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Population Growth and Production of *Apocyclops dengizicus* (Copepoda: Cyclopoida) Fed on Different Diets

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Abstract

This study was carried out to investigate the effects of various diets: 4 monoalgal diets: *Nannochloropsis oculata* (N), *Isochrysis galbana* (I), *Chaetoceros calcitrans* (C), and *Tetraselmis tetrathele* (T); 4 mixed algal diets: N+I+C+T(NICT), N+I+C(NIC), C+T(CT), and I+T(IT); and 2 nonalgal diets: baker's yeast (BY) and prepared shrimp feed (SF) on population growth and density of *Apocyclops dengizicus*. The type and density of diet had significant effects on the growth and density of *A. dengizicus* ($P < 0.01$). Of tested diets, T and CT were optimum diets due to higher density and growth rate of *A. dengizicus* compared to other diets. Their high dietary value was related to the higher contents of polyunsaturated fatty acid, particularly docosahexaenoic acid (22:6n-3), eicosapentaenoic acid (20:5n-3), and arachidonic acid (20:4n-6) compared to *A. dengizicus* cultured on other diets. The results of the present study illustrated that *T. tetrathele* was the most suitable food for the culture of *A. dengizicus*.

The most widely used live foods in aquaculture industry are the brine shrimp, *Artemia* sp., nauplii and the rotifer, *Brachionous plicatilis*. They are fed to penaeid shrimp and fish larvae (Sorgeloos 1980; Chu and Shing 1986; Loya-Javellana 1989). Some researchers (Chu and Shing 1986) reported problematic role of rotifer for feeding of mysis and postlarvae stages of penaeid shrimp because rotifers are small in size and their nutritional values are poor and variable. The abundance of *Apocyclops dengizicus* (Copepoda: Cyclopoida) in Malaysian coastal tropical waters and marine shrimp ponds (Farhadian 2006) makes it an important species as a live food item for shrimp postlarvae. *Apocyclops dengizicus* has six naupliar (85–240 μm size) and five copepodid (320–680 μm size) stages before become adult (Alvarez Valderhaug and Kewalramani 1979; Farhadian 2006). This wide spectrum of sizes enables this live food to become a suitable prey for carnivorous animals.

In addition, this species is easily cultured at a salinity range of 20 to 30 ppt and temperature of 25 to 35 C (Farhadian 2006). All mentioned stages of *A. dengizicus* definitely ingest the cultured marine microalgae of *Nannochloropsis oculata*, *Isochrysis galbana*, *Chaetocerous calcitrans*, and *Tetraselmis tetrathele* (Farhadian 2006).

The high nutritional values of *A. dengizicus* compared to newly hatched *Artemia* nauplii and improved survival and growth rates of *Penaeus mondon* postlarvae (PL1–PL15) indicated that this copepod has a good potential as a new live food for aquaculture marine shrimp industry (Farhadian et al. 2007). Some of the nutritional characteristics are as follow: protein = 55.3% dry weight, lipid = 20.40% dry weight, carbohydrate = 12.24% dry weight, docosahexaenoic acid (DHA) = 20.24%, eicosapentaenoic acid (EPA) = 8.76%, and arachidonic acid (ARA) = 1.46% (Farhadian 2006).

Types and concentrations of diets are two important aspects of *A. dengizicus* rearing experiments because they affect mostly on

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population growth, density, and population doubling time. The objectives of this study were to compare the population growth and density of *A. dengizicus* when fed separately on eight algal and two nonalgal diets.

Materials and Methods

Experimental Design

Two separate experiments were performed to test the effects of monoalgal diets, mixed algal diets, and nonalgal diets on population growth rates and density of *A. dengizicus*. In the first experiment, the *A. dengizicus* were fed with monoalgal diets including *N. oculata* (N), *I. galbana* (I), *C. calcitrans* (C), and *T. tetrathele* (T) and mixed algal diets N +I+C+T (NICT), N +I+C (NIC), C+T (CT), and I +T (IT) in the same ratio at low (2.5×10^4 cells/mL) and high (5×10^6 cells/mL) algal densities. This experiment was designed as factorial design of 8 (diets) × 2 (algal densities). The second experiment was designed as factorial of two diets: baker's yeast (BY) and prepared shrimp feed (SF) each at 3 levels (3, 7, and 10 mg/d) with three replicates.

Copepod Stock

Apocyclops dengizicus was collected and isolated from a shrimp pond in Kuala Selangor (3°17′N, 101°17′E), and the culture was maintained at the Aquatic Research Laboratory at Universiti Putra Malaysia (UPM). They were fed with mixed microalgae including *N. oculata*, *I. galbana*, *C. calcitrans*, and *T. tetrathele* (1:1:1:1 in ratio by number) at least 2 wk before the experiments. During stock maintenance, each culture was examined daily, all exuvia and any dead individuals were removed, and up to 30% of water was also changed daily.

Microalgae Culture

Four algal species: *N. oculata*, *I. galbana*, *C. calcitrans*, and *T. tetrathele*, were grown in Conway medium (Tompkins et al. 1995), at 29 ± 1 C, salinity 30 ppt, 12 : 12 h light : dark cycle, and 40 μmol photons/m²/s light intensity in 10-L carboys separately with mild, continuous aeration for 10 d. The phytoplankton concentration in each beaker was determined using an improved Neubauer hemocytometer (0.25 $mm^2 \times 0.1$ mm) under a phase contrast microscope (Nikon/Eclipse 600, Nikon Corp. Ltd., Kawasaki, Kanagawa, Japan) according to Martinez and Chakroff (1975) after the samples were fixed in Lugol's iodine solution (0.1 mL for 3-mL sample).

