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TEKNOLOGI PERTANIAN

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Diterbitkan oleh :
Kedeputian Bidang Pengkajian Kebijakan Teknologi
Badan Pengkajian dan Penerapan Teknologi
Jakarta

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Berdasarkan Surat LIPI No. 2585/V.2/KP/96, tanggal 3 Mei 1996, Majalah Analisis Sistem diklasifikasikan sebagai Majalah Ilmiah

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Masalah pakan dari hasil produksi pertanian, menjadi masalah pokok yang perlu diperdayakan di masyarakat. Hal ini karena sebagian besar masyarakat bermata pencaharian sebagai petani dan sektor pertanian umumya lebih dominan memanfaatkan sumberdaya lokal.

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KATA SAMBUTAN

Puji dan syukur kita panjatkan kehadirat Allah SWT, karena berkat rahmat dan hidayah-Nya, maka Edisi Khusus Majalah Analisis Sistem yang mengambil topik utama "Teknologi Pertanian" ini dapat diterbitkan. Saya menyambut gembira penerbitan ini karena sektor pertanian menjadi salah satu prioritas utama riset dan teknologi di Indonesia dan diharapkan menjadi andalan dalam ketahanan pangan. Konsekwensi dari pilihan tersebut adalah bahwa berbagai upaya perlu dikerahkan untuk mendukung pembangunan sektor pertanian. Pusat Pengkajian dan Penerapan Teknologi Budidaya Pertanian, sesuai mandat dan kompetensinya telah berusaha untuk memberikan kontribusinya. Salah satu bentuk kontribusi yang berharga adalah penerbitan berbagai hasil kajian di bidang Teknologi Pertanian dalam bentuk tulisan ilmiah yang di wadahi dalam Majalah Analisis Sistem.

Keberhasilan pembangunan sektor pertanian sangat tergantung pada keberhasilan kita dalam mengembangkan dan menerapkan teknologi yang tepat melalui pendayagunaan sumber daya yang dimiliki. Arah pengembangan teknologi tersebut adalah pada upaya intensifikasi di Pulau Jawa, ekstensifikasi untuk kawasan luar Jawa, peningkatan inovasi teknologi, dan diversifikasi produk. Pemanfaatan teknologi tersebut diharapkan menghasilkan berbagai luaran yang akan dapat (1) meningkatkan produktivitas, (2) meningkatkan efisiensi dengan menghemat atau menurunkan faktor produksi, dan (3) meningkatkan kualitas hasil dan nilai tambah produk sehingga meningkatkan daya saing produk, sesuai dengan kebutuhan dan permintaan pasar.

Akhirnya saya ingin menyampaikan apresiasi yang tinggi atas upaya untuk memasyarakatkan hasil-hasil kajian di bidang teknologi pertanian, baik dalam aspek budidaya maupun agroindustri, dalam Edisi Khusus Majalah Analisis Sistem ini.

Semoga penerbitan ini dapat memberikan manfaat yang sebesar-besarnya bagi pengembangan khazanah ilmu pengetahuan dan teknologi di bidang pertanian dan agroindustri.

Jakarta, Desember 2004
Direktur P3 Teknologi Budidaya Pertanian,

Ir. Iding Chaidir, M.Sc.

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BLOOD MEAL AS PROTEIN SOURCE FOR *Marsupenaeus japonicus* JUVENILES

Oleh : *Ophirtus Sumule* *) dan Agung Sudaryono **)

ABSTRACT

The use of blood meal (BM) as protein source for Kuruma shrimp Marsupenaeus japonicus juveniles (0.9 g initial wet weight) was determined by supplementing BM in the diet at 0% (control diet), 6% , 12%, 18% and 24% with reduced fishmeal content from 30% to 0%. These diets were made isoenergetics and isonitrogenoeus by changing lipid and carbohydrate levels. These diet were fed to animal test for 50 days in triplicate per diet (15 shrimp per replicate) once a day at 4 to 6% BW Apparent digestibility of dry matter, protein and lipid were measured after determination of feeding trial. The weight gain (BWG) and specific growth rate (SGR) of shrimp fed BM-75 and BM-100 were similar but lower compare to the groups fed control diet, CBM-25 and CBM-50. Protein and lipid digestibility of diet up to 50% CBM was similar to control diet and statistically higher compare to diet CBM 75 and CBM100. This study concluded fishmeal in practical diet of M. japonicus juvenile can be replaced about 50% by bloodmeal without adverse in growth and digestibility

