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Optimization of Stocking Density for the Sea Cucumber, *Apostichopus japonicus* Selenka, Under Feed-Supplement and Non-Feed-Supplement Regimes in Pond Culture

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Abstract Optimal stocking densities were investigated for the sea cucumber *Apostichopus japonicus* Selenka under feed-supplement and non-feed-supplement regimes in net enclosures for 333 d. Substantial weight loss occurred during the aestivation phase (AE). Decreased growth rates were also observed during the winter phase (WT). In contrast, sea cucumbers showed rapid growth during the spring (SP) and autumn (AU) phases. Feeding regimes considerably influenced the growth performance, *i.e.*, sea cucumbers grew faster under feed-supplement regime than under non-feed-supplement regime ($P<0.05$). The average survival rates of sea cucumbers under feed-supplement regime were higher than those under non-feed-supplement regime for both the autumn phase and spring phase, but the differences were only significant for the latter phase ($P<0.05$). The fitted B-N curves showed that the optimal stocking densities, in terms of net production, were 22.3 ind. m^{-2} for feed-supplement regime and 14.1 ind. m^{-2} for non-feed-supplement regime.

Key words sea cucumber; *Apostichopus japonicus*; stocking density; feed-supplement; non-feed-supplement; yield

1 Introduction

Since the 1980s, the increasing demand for the sea cucumber (*Apostichopus japonicus* Selenka) in China has led to intensive research and development of sea cucumber seed production and grow-out farming (Conand and Byrne, 1993; Ramofafia *et al.*, 1997). Stimulated by great profits, capital investment has been continuously flowing into this sector and farming areas have sharply expanded to 17000 hm^2 in China (Chen, 1990; Chen, 2004), and especially in northern China, mariculture of sea cucumber has quickly become a prosperous aquaculture sector (Chen, 2004) with an annual beche-de-mer production increased from 38952 t in 2003 to 75725 t in 2006 (FAO, 2008). Sea cucumbers live on microorganisms associated with both organic and inorganic materials from sediment (Sloan and von Bodungen, 1980; Yingst, 1976). There are mainly three methods used for farming sea cucumbers, namely, pond culture, pen culture and sea ranching (Chen, 2003; Jia and Chen, 2001). Extensive culture in earthen ponds is the main culture method in China, and supplement feeding (mixed macroalgal meals with sea mud or formulated feed) to enhance sea cucumber production is applied more and more nowadays. However, little scientific information on sea cucumber stocking density is available so far and farmers usually decide the stocking density of sea cucumber in earthen ponds based on practical experiences.

Stocking density is one of the most important production factors that should be considered in aquaculture because it directly influences survival, growth, health, feeding, and yields of cultured organisms (Rowlanda *et al.*, 2006). Overcrowding of cultured organisms may induce slow growth rates and great variabilities in body size, as well as cause deformities, ‘rotten-stomach’ and low metamorphosis rates of sea cucumber, while low density may cause an unnecessary waste of space in aquaculture ponds (Li *et al.*, 2007; Liu *et al.*, 2002). At high stocking intensity, the shortage of food (natural diet) might be a limiting factor on the growth of sea cucumber, and yet increased production output can be achieved *via* providing artificial food. The objective of this study was to investigate optimal stocking densities of the sea cucumber *A. japonicus* under the two feeding regimes, *i.e.*, with or without supplementary feed, in order to improve growth, survival and production of sea cucumber farming.

2 Materials and Methods

2.1 Earthen Pond and Enclosures

This study was conducted at a sea cucumber farm in

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Weihai City, Shandong Province, P. R. China (36°55'N, 122°10'E) from May 20, 2007 to April 20, 2008. Thirty net enclosures were built in a rectangular earthen pond with an area of three hectares and an average water depth of 1.8 m and ensured uniform sediment, water quality and other factors among different enclosures at the beginning of the study. The pond was connected to the main waterways *via* inlet and outlet sluices. The enclosures were made of polyethylene netting of 0.5 cm mesh size. For each net enclosure (8 m long ×8 m wide ×2 m high), eight bamboo sticks were vertically inserted 50 cm deep into the sediment and eight additional bamboo sticks were buried horizontally 30 cm below the seabed surface. The net was fixed onto these bamboo sticks. The distance between the enclosures was 8 m measured from the two directly adjacent sides. Four pieces of plastic tubes (20 cm in diameter ×40 cm long) were bundled together by nylon threads and placed horizontally inside the enclosures to provide shelters for the sea cucumbers. The density of the plastic tubes was 64 bundles per net enclosure or one bundle per square meter.

