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Growth, Survival, and Biochemical Composition of Freshwater Prawns *Macrobrachium rosenbergii* Fed Natural Food Organisms Under Controlled Conditions

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Abstract

Under production conditions freshwater prawns *Macrobrachium rosenbergii* are supplied nutrients by a combination of prepared diets and natural pond organisms. For maximum production efficiency and profitability it is important that the nutritional contributions of natural foods be identified and quantified. In this study the relative importance of forage organisms previously identified as important natural foods for prawns in ponds were evaluated under controlled conditions. Juvenile prawns (average weight 1.80 ± 0.06 g) were stocked into 18 37.5-L aquaria at a density of 10 prawns per tank. The six dietary treatments tested were: 1) unfed (negative control); 2) commercially prepared diet (positive control); 3) oligochaetes; 4) chironomids; 5) zooplankton; and 6) a combination of the latter three. Each treatment was evaluated in triplicate aquaria for 7 wk. The growth rate of prawns in the unfed treatment was statistically lower than in fed treatments ($P < 0.05$). There were no significant differences ($P > 0.05$) between growth rates and survivals among prawns in the five fed treatments. Selective retention of arachidonic acid (20:4n-6), eicosapentanoic acid (22:5n-3), and docosahexanenoic acid (22:6n-3) in unfed prawns likely indicates the relative nutritional importance of these fatty acids. Comparisons of whole-body fatty acid and amino acid concentrations of prawns and food organisms indicate that zooplankton and oligochaetes may have the most appropriate biochemical compositions as prawn food sources.

Feed is often the major expense in pond production of freshwater prawns *Macrobrachium rosenbergii*, representing as much as 40–60% of operating costs (D'Abramo and Sheen 1991). However, even when prepared diets are provided, pond flora and fauna are significant sources of nutrition for prawn growth (Moore 1986) providing up to one-third of the prawns' nutrition (Tidwell et al., in press). Although the nutritional role of natural foods is obviously important, it remains ill-defined (Corbin et al. 1983).

Currently, little or no information exists concerning the dietary nutrient requirements of crustaceans under practical, semi-intensive pond conditions (Tacon 1995). This is largely due to the reluctance of laboratory-based crustacean nutritionists to work under applied field conditions and to the difficulty of merging nutrition and ecology in the quantification of the contribution of natural food organisms in the overall nutritional budget of pond raised crustaceans (Tacon 1995). This lack of understanding limits the potential of developing management procedures for maximizing the availability of desirable forage organisms (Corbin et al. 1983).

Studies of food habits of crustaceans are difficult (D'Abramo and Sheen 1991). Sampling of crustacean stomachs may be misleading due to small stomach and sample sizes, small size of prey items, mastication of food items, and a rapid rate of catabolism and stomach emptying (Brown et al. 1992). Tidwell et al. (1995; in press) monitored population densities of benthic macroinvertebrates in prawn ponds with different feed and fertilization regimes and found that oligochaetes and chironomids are likely major contributors to prawn nutrition under pond conditions.

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TABLE 1. *Means (±SE) for proximate composition of the prepared diet and natural foods. Values are means and SE of two samples per food. Means in a row bearing different letters are significantly different* (P = *0.05).*

	Food			
Composition	Diet	Oligochaetes	Chironomids	Zooplankton
Moisture	11.6 ± 0.1[w]	81.7 ± 0.1[x]	87.3 ± 0.0[y]	88.7 ± 0.1[z]
Lipid[a]	8.0 ± 0.1[z]	4.8 ± 0.8[y]	6.2 ± 0.5[y]	4.3 ± 0.2[x]
Protein[a]	30.7 ± 0.1[x]	51.2 ± 0.2[y]	52.6 ± 0.3[y]	58.1 ± 2.1[z]
Carbohydrate[a,b]	51.5 ± 0.2[z]	38.6 ± 1.6[y]	26.9 ± 1.3[x]	17.6 ± 0.8[w]
Ash	9.8 ± 0.1[x]	5.4 ± 0.6[w]	14.4 ± 0.4[y]	18.8 ± 0.9[z]

[a] Dry weight basis.
[b] Carbohydrate (Nitrogen-free extract; NFE) was estimated by subtracting percentages of moisture, crude protein, lipid, crude fiber, and ash from 100%.

