Feed intake assessment on sub-adult spiny lobster *Panulirus homarus* fed a range of pellet and fresh feeds under a variety of feeding regimes

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Abstract. Low feed intake has been considered a major problem in applying formulated feed for grow-out of spiny lobster (Panulirus homarus). This study was conducted to evaluate the effects of fishery hydrolysate in feed formulation and the co-feeding of formulated feed with fresh food on the feed intake of spiny lobster. Five experiments of feed intake assessment were conducted using either different sources (tuna, crustacean and mussel) or levels (0%, 1%, 2% and 3%) of hydrolysates, and experiments on different feeding schemes where lobsters fed either only formulated feed or formulated feed and fresh food. Each experiment was performed for 2 weeks using 16 fiberglass tanks (300 L) filled with seawater and each experiment has 4 treatments with 4 replicates. The experiment showed that including fish hydrolysate 2%, crustacean hydrolysate 1% (dry) or 2% (liquid), or 2 % tuna hydrolysate in feed increased the feed intake of spiny lobster. Feed intake of formulated feed decreased when they fed in combination with mixed fresh food. Feed intake of lobster fed mixed fresh food was lower than that fed either mussel or crab only, and lobster fed only fish exhibited the lowest feed intake.

1 Introduction

A diet formulation for any animal's particular nutrient requirements is crucial to achieve optimal growth. Crustaceans, including lobster have slow feeding habits, and their feeding responses are difficult to observe compared to finfish. Fish consume intact pellets, while crustaceans tend to manipulate and pull a part pellet during feeding. In fact, the nutrient contents of pellet feed quickly deteriorate when it is submerged in water. Therefore, it is critical that pellet feed be attractive (to be found quickly) and palatable (once it is found, it stimulates the animal to prey until satiated).

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Low feed intake had been considered as a major hindrance in the development and application of formulated feeds for the aquaculture of spiny lobsters, *Panulirus homarus* [1, 2]. A study stated that feed intake could be increased by inclusion of ingredients such as stimulants or feeding incitants and attractants in feed formulations [3]. Attractants and feeding stimulants are commonly used in shrimp farming to promote quick identification and consumption of the feeds. Attractants and feeding stimulants allow to maximize feed intake in fish or shrimp and further reduce feed waste [4]. Fishery byproduct such as hydrolysates from fish or squid processing industries which they rich in free amino acids and nucleotides are commonly used as attractants and feeding stimulants for shrimp [5, 6]. Squid meal which contains small peptides, certain free amino acids, and betaine had been reported to have growth-promoting and attractant properties for fish and shrimp culture [6, 7]. Squid paste that produced from squid viscera by low temperature cooking or fermentation was very common to be used as an attractant and feeding effector in aquaculture feeds in China. Recently, it has been reported that use of a commercial palatability enhancer in shrimp feed improved feed intake and growth of white shrimp, Litopenaeus vannamei [8]. Supplementation of 30 g skipjack tuna viscera hydrolysate in 1 kg diet was reported to significantly improve feed intake of asian seabass, Lates calcarifer [9]. However, there has been limited information on the application of attractants and feeding stimulants in spiny lobster culture.

ACIAR project FIS/2014/059 has developed a benchmark feed formulation for grow-out of a spiny lobster, *P. homarus*. Following the development of the benchmark feed, the present study was designed to formulate diet that promotes lobster to ingest higher amount of feed than the benchmark feed. This study was performed through five experiments with the broad objective of improving feed intake of lobsters by assessing the feed intake of sub-adult spiny lobster, *P. homarus* fed a range of pellets supplemented with fishery hydrolysates, and fresh fishery feed under a variety of feeding regimes.

2 Methods

2.1 Experimental animals and systems

Sub-adult spiny lobster, $P.\ homarus$ were obtained from Pekutatan (south Bali) and Grajagan (Banyuwangi-East Java) coastal areas. Feed intake experiments were performed in an indoor system using 16 fiberglass tanks (300 L) without shelter, supplied with ambient flow-through seawater and equipped with individual air-stones. Seawater was filtered through sand filter. Data on feed intake were recorded for an experiment duration of two weeks unless otherwise stated. The experiment series was run as four treatments: four replicates each. The lobsters were individually weighed to obtain a population average weight. Initial carapace length was also recorded. Five lobsters in the calculated ideal weight range (population mean weight \pm 1 standard deviation) were allocated to each tank.

