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Effects of feeding practical diets containing various protein levels on growth, survival, body composition, and processing traits of Australian red claw crayfish (*Cherax quadricarinatus*) and on pond water quality

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Abstract

A 117-day feeding trial was conducted in ponds with juvenile Australian red claw crayfish (*Cherax quadricarinatus*) to evaluate the effects on growth, survival, body composition, and processing traits when fed diets containing three different protein levels (22%, 32%, and 42%), and the effects of feeding these diets on pond water quality. Juvenile crayfish (mean weight of 4.6 ± 2.2 g) were randomly stocked into nine 0.02-ha ponds at a rate of 500 per pond (25 000 ha^{-1}), and each diet was fed to three ponds. There were two feedings per day, each consisting of one-half of the total daily ration. At harvest, there were no significant differences ($P > 0.05$) in the individual weight, percentage weight gain, or specific growth rate among treatments, which averaged 75.3 g, 1535%, and 2.38% day^{-1} respectively. Red claw fed the 42% crude protein diet had significantly higher ($P < 0.05$) feed conversion ratio (7.34) compared with crayfish fed diets containing 22% (5.18) or 32% (5.13) crude protein, and had significantly lower percentage survival (46.1%) compared with red claw fed diets with 22% (61.1%) or 32% (58.2%) protein. Total yield was significantly lower ($P < 0.05$) in red claw fed the 42% protein diet (640 kg ha^{-1}) compared with red claw fed diets containing 22% (920 kg ha^{-1}) or 32% (904 kg ha^{-1}) protein. Mean total ammonia nitrogen (TAN) levels were significantly higher ($P < 0.05$) in ponds with red claw fed the 42% protein diet (0.55 mg L^{-1}) compared with ponds with red claw fed diets containing 22% (0.32 mg L^{-1}) or 32% (0.38 mg L^{-1}) protein. Mean total nitrite concentrations in ponds with red claw fed the 42% protein diet was significantly higher (0.05 mg L^{-1}) compared with red claw fed diets containing 22% (0.01 mg L^{-1}) or 32% (0.02 mg L^{-1}) protein. These results indicate that a practical diet containing 22% (as fed basis) protein may be adequate for pond production of red claw when stocked at the density used in this study, and that a diet containing 42% protein adversely affected levels of TAN and nitrite, possibly reducing overall survival of red claw. Use of a diet with 22% protein may allow red claw producers to reduce diet costs and thereby increase profits.

Keywords: red claw, *Cherax quadricarinatus*, protein, growth, survival, diets, water quality

Introduction

The Australian red claw crayfish, *Cherax quadricarinatus*, is attracting interest in the United States. Although still a relatively new aquaculture species, red claw are gaining attention due to their potential large size (> 600 g), rapid growth rates (90–120 g in 6 months), potential to be marketed as small freshwater lobsters, tolerance of a wide range of water temperatures (15–32 °C) and low dissolved oxygen concentrations (as low as 1 mg L^{-1}), lack of complex larval stages with no saline water needed, tolerance to high culture densities, and their ability to readily accept prepared diets (Masser & Rouse 1993; Webster, Goodgame-Tiu, Tidwell & Rouse 1994).

Protein is one of the most expensive ingredients in prepared aquaculture diets. Lim (1997) stated that protein is the most critical ingredient in shrimp diets from the standpoint of cost and growth response;

however, Kureshy & Davis (2002) stated that excess dietary protein can reduce water quality associated with nitrogen excretion.

In an effort to reduce diet costs, nutritionists have determined minimal protein levels for several species of freshwater crustaceans. Summerlin (1988) reported that growth of juvenile freshwater prawn, *Machrobrachium rosenbergii*, fed a diet containing 35% protein was significantly higher than that of prawns fed lower (25% and 30%) protein levels. Tidwell, Webster, Clark & D'Abramo (1993) found that freshwater prawn stocked in ponds at a density of 19 760 ha^{-1} could be fed a 29% protein diet. Jones, Silva & Mitchell (1996) evaluated two other Australian species (*Cherax albidus* and *Cherax destructor*), both commonly known as yabbies, and reported that mean weight, percentage weight gain, and specific growth rate were substantially higher for crayfish fed a diet containing 30% versus 15% protein. It has been reported that a diet containing 30% protein was suitable for red swamp crayfish, *Procambarus clarkii* (Hubbard, Robinson, Brown & Daniels 1986; Lochmann, McClain & Gatlin III 1992). Other studies found that the dietary protein requirement for red swamp crayfish grown in a recirculating system was between 25% and 35% (Tarshis 1979; Huner & Barr 1984; Wiernicki 1984). In one of the first studies to evaluate prepared diets for red claw, Webster *et al.* (1994) found that a practical diet containing 33% crude protein appeared suitable for good growth and survival of small (0.02 g) juvenile red claw when grown communally in an indoor recirculating system. Manomaitis (2001) reported that the recommended crude protein level in the diets for 0.02 g juvenile red claw was 40%, while larger (3.03 g) juveniles could be fed a diet with 30% protein.