2.2 Pond Preparation and Experiment Design

The pond was drained and exposed to sunlight for 7 d prior to the experiment. The plastic shelters were placed inside the enclosures parallel to the direction of water current. Five days prior to the initial stocking, the shelters were inoculated with microorganisms to provide natural food for the sea cucumbers. During the period of sea cucumber culture, pond water was exchanged in a period of consecutive seven spring tide days, with a daily water exchange rate of 20%–50% of the total pond water volume.

Table 1 Survival and growth of sea cucumbers reared under different stocking densities and feeding regimes on Nov. 20, 2007

Stocking density		Sr (%)	Wf (g)	Feeding rate (g d^{-1} m^{-2})
Feed-supplement	5	30.7±0.5	23.7±3.9^a	43.3±0.0
	10	35.0±3.9	16.9±1.2^b	50.5±8.7
	15	33.0±5.3	16.6±3.7^b	100.0±0.0
	25	29.7±1.9	12.1±1.4^c	160.0±0.0
	35	33.9±1.5	11.2±0.9^c	200.0±0.0
Non-feed-supplement	5	29.9±0.8	20.5±0.6^a	Excluded
	10	27.6±0.8	14.2±1.6^b	Excluded
	15	29.5±10.6	13.7±1.0^b	Excluded
	25	28.1±2.7	10.1±1.8^c	Excluded
	35	32.9±11.1	7.5±1.2^c	Excluded
Two-way ANOVA				
Feeding regime (F)		NS (0.145)	S (< 0.05)	–
Stocking density (S)		NS (0.348)	S (< 0.05)	–
F × S		NS (0.733)	NS (0.863)	–

Notes: Sr and Wf denote the survival rate and final individual wet weight of sea cucumbers, respectively. Data are means ± S.D. of three replicate net enclosures. Within each feeding regime, values in the same column having the same superscript (a, b, or c) are not significantly different ($P > 0.05$). Decimal fraction within each bracket denotes the P value of two-way ANOVA results. S: significant ($P<0.05$); NS: not significant ($P>0.05$).

Juvenile sea cucumbers were provided by a local sea cucumber farm. Sea cucumbers (average weight was 5.0 g ±2.0 g, $n>600$) were stocked randomly at densities of 5, 10, 15, 25 and 35 ind. m^{-2} or 1.6, 3.2, 4.8, 8.0 and 11.2 kg of biomass at different enclosures. Three replicate enclosures were set and assigned randomly for each density treatment. The enclosures were cleaned regularly off fouling organisms to prevent mesh clogging at least every 3–4 weeks or when necessary.

Granulated commercial feed (ø25 mm, Dahaidi Fishery Feeding Stuff Co., Ltd., Qingdao, China) was provided to the sea cucumbers when water temperature was over 10℃. The content of crude protein, crude fat, coarse fiber, crude ash, water content, salt, calcium, phosphor and lysine of the feed was 15.0%, 2%–5%, 10%, 45%, 11%, 0.5%, 1.5%–4.5%, 0.6% and 1%, respectively. The leaching rate of the feed was less than 30% in 6 h. The amount of feed provided (Tables 1 and 2) was adjusted daily for each enclosure, depending on the amount of uneaten feed observed in experimental feeding trays (100 cm in diameter). However, there was no additional feeding during the aestivation (summer) and hibernation (winter) periods for all the feed-supplement treatments.