The selection of lobster for the experiments was made based on careful considerations. Lobsters that were male, robust, and physically intact were stocked preferentially. Lobsters that were weak, had significant fouling of the carapace, or were missing multiple appendages were not used in the experiment. Non-berried females were used in the experiments, providing the number of females stocked is equally distributed among treatment and replicate tanks.

2.2 Feeding and experiment maintenance

The lobsters were fed twice daily at 09:00 and 15:30. The amount of feed at each feeding was recorded. Daily uneaten feed was collected using a 250 µm screen at 08:00 and 15:00. Uneaten pellet feed was then oven dried, weighed and recorded. Uneaten fresh feed was

drained on tissue paper to remove water on its surface, weighed and recorded. Feed intake is the difference between the amount of feed given and the amount of uneaten feed and presented as percent of body weight of lobster. The feeds were kept in a chiller (<4 °C) except when feeding or weighing. Lobsters were weighed at initiation (Day 0) and termination (Day 14). Molts and mortalities were removed and recorded daily. A non-invasive count for each tank's remaining lobsters was recorded daily. The oxygen and temperature were recorded daily, while the salinity was monitored weekly.

2.3 Formulated feed preparation

Formulated feed for the experiments were produced at IMRAFE's Feed Technology Laboratory in Gondol-Bali, Indonesia using ingredients from a local feed company, whereas CSIRO, Australia, provided fishery hydrolysates. Feed formulations for the experiments are presented in Table 1 - 6. Fresh ingredients, such as mussel, fish flesh and crab were placed at -20 °C until semi-frozen, and then extruded through a 3 mm die plate of a semi-commercial meat grinder to form a homogenous mince. The dry ingredients were finely ground using a hammer mill (Mikro Pulverizer) for bulk ingredients. The fresh ingredients and binder were thoroughly mixed using a domestic electric hand mixer before adding fish oil, then the remaining dry ingredients were added. This is followed by a further 10 minutes of mixing to form a dough of approximately 40–50% moisture content. The dough was extruded through a 3 mm die plate to make pelleted feed. The feeds were oven dried (<93% DM) and then stored at -20 °C before feeding.

2.4 Feeding experiments

2.4.1 Experiment 1: Feed intake assessment on sub-adult lobster fed the benchmark feed supplemented with tuna hydrolysate (SL5) at 0, 1, 2, and 3%.

This trial assessed the inclusion of tuna hydrolysate (SL5) in the benchmark formulation at 0, 1, 2, and 3 % (Table 1). Pellet feed was prepared in dry form. Five lobsters (all male) with initial weight of 217.7±13.2 g and initial total length of 19.2±1.2 cm were allocated in each tank. A feed intake trial was carried out for 13 days.

Table 1. Feed formulation for feed intake assessment on the effects of tuna hydrolysate (SL5) at 0, 1, 2, and 3 % inclusion

T P	Experimental diets					
Ingredients	Diet-A	Diet-B	Diet-C	Diet-D		
Fish meal	65.3	64.3	63.3	62.3		
Tuna hydrolysate (SL5)	0	1	2	3		
Cholesterol	0.5	0.5	0.5	0.5		
Wheat flour	6	6	6	6		
Wheat gluten	6	6	6	6		
MOS	0.5	0.5	0.5	0.5		
Fish (fresh)	6	6	6	6		
Mussel (fresh)	6	6	6	6		
Crab (fresh)	1	1	1	1		
Fish Oil	2.6	2.6	2.6	2.6		
Carophyl pink	1	1	1	1		
Lecithin	1.7	1.7	1.7	1.7		

T P (Experimental diets						
Ingredients	Diet-A	Diet-A Diet-B Diet-C D						
Mineral premix	0.6	0.6	0.6	0.6				
Vitamin premix	1.1	1.1	1.1	1.1				
Stay C	0.4	0.4	0.4	0.4				
Binder (CMC)	1.3	1.3	1.3	1.3				
Total	100	100	100	100				

2.4.2 Experiment 2: Feed intake assessment on sub-adult lobster fed the benchmark feed supplemented with fishery hydrolysates at either 0, 1 or 2%

Crustacean hydrolysate (HP2, dry) and crustacean hydrolysate (ML4, liquid), as well as mussel hydrolysate (NZ) were assessed for feed intake of lobster. Crustacean hydrolysate (HP2) was included at 1 % as a dry ingredient (Table 2). The test lobsters' initial weight and carapace length were 214.7±3.4 g and 8.23±0.1 cm, respectively.