Presently, little information exists on the optimum dietary protein level for red claw when grown in ponds. The purpose of the study was to determine the minimum dietary protein percentage for pond-cultured red claw in an effort to reduce production costs.

Materials and methods

Description and stocking of ponds

Juvenile red claw were obtained from a commercial supplier (Central Queensland Crayfish, Queensland, Australia). Upon arrival to the Aquaculture Research Center, the styrofoam shipping containers were opened and the red claw inspected. Red claw were stocked into nine 0.02-ha ponds, average water depth approximately 1.1 m, located at the Aquaculture Research Center, Kentucky State University, Frankfort, KY, USA on 1 June 2002 at a rate of 500 red claw per pond (25 000 ha^{-1}). Groups of 100 live red claw were hand counted into nine 0.02-ha ponds at random until all ponds had been stocked with 500 red claw. This procedure ensured that each pond was stocked with red claw that were of similar shipping/transport times. Mean stocking weight ($\pm$ SD) was 4.6 $\pm$ 2.2 g. On the second day after stocking, dead red claw seen in the pond were removed, counted, and replaced with live red claw of similar weight. No other replacements were made after this time. Three replicate ponds were randomly assigned to each of the three diets containing various levels (22%, 32%, and 42%) of protein.

Experimental diets and feeding rates

Experimental diets were formulated to contain 22%, 32%, and 42% protein (as fed). Dietary ingredients were processed into 5-mm sinking pellets by a commercial feed mill (Farmers Feed, Lexington, KY, USA; Table 1). Two feedings, each consisting of one-half of the total daily ration, were distributed over the entire surface of each pond between 08:00 and 08:30, and between 15:30 and 16:00 hours for 117 days. Red claw were fed a percentage of body weight (pond biomass) based upon a feeding schedule devised by C. D. Webster (unpubl. data). Survival was assumed to be 100% and feeding rates were adjusted every 2 weeks. Red claw were fed 10% of estimated body weight during the first 14 days of the study, followed by 4% until harvest with an assumed harvest weight of 100 g.

Diet analysis

Diets were analysed for proximate composition (Table 1) as follows: Moisture was determined by placing a 2-g sample into a convection oven (135 °C) for 2 h until constant weight (AOAC procedure 930.15); protein was determined by combustion (AOAC procedure 990.03); lipid was determined by the acid hydrolysis method (AOAC procedure 954.02); fiber was determined by using the fritted glass crucible method (AOAC procedure 962.09); and ash was determined by placing a 2-g sample in a muffle furnace to (600 °C) for 2 h (AOAC 942.05) (AOAC 1990). Carbohydrate (NFE) was determined by difference. Available energy (AE) was calculated from physiological

Table 1 Ingredient and chemical composition (%) of three practical diets containing different levels (22%, 32%, and 42%) of protein fed to red claw crayfish

	Diets (% crude protein)		
Ingredient	1 (22%)	2 (32%)	3 (42 %)
Menhaden fish meal (67%)	6.9	10.0	13.2
Soybean meal (50%)	11.0	34.0	56.0
BGY (35%)*	5.0	5.0	5.0
Wheat flour (14%)	59.2	33.6	8.9
Menhaden oil	5.0	4.5	4.0
Corn oil	2.0	2.0	2.0
Vitamin mix†	3.0	3.0	3.0
Mineral mix‡	1.0	1.0	1.0
Wheat gluten (41%)	5.0	5.0	5.0
Other§	1.9	1.9	1.9
Moisture (%)	12.5	11.8	11.7
Protein (%)¶	25.9	37.5	48.3
Lipid (%)¶	13.0	10.8	10.6
Fiber (%)¶	7.5	6.0	4.6
Ash (%)¶	7.3	7.9	8.6
NFE‖	46.3	37.7	28.0
Available energy**	4.1	4.0	4.0