Table 2 Survival and growth of sea cucumbers reared under different stocking densities and feeding regimes at the end of the study

Stocking density ind. m^{-2}		Sr (%)	Wf (g)	Feeding rate (g d^{-1} m^{-2})
Feed-supplement	5	29.2±0.5^{a,b}	61.2±3.1^a	30.0±0.0
	10	33.3±3.7^b	48.7±7.3^b	41.0±5.0
	15	33.9±4.9^b	45.8±2.3^c	60.0±0.0
	25	26.6±2.3^a	35.0±5.9^d	85.0±0.0
	35	29.3±1.9^{a,b}	28.9±0.6^e	100±0.0
Non-feed-supplement	5	28.4±0.7^{a,b}	53.5±2.3^a	Excluded
	10	26.2±0.7^b	41.4±1.1^b	Excluded
	15	28.4±5.3^b	37.2±2.0^c	Excluded
	25	24.5±1.8^a	25.8±0.1^d	Excluded
	35	28.7±3.1^{a,b}	18.1±1.0^e	Excluded
Two-way ANOVA				
Feeding regime (F)		S (<0.05)	S (<0.01)	–
Stocking density (S)		S (<0.05)	S (<0.01)	–
F×S		NS (0.258)	NS (0.829)	–

Note: For explanations see footnotes to Table 1.

2.3 Sample Protocols

According to the literature on the physiology and ecology of sea cucumbers (Sui, 1990; Yu and Chang, 2008; Yu and Song, 1999) and the water temperature measured in this study (Fig.1), the whole culture period was divided into four phases. The first one was the aestivation (summer) phase (AE) which extended from May 20, 2007 (the start of the experiment) to September 20, 2007. In this phase, the average water temperature was above 20℃ and low growth rate was expected. Therefore, sea cucumbers were sampled for weighing every 30 d. The second phase was the autumn phase (AU) that lasted from September 21, 2007 to November 20, 2007, and sea cucumbers were sampled every 15 d. Sea cucumbers were fished by SCUBA diving and measured for individual weight, survival rate and yield in each enclosure. Sea cucumbers were restocked in the same enclosure after measurement. The third phase was the winter phase (WT) from November 21, 2007 to March 1, 2008. During this period, no sea cucumbers were sampled because no growth would be expected due to the low water temperature (−1.8−5℃) (Yu and Chang, 2008). Water temperature increased from 5℃ to 18℃ in the fourth or spring phase (SP) (March 1, 2008 to April 20, 2008). At the end of the spring phase (the end of the study), all sea cucumbers were collected by SCUBA diving. Growth, survival and yield were recorded.

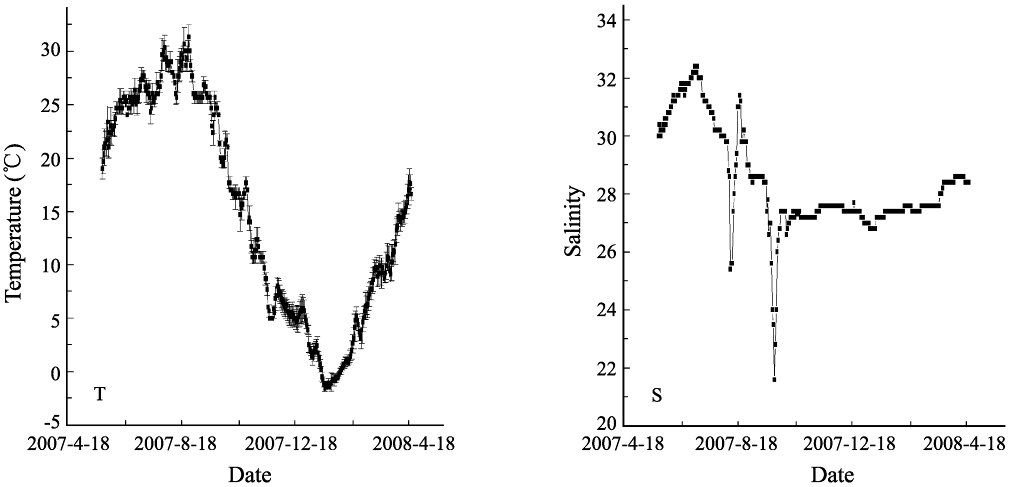

Fig.1 Fluctuations of temperature and salinity during the culture period. T, temperature; S, salinity.