Table 2. Feed formulation for feed intake assessment on the effects of different fishery hydrolysates at either 0, 1 or 2 % inclusion

	Experimental diets					
Ingredients	Diet-A	Diet-B	Diet-C	Diet-D		
Fish meal	65.3	64.3	63.3	63.3		
Crusteans hydrolysate (HP2, dry)		1				
Crusteans hydrolysate (ML4, liquid)			2			
NZ (Mussel hydrolysate)				2		
Cholesterol	0.5	0.5	0.5	0.5		
Wheat flour	6	6	6	6		
Wheat gluten	6	6	6	6		
MOS	0.5	0.5	0.5	0.5		
Fish (fresh)	6	6	6	6		
Mussel (fresh)	6	6	6	6		
Crab (fresh)	1	1	1	1		
Fish Oil	2.6	2.6	2.6	2.6		
Astaxanthin	1	1	1	1		
Lecithin	1.7	1.7	1.7	1.7		
Mineral premix	0.6	0.6	0.6	0.6		
Vitamin premix	1.1	1.1	1.1	1.1		
Stay C	0.4	0.4	0.4	0.4		
Binder (CMC)	1.3	1.3	1.3	1.3		
Total	100	100	100	100		

2.4.3 Experiment 3: Feed intake assessment on sub-adult lobster fed the benchmark feed supplemented with fishery hydrolysates at either 0 or 2 %

This experiment assessed feed intake of lobster fed the benchmark feed supplemented with carp hydrolysates (Sampi), tuna hydrolysate (Sampi), and tuna hydrolysate (SL5) at either 0

or 2 % (Table 3). Initial weight of the selected lobsters was 217.2±5.7 g, and the carapace length was 8.38±0.6 cm.

Table 3. Feed formulations for feed intake assessment on the effects of different fish hydrolysates at either 0 or 2 % inclusion

I P An	Experimental diet					
Ingredients	Diet-E	Diet-F	Diet-G	Diet-H		
Fish meal	65.3	63.3	63.3	63.3		
Carp hydrolysate (Sampi)		2				
Tuna hydrolysate (Sampi)			2			
Tuna hydrolysate (SL5)				2		
Cholesterol	0.5	0.5	0.5	0.5		
Wheat flour	6	6	6	6		
Wheat gluten	6	6	6	6		
MOS	0.5	0.5	0.5	0.5		
Fish (fresh)	6	6	6	6		
Mussel (fresh)	6	6	6	6		
Crab (fresh)	1	1	1	1		
Fish Oil	2.6	2.6	2.6	2.6		
Astaxanthin	1	1	1	1		
Lecithin	1.7	1.7	1.7	1.7		
Mineral premix	0.6	0.6	0.6	0.6		
Vitamin premix	1.1	1.1	1.1	1.1		
Stay C	0.4	0.4	0.4	0.4		
Binder (CMC)	1.3	1.3	1.3	1.3		
Total	100	100	100	100		

2.4.4 Experiment 4: Feed intake assessment on sub-adult lobster fed the benchmark diet, benchmark diet co-fed with a mixed fresh fishery feed, benchmark diet fed with alternating fresh fishery feed or a mixed fresh fishery feed

This experiment presumed that feed intake of lobster could be improved by varying the types of diet to be fed. The benchmark diet was formulated as presented in Table 4. Feed intake of lobster was assessed for the benchmark diet (100%), benchmark diet (70%) co-fed with a mixed fresh fishery feed (30% in total), benchmark diet (70%) fed with alternating fresh fishery feed (30%), and only a mixed fresh fishery feed (Table 5). Five lobsters (3 male and 2 female) with initial weight of 206.6 ± 1.4 g and carapace length of 8.17 ± 0.5 cm were stocked in each tank. This experiment was performed for 10 days.