Dry weights of microalgal cells were determined by filtering and drying algae from aliquots of culture of known concentration according to the method described by Lavens and Sorgeloos (1996). They include: *N. oculata* (2–4 μm, 0.500×10^{-5} μg dry wt/cell), *I. galbana* (4–7 μm, 7.097×10^{-5} μg dry wt/cell), *C. calcitrans* (6–9 μm, 1.313×10^{-4} μg dry wt/cell), and *T. tetrathele* (10–16 μm, 1.566×10^{-4} μg dry wt/cell) (Farhadian 2006). Harvesting was done when the microalgal growth reached the stationary phase using a laboratory centrifuge Sigma 4-15 (Montreal Biotech Inc., Dorval, Canada) (10.38 g) with 250-mL bottles for 10 min. Similar centrifugation method was used to remove salt and Conway medium from microalgal cultures. The microalgal pellet was then resuspended in autoclaved, filtered seawater, and then recentrifuge-harvested microalgae were chilled to 4 C (or −20 C for *T. tetrathele*) and stored for 1 wk before an experiment (Heasman et al. 2000). The microalgal density was controlled during copepod rearing to prevent any contamination as well as to keep the microalgal density constant for both sets of experiment. The mixed microalgal diets (NICT, NIC, CT, and IT) were prepared with equal ration (by number). Weekly algal preparation using the best algal growth phase seemed very suitable for copepod growth.

Experimental Procedure

Experiments were run in 48 glass beakers (diameter 7.5 cm) for microalgal diets and 18 glass beakers for nonalgal diets each with 300 mL. Five gravid *A. dengizicus* females carrying eggs (from laboratory copepod stock) were placed in each beaker filled with culture medium. The feeding frequency was twice per day (morning/evening). The beakers were manually shaken twice a day to ensure homogenous condition inside the beakers.

Population growth rate of *A. dengizicus* was studied for a 30-d period. Every other day, three samples (5–10 mL) from each beaker were removed, and number of copepods of all stages, including nauplii, copepodids, and adults in each sample were counted under a dissecting microscope and then returned to culture. The culture medium was changed every other day by passing the *A. dengizicus* culture through a 40-μm-plankton net which was small enough to retain the copepods but large enough to remove most of the detritus and other solid wastes. Approximately every 4 d, the contents of the rearing containers were transferred to clean beakers to prevent bacterial and algal growth on the walls and bottom.

The feeding conditions were: temperature 27.1 C, salinity 20 ppt, light intensity of 40 μmol photons/m²/s, and photoperiod 12 h light : 12 h dark regimen. Salinity was measured using a hand refractometer (ATAGO, Japan), temperature with a mercury thermometer (Strengthened, England), and light intensity with a light meter (LI-COR LI-189).

The specific population growth rate (K) of *A. dengizicus* was calculated using the following formula (Omori and Ikeda 1984; Hada and Uye 1991):

$$K = (\ln N_t - \ln N_0)/t$$

here, t is the culture days and N_0 and N_t are the initial and final (highest) density of copepods, respectively. In addition, doubling time (Dt) was calculated by dividing $\log_e 2$ by the population growth rate (K) according to the following formula (James and Al-Khars 1986):

$$Dt = (\log_e 2)/K$$

Statistical Analysis

Data were analyzed using two-way analysis of variance (ANOVA) with Fisher's Exact Test, a contingency table procedure. Differences in treatment means were compared by Duncan's New Multiple Range Test. The maximum population growth rates (K) were arcsine square root transformed to ensure a normal distribution (Zar 1984) and tested for statistical significance ($P < 0.01$) by two-way ANOVA. All statistical analyses were carried out using Statistical package for social science (SPSS 2002, version 11.5).

Results

Population Growth and Density of A. dengizicus *Fed on Microalgal Diets*

Two-way ANOVA showed that mono- and mixed microalgal diets have significant effects on density nauplii, copepodids, and adults of *A. dengizicus* as well as population growth rate (K) and population doubling time (Dt) (df = 15; $P < 0.0001$; Table 1). Likewise, there were significant interactions between algal diets and *A. dengizicus* because of its better density at higher algal density (df = 7; $P < 0.01$; Table 1).

The average density of *A. dengizicus* ranged from 97 to 753 ind./female on 30th culture day, the highest with CT and T diets and the lowest with I and N diets at both low (2.5×10^4) and high (5×10^6 cells/mL) microalgal densities (Fig. 1).

The population growth rate (K) and doubling time (Dt) for *A. dengizicus* fed on microalgal diets at both low and high densities are shown in Figures 2C and 2D. The population growth rate (K) ranged from 0.152 to 0.221, the lowest and the highest was observed in N and CT diet, respectively. Our findings showed that *A. dengizicus* fed on CT, T, C, NIC, NICT, I, and N diets required 3.14, 3.22, 3.31, 3.45, 3.32, 3.37, 3.42, and 3.79 d at high algal density, respectively, to double their population, while correspondingly at low algal density, needed 0.36, 0.56, 0.51, 0.50, 0.57, 0.52, 0.61, and 0.76 d longer, respectively.

