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Effect of Reducing the Conventional Feeding Rate on Water Quality and Shrimp Production Variables in *Penaeus monodon* Shrimp Culture with Zero Water Exchange Model Using Molasses as Carbon Source

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Abstrak

Dibudidaya perairan terutama untuk budidaya ikan telah diperlihatkan bahwa penggunaaan karbon membawa manfaat dari sisi kualitas air, produksi ikan dan peningkatan dalam biomassa bakteri. Biomassa bakteri digunakan untuk mengantikan sebagian kebutuhan makanan ikan sehingga dapat menghemat biaya dan menguntungkan bagi petani. Tujuan studi ini adalah untuk mengevaluasi pengaruh pengurangan jumlah pakan konvensional terhadap variable kualitas air dan produksi udang dalam budidaya udang Penaeus monodon *dengan sistem tanpa pergantian air menggunakan molasses. Hasil penelitian menunjukkan bahwa penggunaan molasses dengan level ratio C:N = 20.0:1 dan pemberian pakan 75 % dari jumlah pakan konvensional merupakan pola yang sangat menjanjikan bagi system budidaya tanpa pergantian air sebab perlakuan tersebut dapat meningkatkan biosecurity, mengurangi biaya pakan, limbah dan penggunaan air.*

Kata kunci: *Jumlah pakan,kualitas air,model tanpa pergantian air, molasses*

Abstract

In aquaculture particularly for fishes culture it was shown that the application of carbon had brought about good benefits in terms of water quality, fishes production variables and increase in bacterial biomass. Moreover, bacterial biomass used to replace part of the feed requirement of fishes resulting a considerable cost saving and the boasting profit for the farmer. Therefore, the aim of this study was to evaluate the *effects of reducing conventional feeding rate on water quality and shrimp production variables within* Penaeus monodon *shrimp culture with zero water exchange model using molasses. Study shows that using molasses with the level of C: N ratio = 20.0:1 at 75 % of conventional feeding rate is the most promising features of zero water exchange culture system because it offers increased biosecurity, reduced feed costs, waste and water use.*

Key words: *Feeding rate, water quality, zero water exchange model, molasses*

Introduction

Controlling inorganic nitrogen by manipulating the carbon: nitrogen ratio is a potential control method for aquaculture systems (Avnimelech, I999) through growing heterotrophic bacteria (Burford *et al*., 2003; Burford & Lorenzen, 2004). The addition of carbon material can reduce inorganic nitrogen in fish experimental containers (Avnimelech, I999), simultaneously produce single cells protein for fish (Avnimelech & Mokady I988; Avnimelech *et al*., I989; Kirchman *et al*., 2000), reduce feeding and pumping costs (Avnimelech *et al*., 1992; 1994; Kochva *et al*., 1994; Avnimelech, I999; Decamp *et al.,* 2003; Lemmonier et al., 2003). The sources of carbon matter used in ponds can be sorghum and wheat meal (Avnimelech *et al.,* I994). Further Stuart *et al*. (2009) raised tiger shrimp *Penaeus monodon* in zero water exchange model using a daily carbon source (tapioca powder) to promote the microbial community and improve water quality

Rosenberry (2001) revealed that bacteria grown up in shrimp pond with zero water exchange model consume shrimp waste products and shrimps in turn consume them. Furthermore, heterotrophic communities in zero water exchange shrimp pond developed flocs of composing bacteria cells (McIntosh, 2000; Chamberlain, 2001), and flocculation of the cells can be alone or in combination with clay or feed particles (Harris & Mitchell, I973; Avnimelech *et al*., I989). Flocs containing high protein, amino acids and

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certain microelements, can be directly consumed by ommivorus shrimp like white shrimps (McIntosh *et al*., 2000; Lopez *et al*., 2002; Tacon *et al*., 2002). Consumption of these flocs by shrimps (or fishes, see Schroeder, I978) contributed to both the nutrition of the shrimp and efficient recycling of pond nutrient into shrimp biomass (McIntosh, 2000; 2001; Chuntapa, 2003; Thakur & Lin, 2003). Rosenberry (2001) stated that zero water exchange system produced ten times higher white shrimps than typical semi-intensive ponds and forty times higher white shrimps than typical extensive ponds.

