Nutritional Quality and \textit{in vitro} Digestibility of Shrimp Meal Made of Heads and Hulls of Black Tiger (\textit{Penaeus monodon}), White Leg (\textit{Litopenaeus vannamei}) and Argentine Red (\textit{Pleoticus muelleri}) Shrimps

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The present study was performed to measure the chemical composition and \textit{in vitro} digestibilities of heads and hulls in three different species of the edible shrimp, and discussed their nutritional quality from the view point of practical use of shrimp meal (SM). Heads and hulls of black tiger (\textit{Penaeus monodon}) (BT), white leg (\textit{Litopenaeus vannamei}) (WL) and argentine red (\textit{Pleoticus muelleri}) (AR) shrimps were prepared: 2 sizes of specimen were used in BT. Their inedible parts, such as heads and abdominal exoskeleton with tails (hulls) were removed, dried at 55°C and ground to pass through 1.0 mm aperture and then used as SM. Compared with hulls, heads were significantly rich in crude protein (CP) and ether extract (EE), and poor in crude fibre (CF), crude ash (CA) and chitin. Among head groups, CP level was the greatest in WL, EE level in AR, CF level in large and small BT, CA level in 3 species other than WL, chitin level in large BT. Among hull groups, CP level was the greatest in WL, CF level in large and small BT and chitin level in large BT: EE level in hulls were extremely low in all group. Overall, \textit{in vitro} dry matter (DM) and CP digestibilities were significantly greater in heads than in hulls, which is reasonable because the level of chitin, non-digestible amino polysaccharide, was greater in hulls than heads in all species. There is no great difference in parameters measured between heads and heads + hulls. The results obtained here suggest that the heads of WL can be a more nutritious source of SM in poultry diets.

\textbf{Key words:} \textit{in vitro} digestibility, nutritional quality, shrimp meal, shrimp species


Introduction

In Southeast Asian countries including Bangladesh, a significant amount of shrimp waste is generated in aquaculture industries and disposed as a little-used by-product. However, this by-product can be the potential alternative source of protein in poultry diets: although several studies have been conducted to use the waste as a protein source for poultry diets, the results did not necessarily show the similar tendency. For instance, Khempaka \textit{et al}. (2011) reported that broilers given a diet containing shrimp meal (SM) made of heads of white leg (\textit{Litopenaeus vannamei}) (WL) up to 15% did not show any negative effects on body weight, feed intake and feed efficiency, and Islam \textit{et al}. (1994) and Rosenfeld \textit{et al}. (1997) reported similar observation even when chickens received diets containing 14.3 % and 31.6% of SM, respectively: unfortunately they did not mention the source of SM. On the other hand, Khempaka \textit{et al}. (2006a) reported that broilers given a diet containing SM made of hulls of black tiger (\textit{Penaeus monodon}) (BT) more than 4% showed decreased feed intake, body weight gain and digestibility, and Fanimo \textit{et al}. (1996) also showed that broilers given diets containing SM (unknown source) at 9.9% (in a starter diet) and 8.2% (in a finisher diet) exhibited decreased performance. Similar observation was reported by Oduguwa \textit{et al}. (2004).

The variation in performance may possibly be due to the variation of the nutritional values of SM, which can be altered by species (Ngoan \textit{et al}., 2000; Heu \textit{et al}., 2003), and/or portion (Meyers, 1986) of shrimps, but the data in these reports are fragmentary and hence, do not provide a clue how to improve the nutritional value of SM. The aim of the present study was to measure the proximate composition
and in vitro digestibility of heads and hulls of BT, WL and argentine red (Pleoticus muelleri) (AR), and to discuss the data from the view point of practical use of SM: in BT, large and small sizes of shrimps were examined, because this is the major aquacultured species in Bangladesh.

Materials and Methods

Preparation of SM

Large (about 18.4 cm) and small (about 15.1 cm) sizes of BT, WL (about 14.3 cm) and AR (about 16.5 cm) shrimps were purchased commercially in frozen form: BT and WL were aquacultured in Vietnam and Thailand, respectively, and AR was caught in Argentina. The frozen shrimps were thawed under running water and blotted by paper (Kimwipes S-200). Thereafter, inedible parts, such as heads and hulls of abdomens were removed carefully. After that, they were dried separately in an electric oven at 55°C for about 8 to 10 hours. The dried waste was ground to pass through 1.0 mm aperture and then used as SM. In addition, parts of heads and hulls were taken and mixed according to the weight ratio of individual head: individual hull ranging from 2.5:1 to 3:0:1 (see Results section). Consequently, 12 kinds of SM were prepared (4 kinds of shrimp×3 kinds of portion). Each sample was divided into 3 aliquots for triplicate measurements.