I. INTRODUCTION

The development of commercial aquaculture feeds has been traditionally based on fishmeal (FM) as the main protein source due to its high content of essential amino acids and fatty acids, which are usually well digested for these macro-nutrients and provide vitamins and minerals (El-Sayed 1999). With the increasing world population and increased fishing pressure, the global production of FM has been in a state of decline in the last decade (Starkey, 1994). The shortage is coupled with increased demand for man and livestock which cause the price of FM to substantially increase (Tacon, 1998). It is evident, on the long-run, that many developing countries will be unable to depend on FM as a major protein source in aquafeeds. Therefore, it is important to look for alternative protein sources which are less expensive and locally available for the development of cost-effective of aquafeeds. The search for alternative sources for fishmeal is an international research priority (Manzi, 1989; Hardy and Kissil, 1997).

Blood meal (BM) is a promising ingredient for replacing FM due to the relatively high protein content, high digestibility and predicted to be stable in supply since blood animal is only being discarded as wastes from slaughtering houses. Unfortunately, blood meal contains some limiting nutritional factors, such as isoleucine and methionine (NRC, 1992, Tacon and Jackson, 1985). However, if the proper ration between this product and other source of protein is maintained in the diet, the Essential Amino Acids (EAA) imbalance could be overcome and the quality of such diet is likely to improve (Davies et al. 1989). Several studies have been conducted and using blood meal as a part of diet ingredients for teleost fish such as tilapia, (Elsayed, 1999), grouper (Millamena, 2002), rainbow trout (Bureau, 1999), humpback grouper (Laining *et al*, 2003), rockfish (Lee, 2002) but none for *Marsupenaeus japonicus* shrimp.

In the present study, various levels of BM were included in practical formulated shrimp diets, while the level of fish meal was reduced from 100% to 0% as the main source of protein. Thus, the aim of this study was to evaluate the nutritional value of blood meal for kuruma shrimp , *M. japonicus*, which is one of the most important aquaculture crustaceans in the world

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II MATERIAL AND METHODS

A Test Diets

Blood meal was collected from a s mall slaughtering house in Central Java, Indonesia. The blood were coagulated by boiling for 15 minutes and than dried at room temperature. The coagulated blood was then sieved through a 150 μm mesh to get a homogenous BM particles. Five test diets containing 0% (control), 25% (BM-25), 50% (BM-50), 75% (BM-75) and 100% (BM-100) blood meal were formulated replacing FM.These diets were prepared to be isoenergetic and isocaloric based on gross energy content in each ingredients. The test diets were produced according to the method described by Sakakura *et al.* (1998) with a slight modification. Dry ingredients and lipids with fat-soluble vitamins were well mixed with distilled water using a food mixer (Model KMS, Kitchen Aid Inc.,St. Joseph, Michigan, US) and then extruded using a meat grinder (Royal, Japan; type 22VR-1500) with 1.2 mm diameter die. The diets were dried in the oven at a temperature 70°C for 1 hour, and were then steaming for 1.5 minutes to increase its melting surface. Diets were stored at 28°C, until use. Diet composition are listed in Table 1, while its proximate and amino acid profiles are presented in Table 2.