Brown et al. (1992) stated that an experimental feeding study might be a useful approach in defining nutritionally important food organisms for crustaceans. Coyle et al. (1996) conducted such a study and found that oligochaetes, chironomids, and gastropods were suitable food organisms for prawns. They also found that juvenile prawns, of sizes larger than previously thought, could efficiently utilize live zooplankton as a food source. However, this study was of short duration and used some food organisms that were terrestrial species or had been stored frozen. It would be advantageous to evaluate benthic macroinvertebrates, identified as important natural forages in ponds, under controlled conditions where consumption and nutritional contributions could be identified and quantified. This could lead to the development of optimal pond preparation procedures and feeding practices to maximize the nutritional contribution of natural food items.

The objective of this study was to evaluate the relative acceptance and nutritional value of live benthic macroinvertebrates, identified as important forage organisms in ponds, and to quantify their nutritional contribution based on chemical analyses of the organisms, and prawns fed those organisms.

Materials and Methods

Rearing Units and Procedures

The study was conducted at the Aquaculture Research Center, Kentucky State University, Frankfort, Kentucky, USA. On 7 July 1995 18 37.5-L aquaria were stocked at a density of 20 prawns per aquarium. During this 1-wk conditioning period the prawns were fed a commercially prepared 32% protein diet at a rate of 10% of body weight, once daily. On 14 July immediately prior to initiating test diets, prawn numbers in each aquarium were reduced to 10 shrimp of similar size ($\bar{x}$ ± SD, 1.81 ± 0.06 g).

Each aquarium was supplied with a continuous flow of dechlorinated tap water at a rate of 1.0 L/min. To provide refuge for the prawns and help prevent cannibalism, each aquarium contained one 100-cm × 30-cm plastic screen (1-cm mesh) folded upon itself in an S-shape and secured with 12-cm cable ties, and four "habitat" units consisting of five 8-cm segments of 2.5-cm diameter PVC pipe glued two on top of three.

Feeds and Feeding Rates

The six dietary treatments tested were: 1) unfed (negative control); 2) fed a commercially prepared diet (positive control); 3) chironomids; 4) oligochaetes; 5) zooplankton; or 6) fed a combination of the latter three. Three replicate aquaria were randomly assigned to each treatment. The prepared diet was commercially processed into 5-mm sinking pellets and formulated to contain 32% protein. Ingredient composition of the formulated diet was similar to that of the

TABLE 2. *Amino acid composition (% of total amino acids) of the diet, food organisms, and prawn tail muscle.*

Amino acid	Food				Tail muscle[a]
	Diet	Oligochaetes	Chironomids	Zoo plankton	
Arg	5.68	7.40	5.25	6.68	9.63
Cys	1.46	1.56	0.94	1.39	1.11
His	3.90	3.41	3.38	3.69	2.70
Ile	3.82	4.27	5.07	4.53	4.32
Leu	9.50	8.12	6.44	7.72	7.37
Lys	4.85	8.25	7.13	7.72	8.57
Met	2.05	2.42	2.25	2.18	3.17
Phe	4.57	4.69	6.57	4.87	3.88
Thr	4.57	5.97	5.30	5.70	4.21
Tyr	2.80	3.41	2.44	4.36	3.12
Val	4.47	5.26	6.00	5.87	4.43

[a] Muscle values are from Tidwell et al. (1993) for compariative purposes and are not included in statistical analysis.

TABLE 3. *A/E ratios of the diet, food organisms, and prawn tail muscle.*

Amino acid	Food				Tail muscle[a]
	Diet	Oligochaetes	Chironomids	Zoo plankton	
Arg	121.5	139.0	104.5	128.5	199.4
His	83.5	64.2	67.2	69.0	55.9
Ile	81.9	80.2	100.7	84.6	89.5
Leu	203.4	152.4	138.1	144.2	152.7
Lys	103.8	155.1	141.8	144.2	177.4
Met	43.9	45.5	44.8	40.8	65.7
Phe	97.9	88.2	130.6	90.9	80.4
Thr	97.9	112.3	104.5	106.6	87.3
Val	106.3	98.9	119.4	109.7	91.8
Try	59.9	64.2	48.5	81.5	—

[a] Muscle values are from Tidwell et al. (1993) for comparative purposes and are not included in statistical analysis.

diet used by Tidwell et al. (1995). The chironomid, oligochaete, and zooplankton treatments were selected based on previous pond (Tidwell et al. 1995; in press) and aquarium investigations (Coyle et al. 1996). Chironomids and oligochaetes were collected every other day from the benthic layer of research ponds using a large (36 × 50 cm) No. 30 U.S. Series Screen. Organisms were rinsed clean of mud and debris and stored live for ≤3 d in a water-filled basin (35 × 30 × 15 cm) at 14 C until fed. The mixed zooplankton, primarily *Daphnia spp.*, were collected daily from the same ponds using a fine mesh aquarium net. Zooplanktors were held alive in aerated tanks until being weighed and added to prawn tanks.

