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Population characteristics of *Macrobrachium rosenbergii* fed diets containing different protein sources under coolwater conditions in earthen ponds

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Abstract

The population structure and weight distribution of *Macrobrachium rosenbergii* raised in monoculture and fed diets containing different protein sources in earthen ponds under coolwater conditions were studied. Prawns weighing 0.5 (± 0.3 g) were stocked into nine 0.02-ha ponds at $4/m^2$ and fed at body-weight-dependent rates for 110 days. At harvest, percentage distribution by number and weight of three male morphotypes (blue claw, orange claw, and small male) and two of the three female morphotypes (berried and open) were not significantly affected by diet. The percent contribution of virgin females by weight was significantly higher in prawns fed a diet containing 15% fish meal than in prawns fed a diet containing 7.5% fish meal. The male/female ratio ranged from 2.5 to 5.3 among treatments and averaged 3.0 overall. Combined data collected from all 9 ponds indicate percentage contribution of biomass by small males was low (1.1%) and by orange claw males was high (71.0%). Among harvested prawns, > 88% exceeded 30 g and 96% exceeded 20 g wet weight. These results differ markedly from those of previous studies conducted in areas with higher mean water temperatures. Pond water temperatures in this study were apparently high enough to support rapid growth but below that necessary to stimulate reproductive changes within individuals in the population, thereby disrupting the development of a population structure normally encountered in harvested populations.

Keywords: Freshwater prawns; Population characteristics; Protein; Coolwater

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1. Introduction

Pond-cultured populations of the freshwater prawn (*Macrobrachium rosenbergii*) exhibit a complex population structure involving three male morphotypes: small (SM), orange-clawed (OC), and blue-clawed (BC), and three female morphotypes: berried (BE), open (OP), and virgin (VG) (Kuris et al., 1987). Size differences in male morphotypes of similar age are usually especially pronounced, with large BC males actually suppressing the growth of SM males (Karplus et al., 1992). The resulting wide size range of prawns is considerably disadvantageous under commercial conditions as small males may not reach marketable size (D'Abramo et al., 1989). Recent studies have been directed toward evaluating management practices designed to reduce this growth heterogeneity (D'Abramo et al., 1989). Increasing the marketable proportion of the population is extremely important to the commercial producer and can greatly improve the cost-effectiveness of an enterprise (MacLean et al., 1989). The presence of females with eggs may also have a negative impact on consumer acceptance (Tidwell et al., 1993a).

Several factors are known to affect population structure in freshwater prawns. Karplus et al. (1986) reported that as stocking density increased, the proportion of SM at harvest increased while the proportion of BC males decreased. The proportions of female morphotypes showed no significant changes. Hulata et al. (1990) found that age differences at stocking affected frequencies of morphotypes at harvest, but size at stocking exerted a less pronounced effect. MacLean et al. (1989) stated that nutritional regime may affect the structure of prawn populations.

The present study was originally designed to evaluate the effects of fish meal replacement on growth and survival of freshwater prawns raised in ponds under coolwater conditions. Production and nutritional aspects of the study are presented in Tidwell et al. (1993b). Samples of prawns from all ponds were morphotyped at harvest and effects of diets and culture conditions were evaluated. Large differences in population characteristics from previous reports from other regions were noted and these data are presented here.

2. Materials and methods

Description, preparation, and stocking of ponds

Less than 1 week prior to the anticipated stocking date, ponds were filled with water from a surface reservoir and treated with two applications of liquid fertilizer (10:34:0) at an initial rate of 9.0 kg/ha of phosphorus to achieve an algal bloom. Surface area of all experimental ponds was 0.02 ha and average water depth was approximately 1.1 m. The ponds were located at the Aquaculture Research Center, Kentucky State University, Frankfort, KY. One 7.6-cm air-lift pump operated continuously in the deepest area of each pond to prevent thermal stratification.

Juvenile prawns were shipped by air from a commercial hatchery (Aquaculture of Texas, Weatherford, TX) on 3 June 1992 and held overnight in two 3000-liter fiberglass tanks, partially filled with plastic netting to provide substrate. On the stocking date (4 June, 1992) the mean stocking weight was determined from a sample of 45 prawns that were lightly blotted to remove surface water and individually weighed (mean $\pm$ s.d. $= 0.51 \pm 0.27$ g).

Three replicate ponds were randomly assigned to each of the three experimental diets containing different levels (15, 7.5, and 0%) of fish meal. Prawns were hand-counted and distributed to the ponds at a density of $4/m^2$.