Table 4. Feed formulation for feed intake assessment on the effects of co-feeding with fresh fishery feed

Ingredients	Total (%)
Fish meal	65.3
Cholesterol	0.5
Wheat flour	6
Wheat gluten	6

Ingredients	Total (%)
MOS	0.5
Fish (fresh)	6
Mussel (fresh)	6
Squid (fresh)	1
Fish Oil	2.6
Astaxanthin	1
Lecithin	1.7
Mineral premix	0.6
Vitamin premix	1.1
Stay C	0.4
Binder (CMC)	1.3
Total	100

Table 5. Feeding regimes for feed intake assessment on lobster. The fresh fishery feed were provided as 30% of the total diet when co-fed with pellet (treatments B and C)

Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet
100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet
70%	70%	70%	70%	70%	70%	70%	70%	70%	70%
Mixed	Mixed	Mixed	Mixed	Mixed	Mixed	Mixed	Mixed	Mixed	Mixed
fresh	fresh	fresh	fresh	fresh	fresh	fresh	fresh	fresh	fresh
fishery	fishery	fishery	fishery	fishery	fishery	fishery	fishery	fishery	fishery
feed	feed	feed	feed	feed	feed	feed	feed	feed	feed
(30%)	(30%)	(30%)	(30%)	(30%)	(30%)	(30%)	(30%)	(30%)	(30%)
Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet	Pellet
70%	70%	70%	70%	70%	70%	70%	70%	70%	70%
Fresh fish (30%)	Fresh fish (30%)	Fresh fish (30%)	Fresh fish (30%)	Fresh mussel (30%)	Fresh mussel (30%)	Fresh mussel (30%)	Fresh crab (30%)	Fresh crab (30%)	Fresh crab (30%)
Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed	Mixed fresh fishery feed (100%)
	Pellet 100 % Pellet 70% Mixed fresh fishery feed (30%) Pellet 70% Fresh fish (30%) Mixed fresh fishery	Pellet 100 % 100 % Pellet 70% 70% Mixed fresh fishery feed (30%) (30%) Pellet 70% 70% Pellet 70% fishery feed (30%) (30%) Mixed Fresh fish fish (30%) (30%) Mixed fresh fishery feed feed feed	Pellet 100 %	Pellet Pellet Pellet Pellet 100 % 100 % 100 % 100 % 100 % 100 % Pellet Pellet 70% 70% 70% 70% 70% Mixed fresh fresh fresh fishery fishery feed feed feed (30%) (30%) (30%) (30%) (30%) Pellet Pellet 70% 70% 70% 70% Fresh Fresh Fresh fish fish fish fish fish fish fish fi	Pellet Pellet 100 % 100	Pellet Pellet Pellet Pellet Pellet Pellet 70% 70% 70% 70% Fresh Fresh Fresh Fish fish fish fish fish fish fish fish f	Pellet Pellet Pellet Pellet Pellet Pellet Pellet Pellet 70% 70% 70% 70% 70% 70% 70% 70% 70% 70%	Pellet 100 %Pellet 100 %Pelle	Pellet 100 %Pellet 100 %Pelle

2.4.5 Experiment 5: Feed intake assessment on sub-adult lobster fed a mixed fresh fishery feed, or single source fresh fishery feed of either fish, mussel or crab

This experiment was designed to investigate the maximum feed intake (% BW/day) achieved by feeding with fresh fishery feed. Lobsters were fed a mixed fresh fishery feed (A) and a single source fresh fishery feed of fish (B), mussels (C), and crab (D) (Table 6). Lobsters with an average weight of 206.4 ± 3.7 g and carapace length of 8.55 ± 0.8 cm were used in this trial, where five lobsters (3 male and 2 female) were stocked in each tank. This trial was done for 14 days.

E. d.	Feeding schemes						
Feed types	A B C						
Fish (flesh)	33 %	100 %					
Mussel (flesh)	33 %		100 %				
Crab (flesh)	33 %			100 %			
Total	100	100	100	100			

Table 6. Feed types and feeding schemes for feed intake assement on lobster

2.5 Data analysis

A one-way analysis of variance (ANOVA) was used to analyze the feed intake data. When ANOVA confirmed the significant effects of treatments on feed intake, a Tukey's post-hoc test was carried out to examine significant differences among treatments. Statistically significant differences in means among treatments were accepted at a significant level of $\alpha = 0.05$.