Avnimelech *et al.* (1994) observed that the consumption of bacterial biomass is significant since it is possible to reduce the feeding rate by 66 % of control without reduction in the growth rate of fishes. Therefore, there is a substantial questions which can be answered in this study: Is it possible to reduce feeding rate of *Penaeus monodon* shrimps by applying molasses without detrimental effect on shrimps

The aim of this study was to evaluate the effects of reducing conventional feeding rate on water quality, shrimp growth rate, percentage weight gain and feed conversion ratio of shrimp culture with zero water exchange model using molasses.

Materials and Methods

The experimental shrimp culture

This study were composed of two experiments carried out at the aquaculture outdoor laboratory of Charles Darwin University, Australia. Experiment one was carried out for smaller shrimps (RFS) and treatments evaluated in the experiment one were: without molasses with 100% conventional feeding rate ($RFSC_{100}$); using molasses with 25% of conventional feeding rate (RFS_{25}); using molasses with 50% of conventional feeding rate (RFS_{50}); using molasses with 75% of conventional feeding rate (RFS_{75}); and using molasses with 100% of conventional feeding rate (RFS_{100}). While experiment two was conducted for bigger shrimps (RFB) and treatments evaluated were without molasses with 100 % of conventional feeding rate ($RFBC_{100}$), using molasses with 25 % of conventional feeding rate (RFB_{25}), using molasses with 50 % of conventional feeding rate (RFB_{50}), using molasses with 75% of conventional feeding rate (RFB_{75}), using molasses with 100% of conventional feeding rate (RFB_{100}).

The both experiments were carried out in 160 litre tanks. All treatments had three replicates and allocation for each treatment was completely randomized. Experimental tanks were mixed and aerated with two pieces of air stone suspended in bottom of water column in each tank. The all tanks were filled with 160 litres of sea water and there was no water exchange during the study. The volume of water in each tank was maintained constant by adding 2 litres of freshwater (tap water) weekly to replace lost water due to evaporation. Water temperatures and salinity in experiment one for shrimp growing experiments ranged 21.7-24.9° C and 7.50-28.70 ppt respectively. While water temperatures and salinity in experiment two ranged 26.4-30.9°C and 27.00-33.50 ppt respectively. The level of salinity and temperature in experiment one was lower than in experiment two because experiment one was conducted in rain season while experiment two was conducted in dry season.

Each experimental culture was stocked with 4 *Penaeus monodon* shrimps or equal to 30 shrimps m^{-2} (Allan & Manguire, 1992) and the averages of initial shrimp weight stocked in experiment one and two were 0.359±0.033 gram and 9.902±0.169 gram respectively. Shrimps were obtained from nursery of Charles Darwin University, Australia. The feed used was commercial feed (Taiwan Company Product) with 38 % crude protein. The conventional feeding rate used in experiment one was 10 % of body weight per day until the first six week and 7.5 % for the rest of experiment and in experiment two was 5 % of body per day for the whole period (as recommended by the feed company). It was applied three time daily (at 06.00;12.00 am and 06.00 pm). The amount of molasses (2.669 gram per gram feed) daily was added to bring the feed C: N ratio to 20.0:1 (McIntosh, 2000). Application of molasses was conducted once daily at 06.00 am.

Water quality measurements and analyses

Salinity, temperature, pH and dissolved oxygen in water were measured using a Horiba water quality checker (U-10 Model). Before measurement of those parameters, the Horiba water quality checker was manually calibrated as described in the instruction manual.

Ammonia, nitrite and nitrate concentrations in water were measured photometrically using a Palintest Photometer, based on indophenols method, diazotization method, and cadmium reduction/ diazotization method for ammonia, nitrite and nitrate respectively.