Beginning 14 July (after the 1 wk conditioning period) prawns were fed their respective foods once daily for 7 wk. Prawns in each aquarium were initially fed at a rate of 10% of their body weight, on a dry weight basis. Consumption of food organisms was measured daily by siphoning and weighing uneaten organisms from each tank. After Day 1, prawns in individual aquaria were fed 120% of the amount consumed the previous day. The amounts of zooplankton and commercial feed consumed could not be accurately measured due to difficulty in recapturing the zooplankton and disintegration and leaching of feed pellets. Therefore, the amounts of zooplankton and the prepared diet fed were provided at 10% of the prawns body weight daily. Rates were adjusted weekly based on updated weights of prawns and were near or slightly above consumption in all tanks. Prawns in the combination treatment were fed zooplankton at a set rate of 3.3% of body weight, on a dry weight basis, and 120% of the previous days' consumption of oligochaetes and chironomids. Prawns in each tank were group weighed weekly to the nearest 0.1 g and counted for determination of average weight and survival.

Water Quality

Water quality analyses were conducted weekly. Water temperature and dissolved oxygen were measured using a YSI Model 55 dissolved oxygen meter (YSI Instruments Co., Yellow Springs, Ohio, USA), pH was measured using an electronic pH meter (Omega Engineering Inc., Stanford, Connecticut, USA), and total ammonia-N using a Hach DR/2000 spectrophotometer

TABLE 4. *Means (±SE) for fatty acid composition of the diet and natural foods (percentage of total fatty acids). Values are the mean and SD for two samples per food. Means in a row bearing different letters are significantly different.*

	Food			
Fatty acid	Diet	Oligochaetes	Chironomids	Zooplankton
16:0				
16:1n-7	5.76 ± 0.07^{x}	4.85 ± 0.59^{w}	23.31 ± 1.27^{z}	19.76 ± 0.10^{y}
18:1n-9	1.55 ± 0.09^{w}	3.70 ± 0.10^{x}	12.59 ± 0.44^{z}	8.90 ± 0.26^{y}
18:2n-6	23.69 ± 0.08^{z}	10.76 ± 0.54^{w}	16.68 ± 1.02^{x}	19.88 ± 0.18^{y}
18:3n-3	45.53 ± 0.02^{z}	6.02 ± 1.41^{w}	14.28 ± 0.36^{y}	8.66 ± 0.16^{x}
20:4n-6	2.64 ± 0.25^{y}	3.78 ± 0.66^{y}	9.85 ± 0.69^{z}	11.15 ± 0.22^{z}
20:4n-6	0.35 ± 0.0^{x}	7.41 ± 0.29^{z}	1.50 ± 0.14^{y}	7.19 ± 0.68^{z}
20:5n-3	1.09 ± 0.04^{z}	5.03 ± 0.45^{zy}	3.12 ± 0.08^{yx}	7.12 ± 1.59^{z}
22:6n-3	0.38 ± 0.06^{z}	1.41 ± 0.40^{z}	0.00 ± 0.0^{z}	1.24 ± 1.75^{z}
Saturates	20.84 ± 0.07^{x}	17.28 ± 0.79^{w}	36.51 ± 1.48^{z}	31.29 ± 0.01^{y}
Monoenes	25.69 ± 0.05^{y}	25.73 ± 0.01^{y}	31.48 ± 1.61^{z}	31.21 ± 0.39^{z}
Dienes	45.53 ± 0.02^{z}	8.48 ± 1.76^{x}	14.28 ± 0.36^{y}	8.66 ± 0.16^{x}
Trienes	2.64 ± 0.25^{x}	5.47 ± 0.81^{y}	10.12 ± 0.30^{z}	11.15 ± 0.22^{z}
PUFA	51.66 ± 0.14^{z}	30.00 ± 1.58^{x}	29.02 ± 0.12^{x}	35.35 ± 0.57^{y}
n-3	5.57 ± 0.12^{w}	11.34 ± 0.30^{x}	12.97 ± 0.77^{y}	19.50 ± 0.06^{z}
n-6	45.75 ± 0.02^{z}	18.66 ± 1.29^{y}	14.55 ± 0.75^{x}	15.85 ± 0.52^{x}
n-3/n-6	0.12 ± 0.0^{w}	0.61 ± 0.03^{x}	0.89 ± 0.10^{y}	1.23 ± 0.04^{z}