Table 1. Diet Compositions (g/kg diet) and Its Proximate Values

Ingredients	Dietary Treatments				
	BM-0	BM-25	BM-50	BM-75	BM-100
Fish meal	300	225	150	75	0
Cow blood meal	0	60	120	180	240
Defatted soybean meal	150	150	150	150	150
Squid meal	80	80	80	80	80
Krill meal	120	120	120	120	120
Pollack liver oil	70	73	75	78	80
Cholesterol	10	10	10	10	10
Soybean lecithin	30	30	30	30	30
Dextrin	50	50	50	50	50
α-Starch	50	50	50	50	50
Vitamin mix[*1)]	30	30	30	30	30
Mineral mix[*2)]	30	30	30	30	30
Gluten	50	50	50	50	50
Filler (α-cellulose)	30	42	55	67	80
Proximate composition					
Protein[*3)]	49.7	50.2	50.0	49.8	49.8
Lipid[*3)]	16.4	15.6	15.5	15.2	16.8
Moisture	6.2	8.1	8.8	7.8	9.1
Ash[*3)]	10.3	9.6	8.2	7.4	6.3

[*1)]Vitamin mix (mg/100 g diet) : ρ-Amino benzoic acid 6.77, Biotin 0.27, Inositiol 269.85, Nicotinic acid 26.98, Ca-pantothenate 40.48, Pyridoxine-HCl 8.098, Riboflavin 5.39, Thiamine-HCl 2.7, Menadione 2.70,β-Carotine 6.48, Alpha-Tochoperol 13.49, Cyanocobalamine 0.06, Calciferol 0.80, L-ascorbyl-2 phosphate-Mg 210.64, Folic acid 0.54, Choline chloride 404.77

[*2)]Meneral mix (g/100 g diet) : K_2HPO_4 0.23, $Ca_3(PO_4)_2$ 0.32, $MgSO_4.7\ H_2O$ 0.36, $NaH_2PO_4.2\ H_2O$ 0.09

[*3)] dry weight basis

Table 2. Amino Acid Composition of Experimental Diets (% of total amino acid analyzed)[a]

Amino acid	Diet				
	Control	**BM 25**	**BM 50**	**BM 75**	**BM 100**
EAA[b]					
Threonine	4.07	4.47	4.38	4.51	4.57
Valine	4.69	5.21	5.90	6.03	6.27
Methionine	2.14	1.80	1.43	0.66	0.76
Isoleucine	4.14	3.72	3.25	2.74	2.41
Leucine	7.71	8.09	8.87	9.42	9.75
Phenylalanine	4.66	4.74	5.21	6.52	6.40
Histidine	4.80	3.74	3.94	4.15	4.28
Lysine	7.8	7.60	8.02	8.42	8.35
Tryptophan	tr	tr	tr	tr	tr
Arginine	6.45	6.83	6.18	5.85	5.82
Σ EAA	46.46	46.20	47.18	48.3	48.61
NEAA[c]					
Taurine	0.64	0.53	0.39	0.36	0.16
Hydroxyproline	tr	tr	tr	tr	tr
Aspartic acid	8.56	9.26	8.99	9.12	9.33
Serine	4.23	4.64	4.54	4.79	4.84
Glutamic acid	20.36	19.68	19.19	18.06	17.63
Proline	5.45	5.47	5.37	5.28	5.20
Glycine	4.90	4.67	4.25	4.17	4.11
Alanine	5.28	5.37	5.90	5.96	6.14
Tyrosine	4.12	4.19	4.20	3.97	3.97
Σ NEAA	53.54	53.81	52.83	55.71	51.38
TOTAL	100	100	100	100	100

[a] means from duplicate homogenize samples
[b] essential amino acids
[c] non essential amino acid
tr : trace

B Feeding Trials

Shrimp juveniles (average wet weight 0.9g) were randomly selected from the holding tank and weighed 5 juveniles at a time. Fifteen juveniles per tank in triplicate per treatment were stocked in 30-L rectangular PVC tanks (37 x 25 x 23 cm) with sand-bottoms (3 cm in thickness), filled with filtered seawater (salinity 33 ppt, temperature 14-18°C) and aerated with air stones, cultured for 50 days. Filtered seawater was continuously supplied in flow through system at the rate of 0.1 liter/min through the bottom sand and net filter. The amount of water in each tank was maintained at 25-L. The feeding trial was conducted under 12 h dark: 12 h light photo period. The shrimp were fed once a day at 17:00 on a ration equal to 4-6%

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of BW/day for 50 days. The ration was adjusted after every 10 days after weighing of test animals. The tanks, sand and nets were cleaned when weight measurements are taken. Uneaten diets were siphoned out from the tank every morning and oven-dried at 100°C to constant weight for determination of actual diets intake. All shrimps were blotted on paper towels for 30 seconds to remove the excess water then weighed in an electronic balance.