3 Results and Discussion

3.1 Experiment 1: Feed intake assessment on sub-adult lobster fed the benchmark feed supplemented with fish hydrolysate (SL5) at 0, 1, 2, and 3 %

Results of experiment 1 showed that inclusion of tuna hydrolysate SL5 biologically increased feed intake at all levels when compared to the control feed. However, these values were not significantly different (p>0.05) (Figure 1). Inclusion of tuna hydrolysate SL5 at 1-2%increased feed intake of lobster. However, feed intake of lobster was decreased when the inclusion of tuna hydrolysate SL5 was increased to 3%. Growth, in terms of percent weight gain (WG) of lobster during 2 weeks feeding experiment, was 0.53%, 0.90%, 1.42%, and 1.26% for treatments A, B, C, and D, respectively. These growth data match with feed intake of lobster, where higher feed intake resulted better growth. Therefore, this study recommended that inclusion of 2% tuna hydrolysate SL5 could be used as a modification to the benchmark diet. There had been suggested that inclusion of protein hydrolysate which is rich in small peptides and feeding stimulatory free amino acids (FAAs) might be a practical method to improve the productivity of juvenile lobster P. ornatus fed on dry pellets [10]. Fish hydrolysate at 2% inclusion in feed for shrimps also resulted in significantly higher daily feed intake (4.6% BW/day) than the control feed (2.4% BW/day) [3]. A considerably higher feed intake in shrimp Litopenaues vannamei was obtained in groups fed > 1%, including lowmolecular-weight fish hydrolysate, compared to the control group [11].

The use of fish protein hydrolysate in aqua-feeds should be at the appropriate level since higher inclusion of fish hydrolysate could negatively impact the feed utilization and growth of fish [12]. Growth of shrimp *Penaues vannamei* fed diet with 4% fish hydrolysate inclusion (3% of total dietary protein replacement) was higher than those fed with 12 and 20% fish hydrolysate inclusion [13]. A significant decline in growth was reported in *Japanese flounder*, *Paralichthys olivaceus* when fish hydrolysate was added in the diet at 16% or over [14]. Fish hydrolysate at 20% inclusion in turbot, *Scophthalmus maximus* diet caused significantly decreased feed utilization and specific growth rate [15]. Growth of barramundi was significantly increased when fish meal was replaced with 5 to 10% tuna hydrolysate. However, growth performance started to decline when it was replaced at higher levels (15 to 20%) [16]. The negative effects of fish hydrolysates at a high inclusion level on the

physiological functions of fish could be because of an excessive amount of free amino acids (FAA) and peptides, which may lead to an imbalance in amino acid absorption and saturation of peptide transportation systems [17].

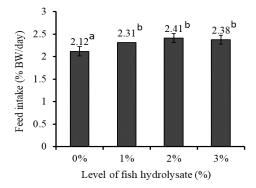


Fig. 1. Feed intake of lobster *Panulirus homarus* as a percentage of body weight, when they fed benchmark feed supplemented with tuna hydrolysate (SL5) at 0, 1, 2, 3%

3.2 Experiment 2: Feed intake assessment on sub-adult lobster fed the benchmark feed supplemented with fishery hydrolysates at either 0, 1 or 2%

Crustacean hydrolysate HP2 (dry) and ML4 (liquid) as well as mussel hydrolysate NZ at either 1 or 2% inclusion gave higher feed intake than the benchmark feed and significantly different (p<0.05) (Figure 2). The figure indicates that the two crustacean hydrolysates were better in promoting feed intake of 200-g lobster than mussel hydrolysate, i.e., 2.91 and 2.92% BW/day compared to only 2.79% BW/day. It was probably that crustacean hydrolysate was more attractive for lobsters than mussel hydrolysate.

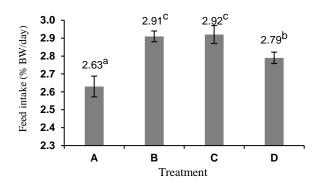


Fig. 2. Feed intake of lobster *Panulirus homarus* as a percentage of body weight, when fed benchmark feed supplemented with fishery hydrolysates at either 0, 1 or 2%. (Treatment A = benchmark feed (control) 0%; B = dry crustacean hydrolysate (HP2) 1%; C = liquid crustacean hydrolysate (ML4) 2%; D = mussel hydrolysate (NZ) 2%)

Growth data indicated that WG of lobster fed with control diet, dry crustacean hydrolysate (HP2) 1%, liquid crustacean hydrolysate (ML4) 2% and mussel hydrolysate (NZ) 2% supplemented diet were 0.85%, 2.52%, 2.34% and 1.46%, respectively. Low value of WG observed in this experiment might due to short period of the experiment, only two weeks. It was also reported that scallop hydrolysate was not attractive to shrimp *L. vannamei*

since it did not improve feed intake and growth of the shrimp [18]. On the other hand, shrimp and krill hydrolysate supplemented in feeds resulted in significantly better feed utilization and growth performance on olive flounder *Paralichthys olivaceus* compared to the fish fed low fishmeal diet [19]. Feed utilization and growth performance of red sea bream were also significantly higher when they fed diets with krill and shrimp hydrolysate at approximately 5% inclusion in diets. Feeds with hydrolysates indicated considerably higher digestibility of dietary protein than the control diet [20]. Another study showed that krill hydrolysate supplementation in feeds positively affected growth performance and feed intake of *Salmo salar* fed low fish meal and high plant protein diets [21].