The sea cucumbers were weighed according to the method modified from Sewell (1987) and Purcell and Kirby (2006). The anterior end of the sea cucumbers were gently pressured to remove the fluid in the respiratory trees, and the body surface was dried with a towel prior to weighing. The individuals were weighed on-site with an electronic balance (ACF-CF, Huade Balance Co., Ltd., China) to the nearest 0.1 g, and 50 individuals were selected randomly per net enclosure. Water temperature was measured daily at 7:00, 13:00 and 17:00. Salinity and dissolved oxygen were measured using a data logger YSI-6000 (YSI, Yellow Springs, OH, USA) during the study period.

2.4 Data Calculation and Statistics

Final mean wet weight (*Wf*) and survival rate (*Sr*) were calculated as follows:

$$Wf(g) = \frac{Tw}{Nf}, \ Sr(\%) = \frac{100Nf}{Ni},$$

where *Tw* is the total weight of sea cucumbers; *Nf* and *Ni* are the final and initial number, respectively.

The specific growth rate (SGR, % day^{-1}) of a particular culture period was estimated by the formula:

$$SGR = \frac{100\,(\ln W_t - \ln W_0)}{t},$$

where W_0 and W_t are the initial and the final mean wet weight (g), respectively; and *t* is the duration of culture period in days.

The relationship between the total yield and stocking densities of sea cucumbers was described with the B-N model (Fréchette, 2005; Fréchette *et al.*, 1996, 2000, and 2005; Westoby, 1984):

$$B = \frac{gN}{1+N^k},$$

where *B* is the total yield of sea cucumbers per m^2; *N* is the number of sea cucumbers per m^2; *g* and *k* are the constants of the equation.

The m-N model was used to describe the relationship between sea cucumber body weight and stocking densities (Fréchette and Bacher, 1998):

$$m = \frac{a}{b+N},$$

where *m* is the final mean wet weight of sea cucumbers (g); *N* is the number of sea cucumbers per m^2; *a* and *b* are the constants of the equation (Fréchette and Bacher, 1998).

The optimal stocking densities were calculated with the Bn-N model (Fréchette and Bacher, 1998).

$$Bn = aN^2 + bN + c,$$

where *Bn* is the net yield of sea cucumbers per m^2 (g); *N* is the number of sea cucumbers per m^2; *a*, *b* and *c* are the constants of the equation.

Experimental results were compared using two-way ANOVA with SPSS for Windows release 15.0 (SPSS, Chicago, IL, USA), followed by Duncan's New Multiple Range test for comparisons between the treatment levels. The statistical significance level of 0.05 was used. The B-N model, m-N model and Bn-N model were estimated and fitted with nonlinear curve fit using Origin 7.5 (OriginLab Corporation, MA, USA).

3 Results

3.1 Water Quality

Water temperature of the pond during the experimental period is shown in Fig. 1T. Daily average water temperature was 17℃ at the beginning of the experiment. Water temperature reached the highest of 32℃ between August 20 and August 28, and then dropped gradually. The lowest water temperature (−1.6℃) occurred on January 20, 2008. From March 1 to April 20, 2008, water temperature increased from 5℃ to 18℃.

Salinity of the pond water fluctuated generally between 27 and 32 (Fig.1S). On August 10 and September 26, 2007, however, it dropped sharply to 25.4 and 21.6, respectively, because of heavy rains, but restored to 27 several days later. Dissolved oxygen was 5−8 mgL^{-1} during the whole experimental period.

3.2 Body Weight and Survival Rate

Mean wet weight of the sea cucumbers during the whole culture phase is shown in Fig.2. Those at the end of the autumn and spring phases (*i.e.* the two growing phases) are listed in Tables 1 and 2, respectively. Compared to the non-feed-supplement regime, supplemental feeding improved body weight of the sea cucumbers significantly at the end of both the autumn phase ($F_{1,30}$=14.821, P<0.05) and the spring phase ($F_{1,30}$=49.183, P< 0.05). Further, body weight of the sea cucumbers reared at the feed-supplement ($F_{4,30}$ = 34.961, P <0.05) and non-feed-supplement regimes ($F_{4,30}$ = 78.797, P<0.05) both decreased as the stocking densities increased (Tables 1 and 2).