Values in parentheses are percentage protein of ingredient.
*Brewer's grains with yeast (F.L. Emmert Company, Cincinnati, OH, USA).
†Vitamin mix contained: thiamin (B_1), 1.01%; riboflavin (B_2), 1.32%; pyridoxine (B_6), 0.9%; nicotinic acid, 8.82%; folic acid, 0.22%; cyanocobalamine (B_{12}), 0.001%; pantothenic acid, 3.53%; menadione (K), 0.2%; ascorbic acid (C), 33.1%; retinol palmitate (A), 4409 IU kg^{-1}; cholecalciferol (D_3), 2 204 600 IU kg^{-1}; α tocopherol (E), 66.2 IU kg^{-1}; ethoxyquin, 0.66%.
‡Mineral mix contained: Mn, 10.0% (as $MnSO_4$); Zn, 10.0% (as $ZnSO_4$); Fe, 7.0% (as $FeSO_4$); Cu, 0.7% (as $CuSO_4$); I, 0.24% (as $CaIO_3$); Co, 0.10% (as $CoSO_4$); Ca, as carrier.
§Other ingredients include: choline chloride, 0.5%; vitamin C, 0.1%; dicalcium phosphate, 1.3%.
¶Dry-matter basis.
‖NFE, nitrogen-free extract.
**Available energy was calculated as 4.0, 4.0, and 9.0 kcal g^{-1} of protein, carbohydrate, and lipid respectively.

Table 2 Amino acid composition of three practical diets containing various percentages (22, 32, and 42) of protein fed to red claw crayfish

	Diets (% crude protein)		
Amino acid	1 (22%)	2 (32%)	3 (42%)
Alanine	1.2 (4.5)	1.6 (4.4)	2.0 (4.1)
Arginine	1.2 (4.7)	1.9 (5.1)	2.6 (5.4)
Aspartic acid	1.7 (6.5)	3.0 (8.1)	3.9 (8.0)
Cystine	0.3 (1.2)	0.4 (1.1)	0.6 (1.1)
Glutamic acid	3.5 (13.7)	5.3 (14.1)	7.0 (14.5)
Glycine	1.1 (4.1)	1.5 (4.0)	1.9 (3.9)
Histidine	0.5 (2.1)	0.8 (2.1)	1.0 (2.1)
Isoleucine	0.8 (2.9)	1.3 (3.4)	1.7 (3.6)
Leucine	1.6 (6.3)	2.4 (6.5)	3.2 (6.7)
Lysine	0.9 (3.5)	1.6 (4.4)	2.3 (4.8)
Methionine	0.4 (1.4)	0.5 (1.4)	0.7 (1.5)
Phenylalanine	0.9 (3.3)	1.4 (3.8)	1.9 (4.0)
Proline	1.3 (4.9)	1.7 (4.6)	2.1 (4.3)
Serine	0.9 (3.5)	1.4 (3.8)	1.8 (3.8)
Threonine	0.8 (2.9)	1.2 (3.2)	1.5 (3.2)
Tyrosine	0.6 (2.4)	1.0 (2.6)	1.3 (2.8)
Valine	1.0 (3.9)	1.5 (4.0)	1.9 (3.9)

Values are percentage of the diet. Values in parentheses are expressed as percentage of dietary protein. Values are means of two replicates per diet.

fuel values of 4.0, 4.0, and 9.0 kcal g^{-1} for protein, carbohydrate (NFE), and lipid respectively (Garling & Wilson 1977; Webster, Tiu & Morgan 1999). Diets were also analysed for amino acid composition (Table 2) by a commercial analytical laboratory (Woodson-Tenent Labs, Des Moines, IA, USA).

Water quality management

Water temperature and dissolved oxygen (DO) were measured in all ponds twice daily (09:00 and 15:30 hours) using a YSI Model 57 oxygen meter (Yellow Springs Instruments, Yellow Springs, OH, USA). A 1/2-HP electric aerator (Air-O-Lator, Kansas City, MO, USA) was positioned in the centre of each pond and run continuously throughout the duration of the study. The pH was measured daily (15:30 hours) using an electronic pH probe (pH pen; Fisher Scientific, Cincinnati, OH, USA). All other water quality parameters were measured three times weekly (13:00 hours), and included total ammonia nitrogen (TAN), nitrite, and alkalinity. TAN and nitrite levels were determined according to the procedures for a Hach DREL/2000 spectrophotometer (Hach Company, Loveland, CO, USA). Alkalinity was measured according to the titration method (Hach Company, Loveland, CO, USA).

Harvest

Ponds were harvested between 23 and 28 September 2002. Three days prior to harvest, the water level in each pond was lowered to approximately 0.5 m at the drain end. On the day of harvest, the standpipe in each pond was lowered so that all water was completely drained and red claw were manually harvested. Total weight and number of red claw (by sex) from each pond were recorded at the harvest.