The *A. dengizicus* mean densities varied from 1.6 to 12.6 ind./mL (Fig. 2) and significantly differed ($P < 0.01$) among eight algal diets. The densities were higher for CT, T, NIC, C, and NICT compared to IT, I, and N diets (Figs. 1, 2). Lowest *A. dengizicus* density for N diet was due to high mortalities which occurred in these cultures.

Algal density is an important factor controlling growth and density of *A. dengizicus*. There was progressive increase in growth and

TABLE 1. *Two-way analysis of variance of the effects of food types (N, I, C, T, NICT, NIC, CT, and IT) and food density levels (2.5×10^4 and 5×10^6 cells/mL) on nauplii, copepodids, adults, total, K, and doubling time of* A. dengizicus.

Factor	Source of variation	df	Sum of squares	Mean of square	*F* ratio	Significance level
Nauplii	Treatments	15	398734.1	26582.3	30.8	**
	Food type	7	128480	18354.3	21.3	**
	Food density	1	241826	241826	280.2	**
	Type × density	7	28428.1	4061.2	4.7	**
	Error	32	27621.3	863.2		
	Total	47	426355.5			
Copepodids	Treatments	15	207534.6	13835.6	49.9	**
	Food type	7	67602.5	9657.5	34.8	**
	Food density	1	119680.2	119680.2	431.4	**
	Type × density	7	20251.9	2893.1	10.4	**
	Error	32	8877.56	277.4		
	Total	47	216412.2			
Adults	Treatments	15	133028.9	8868.6	54.0	**
	Food type	7	43688.4	6241.2	38.0	**
	Food density	1	63816.7	63816.7	388.8	**
	Type × density	7	25523.8	3646.3	22.2	**
	Error	32	5251.8	164.1		
	Total	47	138280.7			
Total	Treatments	15	1903100	126873.3	82.3	**
	Food type	7	602332.3	86047.5	55.8	**
	Food density	1	118811	118811	771.4	**
	Type × density	7	111957	15993.9	10.4	**
	Error	32	49312.5	1541.0		
	Total	47	1952412			
K arcsine root convert	Treatments	15	92.1	6.1	120.5	**
	Food type	7	33.0	4.7	92.6	**
	Food density	1	57.9	57.9	1136.5	**
	Type × density	7	1.2	0.16	3.2	**
	Error	32	1.6	0.051		
	Total	47	93.8			
Doubling time	Treatments	15	6.7	0.44	122.5	**
	Food type	7	2.4	0.34	94.5	**
	Food density	1	3.8	3.8	1052.7	**
	Type × density	7	0.45	0.06	17.7	**
	Error	32	0.12	0.004		
	Total	47	6.78			

** Significant at 1% level.

density of *A. dengizicus* as algal density increased. In this study, results showed that when algal density became 200 folds numerically, the total production of *A. dengizicus* increased by 2.51 (N), 2.56 (I), 2.29 (C), 2.63 (T), 2.47 (NICT), 2.15 (NIC), 1.97 (CT), and 2.26 (IT) times fed on mentioned diets (Figs. 1D, 2A, and 2B).

Population Growth and Density of A. dengizicus *Fed on Nonalgal Diets*

Density of *A. dengizicus* fed on baker's yeast (BY) (181.7 ind./female) was significantly better than shrimp feed (SF) (70.7 ind./female) ($df = 5; P < 0.01$; Table 2). The densities of *A. dengizicus* were 181.7, 87.3, and 42.0 ind./female for low (3), medium (7), and high (10 mg/d) levels of BY diet and correspondingly 70.7, 58.3, and 26.7 inividuals/female for SF diet, respectively. The optimum level of BY and SF for *A. dengizicus* rearing was 3 mg/d (0.01 mg/mL). The population doubling time of *A. dengizicus* fed on low, medium, and high level of BY were 4.00, 4.65, and 5.56 d, while these values at the same condition were 4.88, 5.12, 6.33 d for SF, respectively (Fig. 4D).

