

Analysis of variance (Sokal and Rohlf, 1981) was performed in order to measure the differences among the ratios of the midgut gland and the reproductive system to body weight of the various morphotypes. The SPSS (Statistical Package for the Social Sciences) was used for this analysis.

Results

Ratio of Weight of the Midgut Glands to Weight of the Reproductive System

The ratio between the weight of the midgut glands (MW) and the weight of the reproductive system (RW), including testes, sperm duct, and ampullae, was calculated for each of the morphotypes. Figure 1 shows that for SM, BC, and OBC males, the midgut gland is 2–4.5 times heavier than the reproductive system. In SOC males the MW/RW ratio, 11:1, is significantly higher ($P < 0.001$). The intermediate OC phases, WOC and t-SOC males, show intermediate values, i.e., a ratio of 7.2 and 8.9 of midgut gland weight to reproductive system weight, respectively, both significantly different from SM and BC males ($P < 0.001$).

Weights of the Midgut Glands and the Reproductive System Relative to Body Weight

Since the ratio of the midgut glands to reproductive system weight (MW/RW) showed a significant difference between the three morphotypes, it was of interest to find out whether these differences resulted from enlargement of the midgut glands of SOC males or from enlargement of the reproductive system of SM and BC males.

The relative weights of the midgut glands (RMW) and of the reproductive system (RRW) were calculated as percentages of total body weight (Figs. 2, 3). SOC males are characterized by significantly higher RMW when compared to SM and BC males, 7.03 ± 0.44 versus 2.90 ± 0.57 and 4.90 ± 1.14, respectively ($P < 0.001$). At the same time, the RRW of the SOC males was significantly lower than those observed for SM and BC males, 0.66 ± 0.19 versus 1.32 ± 0.39 and 1.11 ± 0.15, respectively ($P < 0.001$). Thus, SOC males differ from SM and BC males, both by having enlarged midgut glands and by having a relatively small reproductive system. In both parameters, RMW and RRW, the intermediate OC forms, i.e., WOC and t-SOC males, showed intermediate values, significantly

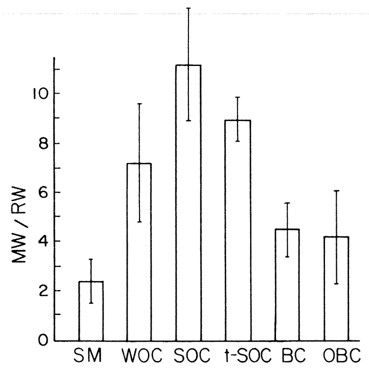

Fig. 1. The relationship between the midgut gland (hepatopancreas) weight (MW) and reproductive system weight (RW) for the different male morphotypes and transitional forms in *Macrobrachium rosenbergii.* Error bars represent SD. Male morphotypes: SM = Small Male; WOC = Weak Orange-Claw male; SOC = Strong Orange-Claw male; t-SOC = transforming SOC into blue-claw male; BC = Blue-Claw male; OBC = Old BC male.

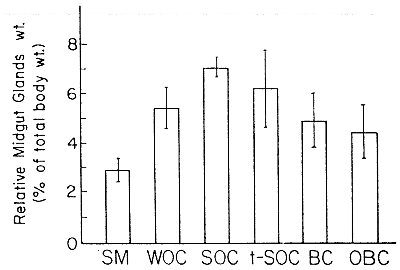

Fig. 2. Midgut glands (hepatopancreas) weight as a percentage of body weight in the different male morphotypes and transitional forms in *Macrobrachium rosenbergii.* Error bars represent SD. Male morphotypes: SM = Small Male; WOC = Weak Orange-Claw male; SOC = Strong Orange-Claw male; t-SOC = transforming SOC into blue-claw male; BC = Blue-Claw male; OBC = Old BC male.

different from those obtained for the SM, SOC, and BC males ($P < 0.001$).