Levels of viable heterotrophic bacteria were determined by counting the colonies which grew on plates of Tryptone Soya Agar (TSA) with 10 % of NaCl (Jorgensen *et al.,* 1993). Before plating each sample onto agar medium, serial dilutions were made in physiological saline solution composed of 9 % NaCl (Sohier and Bianchi, 1985). Levels of bacteria are quoted in colony forming units per ml of water (CFU ml^{-1}) (Smith, 1998).

Shrimp survival rate, growth, percentage weight gain and feed conversion ratio

Experiment of shrimp cultures in laboratory was conducted for eight-week duration. Every two weeks, the total body weight of shrimp (W) was measured for each experimental container. Similarly, the number of live shrimps (N) in each tank was counted. Further, the amount of feed used in each tank (Wf) was recorded. The average body weight (W_a) were calculated by dividing W by N. The overall average values of survival rate (%), growth rate of shrimp (gram/day), percentage weight gains (%), and feed conversion ratios (FCR) as used in common aquaculture studies (Tseng *et al*., 1998).

Data analysis

Data of laboratory study were analysed using Statistica Version 6.1 software and with one-way ANOVA (Steel & Torrie, 1980) to evaluate the effects of each treatment. The homogeneity of variance and normality of all data sets were tested using Cochran's test. Tukey test was used to differentiate among the treatment means of each experiment after ANOVA analysis (Steel & Torrie, 1980).

Results and Discussion

Responses of ammonia and nitrite concentrations to reducing conventional feeding rate are shown in Tables 1 and 2. The results show that addition of molasses have had pronounced effects on levels of ammonia, nitrite and nitrate. These water quality variables remained low and the same in all treatments with molasses regardless of feeding rates and size of shrimps. Other wise, the treatments without using molasses with 100 % of conventional feeding rate in experiment one ($RFSC_{100}$) and experiment two ($RFBC_{100}$) had significantly highest level of ammonia and nitrite. Regression analysis also reveals that concentrations of ammonia at the end of experiment were negatively correlated with numbers of heterotrophic bacteria in experiment one. This result was due to the fact that application of molasses with C: N ratio level was 20.0:1 had effectively removed inorganic nitrogen from the culture medium by heterotrophic bacteria . These results are similar to those obtained in the previous studies by several authors (e.g. Tezuka, 1990; Anvimelech *et al*., 1992; 1994; Avnimelech, 1999; McIntosh, 2000). They proved that the feed C:N ratio level of 15.0:1 or more effectively removed inorganic nitrogen from water.

Concentrations of ammonia and nitrite in the treatments without molasses in experiment one were lower than that of experiment two and this could be due to the application of higher feed amount in experiment two to compensate the larger size of shrimp. The higher amount of feed applied led to the higher production of inorganic nitrogen. Montoya *et al*. (2002) reported that the concentration of inorganic nitrogen such as ammonia and nitrite increased as feeding rate inclined.

Concentrations of dissolved oxygen in the two experiments were significantly affected by reducing conventional feeding rate as shown in Tables 1 and 2. In the treatments using molasses, those were a significantly increased dissolved oxygen levels in response to reducing feeding rate in the both experiments and concentrations of dissolved oxygen were significantly highest in treatments without using molasses in both experiments. This could be due to the increase in numbers of heterotrophic bacteria in response to increase feeding and molasses. It has been earlier reported that dissolved oxygen decreased as increasing feeding rates of shrimp (Chieng *et al*., 1989; Hopkins *et al*., 1991). Heterotrophic bacteria had a great contribution in a reduction of disolved oxygen concentration in shrimp culture water (Visscher & Duerr, 1991; Sun *et al*., 2001).