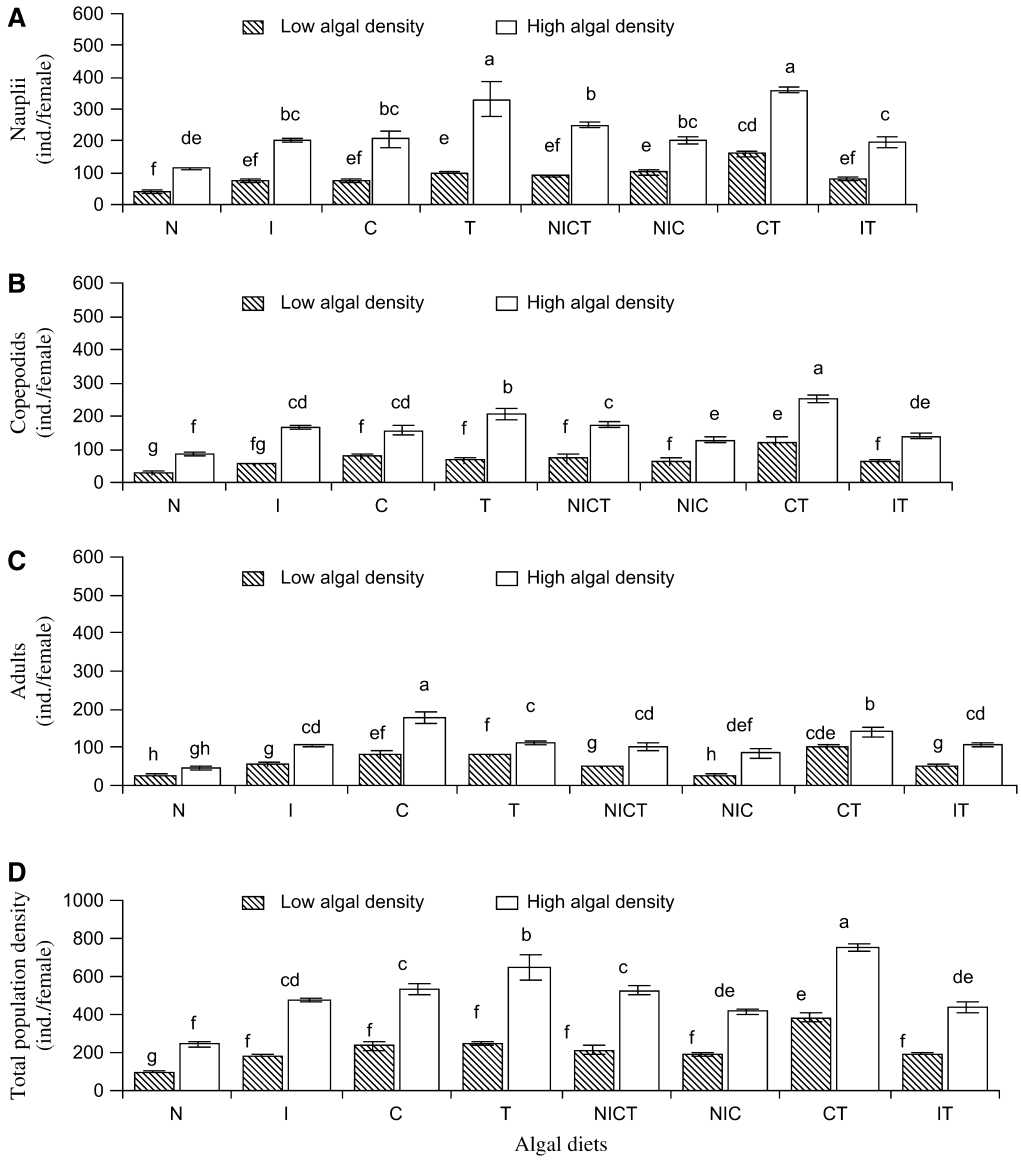

FIGURE 1. *Mean (±SE) nauplii (A), copepodids (B), adults (C), and total (D) population density of* A. dengizicus *in different algal diets in the laboratory conditions. Bars with same letters are not significantly different* ($P > 0.01$). *Low algal density = 2.5 × 10⁴ cells/mL and high algal density = 5 × 10⁶ cells/mL.*

Discussion

Algal Diets

The microalgae cells (from 2 to 16 μm) used in the present study were suitable to be ingested by *A. dengizicus*. For culture of *A. dengizicus* throughout their life history, smaller algal cell like *N. oculata* and *I. galbana* were preferred, particularly for the naupliar stages, while larger algal species such as *C. calcitrans* and *T. tetrathele* were essential for copepodids or the later stages. The total densities of *A. dengizicus* fed on N, I, C, T, NICT, NIC, CT, and IT diets ranged from 1.6–4.1, 3.1–7.9, 3.9–9, 4.1–10.8, 3.5–8.8, 3.2–6.9, 6.4–12.6, and 3.3–7.3 ind./mL, respectively. Of tested algal diets, T, C,