Samples

A 3.2-mm square mesh seine was used to collect a sample of individuals from each pond every 3 weeks during the growing season. Prawns composing the sample were counted, group weighed (wet weight) to the nearest gram, and returned to their respective pond.

Feed and feeding rates

Experimental diets were formulated to contain 32% crude protein. The composition of the control diet (Diet 1) was similar to that of the diet utilized by D'Abramo et al. (1989) and contained 15% fish meal. In the remaining two experimental diets, the level of fish meal was reduced to 7.5 (Diet 2) and 0% (Diet 3). The desired protein level for these diets was maintained by adding distillers' dried grains with solubles (DDGS) at 40% of the formulation, and adjusting the levels of soybean meal. Detailed formulations and diet analyses are presented in Tidwell et al. (1993b).

Two separate feedings, each consisting of one-half of the total daily ration, were distributed over the entire surface of each pond between 09.00 and 10.00 h and between 15.00 and 16.00 h. Prawns were fed a percentage of body weight based on a feeding schedule reported by D'Abramo et al. (1989). Feeding rates were adjusted weekly based on an assumed 3.0 feed conversion (D'Abramo et al., 1989) throughout the study. Every 3 weeks, total pond biomass was calculated from sample weights and feeding rates adjusted accordingly by pond. Survival was assumed to be 100%.

Water quality management

Dissolved oxygen (DO) and temperature of all ponds were monitored twice daily (09.00 h and 15.30 h) by means of a YSI Model 58 oxygen meter (Yellow Springs Instruments, Yellow Springs, Ohio). When the DO level of any pond was predicted (graphically) to decline to below 4.0 mg/l, overnight aeration was provided to that pond using an electric vertical pump aerator. Levels of total ammonia nitrogen (TAN) and nitrite in water samples collected weekly at approximately 13.00 h from each pond were determined according to the outlined procedures for a Hach DREL/5 spectrophotometer (Hach Co., Loveland, Colorado). The pH of each pond was determined daily at 13.00 h using an electronic pH meter (Fisher Scientific, Cincinnati, Ohio). When pH of the water within a pond reached or exceeded 9.5, the pond was flushed overnight with water from a reservoir pond. Comprehensive water quality data are presented in Tidwell et al. (1993b).

Harvest

Harvest occurred after 110 days of culture. One day prior to harvest, the water level in each pond was lowered to approximately 0.9 m at the drain end. Each pond was seined (21 September, 1992) three times using a 1.3-cm square mesh seine. Complete draining of each pond followed and all remaining prawns were manually harvested from the bottom of each pond and transferred to clean water for purging. Total weight and number of prawns from each pond were recorded. A sample of 50 prawns (Smith et al., 1978) per pond were randomly collected, individually weighed, and classified into one of three female morphotypes: berried (egg-carrying; BE), open (previously egg-carrying; OP), and virgin (VG), or one of three male morphotypes, blue-claw (BC), orange-claw (OC), and small (<20 g; SM), as described by Cohen et al. (1981).

Statistical analysis

Growth performance was measured in terms of final mean individual prawn wet weight (g), and total yield. Average weight, number, and total weight of each morphotype were compared among treatments by ANOVA using the SAS ANOVA procedure (SAS, 1988). Duncan's multiple range test was used to compare treatment means. Percentage data were transformed to arc sin values prior to analysis (Zar, 1984). A chi-square test (Dowdy and Wearden, 1983) was used to test if sex ratios significantly varied from the expected (1:1).

3. Results

Population structure

There was no significant difference ($P>0.05$) in survival among prawns fed the three diets. Survival in individual ponds ranged from 65 to 93%, averaging 78% overall. Males comprised 63–84% of sampled populations (by number) and averaged 75%, overall. This 3:1 sex ratio deviated significantly from the expected 1:1 ($P<0.05$). The male/female ratio did not differ significantly ($P>0.05$) among prawns fed the three diets.

Percentage contributions according to number and wet weight of different morphotypes by diet are provided in Tables 1 and 2, respectively. There were no significant treatment differences ($P>0.05$) among prawns fed the three diets except in biomass contributed by virgin females. The virgin class of females fed Diet 1 composed a significantly greater ($P<0.05$) proportion of biomass than virgin females fed Diet 2 (Table 2). Average weight of individuals within each morphotype did not differ significantly ($P>0.05$) among treatments (Table 3).