(Hach Co., Loveland, Colorado, USA). Overall means (±SE) for these variables over the duration of the study were: water temperature 26.6 ± 0.7 C; dissolved oxygen 6.3 ± 1.8 mg/L; pH 7.8 ± 0.1, total ammonia-nitrogen 0.31 ± 0.4 mg/L. These represent conditions suitable for prawn culture.

Biochemical Sampling

Near the end of the study, samples of the diet and freshly captured forage organisms were frozen using liquid nitrogen (−196 C) and stored frozen (−40 C) for proximate analyses (Table 1), amino acid analyses (Tables 2 and 3), and fatty acid analyses (Table 4). At termination of the study, all prawns in each tank were sacrificed, homogenized in a blender, immediately frozen in liquid nitrogen (−196 C) and stored (−40 C) for fatty acid and amino acid analyses.

Statistical Analyses

Data on weekly individual weights (g) were regressed against weeks cultured and tested for significance of regression (Dowdy and Wearden 1983). Slopes of regression lines for the six treatments were compared using Student's t-tests (Steel and Torrie 1980). Survivals and biochemical compositions of prawns in the different treatments were compared using the SAS ANOVA procedure (SAS Institute, Inc. 1987) after transformation to *arc sin* values (Zar 1984).

TABLE 5. *Regression equations developed for prawn weights over time in the six treatments, their coefficients of determination (r^2), and their level of significance.*

Variable	Regression equation	r^2	Level of significance
Unfed	Y = 1.7051 + 0.1661X	0.367	0.0001
Feed	Y = 1.7519 + 0.2967X	0.875	0.0000
Oligochaetes	Y = 1.8074 + 0.331X	0.844	0.0000
Chironomids	Y = 1.7705 + 0.3400X	0.882	0.0000
Combination	Y = 1.8541 + 0.3779X	0.878	0.0000

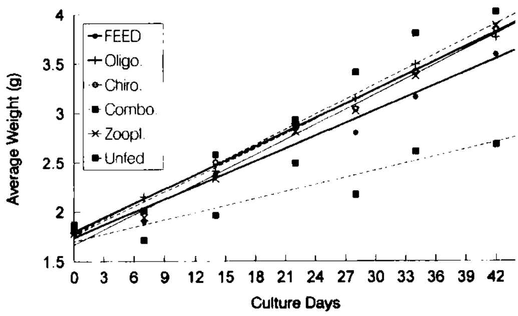

FIGURE 1. *Relationship between prawn body weight and culture period (d). Means represent three replicate aquaria.*

Means were separated using LSD (least significant difference test) (Steel and Torrie 1980).

Results and Discussion

The regression of body weight(g) on culture time (wk) was significant ($P \leq 0.05$) for all six treatments (Table 5) indicating that rates of gain were well described by a straight line (Fig. 1). The slope of the line for unfed animals was significantly different ($P \leq 0.05$) from the slopes of lines for all fed treatments. The slopes of regression lines for the five fed treatments were not significantly different ($P > 0.05$), indicating that differences in growth rates were not statistically significant. This differs from Coyle et al. (1996) who reported that the growth rate of prawns fed zooplankton was significantly greater than those fed oligochaetes. However, oligochaetes in that study were terrestrial species and may have differed from aquatic species, especially in fatty acid profiles. Survival was not significantly different ($P > 0.05$) among the six treatments and averaged 56% overall. This is lower than the 80% survival reported by Coyle et al. (1996). However, the duration of the present study was greater (7 wk vs. 3 wk). D'Abramo et al. (1988) reported that reduced survivals were common in communally reared prawns.

Prawns are known to be opportunistic omnivores and appear to be able to efficiently utilize different types of food items. Even unfed prawns gained 48% during the study period, presumably by grazing on bacterial growth in the tanks. Brown et al. (1992) reported weight gains of 272% in small unfed crayfish *Procambarus clarkii* and at least partially attributed this to grazing on bacteria. Of prawns in fed treatments, those fed the combination of natural foods (oligochaetes, chironomids, and zooplankton) had the highest rate of gain (and highest survival) while those fed the prepared diet had the lowest rate of gain.