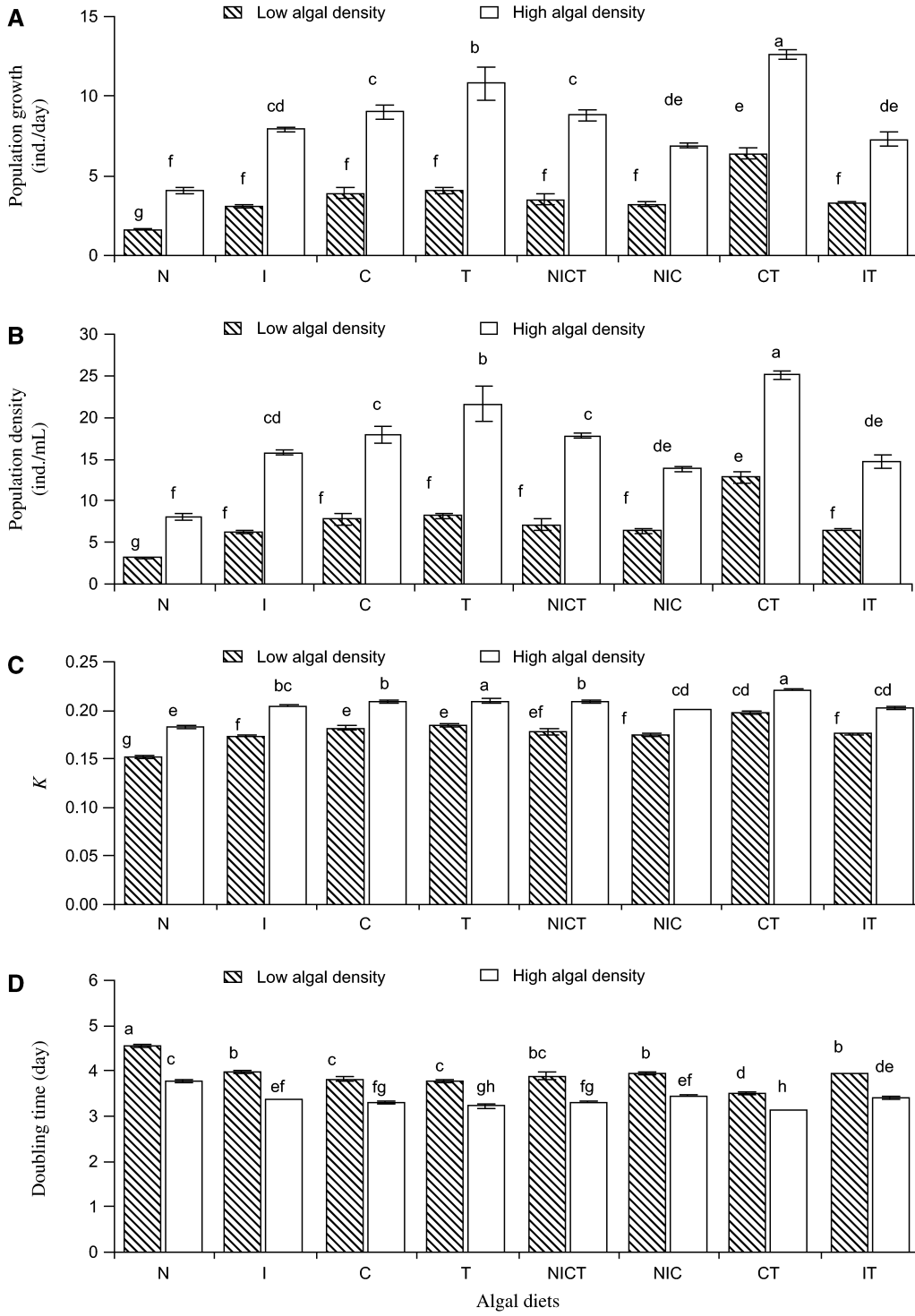

FIGURE 2. *Mean (±SE) population growth (A), population density (B), population growth rate (K) (C), and population doubling time (D) of* A. dengizicus *in different algal diets in the laboratory conditions. Bars with same letter are not significantly* ($P > 0.01$). *Low algal density* = 2.5×10^4 *cells/mL and high algal density* = 5×10^6 *cells/mL.*

TABLE 2. *Two-way analysis of variance of the effects of food types (Baker's yeast and prepared shrimp feed) and food density levels (3, 7, and 10 mg/d) on nauplii, copepodids, adults, total population density, K, and doubling time of* A. dengizicus.

Factor	Source of variation	df	Sum of squares	Mean of square	*F* ratio	Significance level
Nauplii	Treatments	5	12093.8	2418.8	101.7	**
	Food type	1	6904.8	6904.8	290.4	**
	Food density	2	2266.9	1133.4	47.7	**
	Type × density	2	285.3	1461.1	61.4	**
	Error	12	285.3	23.8		
	Total	17	12379.1			
Copepodids	Treatments	5	1530.9	306.2	6.2	**
	Food type	1	660.1	660.1	13.3	**
	Food density	2	817.4	408.7	8.2	**
	Type × density	2	53.4	26.7	0.5	ns
	Error	12	594.7	49.6		
	Total	17	2125.6			
Adults	Treatments	5	5047.2	1009.4	38.4	**
	Food type	1	1334.7	1334.7	50.8	**
	Food density	2	2645.3	1322.7	50.3	**
	Type × density	2	1067.1	533.6	20.3	**
	Error	12	315.3	26.3		
	Total	17	5362.5			
Total	Treatments	5	45615.8	9123.2	105.7	**
	Food type	1	12064.2	12064.2	139.8	**
	Food density	2	25250.1	4015.7	147.9	**
	Type × density	2	8031.5	4015.7	46.28	**
	Error	12	1035.3	86.3		
	Total	17	46651.1			
K arcsine root convert	Treatments	5	53.9	10.8	20.3	**
	Food type	1	13.5	13.5	25.4	**
	Food density	2	39	19.5	36.7	**
	Type × density	2	1.4	0.7	1.3	ns
	Error	12	6.4	0.5		
	Total	17	60.3			
Doubling time	Treatments	5	12.1	2.4	10.3	**
	Food type	1	2.8	2.8	12.0	**
	Food density	2	9.0	4.5	19.1	**
	Type × density	2	0.3	0.2	0.7	ns
	Error	12	2.8	0.2		
	Total	17	14.9			

ns = not significant at 5%.
** Significant at 1% level.

and CT were optimum diets for *A. dengizicus* culture. *A. dengizicus* culture density obtained in this study was 9–12 ind./mL when fed on *T. tetrathele* solely or in combination with *C. calcitrans*. These findings were similar with previous researchers. The densities for *A. distans* (Hsu 1999; Velasquez et al. 2001), *Acartia tsuensis* (Ohno and Okamura 1988), *Acartia* sp. (Schipp et al. 1999), *A. royi* (Cheng et al. 2001), and *Paracyclopina nana* (Lee et al. 2006) were 6.5, 2.0, 7.0, 33, and 96–119 ind./mL, respectively. Velasquez et al. (2001) reported that better growth performance of *A. distans* was due to highest volume and motility of *T. chuii* compared to *N. oculata* and *Dunaliella salina*. The better growth and density of *A. dengizicus* fed on T and CT may be related to feeding behavior of this species, which preferred T due to its motility.