Because of relatively small differences among treatments, results from all ponds were pooled. Mean ($\pm$s.e.) wet weight of males (44.0 ± 13.0 g) was not significantly different ($P>0.05$) from that of females (38.6 ± 11.4 g). Mean weights of BC (54.8 g) and OC males (48.0 g) were significantly higher ($P<0.01$) than that of SM (13.9 g) males but

Table 1
Percent distribution according to number of each morphotype based on a sample of 50 prawns per pond

	Male			Female		
Diet	Blue claw	Orange claw	Small	Berried	Open	Virgin
1	6.2 ± 2.3	57.2 ± 4.6	3.4 ± 1.3	8.6 ± 3.0	4.9 ± 0.7	13.8 ± 1.7
2	6.3 ± 1.2	69.9 ± 3.6	5.6 ± 9.6	7.0 ± 0.5	4.9 ± 1.4	6.3 ± 2.1
3	7.2 ± 1.4	67.2 ± 8.1	1.4 ± 0.7	8.5 ± 4.2	5.0 ± 2.8	12.4 ± 3.8

Values are means $\pm$ s.e. of three replications. Differences between treatment means were not statistically different ($P>0.05$).

Table 2
Percent distribution according to wet weight of each morphotype based on a sample of 50 prawns per pond

	Male			Female		
Diet	Blue claw	Orange claw	Small	Berried	Open	Virgin
1	7.8 ± 3.5	67.3 ± 6.9	1.0 ± 0.2	13.9 ± 3.7	4.3 ± 1.0	10.5 ± 0.9[a]
2	8.6 ± 1.7	74.7 ± 3.0	1.7 ± 1.7	6.3 ± 2.3	4.0 ± 1.3	4.7 ± 1.7[b]
3	8.7 ± 1.0	71.0 ± 7.9	0.5 ± 0.2	7.7 ± 4.3	4.3 ± 2.6	7.9 ± 1.8[ab]

Values are means ± s.e. of three replications. Means within a column having different superscripts were significantly different ($P < 0.05$).

Table 3
Average individual weight of each morphotype based on a sample of 50 prawns per pond

	Male			Female		
Diet	Blue claw	Orange claw	Small	Berried	Open	Virgin
1	47.1 ± 26.1	48.8 ± 6.2	14.2 ± 4.9	41.2 ± 4.3	35.3 ± 2.1	31.9 ± 1.3
2	59.9 ± 10.6	49.0 ± 5.1	13.3 ± 0.0	39.2 ± 0.3	35.1 ± 3.2	31.0 ± 4.1
3	57.3 ± 5.8	48.6 ± 6.3	13.8 ± 0.4	38.0 ± 2.5	37.2 ± 6.1	32.9 ± 4.5

Values are means ± s.e. of three replications. Differences between treatment means were not statistically different ($P > 0.05$).

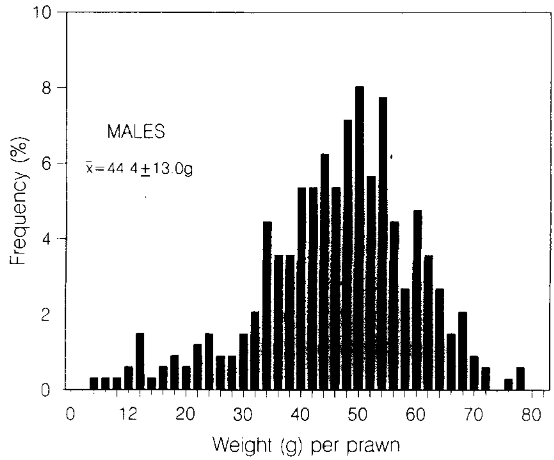

Fig. 1. Weight–frequency distribution of male prawns harvested from 9 monoculture ponds (treatments pooled, $n = 336$).

not significantly different ($P > 0.05$) from each other. There were significant differences ($P < 0.05$) in mean wet weight among all the female morphotypes with BE (39.5 g) > OP (35.9 g) > VG (31.9 g).