Furthermore, dissolved oxygen concentrations were inversely related to numbers of heterotrophic bacteria in experiment one. Dissolved oxygen levels reduced with experiment time in the both experiments. This evident can be supported by earlier studies proved that the concentrations of dissolved oxygen had a significant negative correlation with the numbers of heterotrophic bacteria (Sun *et al*., 2001; Montoya *et al*., 2002).

Values of pH reduced significantly with increasing feeding rate in the both experiments (Table 1 and 2) and levels of pH in experiments untreated with molasses were lowest in the both experiments and this trend could due to the highest activity of nitrifying bacteria which was indicated by the highest concentration of nitrite and nitrate. It has been stated already that oxidizing each mole of ammonia released two hydrogen ions which eventually reduced pH (Hargreaves, 1998; Tacon, 2001). Also Tacon et al. (2002) reported that an increase in nitrite caused decreased pH in shrimp culture.

Table 1. The effect of reducing conventional feeding rate of smaller shrimps (experiment one) on several water quality and shrimp production variables within shrimp culture with ZWEM using molasses daily with feed C:N ratio level = 20.0:1. Values are means and standard deviations of three replicates at the of the eight-week experimental period.

Water Quality Variables	Treatment	Mean		Std. Dev
1. Ammonia (mg/litre)	1. No Molasses with 100 % of Feeding Rate	0.3174	±	0.0253^{a}
	2. Molasses with 25 % of Feeding Rate	0.0397	±	0.0037^{b}
	3. Molasses with 50% of Feeding Rate	0.0457	±	0.0023^{b}
	4. Molasses with 75 % of Feeding Rate	0.0499	±	0.0089^{b}
	5. Molasses with 100% of Feeding Rate	0.0530	±	0.0025^{b}
2.Nitrite (mg/litre)	1. No Molasses with 100 % of Feeding Rate	36.0768	±	0.0005^{a}
	2. Molasses with 25 % of Feeding Rate	0.0000	±	0.0000^{b}
	3. Molasses with 50% of Feeding Rate	0.0000	±	0.0000^{b}
	4. Molasses with 75 % of Feeding Rate	0.0000	±	0.0000^{b}
	5. Molasses with 100% of Feeding Rate	0.0000	±	0.0000^{b}
3.Dissolved Oxygen (mg/litre)	1. No Molasses with 100 % of Feeding Rate	6.13	±	0.06^{a}
	2. Molasses with 25 % of Feeding Rate	5.54	±	0.17^{b}
	3. Molasses with 50% of Feeding Rate	5.21	±	0.07^{c}
	4. Molasses with 75 % of Feeding Rate	4.89	±	0.07^{d}
	5. Molasses with 100% of Feeding Rate	4.71	±	0.03^{d}
4.pH	1. No Molasses with 100 % of Feeding Rate	7.56	±	0.08^{a}
	2. Molasses with 25 % of Feeding Rate	8.16	±	0.06^{b}
	3. Molasses with 50% of Feeding Rate	8.00	±	0.01^{c}
	4. Molasses with 75 % of Feeding Rate	7.82	±	0.02^{e}
	5. Molasses with 100% of Feeding Rate	7.74	±	0.03^{e}
5.Number of Heterotrophic Bacteria (CFU/ml)	1. No Molasses with 100 % of Feeding Rate	$1.85 X10^{9}$	±	$5.50 X10^{7}$ ^{a}
	2. Molasses with 25 % of Feeding Rate	$7.33 X10^{9}$	±	$4.53 X10^{8}$ ^{b}
	3. Molasses with 50% of Feeding Rate	$8.84 X10^{9}$	±	$3.28 X10^{8}$ ^{c}
	4. Molasses with 75 % of Feeding Rate	$1.04 X10^{10}$	±	$9.94 X10^{8}$ ^{d}
	5. Molasses with 100% of Feeding Rate	$1.21 X10^{10}$	±	$2.65 X10^{8}$ ^{e}
6. Shrimp Survival Rate (%)	1. No Molasses with 100 % of Feeding Rate	100.00	±	0.00^{a}
	2. Molasses with 25 % of Feeding Rate	100.00	±	0.00^{a}
	3. Molasses with 50% of Feeding Rate	100.00	±	0.00^{a}
	4. Molasses with 75 % of Feeding Rate	100.00	±	0.00^{a}
	5. Molasses with 100% of Feeding Rate	100.00	±	0.00^{a}
7.Shrimp Growth Rate (gram/day)	1. No Molasses with 100 % of Feeding Rate	0.030	±	0.005^{a}
	2. Molasses with 25 % of Feeding Rate	0.014	±	0.004^{b}
	3. Molasses with 50% of Feeding Rate	0.037	±	0.006^{c}
	4. Molasses with 75 % of Feeding Rate	0.052	±	0.001^{d}
	5. Molasses with 100% of Feeding Rate	0.040	±	0.001^{e}
8.Percentage Weight Gain (%)	1. No Molasses with 100 % of Feeding Rate	313.149	±	22.156^{a}
	2. Molasses with 25 % of Feeding Rate	230.088	±	58.911^{b}
	3. Molasses with 50% of Feeding Rate	500.720	±	64.277^{c}
	4. Molasses with 75 % of Feeding Rate	892.326	±	55.200^{d}
	5. Molasses with 100% of Feeding Rate	698.340	±	32.954^{e}
9.Feed Conversion Ratio	1. No Molasses with 100 % of Feeding Rate	3.521	±	0.178^{a}
	2. Molasses with 25 % of Feeding Rate	0.770	±	0.194^{b}
	3. Molasses with 50% of Feeding Rate	1.140	±	0.085^{c}
	4. Molasses with 75 % of Feeding Rate	1.238	±	0.074^{d}
	5. Molasses with 100% of Feeding Rate	1.998	±	0.145^{e}