Though *N. oculata* with cell diameter of 2–4 μm was suitable for nauplii rearing, nauplii of *A. dengizicus* did not develop well when

fed this microalga compared to other diets. Cano et al. (2004) stated that *A. panamensis* could not digest the hard cell wall of *N. oculata*. Low egg and nauplii production and low survival of nauplii during development of *A. dengizicus* fed N diet (Farhadian 2006) could be related to lower population growth. There are a few reports of copepod culture with *N. oculata* as food (Kitajima 1973 on *Tigriopus japonicus*; Kitajima 1973 on *Acartia clause*; and Ohno et al. 1990 on *Acartia tsuensis*). The poor nutritional value of N could be one of the reasons for high mortality in *A. dengizicus* population. Although N is devoid of 22:6n-3 acids, it contains small amounts of 20:3n-3 and 20:4n-6 and considerable amounts of 18:3n-3 and 18:2n-6 acids.

On the other hand, optimum density and growth of *A. dengizicus* fed on T, C, and CT diets could be related to the microalgal higher dietary value. Although Thinh et al. (1999) stated that *T. tetrathele* was rich in essential fatty acid 20:5n-3 (EPA) but contains almost no 22:6n-3 (DHA), our findings showed that *T. tetrathele* has 4.6% DHA (Table 3). Moreover, Lee et al. (2006) concluded that better growth performance in *Paracyclopina nana* fed on *T. suecica* could be attributed to biosynthesis DHA from linolenic acid, 18:3n-3. In our findings, the linolenic acid contents of N, I, C, and T diets were 8.7, 0, 5.2, and 9.8%, respectively, and for BY and SF were 0.8 and 0.3%, respectively.

The better population growth and density of *A. dengizicus* could be related to DHA:EPA:ARA ratio in T and C diets. The DHA:EPA:ARA ratio of T and C were 0.66:1.21:1 and 0.65:2.31:1, respectively. ARA is an important precursor of some prostaglandins and other biologically active compounds that regulate growth and reproductive functions (Barclay and Zeller 1996; Sargent et al. 1997). In addition, several researchers have stated the importance of ARA, which plays a significant role in fish

TABLE 3. *Fatty acid composition (% total fatty acid) of microalgae (N, I, C, T), Baker's yeast, and prepared shrimp feed used for experiment.*

	Nannochloropsis oculata	*Isochrysis galbana*	*Chaetocerous calcitrans*	*Tetraselmis tetrathele*	Baker's yeast	Shrimp feed
C14:0	1.9	19.3	12.1	9.1	0.5	0.4
C15:0	—	—	—	7.3	0.1	—
C16:0	27.2	9.8	10.0	13.2	8.6	26.5
C17:0	4.0	—	1.8	—	—	—
C18:0	8.6	—	5.8	5.2	3.5	16.4
C20:0	—	—	6.2	—	4.4	—
C14:1n	—	—	1.3	—	0.1	—
C15:1n	—	—	1.5	—	—	—
C16:1n-7	5.8	9.3	12.7	9.2	15.5	5.1
C17:1n	1.6	—	2.3	5.8	—	0.2
C18:1n-9	13.9	19.5	5.9	3.3	20.1	12.3
C18:2n-6	14.5	9.5	2.4	6.4	2.2	—
C18:3n-3	8.7	—	5.2	9.8	0.8	0.3
C18:3n-6	5.4	—	—	0.9	—	—
C:20:4n-6	—	—	7.2	7.0	—	0.2
C20:5n-3	—	—	16.6	8.5	—	0.4
C22:6n-3	—	7.6	4.7	4.6	—	6.5
SFA	41.7	29.1	35.9	34.8	17.1	43.3
MUFA	21.3	28.8	23.7	18.3	35.7	17.6
PUFA	28.6	17.1	36.1	37.2	3.0	7.4
Total n-3	8.7	7.6	26.5	22.9	—	7.2
Total n-6	19.9	9.5	9.6	14.3	—	0.2
n-3/n-6	0.44	0.80	2.76	1.60	—	—
DHA:EPA	—	—	4.7:16.6	4.6:8.5	—	6.5:0.4
DHA:EPA:ARA	—	—	0.7:2.3:1	0.7:1.2:1	—	32.5:2:1

SFA = saturated fatty acid; MUFA = monounsaturated fatty acid; DHA = docosahexaenoic acid; ARA = arachidonic acid; EPA = eicosapentaenoic acid; PUFA = polyunsaturated fatty acid; — = not detectable.

larval nutrition. Ogata et al. (2005) stated ARA:EPA ratio of 3.5:1 and DHA:ARA ratio of 2.1:1 as the optimum ratios of brood stock diets in tropical fishes. Chavez et al. (2005) concluded that dietary ARA was more important for development of fry production technology in tropical fishes. In our study, the higher amount of ARA may partly explain better growth and density of *A. dengizicus* fed on T and CT diet.