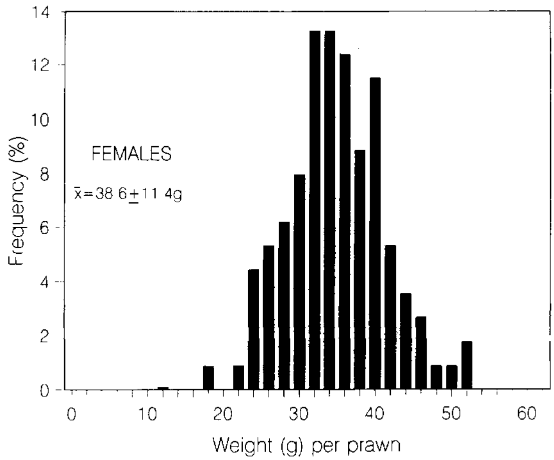

Fig. 2. Weight–frequency distribution of female prawns harvested from 9 monoculture ponds (treatments pooled, $n=114$).

Weight–frequency distributions of male and female prawns from all ponds are illustrated in Figs. 1 and 2, respectively. Prawn weights were subjected to a goodness-of-fit test for normal distribution. The hypothesis that the population is normal could not be rejected for combined sexes ($P=0.682$), females ($P=0.802$), and males ($P=0.082$). The lack of bimodality in the male population of this study was largely due to a comparatively low number of small males (<20 g).

4. Discussion

The percent distribution of the identified prawn morphotypes derived from the combined harvest data of all ponds (Fig. 3) is markedly different from those of other published studies. Especially notable are differences in proportions of SM and OC males. Karplus et al. (1986) found that harvested prawns originally stocked at $4/m^2$ in polyculture consisted of 24% SM males and 15% OC males by number, compared to 4% and 65%, respectively, in this study. D'Abramo et al. (1989) reported that OC males comprised 37% of total harvest biomass by weight in monoculture ponds stocked at $4/m^2$, whereas this amount was 71% in the present study. Fig. 4 illustrates the relative proportions of different morphotypes within the total males harvested in three studies conducted in different climatic regions. Decreased numbers of small males and increased numbers of larger, faster growing OC males increases the marketable proportion of harvested prawns. This could be of utmost importance to the success of a commercial enterprise (MacLean et al., 1989). Pond population data in this study are based on samples representing approximately 8% of the harvested population. It is possible that prawns in the SM classification were under-represented in the sample collected from each pond. However, similar average sizes of the sample population (44 g) and the total harvested population (42 g) for each pond suggest otherwise.

The proportions of prawns in different weight classes are presented in Fig. 5. Some investigators have considered ≥30 g to be a marketable size (D'Abramo et al., 1989).

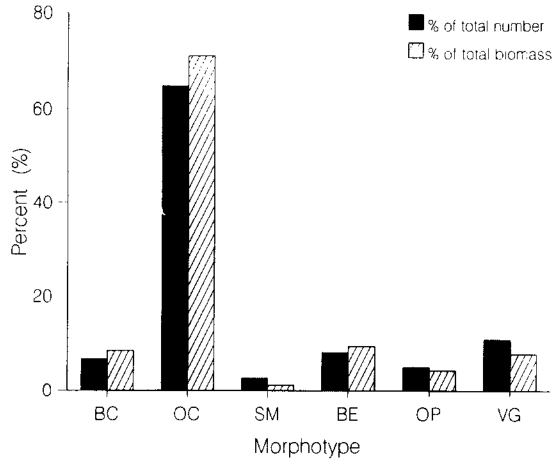

Fig. 3. Proportions of male and female morphotypes as percentage of total number and total biomass. BC = blue claw males; OC = orange claw males; SM = small males; BE = berried females; OP = non-berried females with open brood chambers; VG = non-breeding females with closed brood chambers.

Based on this definition over 88% of the prawns in this study would be of marketable size, a percentage exceeding those encountered in similar studies. In a polyculture study with prawns stocked at $4/m^2$ Karplus et al. (1986) reported that 35% of the harvested prawns achieved weights of ≥ 30 g. Hulata et al. (1990) reported that 84% of prawns reached harvestable weight (≥ 30 g) when stocked at $2/m^2$, half the stocking density in this study.