At the end of the autumn phase, no significant differences were observed in survival rates among different stocking densities ($F_{4,30}$=1.20, P>0.05) and feeding regimes ($F_{1,30}$=2.00, P>0.05) (Table 1). At the end of the spring phase, however, statistical analysis revealed that survival rates of the sea cucumbers were significantly influenced by feeding regimes ($F_{1,30}$ = 8.55, P<0.05) and stocking densities ($F_{4,30}$=2.90, P<0.05) (Table 2). Overall, average survival rates of the sea cucumbers under the feed-supplement and non-feed-supplement regimes were 30.4% and 28.9%, respectively, at the end of the experiment.

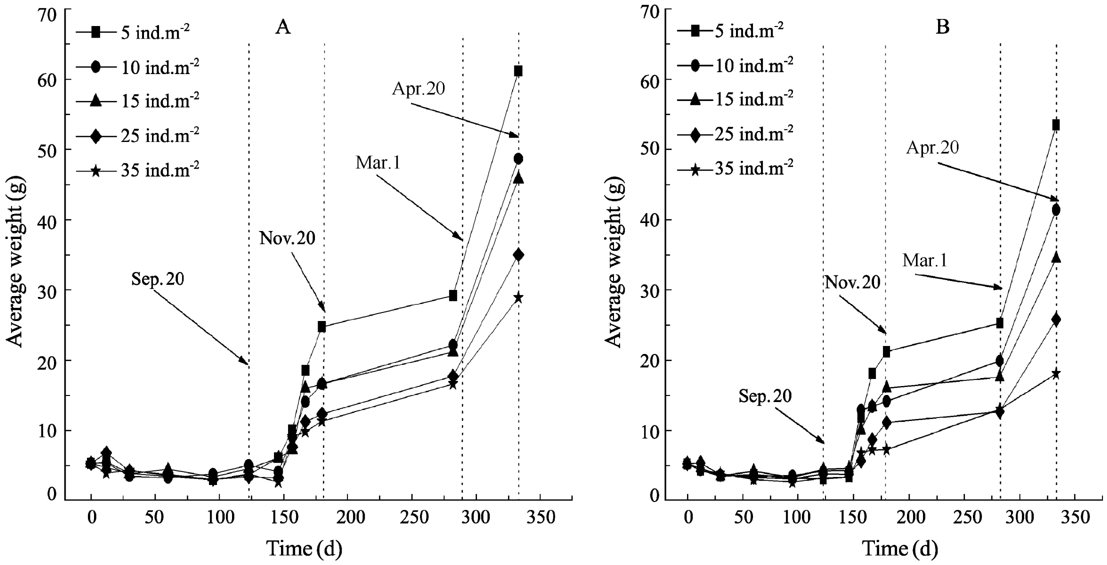

Fig.2 Average wet weight of sea cucumber cultured for 330 d at stocking densities of 5, 10, 15, 25, and 35 ind. m^{-2}. Data are means of three replicate enclosures. A and B refer to feed-supplement and non-feed-supplement regimes, respectively. September and November refer to 2007 and March and April refer to 2008.

3.3 Specific Growth Rate (SGR)

Specific growth rate (SGR) of the sea cucumbers during different culture phases are summarized in Table 3. At the end of the 333-day culture, stocking density ($F_{4,30}$= 113.479, P<0.01), feeding regimes ($F_{1,30}$=70.501, P< 0.01), and their interaction ($F_{1,4}$=4.089, P<0.05) all had significant influence on SGR, *i.e.*, in general, the overall SGR decreased significantly with increasing stocking density and artificial feeding improved the growth of sea cucumbers significantly (Table 3).

In the aestivation phase (AE), sea cucumbers in all the treatments showed little growth and there was no significant difference in SGR between the two feeding regimes ($F_{1,30}$=0.049, P>0.05). However, SGRs of the sea cucumbers at stocking densities 10 and 15 ind.m^{-2} were significantly lower than those at the other stocking densities in both the feeding regimes (P<0.05) (Table 3).

During the autumn phase (AU), sea cucumbers achieved the highest growth rates among all the culture phases, and feeding regimes did not affect growth significantly either ($F_{1,30}$=3.597, P>0.05). SGR of the sea cucumbers at a stocking density of 5 ind. m^{-2} was significantly higher than those at the other four stocking densities in both the feeding regimes (P<0.05) (Table 3).