Determination of exact amino acid requirements in crustaceans is difficult (D'Abramo and Sheen 1991). However,

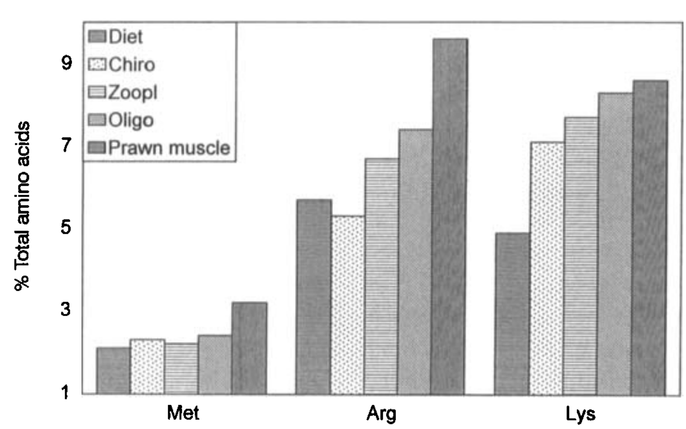

FIGURE 2. *Concentrations of methionine (Met), arginine (Arg), and lysine (Lys) (as % total amino acids) in a prepared diet and natural foods fed to prawns in aquaria. The amount in prawn muscle tissue is from Tidwell et al. (1993).*

TABLE 6. *Means (±SE) for fatty acid composition of whole bodies (percentage of total fatty acids) of prawns either unfed, fed a prepared diet, or natural foods and prawn egg compositions are included as a reference tissue. Values represent means ± SE of three replicate tanks. Each tank was a composite sample of all surviving prawns. Means in a row bearing different letters are significantly different (*$P < 0.05$*).*

	Dietary treatment			
Fatty acid	Unfed	Diet	Zooplankton	Chironomids
16:01	17.67 ± 1.11xf^{y}	18.02 ± 0.34^{y}	18.52 ± 0.34^{y}	19.72 ± 0.54^{z}
16:1n-7	1.29 ± 1.12^{w}	1.09 ± 0.31^{w}	2.62 ± 0.09^{x}	6.25 ± 0.71^{z}
18:0	12.54 ± 1.35^{z}	7.36 ± 0.23^{e}	9.84 ± 0.63^{yx}	10.56 ± 0.44^{y}
18:1n-9	19.29 ± 2.33^{yx}	23.90 ± 0.56^{z}	19.57 ± 0.49^{xy}	20.55 ± 1.07^{y}
18:2n-6	7.80 ± 0.97^{e}	30.77 ± 0.67^{z}	10.16 ± 0.56^{x}	12.30 ± 1.48^{y}
18:3n-3	0.58 ± 1.00^{w}	1.42 ± 0.32^{xw}	6.18 ± 0.81^{z}	4.42 ± 0.28^{y}
20:4n-6	9.92 ± 0.86^{z}	2.06 ± 0.34^{e}	6.98 ± 0.32^{yx}	3.36 ± 0.26^{w}
20:5n-3	17.70 ± 0.66^{z}	5.49 ± 0.73^{w}	10.58 ± 0.30^{y}	8.22 ± 0.66^{x}
22:6n-3	6.65 ± 0.55^{z}	2.64 ± 0.22^{x}	3.45 ± 0.40^{y}	1.56 ± 0.41^{w}
Saturates	31.57 ± 0.34^{xw}	27.86 ± 0.41^{w}	37.38 ± 5.80^{z}	36.22 ± 0.90^{zy}
Monoenes	20.58 ± 2.39^{w}	25.50 ± 0.70^{yx}	23.05 ± 0.58^{xw}	28.93 ± 1.52^{z}
Dienes	7.80 ± 0.97^{w}	31.69 ± 1.33^{z}	10.32 ± 0.83^{x}	12.95 ± 1.45^{y}
Trienes	0.58 ± 1.00^{w}	1.42 ± 0.32^{w}	6.18 ± 0.81^{z}	4.69 ± 0.46^{y}
PUFA	42.65 ± 2.08^{z}	43.26 ± 0.46^{z}	38.30 ± 0.42^{y}	30.79 ± 2.35^{w}
n-3	24.93 ± 0.90^{z}	9.52 ± 1.03^{w}	14.46 ± 0.86^{yx}	9.78 ± 1.04^{w}
n-6	17.72 ± 1.19^{yx}	30.90 ± 5.29^{z}	13.31 ± 1.00^{xw}	8.62 ± 0.17^{e}
n-3/n-6	1.41 ± 0.05^{z}	0.28 ± 0.03^{w}	1.09 ± 0.13^{y}	1.14 ± 0.14^{y}