D'Abramo et al. (1989) stated that economic feasibility would be strengthened by maximizing final harvest weight and minimizing size variation and contribution of economically undesirable morphotypes rather than simply maximizing overall yield. Large differ-

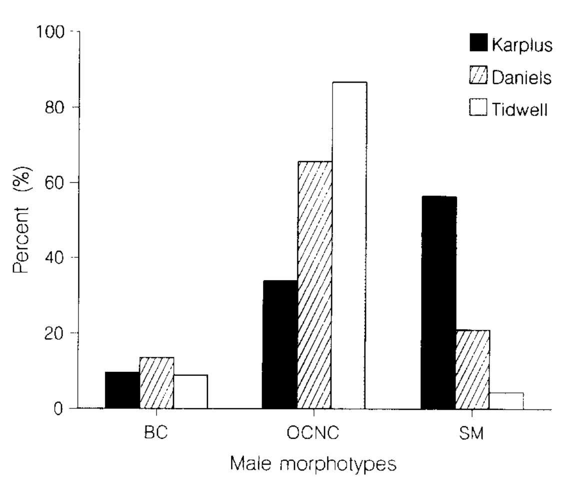

Fig. 4. Proportions of male morphotypes as percentage of total males in three studies with prawns stocked at similar densities. Karplus = Karplus et al. (1986), Daniels = Daniels and D'Abramo (1994), Tidwell = present study. BC = blue claw males; OC = orange claw males; SM = small males.

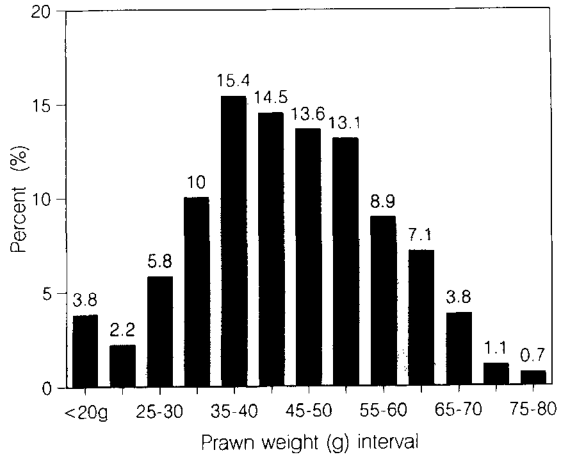

Fig. 5. Proportions of prawns (both sexes) harvested from 9 monoculture ponds within selected 5 g weight intervals.

ences in population structure exist between previous studies and this study despite similar culture densities. However, in some cases management practices and more importantly environment differ. The proportion of orange-claw males, which is the fastest growing and often largest morphotype, was very high in the present study. The proportions of small males and berried females was very low. Small males are not a marketable size and berried (egg-carrying) females are considered unsightly and undesirable by some consumers. Also, the sex ratio at harvest was skewed towards males while most studies have reported male/female ratios of less than one (Willis and Berrigan, 1977; Smith and Sandifer, 1980; Smith et al., 1978, 1980, 1981, 1982; Sandifer et al., 1982; Karplus et al., 1986), even when prawns were graded by size or age prior to stocking (Hulata et al., 1990).

Smith et al. (1978) suggested three mechanisms that might explain the prevalence of females in previously studied prawn populations, these include differences in sex ratio at stocking, environmental conditions that favor the development of females, and selective mortality of males. Similar mechanisms could also possibly explain the predominance of males in the current study.

Due to lack of sexual dimorphism at small sizes, sex and sex ratios at stocking cannot be readily determined. However, in previous studies in which prawns stocked into ponds were either of similar size (Tidwell et al., 1993a,b), graded by size (0.1–0.4 g) (Daniels and D'Abramo, 1994), or divided by size (0.25 and 0.5 g) and age (66 or 129 days) (Hulata et al., 1990), sex ratios skewed toward males have not been reported.

Effects of environmental conditions, especially culture temperature, can also be considered. Sandifer and Smith (1985) reported that satisfactory temperatures for prawn culture ranged from 26 to 31°C. In most aquatic animals optimum temperature ranges and temperature ranges amenable to reproduction are usually more narrow than those for growth (Boyd, 1990). Optimum temperatures for prawn culture are reported to be 28–30°C (Sandifer and Smith, 1985). In this study, during the 110-day culture period, morning temperatures averaged 23°C and afternoon temperatures 25°C. The minimum recorded morning temperature was 18.7°C (June) and maximum afternoon temperature was 30.4°C (July). Monthly

Table 4
Mean monthly water temperatures (± s.e.) of all ponds and the monthly mean (± s.e.) of morning dissolved oxygen level and afternoon pH of all ponds