Relative Length of the Reproductive System

While dissecting the animals, we observed that the reproductive system of SM and BC males was characterized by a relatively elongated sperm duct. We determined the ratio between the length of the reproductive system when extended (RL), and the carapace length (CL) of the individuals (RL/CL ratio) (Fig. 4). Once more, the mean relative lengths of the reproductive system of SM and BC males, 2.71 ± 0.21 and 3.68 ± 0.31, respectively, differ significantly between them, and both are significantly higher than the value obtained for SOC males, 1.63 ± 0.24. It should be noted, however, that the absolute length of the reproductive system was very similar for SM and SOC males, 6.16 ± 0.75 cm and 6.76 ± 1.02 cm, respectively, despite the large difference in body size between the morphotypes (Table 1, Fig. 5).

Discussion

Two alternative reproductive strategies have been described for males of *Macrobrachium rosenbergii.* An undifferentiated juvenile male transforms into an adult SM. The SM animals may remain SM, or may further transform into OC, and eventually, BC morphotypes (Ra'anan and Sagi, 1985).

Males that remain SM cease to grow. They invest a large part of their energy on mating attempts, using sneak copulatory behavior (Ra'anan, 1982; Sagi, 1984; Telecky, 1984). The SM males possess a well-developed reproductive system, having the relative size close to that of the dominant reproductive BC morphotype (Figs. 3, 4). These findings correspond with the intensive sexual activity of the small males. Alternatively, SM

Table 1. Characterization of the male morphotypes in *Macrobrachium rosenbergii.**

Morphotype	Body weight (g) (± SD)	Carapace length (cm) (± SD)	Propodus length (cm) (± SD)	Claw color	Spination
SM	9.01 ± 2.79	2.24 ± 0.30	2.00 ± 0.24	clear	–
WOC	21.38 ± 1.43	3.20 ± 0.09	2.85 ± 0.20	orange	–
SOC	40.74 ± 3.59	4.04 ± 0.15	4.93 ± 0.38	orange	–
t-SOC	50.55 ± 6.37	4.36 ± 0.24	6.87 ± 0.64	orange-green	+
BC	53.85 ± 4.53	4.17 ± 0.14	9.68 ± 0.48	deep blue	++
OBC	30.79 ± 4.66	3.43 ± 0.20	7.26 ± 0.68	deep blue	++

* SM = Small Male; WOC = Weak Orange-Claw male; SOC = Strong Orange-Claw male; t-SOC = transforming SOC into blue-claw male; BC = Blue-Claw male; OBC = Old BC male.

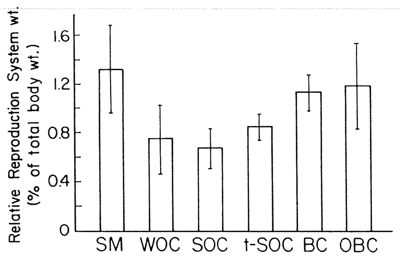

Fig. 3. Reproductive system weight as a percentage of body weight in the different male morphotypes and transitional forms in *Macrobrachium rosenbergii*. Error bars represent SD. Male morphotypes: SM = Small Male; WOC = Weak Orange-Claw male; SOC = Strong Orange-Claw male; t-SOC = transforming SOC into blue-claw male; BC = Blue-Claw male; OBC = Old BC male.

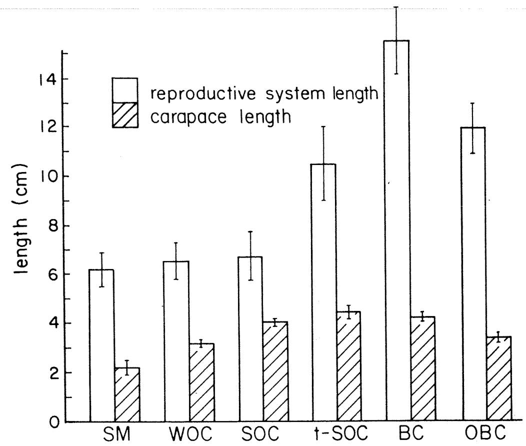

Fig. 5. Reproductive system length and carapace length in the different male morphotypes and transitional forms in *Macrobrachium rosenbergii*. Error bars represent SD. Male morphotypes: SM = Small Male; WOC = Weak Orange-Claw male; SOC = Strong Orange-Claw male; t-SOC = transforming SOC into blue-claw male; BC = Blue-Claw male; OBC = Old BC male.