Sea cucumbers grew relatively slowly in the winter phase (WT), and no significant difference in SGR was found between the two feeding regimes ($F_{1,30}$=0.056, P> 0.05) and among different stocking densities ($F_{4,30}$ = 1.997, P>0.05). In the following spring phase (SP), the overwintered sea cucumbers grew at faster rates. Significant differences were observed in SGR between the two feeding regimes ($F_{1,30}$=8.845, P<0.01) and among different stocking densities ($F_{4,30}$=9.015, P<0.01). In all the above four intermediate phases, however, no interactive effects between stocking densities and feeding regimes were found on SGR of the sea cucumbers ($P>0.05$).

Table 3 Specific growth rates (SGR) of sea cucumbers, reared under different stocking densities and feeding regimes, in different growth periods

Model	Stocking density (ind. m^{-2})	Aestivation phase	Autumn phase	Winter phase	Spring phase	Whole year
Feed-supplement	5	0.33±0.24^{a}	3.43±0.42^{a}	0.22±0.14	1.39±0.13^{a,b}	0.76±0.02^{a}
	10	0.00±0.32^{b}	2.09±0.69^{b}	0.27±0.15	1.48±0.20^{a}	0.69±0.05^{b}
	15	0.02±0.19^{b}	2.39±0.45^{b}	0.26±0.17	1.46±0.13^{a,b}	0.67±0.02^{b,*}
	25	0.41±0.18^{a}	1.80±0.60^{b}	0.38±0.05	1.28±0.16^{b}	0.59±0.05^{c,*}
	35	0.23±0.13^{a}	2.23±0.38^{b}	0.40±0.07	1.05±0.11^{c}	0.53±0.01^{d,*}
Non-feed-supplement	5	0.47±0.09^{a}	3.49±0.15^{a}	0.24±0.05	1.35±0.01^{a,b}	0.72±0.01^{a}
	10	0.05±0.12^{b}	2.23±0.50^{b}	0.31±0.15	1.31±0.19^{a}	0.64±0.01^{b}
	15	0.06±0.14^{b}	1.89±0.22^{b}	0.21±0.18	1.24±0.09^{a,b}	0.61±0.01^{b,*}
	25	0.20±0.12^{a}	1.63±0.62^{b}	0.36±0.15	1.09±0.07^{b}	0.50±0.00^{c,*}
	35	0.28±0.10^{a}	1.12±0.47^{b}	0.45±0.38	0.83±0.29^{c}	0.39±0.02^{d,*}
Two-way ANOVA						
Feeding regime		NS (0.827)	NS (0.072)	NS (0.816)	S (< 0.01)	S (< 0.01)
Stocking density		S (< 0.01)	S (< 0.05)	NS (0.134)	S (< 0.01)	S (< 0.01)
F×S		NS (0.530)	NS (0.160)	NS (0.977)	NS (0.862)	S (< 0.05)

Notes: Data are means ± S.D. of three replicate net enclosures. Within each feeding regime, values in the same column having the same superscript (a, b, or c) are not significantly different ($P > 0.05$). For the whole year data, the means at the same stocking density were compared between feed-supplement and non-feed-supplement regimes, and an * following the means indicates significant difference ($P<0.05$). Decimal fraction within each bracket denotes the P value of two-way ANOVA results. S: significant ($P<0.05$); NS: not significant ($P>0.05$).

3.4 B-N Model and m-N curve

The relationships between total yield and stocking density and between final mean wet weight and stocking density were described with the B-N model (Fig.3) and the m-N model (Fig.4), respectively:

$$B=\frac{78.04N}{1+N^{0.598}},\ R^2=0.80\ \text{(feed-supplement)},$$

$$B=\frac{49.3N}{1+N^{0.563}},\ R^2=0.89\ \text{(non-feed-supplement)};$$

$$m=\frac{1492.2}{19.7+N},\ R^2=0.90\ \text{(feed-supplement)},$$

$$m=\frac{974.2}{13.1+N},\ R^2=0.97\ \text{(non-feed-supplement)}.$$

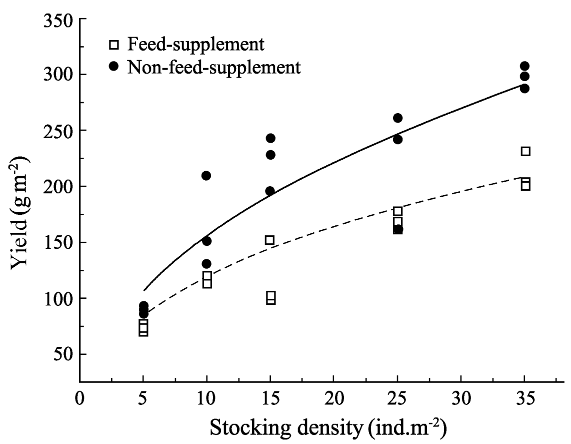

Fig.3 Relationship between yield and stocking density of sea cucumbers, cultured under different stocking densities and feeding regimes, fitted with the B-N model.

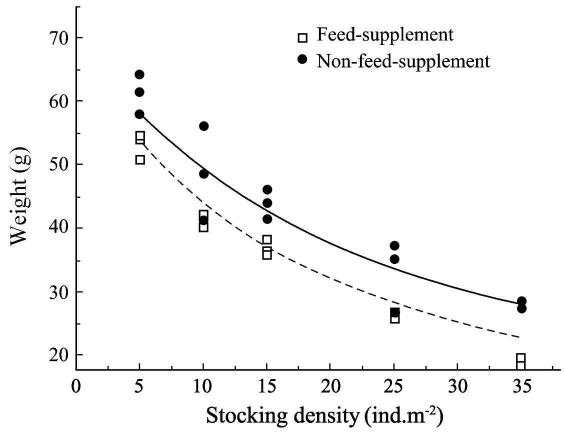

Fig.4 Relationship between body weight and stocking density of sea cucumbers cultured under different stocking densities and feeding regimes, fitted with the m-N model.

The high coefficient between stocking density and animal biomass indicated that stocking density could affect biomass (Table 2).

4 Discussion and Conclusions

4.1 Growth and Survival Rate

Aestivation and hibernation are important adaptive mechanisms of the sea cucumber *Apostichopus japonicus* in response to high and low temperatures, respectively (Liu *et al.*, 1996; Yang *et al.*, 2006). Characteristics of aestivation in *A. japonicus* include fast gut tract degeneration, weight loss and metabolic rate depression (Ji, *et al.*, 2008; Yang, *et al.*, 2006). In the present study, sea cucumbers lost about 40% of their body weight at the end of aestivation, resulting in negative specific growth rates (Table 3). Such results are consistent with those of Yang *et al.* (2006). At the autumn phase, growth of sea cucumbers decreased with increasing stocking density ($P<0.05$), and the SGR decreased from 3.49% d^{-1} (5 ind. m^{-2}) to 1.12% d^{-1} (35 ind. m^{-2}) ($P < 0.05$) under the non-feed-supplement regime (Table 3 and Fig.2) and from 3.43% d^{-1} (5 ind. m^{-2}) to 2.23% d^{-1} (35 ind. m^{-2}) ($P<0.05$) under the feed-supplement regime. For the same stocking density, the SGR of the sea cucumbers under feed-supplement regime were higher than those under the non-feed-supplement regime ($P < 0.05$) (Table 3), indicating that food availability is an essential factor in determining the growth performance of sea cucumbers in the present study. Hence, it might be speculated that the reduced growth rates of sea cucumbers with increasing stocking density under supplementary feeding regime is due to limited food supply. These results indicate that food was a limiting factor for the growth of sea cucumber reared at high stocking intensity. The constants of a and b in the m-N model under the feed-supplement regime were larger than those under non-feed-supplement regime, suggesting that the abundance of food is improved by supplementary feeding. We did not investigate the benthic (sediment) conditions of the culture pond in the present study. However, results of a different study carried out in the same culture pond by Ren *et al.* (personal comm.) showed that the organic carbon loadings of the sediment in the culture pond decreased significantly with increasing stocking density, suggesting limitations of food supply at higher stocking densities.