Month	Water temperature (°C)		Dissolved oxygen (mg/l)	pH
	a.m. (08.00 h)	p.m. (16.00 h)	a.m. (08.00 h)	p.m. (16.00 h)
June	22.3 ± 0.3	24.6 ± 0.2	4.5 ± 0.8	9.0 ± 0.3
July	25.1 ± 0.2	27.0 ± 0.2	4.6 ± 0.4	9.5 ± 0.3
August	22.8 ± 0.3	24.7 ± 0.3	4.1 ± 0.4	9.5 ± 0.3
September	23.6 ± 0.2	25.7 ± 0.2	4.0 ± 0.7	8.4 ± 0.1
Overall	22.9 ± 0.2	24.9 ± 6.2	4.3 ± 0.3	9.1 ± 0.5

No dissolved oxygen concentrations less than (<) 2.6 mg/l were recorded.

means (± s.e.) for water temperature are presented in Table 4. The low incidence of egg-carrying and previously egg-carrying females (8 and 5%, respectively) may indicate that culture temperatures were consistently below that temperature needed to cause females to become reproductively mature. No egg-carrying females were captured in any scheduled seine samples until the last sampling date, approximately 3 weeks prior to harvest. In addition, the small number of ovigerous females captured had many eggs that appeared inviable and some were infested with fungi.

Culture temperatures at the low end of the satisfactory range could also potentially delay progression of males through their developmental pathway by direct environmental influence or indirectly by delaying female maturation. Males are known to adjust their maturation time to correspond to that of the females (Cohen et al., 1981). Late maturation of females could delay development of the final stage (BC) in the male maturation pathway. With development of BC males delayed, growth suppression of SM males would be absent or reduced, allowing them to advance to the OC stage (Ra'anan and Cohen, 1985). With SM males not inhibited from developing to the OC stage, and with no mature females to stimulate development of BC males, a large proportion of OC males could develop.

Table 5
Summary of *M. rosenbergii* monoculture research with prawns stocked at approximately 4/m^2 in different geographical locations

Study	Location	Stocking size (g)	Stocking density (prawn/m^2)	Culture period (days)	Mean temp. (p.m.) (°C)	Sex ratio (M/F)	Avg. size (g)	Production (kg/ha)	Daily yield (kg/ha)	Survival (%)	FCR[3]
Smith et al. (1981)	SC	0.37	4.31	168	26.6	0.73	25.8	995	5.9	86.9	1.2
D'Abramo et al. (1989)	MS[1]	0.17	3.95	140	29.8	1.13	26.0	830	5.9	80.8	3.4
	MS[2]	0.72	3.95	135	29.7	1.13	36.3	894	6.6	92.6	4.7
Present study	KY	0.51	3.95	110	24.9	2.95	42.0	1,268	11.5	78.1	2.9

SC = South Carolina, USA; MS = Mississippi, USA; KY = Kentucky, USA.
[1] Study conducted in 1985.
[2] Study conducted in 1986.
[3] FCR = feed conversion ratio.

A comparison of culture conditions and results of monoculture studies in different regions is given in Table 5. D'Abramo et al. (1989) under similar conditions of stocking size, monoculture, and stocking rate, but with higher average temperatures and a longer growing season reported daily yields of 5.9 and 6.3 kg/ha. In this study daily yields averaged 11.5 kg/ha. With energetic demands of reproduction reduced for both sexes, energy that would otherwise be devoted to reproduction (i.e., egg production in females, aggressive interactions in males) could be devoted to growth. In our study prawns were fed aggressively based on a percentage of biomass, twice daily, with feed distributed uniformly over the pond. Access to, and amounts of, feed should not have been limiting. Lack of reproductive demands, large numbers of OC males, high feed availability, and efficient feed conversion could possibly account for the high daily prawn yields at comparatively low culture temperatures and a short growing season (110 days) reported here. Culture of freshwater prawn at this latitude generally has been considered economically prohibitive due to the short growing season. However, under conditions of lower water temperature shifting of population structure toward desirable morphotypes and increasing percentages of marketable individuals, a potential increase in total revenue may be realized. Culture at this latitude could also offer the prospects of multi-cropping with cool or coldwater finfish species, thereby allowing efficient use of pond resources throughout a calendar year (Tidwell et al., 1991).

Cohen et al. (1981) stated that further studies concerning the dynamics of the interactions among the various morphotypes under different environmental conditions would contribute to the understanding of aspects of the social biology of freshwater prawns and possibly lead to the incorporation of management practices that will improve aquaculture production. Effects of latitude (i.e., water temperature) on population structure in prawns should be further evaluated. Culture of prawns from a single source, using the same feed, stocking density, and pond size at different latitudes could help to increase our understanding of the role of water temperature.

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