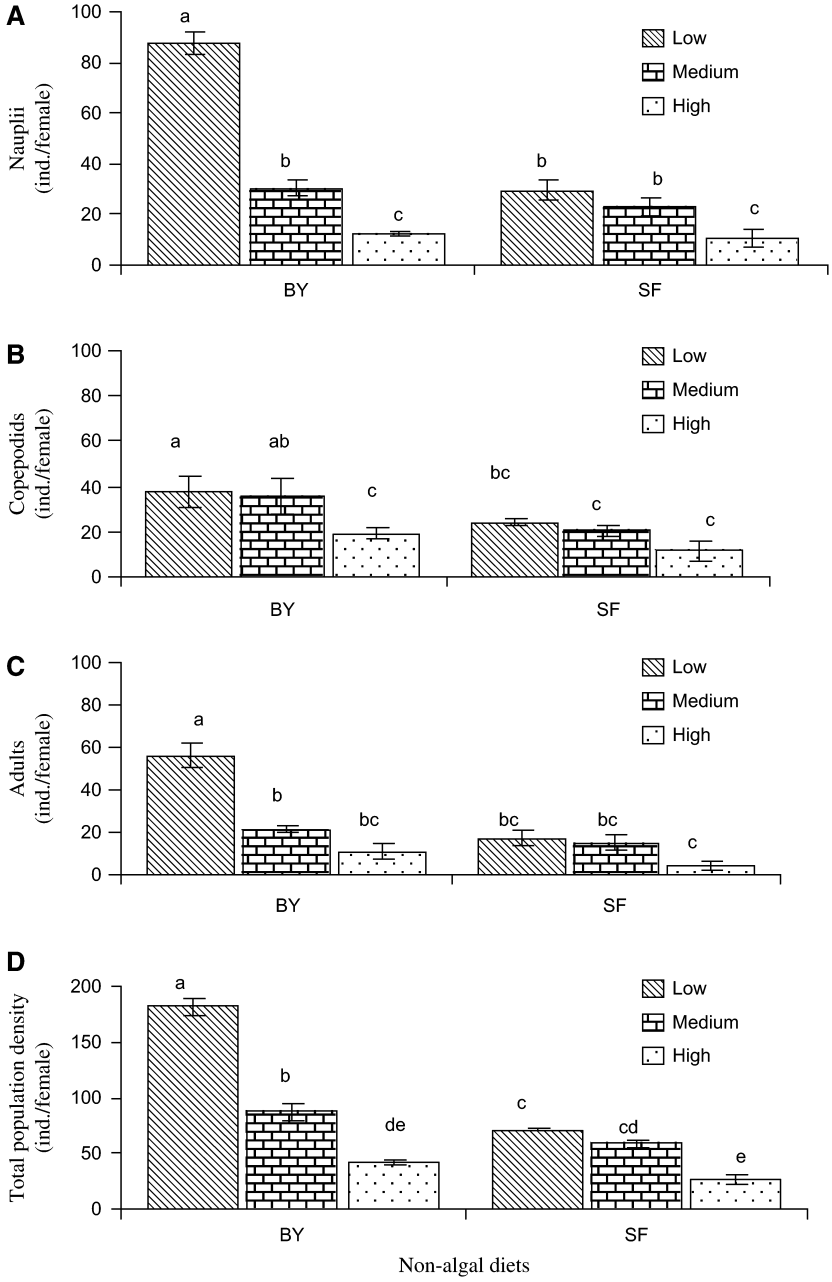

FIGURE 3. *Mean (±SE) nauplii (A), copepodids (B), adults (C), and total (D) population density of* A. dengizicus *fed on baker's yeast and prepared shrimp feed. Bars with same letters are not significantly different* ($P > 0.01$). *Low = 3 mg/d, medium = 7 mg/d, and high = 10 mg/d.*

Although the findings of this study promote the use of *C. calcitrans* diet for *A. dengizicus* culture, it is not in agreement with those results reported by Miralto et al. (1999) and Carotenuto et al. (2002). Miralto et al. (1999) reported that aldehydes found in diatoms arrested embryonic