Growth parameters and feed efficiency were calculated as follows:

SGR (specific growth rate) (% day^{-1}) = $[(\ln W_t - \ln W_i)/T] \times 100$, where W_t and W_i are the final and initial individual weights of the red claw, respectively, and T is the length of the culture period in days;

Weight gain (%) = $100[(W_t - W_i)/W_i]$;

Feed conversion ratio (FCR) = total diet fed (kg)/total wet weight gain (kg).

Ten males and 10 females from each pond were randomly sampled, chill-killed using an ice-water bath, and the tail muscle meat removed from the shell and analysed for percentage moisture, protein, lipid, and ash (Table 6). Samples were stored in polyethylene bags, and frozen (-15 °C) until analysis. Proximate analysis procedures were as described for the diets except for moisture (AOAC procedure 950.46) and lipid was determined by ether extraction (AOAC procedure 960.39) (AOAC 1990).

Processing traits were measured from 20 males and 20 females that were randomly chosen from each pond, chill-killed using an ice water bath, and measured for total weight, claw weight, tail weight, tail muscle weight, and cephalothorax weight (wet-weight basis) (Table 7).

Statistical analysis

Data was calculated for final individual weight (g), percentage weight gain, SGR, FCR, percentage survival, total yield (kg ha^{-1}), proximate composition, and processing data. Pond average values were used as units of observation for statistical analysis. Data were analysed by ANOVA and Duncan's multiple range test was used to compare treatment means using SAS software version 8.0 (SAS 1999). All percentage and ratio data were arcsin transformed prior to statistical analysis (Zar 1984). All statistical computations were performed at the $P \leq 0.05$ probability level.

Results

Water quality

All ponds were continuously aerated throughout the duration of the study and averaged 7.51 mg L^{-1} O_2 for the morning and 9.38 mg L^{-1} O_2 for the afternoon (Table 3). There were no significant differences in morning and afternoon water temperature, or in pH, which averaged 24.8 °C, 26.9 °C, and 8.7 respectively (Table 3). TAN was significantly higher in ponds with red claw fed the 42% protein diet (0.55 mg L^{-1}) compared with ponds with red claw fed diets containing 22% (0.32 mg L^{-1}) or 32% (0.38 mg L^{-1}) protein (Table 3). Mean total nitrite concentrations in ponds with red claw fed a diet containing 42% protein was significantly higher (0.05 mg L^{-1}) than in ponds with red claw fed diets containing 22% (0.01 mg L^{-1}) and 32% (0.02 mg L^{-1}) protein. Moreover, water in ponds with red claw fed diets containing 42% protein had the highest TAN (by month) compared with red claw fed all other treatments (diet), and total NH_3-N levels progressively increased up to 2.33 mg L^{-1} and averaged 1.11 mg L^{-1} in September (Fig. 1). Likewise, higher nitrite levels were found in ponds with red claw fed a diet containing 42% protein, with levels peaking at 0.14 mg L^{-1} in the final month of the growing season (Fig. 2). Alkalinity was significantly higher in ponds with red claw fed a diet with 22% protein (110 mg L^{-1}) compared with ponds with red

Table 3 Means ($\pm$ SE) of water quality parameters measured in ponds of red claw crayfish fed three practical diets containing various percentages (22, 32, and 42) of protein

	Diets (% crude protein)		
	1 (22%)	2 (32%)	3 (42%)
Dissolved oxygen (mg L^{-1}; am)	7.58 ± 0.07^a	7.60 ± 0.06^a	7.36 ± 0.07^b
Dissolved oxygen (mg L^{-1}; pm)	9.21 ± 0.07^b	9.48 ± 0.08^a	9.45 ± 0.09^a
pH	8.7 ± 0.03^a	8.7 ± 0.04^a	8.7 ± 0.04^a
Temperature (°C; am)	24.7 ± 0.18^a	24.6 ± 0.19^a	25.0 ± 0.19^a
Temperature (°C; pm)	26.8 ± 0.22^a	26.7 ± 0.22^a	27.1 ± 0.22^a
TAN (mg L^{-1})	0.32 ± 0.03^b	0.38 ± 0.03^b	0.55 ± 0.05^a
Nitrite (mg L^{-1})	0.01 ± 0.0^b	0.02 ± 0.0^b	0.05 ± 0.01^a
Alkalinity (mg L^{-1})	110 ± 1.7^a	102 ± 1.8^b	103 ± 1.4^b

Means within a row having different superscripts are significantly different ($P < 0.05$).
TAN, total ammonia nitrogen.