Values Note : Values in every water quality variable within the same column that are followed by different superscript are significantly different ($p < 0.05$). Equality of variance and normality of each water quality variable level were checked by Cochran's test.

Table 2. The effect of reducing conventional feeding rate of bigger shrimps (experiment two) on several water quality and shrimp production variables within shrimp culture with ZWEM using molasses daily with feed C:N ratio level = 20.0:1. Values are means and standard deviations of three replicates at the of the eight-week experimental period.

Water Quality Variables	Treatment	Mean	Std. Dev
1. Ammonia (mg/litre)	1. No Molasses with 100 % of Feeding Rate	0.3847	± 0.0031[a]
	2. Molasses with 25 % of Feeding Rate	0.0329	± 0.0035[b]
	3. Molasses with 50% of Feeding Rate	0.0432	± 0.0008[b]
	4. Molasses with 75 % of Feeding Rate	0.0481	± 0.0028[b]
	5. Molasses with 100% of Feeding Rate	0.0491	± 0.0020[b]
2.Nitrite (mg/litre)	1. No Molasses with 100 % of Feeding Rate	47.7061	± 0.3778[a]
	2. Molasses with 25 % of Feeding Rate	0.0617	± 0.0002[b]
	3. Molasses with 50% of Feeding Rate	0.0750	± 0.0001[b]
	4. Molasses with 75 % of Feeding Rate	0.0820	± 0.0002[b]
	5. Molasses with 100% of Feeding Rate	0.0873	± 0.0001[b]
3.Dissolved Oxygen (mg/litre)	1. No Molasses with 100 % of Feeding Rate	5.22	± 0.01[a]
	2. Molasses with 25 % of Feeding Rate	4.81	± 0.04[b]
	3. Molasses with 50% of Feeding Rate	4.74	± 0.02[c]
	4. Molasses with 75 % of Feeding Rate	4.56	± 0.01[d]
	5. Molasses with 100% of Feeding Rate	4.54	± 0.01[d]
4.pH	1. No Molasses with 100 % of Feeding Rate	7.16	± 0.03[a]
	2. Molasses with 25 % of Feeding Rate	7.77	± 0.03[b]
	3. Molasses with 50% of Feeding Rate	7.63	± 0.02[c]
	4. Molasses with 75 % of Feeding Rate	7.57	± 0.01[e]
	5. Molasses with 100% of Feeding Rate	7.42	± 0.02[e]
5.Number of Heterotrophic Bacteria (CFU/ml)	1. No Molasses with 100 % of Feeding Rate	2.22 X10^{9}	± 6.60X10^{7} [a]
	2. Molasses with 25 % of Feeding Rate	8.79 X10^{9}	± 5.44 X10^{9} [b]
	3. Molasses with 50% of Feeding Rate	1.03 X10^{10}	± 2.21 X10^{8} [c]
	4. Molasses with 75 % of Feeding Rate	1.24 X10^{10}	± 6.43 X10^{8} [d]
	5. Molasses with 100% of Feeding Rate	1.45 X10^{10}	± 3.18 X10^{8} [e]
6. Shrimp Survival Rate (%)	1. No Molasses with 100 % of Feeding Rate	100.00	± 0.00[a]
	2. Molasses with 25 % of Feeding Rate	100.00	± 0.00[a]