At the end of feeding trial, fifty percent of shrimp in each tank were sampled for proximate and amino acid composition while the remainder were used for the determination of apparent digestibility coefficient (ADC) of each diet. The parameters: body weight gain (BWG), specific growth rate (SGR), feed Intake (FI), feed conversion ratio (FCR) and protein efficiency ratio (PER) were calculated to evaluate the growth performance of the shrimp juveniles.

C Digestibility

Shrimps were transferred to 30x30x30 cm of 54-L rectangular PVC tank (3 tanks/treatment), filled with 45 L filtered sea water (33 ppt, temperature 12-16°C) under a moderate aeration and predominantly dark condition Prawns were fed diets as those in feeding trials except a portion of α-starch was replaced by Cr_2O_3 (1% in the diet). Prior to the collection of feces, prawns were adapted to the test diet for a week. Prawns which feed test diets at ration 6% of BW, allow to eat for 2 hours. Thereafter, the uneaten feed was siphoned out and the water in tanks was renewed with the new filtered sea water to make sure that there was no leftover of diets given. Feces were then siphoned out from the bottom of the tank every 2 hours for 8 hours during the daytime. Feces were freeze dried immediately after collecting. Chromium contents were analyzed according to Furukawa and Tsukahara (1966).

The apparent digestibility of dry matter, protein and lipid of diets were calculated as follows :

Digestibility of dry matter (%) =
(1- % Cr2O3 in diet/% Cr2O3 in feces) x 100,

digestibility of protein (%) =
{1-(% protein in feces/% Cr_2O_3 feces)/{% protein in diet/% Cr_2O_3 in diet}x100%

digestibility of lipid (%) =
{1-(% lipid in feces/% Cr2O3 feces)/(% lipid in diet/% Cr2 O3 in diet)}/x100%

D Chemical and Statistical Analysis

The protein and lipid contents of samples were determined using Kjeldhal and by cloroform-methanol extraction methods (Bligh and Dyer. 1959), respectively; while ash and moisture were determined according to the Association of Officials Analytical Chemist (1990).

Amino acid profiles were performed by using HPLC according to Teshima *et al.* (1986). Approximately 2 mg of dried samples was weighed and hydrolized with N-methane-sulfonic acid for 22 h at 110°C. The pH of the hydrolysate was adjusted to pH 2.2 and injected to into HPLC unit with an ion exchange resin column. Norleucine was used as an internal standard.

Growth performance data and the apparent digestibility coefficients of diets were compared by one-way analysis of variance (Package super-ANOVA, Abacus Concepts, Berkeley, California, USA). Significant differences between the means were tested by Duncan's Multiple Comparison Test. Probabilities of $p< 0.05$ were considered significant. The optimum dietary BM level was estimated following the broken line-regression metods (Zeitoun *et al,* 1976; Robbins *et al*, 1979)).

III RESULTS

Protein and lipid content of diets were not significantly between the dietary treatment (Table 1). The moisture content in BM-100 is slightly higher then other groups but significantly difference was not detected between the diets. The ash content in diets tend to decrease with the increment of BM in diet. The diet BM-100 contains lower ash compare to other diets

The EAA contents in diet were similar each others except the methionine and isoleucine showed the decrement as BM in diet increase. On the otherhand, leucine and valine shows a reverse trend. The tryptophan would not be measured exactly because of inhibitation by carbohydrate in the diet.

The results of feeding trials were presented in table 3. The survival rate of shrimp were over than 65% among all diets group and not affected by the level of dietary BM. The high mortality considered by the carnivorous as the main character of the *M. Japonicus* (Liao and Chien 1993) and handling effects during the feeding trial. However, there was a general trend that growth of shrimp decrease with BM in diet. The BWG and SGR of shrimp fed BM-75 and BM-100 were similar but lower compare to the groups fed control diet, BM-25 and BM-50.