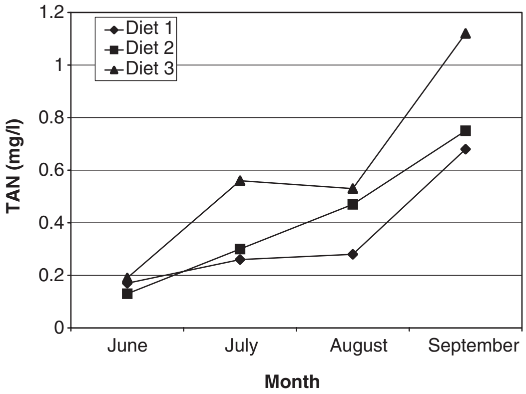

Figure 1 Changes in TAN (mg L^{-1}) in ponds (by month) over the course of the 117-day feeding trial.

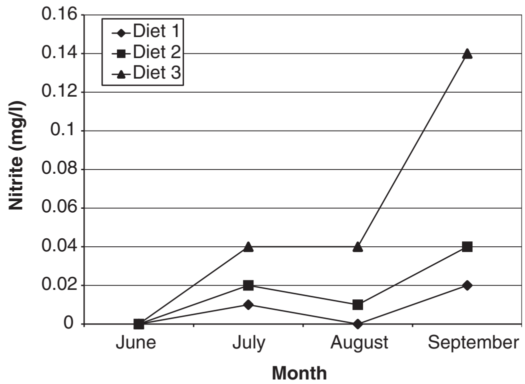

Figure 2 Changes in nitrite (mg L^{-1}) in ponds (by month) over the course of the 117-day feeding trial.

claw fed diets containing 32% (102 mg L^{-1}) and 42% (103 mg L^{-1}) protein.

Growth and production

At harvest, there were no significant differences in the mean final weight, percentage weight gain, and specific growth rate among treatments, which averaged 75.3 g, 1535%, and 2.38% day^{-1} respectively (Table 4). Red claw fed a diet containing 42% protein had significantly higher FCR (7.34) compared with red claw fed diets containing 22% (5.18) and 32% (5.13) protein, and had significantly lower percentage survival (46.1%) compared with red claw fed diets with 22% (61.1%) and 32% (58.2%) protein. Total yield was significantly lower in red claw fed a diet containing 42% protein (640 kg ha^{-1}) compared with red claw fed diets containing 22% (920 kg ha^{-1}) or 32% (904 kg ha^{-1}) protein.

Table 4 Means (± SE) of final weight, percentage weight gain, specific growth rate (SGR), feed conversion ration (FCR), percentage survival, and yield of red claw crayfish fed three practical diets containing various percentages (22, 32, and 42) of protein

	Diets (% crude protein)		
	1 (22%)	2 (32%)	3 (42%)
Final weight (g)	79.83 ± 5.02[a]	75.77 ± 5.57[a]	70.47 ± 1.46[a]
Weight gain (%)	1632 ± 109[a]	1544 ± 121[a]	1428 ± 32[a]
SGR (% day^{-1})	2.43 ± 0.05[a]	2.39 ± 0.06[a]	2.33 ± 0.02[a]
FCR	5.18 ± 0.65[b]	5.13 ± 0.12[b]	7.34 ± 0.65[a]
Survival (%)	61.1 ± 3.8[a]	58.2 ± 3.2[a]	46.1 ± 3.1[b]
Yield (kg ha^{-1})	920 ± 106[a]	904 ± 20[a]	640 ± 52[b]

Means within a row having different superscripts are significantly different ($P < 0.05$).

Percentage of males and females at harvest

There were no significant differences in the percentages of males and females harvested from ponds among treatments (Table 5). The percentage of males averaged 48.5% and the percentage of females averaged 51.5%. There were no significant differences in the final mean weight of males (87.9 g) fed any treatment (diet). Likewise, there were no significant differences in the final mean weight of females (64.2 g) fed any diet.

Body composition and processing traits

Proximate compositions of male and female tail muscle are presented in (Table 6). There were no significant differences in percentage moisture, protein, lipid, and ash (wet-weight basis) in the tail muscle of males among treatments (diet), which averaged 79.0%, 17.2%, 0.2%, and 1.5% respectively. There were no significant differences in percentage moisture, protein, and lipid (wet-weight basis) in the tail muscle of females among treatments, which averaged 78.1, 18.1, and 0.2 respectively. However, female red claw fed the diet containing 32% protein had significantly higher percentage ash (1.6%) in the tail muscle compared with red claw fed diets containing 22% and 42% protein.