	3. Molasses with 50% of Feeding Rate	100.00	± 0.00[a]
	4. Molasses with 75 % of Feeding Rate	100.00	± 0.00[a]
	5. Molasses with 100% of Feeding Rate	100.00	± 0.00[a]
7.Shrimp Growth Rate (gram/day)	1. No Molasses with 100 % of Feeding Rate	0.178	± 0.003[a]
	2. Molasses with 25 % of Feeding Rate	0.085	± 0.006[b]
	3. Molasses with 50% of Feeding Rate	0.188	± 0.006[c]
	4. Molasses with 75 % of Feeding Rate	0.314	± 0.005[de]
	5. Molasses with 100% of Feeding Rate	0.318	± 0.004[de]
8.Percentage Weight Gain (%)	1. No Molasses with 100 % of Feeding Rate	101.366	± 1.497[a]
	2. Molasses with 25 % of Feeding Rate	47.991	± 3.314[b]
	3. Molasses with 50% of Feeding Rate	104.221	± 2.173[c]
	4. Molasses with 75 % of Feeding Rate	179.382	± 1.277[de]
	5. Molasses with 100% of Feeding Rate	178.599	± 1.873[de]
9.Feed Conversion Ratio	1. No Molasses with 100 % of Feeding Rate	3.694	± 0.036[a]
	2. Molasses with 25 % of Feeding Rate	1.610	± 0.093[b]
	3. Molasses with 50% of Feeding Rate	1.747	± 0.015[c]
	4. Molasses with 75 % of Feeding Rate	1.709	± 0.033[d]
	5. Molasses with 100% of Feeding Rate	2.289	± 0.032[e]

Note : Values in every water quality variable within the same column that are followed by different superscript are significantly different ($p < 0.05$). Equality of variance and normality of each water quality variable level were checked by Cochran's test.

Allan *et al.* (1995) found that there was a decrease pH with increasing feeding rate due to the decomposition of excess feed. The present study also shows there was a significant reduction in pH values with enhancing feeding rate in shrimp cultures treated with molasses.

With the addition of molasses, numbers of heterotrophic bacteria were augmented with increasing conventional feeding rate in the both experiments whereas in treatments using no molasses but with 100 % of feeding rate had lowest numbers of bacteria in both experiments as shown in Table 1 and 2. This evident was because higher amount of molasses led to higher numbers of heterotrophic bacteria in shrimp cultures and treatments with higher feeding rate always received higher amount of molasses. The same result has been investigated by several investigators (Parson *et al.,* 1981; Avnimelech *et al.,* 1992; 1994; Kochva *et al.,* 1994; Middleboe *et al.*, 1995; Avnimelech , 1999) who reported that addition of carbon in water created a increase in number of heterotrophic bacteria.