It is likely that SGR and BWG of shrimp was not affected by the inclusion of BM up to 120 g BM in 1 kg of diets. Similar to BWG parameter, the inclusion of BM in shrimp diet has no effect on FI up to the 120 g BM/kg diet (50% of FM replaced). The FCR and PER shows a similar trend whereas no effect up to the inclusion level of 120 g BM/kg diet even the group of shrimp fed BM-50 was not statistically different compare to shrimp group fed BM-75 and BM-100. By using broken-line regression methods, we estimated that 41.7% and 43.2% of fish meal could be replaced by BM from BWG and FI point of view, respectively (Fig 1 and Fig 2).

Dry matter, protein and lipid digestibility, were presented in table 4. Similar to growth parameters, the digestibilities tend to decrease with the increment of BM in diet. Protein and lipid digestibility of diet up to 50% BM was similar to control diet and statistically higher compare to diet BM-75 and BM-100. Dry matter digestibility shows similar tendency although BM-25 has similar with diets BM-50, BM-75 and BM-100.

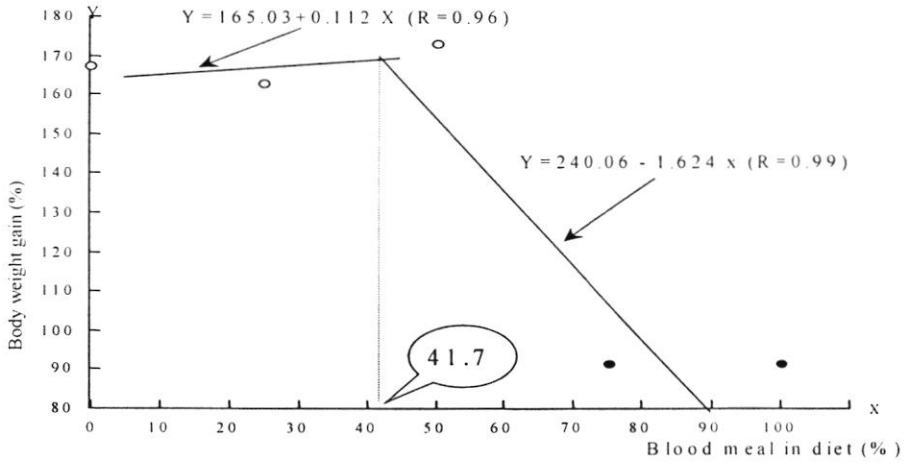

Fig 1. Relationship between BWG (%) of the M. japonicus and the level of FM replaced by BM in diet (%) after 50 days of feeding trial as described by broken line regression model

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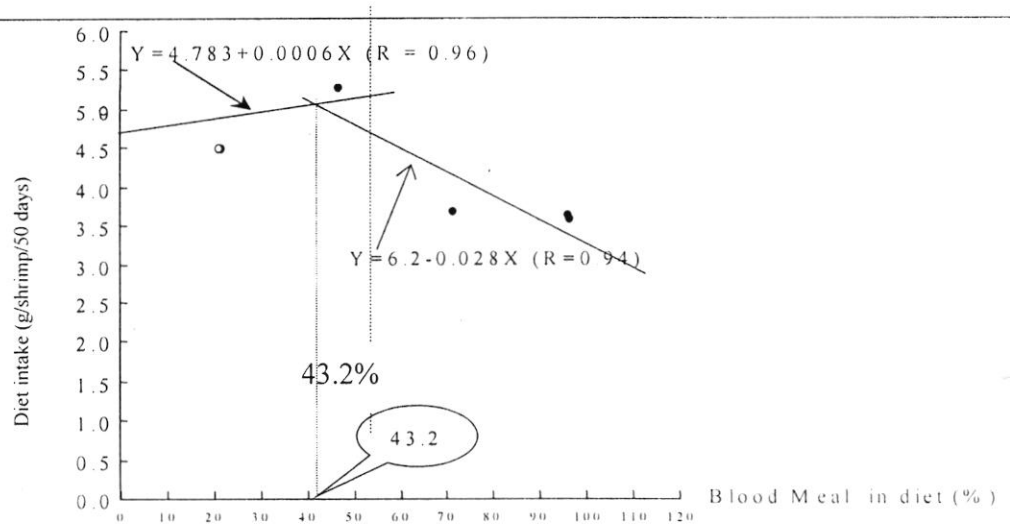