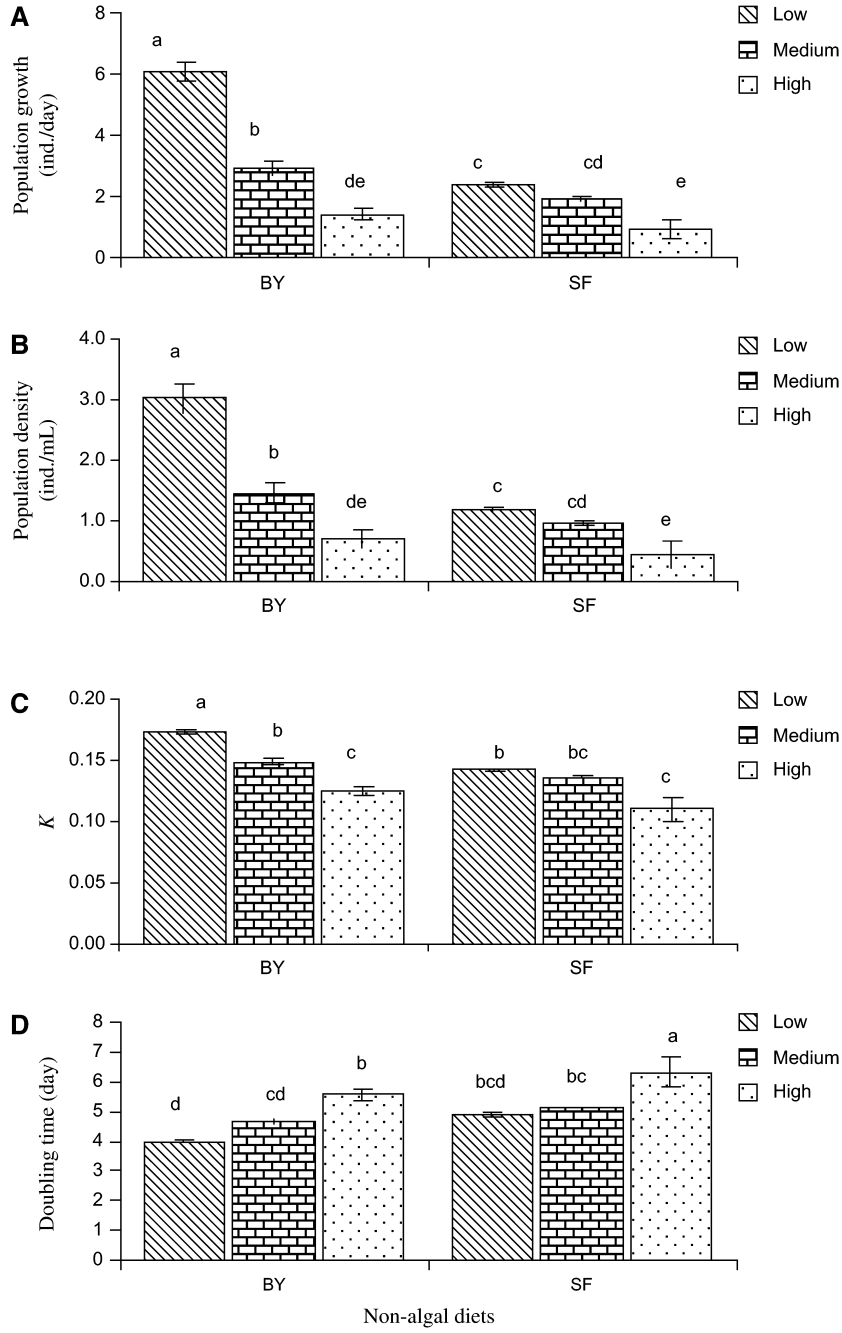

FIGURE 4. *Mean (± SE) population growth (A), population density (B), population growth rate (K) (C), and doubling population time (D) of* A. dengizicus *fed on baker's yeast and artificial shrimp feed. Bars in the same letters are not significantly different (*$P > 0.01$*). Low = 3 mg/d, medium = 7 mg/d, and high = 10 mg/d.*

development in copepod. Similarly, Carotenuto et al. (2002) found that diatoms have deleterious effects on embryonic development and on larval growth of *Temora stylifera.* In addition, Laabir et al. (2001) did not find any improvement in copepod hatching rate with enriched DHA and EPA of diatoms.

While *A. dengizicus* can be cultured on different algal diets under laboratory conditions, microalgal density seemed to be important to determine levels of copepod culture. Nassogne (1969) found that the highest egg production and best feeding efficiencies for *Euterpina acutifrons* was at 5×10^4 cells/mL. Kraul (1989) recommended densities between 5×10^4 and 2×10^5 cells/mL, while Kahan and Azoury (1981) used 1–3 $\times$ 10^5 cells/mL as the best density for *Nitocar spinipies*. Iwasaki and Kamiya (1977) estimated that the optimum densities for *Acartia clausi* at 1:1 mixture of *I. galbana* and *Monochrysis lutheri* at 1×10^6 cells/mL at 15 C and 1.5×10^6 cells/mL at 20 C. Our findings indicated that algal density between 2.5×10^4 to 5×10^6 cells/mL was suitable for *A. dengizicus* rearing.

Nonalgal Diets

Baker's yeast, *Saccharomyces cerevisiae*, can be used as an algal substitute in live food production. There are obvious benefits to this practice, such as the reduction of algal production facilities. However, baker's yeast contains mainly 16:1n and 18:1n-9 fatty acids. It is completely devoid of DHA, EPA, and ARA fatty acids. Yeast, in contrast to algae, completely lacks PUFA (polyunsaturated fatty acid). In addition, BY and SF rejected by *A. dengizicus* may pollute the water, increase ammonia level, and reduce growth rate. Baker's yeast is deficient in essential nutrients that are required for growth of *A. dengizicus*. Therefore, the increase BY and SF significantly decreased *A. dengizicus* population growth and density ($P < 0.01$; Table 3; Figs. 3, 4). The fatty acid composition of *A. dengizicus* is largely dependent on that of their food, indicating that the ingested lipids, are hydrolyzed in the gut, desorbed, metabolized, and incorporated into body lipids.

Population growth of *A. dengizicus* fed on BY and SF was poor, probably due to their lack of PUFA content. The previous studies showed that lipid and fatty acid contents play a major role in determining level of copepod production (Støttrup and Jensen 1990). Although BY gave better growth and production than SF ($P < 0.01$), results suggested that both the diets could not support population of *A. dengizicus* properly. Another possible reason could be related to feeding behavior of *A. dengizicus* because this species, like other cyclopoids, preferred large motile foods.

In conclusion, production of *A. dengizicus* was found to be high with the consumption of all types of microalgae. *Tetraselmis tetrathele* and its combination with *C. calcitrans* had significantly higher population density compared to other diet. The optimum microalgal density was 5×10^6 cells/mL.

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