males may transform into the OC morphotype, which possesses a relatively small reproductive system (Figs. 3, 4); again this corresponds with the reproductively ineffective behavior of the OC males (Ra'anan and Sagi, 1985). During most of the OC phase, i.e., the WOC transition and the SOC morphotype, the reproductive tract does not lengthen, even though somatic growth is evident. (Carapace length changes from 2.24 to 4.04 cm, while the reproductive system length remains a rather constant 6.55 cm to 6.76 cm; Table 1, Fig. 5.) In the t-SOC animals, some growth of the reproductive system is evident (Table 1, Fig. 5). Growth of the reproductive system peaks at the BC stage (Figs. 3, 5). The OC morphotype is a phase of rapid somatic growth and drastic changes in claw shape and coloration (Ra'anan and Cohen, 1985). Claw color changes from the variable clear colors of the SM males, through the bright orange of the SOC males, to the deep blue of the BC males. The above somatic changes are accompanied by the enlargement of the midgut glands, which reach their relatively largest size in the SOC morphotype (Fig. 2).

Upon becoming a BC male, somatic growth almost comes to a standstill. BC males invest most of their energy in reproductive and dominance behavior (Ra'anan and Sagi, 1985). The reproductive system of this morphotypic stage is elongated, reaching its peak relative to carapace length (Figs. 4, 5). Old BC males have a shorter reproductive system when compared to other BC males (as measured in absolute length; Fig. 5). This is due to the fact that these individuals reached the BC status in the early stages of morphotypic differentiation and at a smaller body size (Ra'anan, 1982). Other than in absolute measurements, BC and OBC males did not differ significantly in any of the relative measurements calculated for both hepatopancreas and reproductive systems. This further supports our conclusion that the specific ratios examined are not a function of body weight, but are a function of the morphotype.

In several groups of crustaceans, specific seasons are set apart for reproduction and somatic growth. This phenomenon is described mainly with respect to female re-

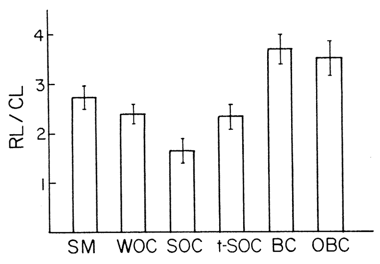

Fig. 4. The relative length of the reproductive system (RL) to carapace length (CL) in the different male morphotypes and transitional forms in *Macrobrachium rosenbergii*. Error bars represent SD. Male morphotypes: SM = Small Male; WOC = Weak Orange-Claw male; SOC = Strong Orange-Claw male; t-SOC = transforming SOC into blue-claw male; BC = Blue-Claw male; OBC = Old BC male.

production (Cheung, 1969; Armitage *et al.*, 1973). In the Brachyura, the period of somatic growth may be temporarily separated from that of reproductive growth (Adiyodi, 1978). Here, an antagonism between energy demands related to somatic growth and those linked to male reproduction could exist. In *Macrobrachium rosenbergii* the reproductive state does not appear to be associated with seasonal and environmental changes. The BC and SM males represent the reproductive, slowly growing phases, while the OC morphotype represents the nonreproductive growth phase.

Although the social and anatomical aspects of the differentiation of morphotypes in male *Macrobrachium rosenbergii* have been partially characterized, the external stimuli and intrinsic mechanisms governing the process of morphotypic differentiation remain to be studied.

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