Fig 2. Relationship between diet intake of M. japonicus juvenile and the level of FM replaced by BM in diet (%) after 50 days of feeding trial as described by broken-line regression model

Table 3. Results of growth trials for kuruma prawns fed diets different contents of Blood meal[*1)]

Parameters measured[*2)]	Diets groups				
	Control	BM-25	BM-50	BM-75	BM-100
Initial BW (g)[*3]	0.90±0.0	0.91±0.1	0.91±0.0	0.92±0.1	0.92±0.1
Final BW (g)[*3]	2.44±0.03^{a}	2.40±0.03^{a}	2.48±0.04^{a}	1.78±0.08^{b}	1.76±0.06^{b}
Survival rate (%)	66±12	75±8	72±2	75±0	67±5
BWG (%)[*4]	167.4±3.3^{a}	163.1±1.8^{a}	173.0±3.8^{a}	91.6±9.2^{b}	91.8±8.3^{b}
SGR (%/day)[*5]	0.86±0.03^{a}	0.78±0.02^{a}	0.80±0.03^{a}	0.52±0.04^{b}	0.52±0.06^{b}
FI[*6]	5.0±0.2 ^{bc}	4.5±0.1^{b}	5.3±0.3^{c}	3.7±0.3 ^{a}	3.6±0.3 ^{a}
FCE[*7]	30.9±0.9^{b}	33.3±0.6^{b}	29.8±2.3^{ab}	23.6±3.4^{a}	23.0±1.9^{a}
PER[*8]	0.62±0.02^{b}	0.66±0.01^{b}	0.56±0.05^{ab}	0.47±0.07^{a}	0.49±0.04^{a}

[*1] All values are means ± s.e of triplicate tanks
[*2)] Values with the same superscripts at the same row are not significantly different at 5% level
[*3] BW : Body weight,
[*4]BWG : body weight gain = (final BW– initial BW) x 100/initial BW,
[*5]SGR : Specific growth rate = 100(Ln final weight – Ln initial weight)/50.
[*6] FI : feed intake (One prawn/g diet/50 days),
[*7] FCE : Food conversion efficiency = Weight gain x 100/diet given.
[*8] PER : Protein efficiency ratio = Weight gain/protein given.

Table 4. Apparent digestibility of dry matter, protein and lipid on kuruma shrimp, *Marsupenaeus japonicus* fed diets containing different level of Blood meal

Diets Groups	Digestibility (%)*[1]		
	Dry matter	Protein	Lipid
Control	72.0±0.7[c]	77.2±0.9[c]	79.9±0.8[b]
BM-25	64.3±2.2[bc]	70.74±1.5[c]	79.8±1.1[b]
BM-50	63.1±4.4[bc]	71.11±0.7[c]	76.4±0.6[b]
BM-75	55.6±1.3[ab]	55.81±0.3[b]	63.2±0.2[a]
BM-100	46.3±5.8[b]	41.6±4.5[a]	65.2±2.7[a]

*[1] Means ± s.e of triplicate tanks
*[2] Values with the same superscripts are not significantly different at 5% level

Discussion

The growth of shrimp fed control diet in this study was lower compare to the results of other study using a juvenile size (Saitoh *et al*, 2000). Those facts may mainly affected by low temperature since this experiment was conducted in temperature range 14 to 18°C. Most shrimp are farmed between 22 and 34°C, as temperature increases, the growth rate also increases. How ever, temperature above 35°C produce mortality (Carvajal, and Nebot., 1998). The maximum growth of *Marsupenaeus japonicus* was obseved at temperature 26°C (Labat, 1974).