When analysed within the same treatment (diet), males fed diets containing 22% and 42% protein had a significantly higher percentage moisture (79.1% and 78.9%, respectively) compared with females. Females fed a diet containing 42% protein had significantly higher percentage lipid (0.2%) in the tail muscle compared with males fed the same

Table 5 Mean (± SE) harvest percentage and final individual weight of male and female red claw harvested from ponds fed three different practical diets containing various percentages (22, 32, and 42) of protein for 117 days

	Male		Female	
Treatment (%)	Harvest (%)	Final weight (g)	Harvest (%)	Final weight (g)
22	47.0 ± 2.82^{a}	95.0 ± 5.43^{a}	53.0 ± 2.82^{a}	66.4 ± 5.83^{a}
32	49.6 ± 1.55^{a}	86.2 ± 7.77^{a}	50.4 ± 1.55^{a}	65.3 ± 2.77^{a}
42	49.0 ± 1.97^{a}	80.3 ± 1.61^{a}	51.0 ± 1.97^{a}	61.0 ± 3.09^{a}

Values in the same column with the same superscript are not significantly different ($P > 0.05$).

Table 6 Means (± SE) percentage moisture, protein, fat, and ash (wet-weight basis) of tail muscle of male and female red claw crayfish fed three practical diets containing various percentages (22, 32, and 42) of protein

	Diets (% crude protein)		
	1 (22%)	2 (32%)	3 (42%)
Males			
Moisture (%)	79.1 ± 0.4^{a,x}	79.0 ± 0.3^{a,x}	78.9 ± 0.1^{a,x}
Protein (%)	17.0 ± 0.6^{a,x}	17.2 ± 0.1^{a,x}	17.4 ± 1.3^{a,x}
Fat (%)	0.2 ± 0.0^{a,x}	0.2 ± 0.1^{a,x}	0.1 ± 0.0^{a,y}
Ash (%)	1.6 ± 0.0^{a,x}	1.5 ± 0.0^{a,x}	1.5 ± 0.1^{a,x}
Females			
Moisture (%)	77.9 ± 0.1^{a,y}	78.2 ± 0.4^{a,x}	78.1 ± 0.2^{a,y}
Protein (%)	17.5 ± 1.0^{a,x}	18.3 ± 0.8^{a,x}	18.4 ± 0.7^{a,x}
Fat (%)	0.2 ± 0.0^{a,x}	0.2 ± 0.0^{a,x}	0.2 ± 0.0^{a,x}
Ash (%)	1.5 ± 0.0^{b,x}	1.6 ± 0.1^{a,x}	1.4 ± 0.0^{b,x}

Means within a row having different superscripts (a, b) are significantly different ($P < 0.05$) among dietary treatments. Means between males and females in the same column for each respective variable with different superscripts (x, y) are significantly different ($P < 0.05$).

diet. All other variables between males and females within each treatment (diet) were not significantly different.

Processing traits of male and female red claw are presented in (Table 7). Male red claw fed a diet containing 22% protein had significantly higher total weight (101.2 g), claw weight (29.2 g), and weight of tails with shell (32.0 g) compared with male red claw fed all other diets. Male red claw fed a diet containing 22% protein had significantly higher weight of tail muscle meat (21.4 g) compared with males fed the 42% protein diet, but not different from males fed a diet containing 32% protein. Male red claw fed a diet containing 22% protein had a significantly higher cephalothorax weight (40.2 g) compared with males fed a diet containing 32% protein, but not different from males fed a diet containing 42% protein.

Female red claw fed a diet containing 22% protein had significantly higher total weight (67.6 g) and weight of tails with shell (25.4) compared with females fed a diet containing 42% protein, but not different from females fed a diet containing 32% protein. Females fed diets containing 22% and 32% protein had significantly higher claw weight (12.0 and 11.9 g, respectively) and weight of tail muscle meat (14.3 and 14.0 g respectively) compared with females fed a diet containing 42% protein. There were no significant differences in the cephalothorax weight among treatments. When analysed within the same diet, males had significantly higher total weight, higher claw weight, tail weight (with shell on), tail muscle meat weight, and cephalothorax weight compared with females fed the same diet.