As reported in some previous studies, stocking densities could affect animal's survival rate, with survival rate often decreasing with an increase in stocking densities (Pomerleau and Engle, 2003; Rahmana *et al.*, 2005; Rowlanda *et al.*, 2006; Yi and Lin, 2001). In the present study, stocking density did not significantly affect survival rate of the sea cucumbers ($P>0.05$) in the autumn phase (Table 2), which might be due to the larger experimental animals used here (Battaglene *et al.*, 1999; Slater *et al.*, 2007). At the end of the spring phase (study), survival rate of the sea cucumbers in the feed-supplement treatments was significantly higher than that in the non-feed-supplement treatments ($P < 0.05$). Chang *et al.* (2004) found that the survival rate of sea cucumbers was 10%–35% at body length of 2–5 cm, 30%–80% at body length of 5–10 cm, and as high as 90% at body length of over 10 cm. The body length of sea cucumbers used was about 5 cm at the beginning of the present study, and the survival rate (Table 2) was within the range reported by Chang (2004). On August 10, 2007 and September 26, 2007, an acute decrease of salinity occurred due to heavy rains. This could affect the survival of sea cucumber significantly because *A. japonicas* is known to be sensitive to low salinity (Chen *et al.*, 2007; Xiao *et al.*, 2004; Yuan *et al.*, 2006). Thus, it is important to prevent the incidence of freshwater runoff to the culture area to ensure high survival. The mean survival rate under non-feed-supplement regime (28.9%) was lower than that under feed-supplement regime (30.4%) ($P < 0.05$), indicating that supplemental feeding is important for improving the survival rate of sea cucumbers after their aestivation and hibernation.

4.2 The Optimal Stocking Density

There are different definitions on optimal stocking densities. Yang *et al.* (1998) introduced a comprehensive efficiency index to evaluate the optimal stocking density and polyculture ratio. Fréchette *et al.* (1996 and 2005) considered that the (total) yield (B) controlled the population density (N) at the beginning of the experiments and the B-N curves may allow estimation of self-thinning functions. The optimal stocking density adopted in the present study was the stocking density at which the maximum net yield was obtained (Fréchette and Bacher, 1998). The relationship of net yield and stocking densities in this study could be described as follows (Fig.5):

$$Bn = -0.21N^2 + 9.37N + 35.07 \quad \text{(feed-supplement)};$$

$$Bn = -0.145N^2 + 4.08N + 37.2 \quad \text{(non-feed-supplement)}.$$

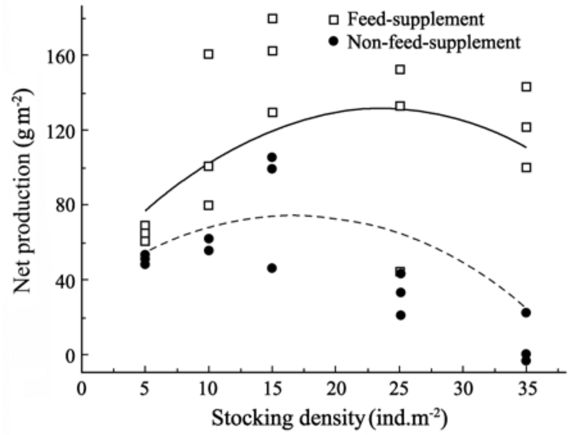

Fig.5 Relationship between net yield and stocking density of sea cucumbers, cultured under different stocking densities and feeding regimes, fitted with the Bn-N model.

It is therefore deduced from the Bn-N curves that the optimal stocking densities of sea cucumbers (with an initial body weight of 5 g $ind.^{-1}$) were 22.3 ind. m^{-2} and 14.1 ind. m^{-2} under feed-supplement and non-feed-supplement regimes, respectively. Considering the possible influence of the enclosures on water exchange, these optimal stocking densities estimated are likely to be underestimated slightly. On the other hand, the present study considered the optimal stocking density only from the net yield point of view. In practice, factors such as body size and the size-related price of the product should also be involved. Thus, prolonged culture period or relatively low stocking density should be implemented to yield larger size and therefore high price animals.

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