This study demonstrated that blood meal could be the possible alternative source of protein in the practical diet for Kuruma shrimp, *Marsupenaeus japonucus* juvenile partially substituting the fish meal based diet up to about 50% or reducing the utilization of fishmeal from 300 g/kg diet to about 150 g/kg diet without retardation effect in term of growth. This present study clearly showed that the retardation of growth of shrimp group fed BM-75 and BM-100 were mainly affected by the reduction of FI associate with the decrement of digestibility and the decreasing of some indispensable amino acids (methionine and isoleucine) as the BM in diet increases.

The scientific report concerning the utilization of blood meal as protein source for Penaeid shrimps juvenile is still limited. Brand and Colvin (1977 reported that a growth depression was found on *Penaeus californiensis*, when fed artificial diet contains 5% to 10% of blood meal. On the other hand, white leg shrimp (*Penaeus vannamei*) did not show significant differences in weight gain, feed conversion and survival when fed diets containing 10% level of differently processed blood meals in diet (Dominy and Ako, 1988). Similar to this latter report, this present study shows that *M. japonicus* juvenile shows no adverse in term of growth when fed diet contains 6 to 12% of BM.

Another product of rendered animal such as rendered meat and bone meals has been recently examined as fishmeal replacement ingredients for Pacific white shrimp *Litopenaeus vannamei* (Forstel *et al*, 2003). The authors demonstrated that the maximum level of rendered meat and bone meals to replace of fishmeal in Pacific white shrimp diet is about 75%. Moreover, the authors found that beef products are more effective in meeting the nutritional needs of shrimp than are either pork or poultry products. In this study, the animal rendered product such as blood meal apparently allowable to replace of fishmeal up to 50% in *M. japonicus* juvenile diet. Rendered products have been use also for *Penaeus monodon*

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The growth retardation found in group of shrimp fed BM-75 and BM-100 respectively compare to the other dietary groups may contributed by lower FI. The feed intake strongly correlates with the acceptability and palatability of diet, which those affected by the essential amino acids such as Arginine (Lovel 1989) and the free amino acids such as betain, glutamic acids, alanine and glycine. (Akiyama, 1986) that contain in diet. This study shows that the glutamic acid and glycine in diet were decreasing as BM in diet increasing (Table 2).

The inclusion of BM up to 120 g/kg diet (i.e: 50% of FM replaced by blood meal) still allowable for juvenile of *M. japonicus* from dry matter, protein and lipid digestibility view of points. This fact consistent with the growth performance data which shows no statistically differences among the group of shrimp fed control, BM-25 and BM-50 respectively. The lower digestibility of protein in group of shrimp fed higher level BM may indicate the presence of indigestible nitrogen compound in this ingredient. The digestibility of this ingredient may increase by proper processing as found in rainbowtrout (NRC, 1991) and chinook salmon (Hajen et al, 1993). Some results shows that protein digestibility of blood meal were variable (Meng, 2002). Differences in digestibility of blood meal among studies may be due to differences in animal species, meal processing condition or meal quality.

Protein quality of dietary protein sources depends on the amino acid composition and their availability. The similar growth of shrimp fed BM-0, BM-25 and BM-50 indicate that the essential amino acid in those diets are still allowable although found that there was a variability of their essential amino acid compositions. In case of group of shrimp fed diet BM-75 and BM-100, which have lower growth may affected by the decrement of methionine as BM increase in diet as reflected in essential aminoacid composition of whole body (Table 2). It has been reported that BM is deficient in methionine and isoluecine but rich in leucine (Hertrampf and Pascual, 2000, NRC, 1992, Tacon and Jackson, 1985). Teshima et al (2002) established the isoleucine and methionine requirement of M. japonicus were in the range 2.3-2.9% and 1.3-1.6% respectively. In our data we calculated that the isoleucnie and methionine were 1.4 and 0.3 for BM-75 and 1.2 and 0.38 for BM-100, far below than those value sugested.

In summary, fishmeal in practical diet of *M. japonicus* juvenile can be replaced 50% by bloodmeal without adverse in growth and digestibility.

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