Discussion

Results of the present study indicate that dietary protein level can be reduced to 22% for red claw grown in ponds at the size and stocking densities used here, thus reducing diet costs. Red claw fed the 22% protein diet had similar growth and survival compared with red claw fed diets containing 32% and 42% protein. Webster *et al.* (1994) reported that small (mean weight 0.022 g) juvenile red claw required a 33% crude protein diet for rapid growth and survival when grown communally in an indoor recirculating aquarium system. Manomaitis (2001) evaluated the crude protein requirements of juvenile red claw crayfish utilizing two age classes and reported that small (0.02 g) red claw required a 40%-protein diet, whereas larger (3.03 g) red claw could be fed a diet containing 30% protein when grown indoors. Red claw stocked in the present study were larger (4.6 g) and stocked into ponds compared with studies by Webster *et al.* (1994) and Manomaitis (2001) which may explain why the present study indicates that a lower protein level may be sufficient.

In the present study, natural food items in the pond may have satisfied part of the protein requirements of red claw, which are omnivorous detritivores.

Table 7 Means (± SE) total weight, claws, tail, tail muscle, and cephalothorax (wet-weight basis) of male and female red claw crayfish fed three practical diets containing various percentages (22, 32, and 42) of protein

	Diets (% crude protein)		
	1 (22%)	2 (32%)	3 (42%)
Males			
Total weight (g)	$101.2 \pm 4.3^{a,x}$	$87.8 \pm 3.9^{b,x}$	$85.4 \pm 4.1^{b,x}$
Claws (g)	$29.2 \pm 2.2^{a,x}$	$23.3 \pm 2.1^{b,x}$	$21.3 \pm 1.5^{b,x}$
Tail (g)	$32.0 \pm 1.0^{a,x}$	$28.6 \pm 0.9^{b,x}$	$26.5 \pm 1.2^{b,x}$
Tail muscle (g)	$21.4 \pm 0.8^{a,x}$	$19.2 \pm 0.7^{ab,x}$	$17.1 \pm 1.0^{b,x}$
Cephalothorax (g)	$40.2 \pm 1.4^{a,x}$	$35.7 \pm 1.3^{b,x}$	$37.6 \pm 1.6^{ab,x}$
Females			
Total weight (g)	$67.6 \pm 2.0^{a,y}$	$65.7 \pm 2.1^{ab,y}$	$61.3 \pm 2.0^{b,y}$
Claw (g)	$12.0 \pm 0.5^{a,y}$	$11.9 \pm 0.5^{a,y}$	$9.8 \pm 0.5^{b,y}$
Tail (g)	$25.4 \pm 0.8^{a,y}$	$24.0 \pm 0.7^{ab,y}$	$22.2 \pm 0.7^{b,y}$
Tail muscle (g)	$14.3 \pm 0.7^{a,y}$	$14.0 \pm 0.6^{a,y}$	$12.0 \pm 0.6^{b,y}$
Cephalothorax (g)	$30.2 \pm 0.9^{a,y}$	$29.5 \pm 1.0^{a,y}$	$29.1 \pm 1.0^{a,y}$

Means within a row having different superscripts (a, b) are significantly different ($P<0.05$) among dietary treatments. Means between males and females in the same column for each respective variable with different superscripts (x, y) are significantly different ($P<0.05$).

However, no measurement of pond organic matter was made during the present study. Thus, it is difficult to state the impact that natural foods contributed to the diet of red claw. At harvest, six of the nine ponds were devoid of aquatic vegetation and the pond bottoms were bare mud.

Average final weights in the present study were similar to or higher than those stated in other reports. Brummett & Alon (1994) reported an average final individual weight of 56.1 g when red claw were stocked (initial weight of 2.0 g) into earthen ponds at a rate of 25 000 ha^{-1} and grown for 170 days. Webster, Thompson, Muzinic, Yancey, Dasgupta, Xiong, Rouse & Manomaitis (in press) reported an average final weight of 64.7, 57.0, and 53.5 g for red claw (8.1 g) stocked at 12 000, 18 000, and 24 000 ha^{-1} when grown in 0.02-ha earthen ponds for 70 days. Karplus, Barki, Cohen & Hulata (1995) stated that red claw males (3 g initial weight) grew to an average weight of 34.5 g, while females were 31.9 g, after a 92-day grow-out period in ponds. Salame & Rouse (2000) reported an average size of 30 g after 90 days when red claw (1–2 g initial weight) were stocked at 40 000 ha^{-1} when fed diets containing 28–35% protein. Rouse & Kahn (1998) reported an average weight of 76 g when red claw (7 g initial weight at 20 000 ha^{-1}) were grown in monoculture for 135 days in ponds and fed a 32%-protein sinking fish diet with added supplemental forage (250 kg ha^{-1} dried alfalfa hay).