3.3 Experiment 3: Feed intake assessment on sub-adult lobster fed the benchmark feed supplemented with fishery hydrolysates at either 0 or 2%

The fishery hydrolysate at 2% inclusion which promoted the highest feed intake was tuna hydrolysate (Sampi) (3.02% BW/day) as shown in (p<0.05) Figure 3. Carp and tuna hydrolysate (SL5) resulted in similar feed intake, 2.84 and 2.83% BW/day, respectively. The benchmark feed had the lowest feed intake (2.7% BW/day). Weight gain (WG) of lobster fed tuna hydrylysate (Sampi) was 2.08%, higher compared to other treatments, i.e., 0.48%, 1.85% and 0.98% for treatment E, F, and H, respectively. Data in these studies showed that data of feed intake was not in line with growth data, especially for treatment F with WG only 0.98% even though its feed intake was 2.83%. Lobster growth occurs through the process of molting, and the body weight of lobster increases after the process succeeds. This experiment was conducted only for two weeks (a short period), so no molting occurred. Results of the present study suggested the use of tuna hydrolysate (Sampi) in addition to the benchmark feed to improve the feed intake of lobster.

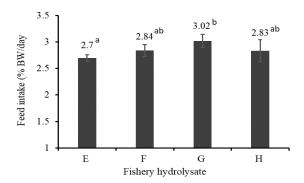


Fig 3. Feed intake of lobster *Panulirus homarus* as a percentage of body weight, when fed benchmark feed supplemented with fishery hydrolysates at either 0 or 2 %. (Treatment E = benchmark feed (control) 0%; F = carp hydrolysate (Sampi) 2%; G = tuna hydrolysate (Sampi) 2%; H = tuna hydrolysate (SL5) 2%

A previous study showed that tuna hydrolysate supplementation in feeds significantly affected feed utilization and growth performance of juvenile barramundi. Higher feed intake, final body weight and weight gain were observed in fish fed diets supplemented with tuna hydrolysate (at 10%) when compared to the control [22]. Tuna viscera protein hydrolysate at 0.5 and 1.64% inclusion in feed could be used as a feeding stimulant in *Lates calcarifer*. The inclusion of tuna viscera protein hydrolysate in feeds improved feed palatability because the free amino acids perform as flavor enhancers, which promptly increase feed intake and growth performance in *L. calcarifer* [9].

3.4 Experiment 4: Feed intake assessment on sub-adult lobster fed the benchmark diet, benchmark diet co-fed with a mixed fresh fishery feed, benchmark diet fed with alternating fresh fishery feed or a mixed fresh fishery feed

Results of experiment 4 showed that lobsters fed only the benchmark diet (pellet) had a feed intake of 2.8% BW/day. Lobster fed the benchmark feed co-fed with a mixed fresh fishery feed showed feed intake of the benchmark diet 2.6% BW/day and fresh fishery feed of 1.4% BW/day. Lobster fed the benchmark diet with alternating fresh fishery feed resulted in feed intake of the benchmark 2.5% BW/day and fresh fishery feed 1.5% BW/day. The highest feed intake was obtained from mixed fresh fishery feed (5.2% BW/day). However, feed intake values for fresh fishery feed was based on fresh or wet weight (Figure 4). Mixed fresh fishery feed probably retained their attractiveness throughout the feeding period and promoted lobster's feeding response. The results of this experiment showed that addition of fresh fishery feed to the benchmark diet increased the feed intake of sub-adult lobster. A previous study reported that mussel intake on a dry matter basis was higher than pellet intake for post-puerile and year-1 juvenile *P. cygnus*. Mussel intake was double over pellet intake for post-puerile [23]. All lobsters in this experiment showed low growth during the two week feeding experiment. Weight gain (WG) of lobster for treatment A, B, C and D were 0.14%, 0.31%, 0.83% and 0.12%, respectively.