Levels of shrimp survival were (100%) in the all treatments of the two experiments and were unaffected by reducing the conventional feeding rate. Whereas reducing conventional feeding rate had a significant effect on the growth rates, percentage weight gains and feed conversion ratios in the both experiments as described in Table 1 and 2.

Treatments with 25 % of conventional feeding rate in the two experiments had significantly lowest growth rate and percentage weight gain among treatments. Likewise, in the two experiments using molasses, levels of growth rate and percentage weight gain were lower in treatments with 50 % of conventional feeding rate compared with 75 % and 100 % of feeding rate treatments. These results reveal that using 25 % and 50 % of conventional feeding rate was insufficient for shrimp growth, despite application molasses with the level of C:N ratio = 20 was conducted in those treatments. These results imply that those treatments show underfed shrimp. This view was supported by daily observation shown that no uneaten feed found in shrimp culture treated with 25 and 50 % ofmconventional feeding rate. These findings also indicates that natural food including heterotrophic bacteria was incapable supplementing reducing feeding rate of 50 or more in sustaining shrimp growth.

Interestingly, in the two experiments, there was a decrease in feed conversion ratios in response to decreasing feeding rate. Therefore, differences in values of feed conversion ratio among the test treatments reflect feed consumption rather than feed quality, as the feed used was the same. It was stated that feed conversion ratio decrease as the amount of feed decrease and increases with an increase in the amount of feed fed (Halver, 1989). Similarly, Abdelghany & Ahmad (2002). pointed out that feed conversion ratio of fish was lower in treatment with lower feeding rate. This result implies that efficiency of shrimp culture should not be evaluated by only feed conversion but also by growth rate and percentage weight gain.

Further the treatment without using molasses with 100 % of conventional feeding rate had significantly lower growth rate and percentage weight gain and higher feed conversion ratio compared to treatments using molasses with 50 % or more of conventional feeding rate. Furthermore, daily observation shown that the remain of unfed feed was relatively high in treatment without using molasses at 100 % of conventional feeding rate particularly after week six it was likely to be associated with stressed level in shrimp due to high concentration of nitrite.

Numbers of heterotrophic bacteria in treatments without using molasses which were lower compared to those obtained in treatments using molasses could be one explanation for lower growth rates and percentage weight gains and higher feed conversion ratios in treatments with no molasses than treatments using molasses receiving 50 % or more of conventional feeding rate. Further investigation shown that numbers of heterotrophic bacteria were positively correlated with growth rates and percentage weight gains and were negatively with feed conversion ratio. The product of microbial protein has been proved to be an efficient source of proteins for fish (Avnimelech & Mokady, 1988; Avnimelech *et al.*, 1989). Also Avnimelech *et al.* (1994) reported that growth rate of fish was higher in treatment with lower conventional feeding rate using carbon source compared to those obtained in treatment with higher of conventional feeding rate with not using carbon source.

The results strongly suggest that bacterial flocculation can be used as feed for shrimps and can be partially replaced the feed in RFS_{75} and RFB_{75} treatments, although the present study did not prove directly use of bacterial flocculation by shrimps. It has been reported by several authors (Harris & Mitchell, 1973; Schroeder, 1978; Avnimelech *et al.*, 1982) who revealed that flocculation of heterotrophic was consumed by fish and had significant contribution in fish production. Likewise, Tacon *et al.* (2002) reported that the growth rates of white shrimp in zero water exchange model was very high due to the endogenous production and availability of microbial food organisms (bacterial flocculation). Bacterial flocculation constituted high amino acids such as threonine, valine, isoleucine and phenylalanine which is required by shrimps (Tacon *et al.,* 2002).

Conclusions

Based on evaluation of shrimp growth rate, percentage weight gain, feed conversion ratio and water quality variables, the present study clearly shows that the best treatment in the shrimp culture of zero water exchange model for *Penaeus monodon* was the treatment using molasses with 75 % of conventional feeding rate at the level of feed C: N ratio = 20.0:1.

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