[a] Egg values are from Tidwell et al. (1993) for comparative purposes and were not included in statistical analyses.

amino acid requirements can be estimated by examining the amino acid content of the animal (Wilson and Poe 1985). Compared to prawn tail muscle (as an indicator tissue) oligochaetes had lower levels of methionine and arginine, chironomids had lower levels of methionine, leucine, lysine, and arginine, while zooplankton had lower levels of methionine, lysine, and arginine (Table 2). The prepared diet appeared to have the poorest match with the amino acid profile of prawn tissue (Fig. 2), being lower in methionine and isoleucine, and much lower in lysine and arginine. Arginine deficiency has been implicated in cannibalism in crustaceans (Brown et al. 1992). In this study, the fed treatments with the lowest prawn survivals (57%, 51%, and 53%) were also the ones with the lowest arginine levels (zooplankton, feed, and chironomids, respectively).

Arai (1981) proposed the use of A/E ratios (the ratio of individual essential amino acids and total amino acid content) to estimate amino acid requirements and identify potential amino acid deficiencies. Comparisons of A/E ratios of the natural foods with those of the prawn tail muscle (Table 3) largely support the relationships identified by absolute composition data (Table 2). The A/E ratios of the prepared diet were less similar to the A/E ratios of prawn muscle than other food items, being lower in arginine, methionine and extremely low in lysine. Chironomids were also lower in leucine, lysine, methionine, than prawn muscle, and extremely low in arginine. Zooplankton had A/E ratios more similar to prawn muscle than the diet and chironomids but still had lower levels of arginine, leucine, lysine, and methionine than prawn muscle. Oligochaetes appeared to have the most suitable A/E ratios of the foods evaluated, having higher ratios for most amino acids, though ratios for arginine, lysine, and methionine were still lower than in prawn muscle.

Qualitative dietary fatty acid requirements of crustaceans have been identified

TABLE 6. *Extended.*

Dietary treatment		
Oligochaetes	Combination	Prawn eggs[a]
14.37 ± 0.25^{w}	16.61 ± 0.29^{x}	17.85 ± 0.64
3.95 ± 0.24^{y}	4.01 ± 0.75^{y}	3.84 ± 0.65
8.42 ± 0.39^{we}	9.06 ± 0.24^{xw}	6.79 ± 0.25
16.48 ± 0.67^{w}	17.66 ± 0.18^{xw}	26.82 ± 2.31
7.83 ± 0.59^{we}	9.35 ± 0.38^{xw}	24.79 ± 2.35
2.42 ± 0.65^{x}	3.82 ± 0.09^{y}	1.51 ± 1.01
7.89 ± 0.39^{y}	6.60 ± 0.90^{x}	2.10 ± 0.15
8.73 ± 0.55^{x}	8.87 ± 1.34^{x}	3.16 ± 6.40
3.00 ± 0.26^{yx}	2.93 ± 0.31^{yx}	1.55 ± 0.06
29.18 ± 0.32^{xw}	32.27 ± 0.69^{yx}	27.73 ± 0.65
25.85 ± 0.80^{y}	25.19 ± 1.54^{yx}	32.08 ± 2.97
9.71 ± 0.59^{x}	10.64 ± 0.38^{x}	26.41 ± 2.49
3.19 ± 0.84^{x}	4.19 ± 0.45^{yx}	3.56 ± 1.07
34.15 ± 0.91^{x}	34.53 ± 2.02^{x}	36.17 ± 3.34
15.16 ± 0.16^{y}	12.65 ± 1.74^{x}	7.06 ± 1.37
20.98 ± 2.71^{y}	12.53 ± 0.33^{we}	29.06 ± 2.63
0.73 ± 0.10^{x}	1.01 ± 0.12^{y}	0.24 ± 0.04

using a variety of methods, including tissue analysis (D'Abramo and Sheen 1993). High levels of individual fatty acids in tissues relative to dietary levels may indicate relative importance, as can selective retention during nutritional stress (Reigh and Stickney 1989). Prawns in the unfed treatment had significantly higher levels ($P \leq 0.05$) of arachidonic acid (AA; 20:4n-6), eicosapentanoic acid (EPA; 20:5n-3), and docosahexanenoic acid (DHA; 22:6n-3) than prawns fed other foods (Table 6), indicating selective retention and possibly relative importance.