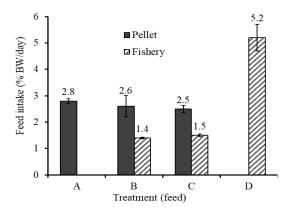


Fig. 4. Feed intake of lobster *Panulirus homarus* as a percentage of body weight, when either fed the benchmark diet, fresh fishery feed or co-fed a combined diet. (Treatment A = benchmark diet; B = benchmark diet + alternating fresh fishery feed; C = benchmark diet + a mixed fresh fishery feed; D = a mixed fresh fishery feed). Feed intake for fresh fishery feed was calculated based on fresh (wet) weight

Study on lobster *P. ornatus* found that formulated feeds promoted a feeding response of lobster within only the first 2–3 hours after feeding, but mussels remain attractive along the feeding period [24]. Another study on lobster *Jasus edwardsii* juvenile also showed that a high level of feed intake could not be achieved by feeding with formulated feed due to foregut capacity being small. In addition, formulated feeds enter midgut directly, resulted in a prolonged gut evacuation time and appetite revival [2]. Further, difficulties in digestive processing had been reported on lobster fed formulated feeds leading to a significant decrease in digestive capacity of lobster [25]. However, a contradictory result was found in a study on lobster *P. ornatus* juveniles which concluded that a single diet of frozen half shell green-lip mussel *Perna canaliculus* caused lower growth than formulated feeds. This was due to nutritional inadequacies of mussel meat for lobster [1].

3.5 Experiment 5: Feed intake assessment on sub-adult lobster fed a mixed fresh fishery feed, or single source fresh fishery feed of either fish, mussel or crab

Feed intake of lobster varied when they were fed fresh fishery feed with different compositions. The highest feed intake was achieved from feeding with a single source of fresh fishery feed, i.e., mussel (7.3% BW/day) and crab (6.9% BW/day). Interestingly, feed intake of lobster fed mixed fresh fishery feed was lower than they fed either mussel or crab only. Lobster fed only fresh fish exhibited the lowest feed intake (4.4% BW/day) (Figure 5). Growth (WG) of lobster during 2 weeks feeding experiment were 1.67%, 0.82%, 3.62% and 3.68% for treatments A, B, C and D, respectively. These growth data are in line with values of the feed intake of lobster, where higher feed intake results in better growth. This study revealed that mussels as well as crabs stimulate the feeding response of sub-adult *P. homarus*. Another experiment showed that molluscan flesh is the most desirable fishery feed for lobsters. The attractiveness of molluscans prolonged even >10 hours of immersion in water [10]. A significantly higher growth of lobster *P. argus* juveniles was achieved when they fed on frozen shrimp, clams, oysters and squid at 100% of BW once at dusk than when they fed at 50 % of BW twice daily [26].

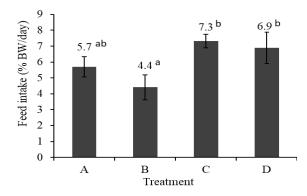


Fig. 5. Feed intake of lobster *Panulirus homarus* as a percentage of body weight, when they fed a mixed fresh fishery feed, or single source fresh fishery feed of fish, mussel or crab. (Treatment A = mixed fresh fishery feed; B = fish; C = mussel meat; D = crab)

4 Conclusion

Inclusion of fishery hydrolysates in feed formulation increased feed intake of spiny lobster *P. homarus*. Inclusion of either tuna hydrolysate (SL5) 2%, crustacean hydrolysate 1% (dry) or 2% (liquid), and 2% tuna hydrolysate (Sampi) in feed could be used to improve feed intake of lobster. Feed intake of formulated feed decreased when the lobster fed formulated feed which co-fed with a mixed fresh fishery feed. However, total feed intake (formulated feed and fresh fishery feed) increased when compared to only pellet diet. Feed intake of lobster varied when they fed fishery diets with different compositions. Feed intake of lobster fed mixed fresh fishery feed was lower than they fed either mussel or crab only, and lobster fed only fish exhibited the lowest feed intake.

This study was financially supported by the ACIAR Project FIS/2014/059 "Research for Development of Lobster Growout Technology in Indonesia". The authors gratefully thank to the all technicians of Lobster Research Activities and technicians of Nutrition and Feed Development of IMRAFE-Gondol for their technical supports during the experiment.

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