FCR values reported in the present study (5.13–7.34) were similar to previous reports for red claw. Webster *et al.* (in press) reported a range from 6.0 to 9.3 after 70 days of culture. Rouse & Kahn (1998) reported FCR values of 8.0 when red claw were fed a 32%-protein diet, while Jones & Ruscoe (2000) stated FCR values between 1.4 and 7.4 for different sizes of red claw stocked into pens and fed a prepared diet.

At harvest, red claw populations (all treatments) averaged 48.5% males and 51.5% females. There were very few (<1%) intersex animals and these were counted as males. Intersex red claw are those animals that exhibit both male and female secondary sexual characteristics (Medley & Rouse 1993). Similar sex ratios have been reported by other investigators (Karplus *et al.* 1995; Pinto & Rouse 1996; Webster *et al.* 2004).

Mean survival rates (average of 55%) and total yield (average of 821 kg ha^{-1}) of red claw in the present study were similar to or higher than other reports (Brummett & Alon 1994; Rouse & Kahn 1998; Salame & Rouse 2000; Webster *et al.* 2004).

Excess dietary protein reduces pond water quality due to an increase in nitrogenous wastes. Tomasso (1994) reported that ammonia can be toxic to crustaceans if allowed to accumulate in the water. During the course of the 117-day feeding trial, red claw fed a diet containing 42% protein averaged the highest TAN and nitrite levels (0.55 and 0.05 mg L^{-1} respectively). Masser & Rouse (1997) reported that the recommended range of total ammonia nitrogen for hatchery production is 0.5 mg L^{-1} or less for red claw. The increased levels of TAN and nitrite in ponds where red claw were fed a diet containing 42%

protein, especially in the last 4 weeks of the feeding trial, may have reduced survival. In the present study, all ponds were fed the same feeding rate daily. Thus, diet allowance was not a factor. However, since Diet 3 contained more protein than the other two diets, more protein nitrogen was fed to red claw. McIntosh, Samocha, Jones, Lawrence, Horowitz & Horowitz (2001) reported that ponds with Pacific white shrimp, *Litopenaeus vannamei*, fed diets with 21% or 31% protein, had no significant difference in water quality (total ammonia-N and nitrite-N). However, Samocha, Lawrence, Horowitz & Horowitz (1998a, b) reported a significant increase in ammonia and nitrite when *Penaeus setiferus* were fed a 45%-protein diet compared with a 20%-protein diet.

Data from the present study indicate that male red claw were larger than the females in all dietary groups when grown in a mixed-sex population. This is in agreement with other reports. Curtis & Jones (1995) found that males from a mixed-sex population attained greater mean weight than females of red claw grown for 10 months in earthen ponds. Sagi, Milstein, Eran, Joseph, Khalaila, Abdu, Harpaz & Karplus (1997) also demonstrated improved growth performance for males (mean weight 93.7 g for specimens > 50 g) compared with females (mean weight 66.9 g for specimens > 50 g) of red claw produced in mixed-stocking ponds.

Data from the present study indicates that males have a higher percentage of their body weight comprised of claw (chelae) weight compared with females. This is in agreement with Gu, Mather & Capra (1994), who reported that mature red claw males had wider chelae, more chelar muscle, longer chelipeds (claws), and increase in growth of these characters than mature female crayfish. However, in cambarid crayfish, allometric growth of the claws occurs only in breeding males; after breeding, the males moult to smaller claws (Romaire, Forester & Avault Jr 1977). Data from the present study indicate that this does not seem to be characteristic of male red claw crayfish. Ordinarily, the function of the claws (chelae) is to capture and manipulate prey and serve as a defence or safeguard against predators (Hartnoll 1974).

Further data from the present study indicates that males also have a higher percentage of their body weight comprised of tail (outer shell) and tail muscle (shell off) than female red claw. This is in agreement with Harlioglu & Holdich (2001) who reported that male freshwater crayfish, *Astacus leptodactylus*, had more tail meat in the winter months compared with females, however, the reverse was found in the summer. Red claw were grown during the summer growing season only in the present study.

In conclusion, it appears that red claw grown in a single season in temperate-climate ponds can be fed a diet containing 22% protein without adverse effects on growth, survival, or water quality. Using a 22% protein diet will reduce diet costs and thereby increase profits. Stocking all-male populations may also allow producers to grow larger individuals as it appears that males grow larger and more rapidly than females. Further research is needed to determine the minimum dietary protein level that can be used for feeding red claw in ponds, and on the optimum formulation for production diets.

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