Prawns fed the prepared diet had significantly higher ($P \leq 0.05$) levels of linolenic acid (18:3n-3) than prawns in other treatments (Table 6), probably reflecting high levels in the diet (Table 4). Arachodonic acid (AA), EPA, and DHA were 489, 404, and 595% higher, respectively, in the whole body of prawns fed the prepared diet than in the diet itself, supporting data from unfed prawns indicating their relative importance. Prawns fed chironomids had significantly higher ($P \leq 0.05$) levels of palmitoleic acid (16:1n-7) than prawns fed other diets (Table 6), reflecting the high levels of 16:1n-7 in the chironomids being consumed (Table 4). Tissue levels of AA, EPA, and DHA in prawns fed chironomids exceeded levels in the chironomids themselves. The chironomids analyzed contained no DHA; however, prawn growth was not significantly reduced ($P > 0.05$). D'Abramo and Sheen (1993) demonstrated that AA (20:4n-6) could be equally effective as DHA (22:6n-3) when DHA was not provided. However, chironomids were also not rich sources of 20:4n-6, compared to other live foods. Although the chironomids contained no DHA (Table 4), prawns fed those chironomids as a sole food source contained 1.1% DHA (Table 6). Prawns cannot biosynthesize DHA from linolenic acid (D'Abramo and Sheen 1991), so it is likely that the DHA in prawns fed chironomids came from grazing bacteria in the tank. Mims et al. (1991) reported that *Daphnia* spp. grown in a bacterial medium contained significant levels of DHA (1.6%), suggesting bacteria could serve as a source of dietary DHA.

Prawns fed live zooplankton had significantly higher levels ($P \leq 0.05$) of linolenic acid (18:3n-3) than prawns fed other foods (Table 6), likely due to high 18:3n-3 levels in the zooplankton (Table 4). Prawns fed zooplankton had tissue levels of EPA and DHA that were 49 and 178% greater, respectively, than levels in the zooplankton. However, unlike prawns fed the prepared diet or chironomids, tissue levels of arachodonic acid were not elevated above dietary levels. This may indicate that zooplankton contain sufficient dietary levels of 20:4n-6 for prawns. This was also true in prawns fed oligochaetes which had tissue levels of arachodonic acid that were not significantly different ($P > 0.05$) from prawns fed zooplankton, but were significantly greater ($P \geq 0.05$) than in prawns fed chironomids or the prepared diet. In prawns fed oligochaetes, tissue levels of EPA and DHA

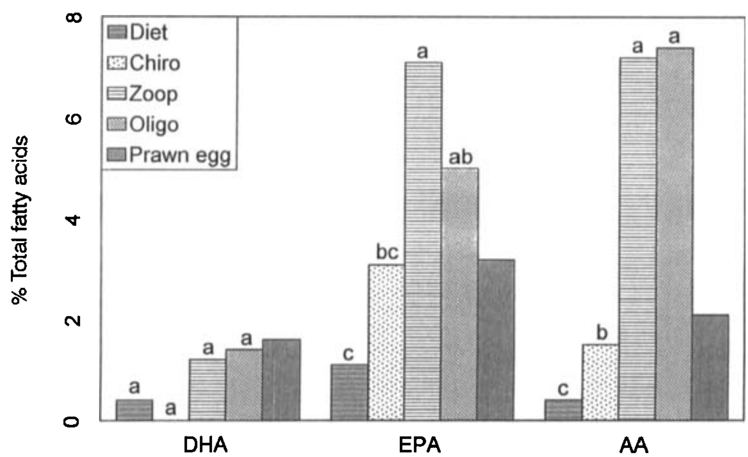

FIGURE 3. *Concentrations of docosahexanenoic acid (DHA; 22:6n-3), eicosapentanoic acid (EPA; 20:5n-3), and arachidonic acid (AA; 20:4n-6) in a prepared diet and natural foods fed to prawns in aquaria. The amount in prawn eggs is from Tidwell et al. (1993).*

were 74 and 113% above dietary concentrations.

These data would support the essentiality of AA, EPA, and DHA for prawns. If levels deposited in eggs are good indicators of nutritional importance, these data would indicate that the prepared diet has levels lower than egg concentrations for all three of these fatty acids (Fig. 3). Chironomids had lower levels of AA and EPA than prawn eggs, and were in this case devoid of DHA. Zooplankton and oligochaetes appeared to have the most desirable fatty acid profiles with AA and EPA levels being higher than prawn eggs and DHA concentrations being only slightly lower. D'Abramo and Sheen (1993) reported that diets containing either 22:6n-3 or 20:4n-6 HUFA's at levels ranging from 0.075% to 0.60% of the diet were equally effective and stated that investigations to determine minimum dietary requirements for 20:4n-6, 20:5n-3, and 22:6n-3 were necessary. These data appear to indicate that prawns fed zooplankton and oligochaetes had satisfied their dietary requirements for AA which was present in those foods at 0.035% of the diet. However, requirements for EPA and DHA did not appear to be met when present at 0.044% and 0.012% of the diet, respectively.

D'Abramo and Sheen (1993) suggested that a combined percentage of at least 15% $C > 20$ PUFA's of the n-6 and n-3 families in whole body tissues of an animal could serve as an indicator that optimal growth requirements for essential fatty acids had been met. The prawns fed the prepared diet had a combined level of these fatty acids of 10.2%, while those fed chironomids had 13.1%. Prawns fed the combination diet, oligochaetes, or zooplankton had levels of 18.4, 19.6, and 21.0%, respectively.

For oligochaetes and chironomids, consumption was measured daily and recorded as a percentage of body weight. (Consumption of feed and zooplankton could not be determined due to difficulty in recovering uneaten portions). Prawns fed oligochaetes consumed an average of 3.70% of their bodyweight daily (dry weight basis), while those fed chironomids consumed 3.84%. Based on proximate analysis of food organisms (Table 1) this equates to protein intakes of 0.24 and 0.22 g/d. Using physiological fuel values of 9.44, 5.64, and 4.11 kcal/g for lipid, protein, and nitrogen free extract, respectively (NRC 1993), the prawns fed oligochaetes consumed 1,542 kcal/d and those fed chironomids 1,430 kcal/d. Protein/energy ratios (P/E) for prawns consuming oligochaetes (155.6 mg/kcal) and chironomids (153.8 mg/kg) agree well with the findings of Summerlin (1988) who reported optimal P/E ratios for prawns to be between 130–158 mg/kcal. Bautista (1986) reported that growth rates of *Penaeus monodon* were highest when fed diets containing a P/E ratio of 120 to 174 mg/kcal. Feed conversion ratios (dry weight diet per unit wet-weight gain) (Hardy 1989) for prawns fed oligochaetes or chironomids were 1.55 and 1.61, respectively.

Over 80% of crustacean aquaculture is conducted under semi-intensive pond-based farming systems in which their nutrients are supplied by a combination of artificial diets and live food organisms, produced endogenously within the pond ecosystem (Tacon 1995). To truly understand prawn nutrition, reduce production costs, and maximize the economic benefit of extensive or semi-intensive farm systems, it is as important to identify and quantify the nutritional contribution of natural foods as it is to develop complete feeds. Tidwell et al. (in press) found that in ponds natural foods contribute as much as 34% of the prawn's nutrition, even when high quality diets are fed.

The data presented here indicate that formulated diets may not closely match the nutritional requirements of prawns, possibly being low in arginine, methionine, and lysine. Higher levels of AA, EPA, and DHA in the prepared diet could possibly be advantageous. All natural foods supported higher growth rates than the prepared diet. Oligochaetes and zooplankton appeared to match the biochemical composition of the prawns, and presumably their nutritional requirements, most closely. Enhancement of the numbers of, and access to, these organisms in pond systems could prove advantageous to the economic production of prawns by decreasing feed, and thereby, production costs. To achieve high production rates (> 500–1,000 kg/ha) the supply of natural food organisms is normally not sufficient, even in fertilized systems. This makes the feeding of prepared diets essential, especially during the later stages of the production cycle when biomass densities are high. Future studies should evaluate diet formulations more closely matching amino acid profiles of prawns and amino acid and fatty acid profiles of preferred prey species.

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