Chemical Analysis

Proximate composition was analyzed according to standard methods (AOAC, 1990). The chitin extraction process was done according to Ghanem et al. (2003), which is summarized briefly as follows: about 1.0 g of dried SM mixed with 12.5 ml of 2.5 N NaOH, placed in an oven at 75°C for 6 hrs and then filtrate. The filtrate residue, crude chitin, was dried at 105°C in an electric oven for 1 hour. After drying, about 1.0 g of crude chitin was mixed with 10 ml of 1.7 N HCl for 6 hrs and filtered. The filtrate residue was washed with 95% ethanol followed by a final washing with distilled water. The filtrate residue was dried and weighed as chitin.

Digestibility Measurements

The in vitro dry matter (DM) and crude protein (CP) digestibilities of SM were determined according to Saunders et al. (1973) with slight modifications: briefly stated, about 250 mg of dried SM sample was suspended in 15 ml of 0.1 N HCl containing 1.5 mg pepsin (10,000 U/mg protein) (Nacalai Tesque Inc., Kyoto, Japan), and gently shaken at 37°C for 3 hours. After neutralisation with 0.5 N NaOH, the digesta was mixed with 7.5 ml of phosphate buffer at pH 8.0 containing pancreatic (amylase activity 3,220 U/g, protease activity 38,500 U/g and lipase activity 1,600 U/g) (Nacalai Tesque Inc., Kyoto, Japan) and shaken at 37°C for 24 hours. The solution was then centrifuged at 240×g for 10 min, washed with distilled water, filtered and dried.

The DM and CP digestibilities of SM were determined as follows:

\[
\text{DM digestibility} = \left(\frac{\text{Dried sample weight} - \text{Dried residue weight}}{\text{Dried sample weight}}\right) \times 100
\]

\[
\text{CP digestibility} = \left(\frac{\text{Total N in sample} - \text{N in residue}}{\text{Total N in sample}}\right) \times 100
\]

Statistical Analysis

The obtained data were analysed by two-way ANOVA. Contrasts between group means were evaluated by Tukey’s test at a significance level of 5%.

Results

Weight of Head and Hull

In dry matter basis, the weights of heads and hulls were 3.8 g and 1.4 g in large BT, 2.1 g and 0.7 g in small BT, 1.8 g and 0.7 g in WL and 4.2 g and 1.6 g in AR, respectively, indicating that heads are much heavier than hulls in all species (data not shown). Their weight ratio of head: hull was 2.7:1 in large BT, 3.0:1 in small BT, 2.5:1 in WL and 2.6:1 in AR.

Proximate Composition and Chitin (Table 1)

Major components of SM was CP. CP level in heads ranged from 42.4% (AR) to 54.4% (WL), and that in hulls from 40.9% (large BT) to 51.3% (WL), which showed that CP levels in both portions were the greatest in WL. CP level in heads was greater than that in hulls in all groups excepting AR. Ether extract (EE) was a minor component in large and small BT and WL, but this was extremely rich and second major component in heads of AR. EE level in heads was greater than that in hulls in all groups. Crude fibre (CF) level in heads ranged from 6.7% (AR) to 10.8% (large BT), and that in hulls from 16.8% (AR) to 21.5% (small BT). Crude ash level in heads ranged from 15.8% (WL) to 20.4% (large BT), and that in hulls from 19.1% (WL) to 30.2% (AR). Chitin level in heads ranged from 8.4% (AR) to 14.1% (large BT), and that in hulls from 18.9% (AR) to 24.6% (large BT) indicating that both portions in large BT were rich in chitin. Levels of CF, crude ash and chitin in heads were smaller than those in hulls in all groups. Effect of body size in BT was found in EE, crude ash and chitin: in most case, greater value was found in large BT. All values measured in heads + hulls were similar to the corresponding values in heads, because heads comprised more than 70% of heads + hulls on weight basis in all groups.

Digestibility (Table 2)

DM digestibility in heads ranged from 51.8% (AR) to 65.3% (WL), and that in hulls from 42.7% (small BT) to 53.3% (WL), showing that in both portions, DM of WL was the most digestible. CP digestibility in heads ranged from 72.6% (AR) to 81.6% (large BT), and that in hulls from 64.9% (AR) to 76.1% (WL): CP digestibility of heads in WL was comparable to that in large and small BT. Both digestibilities in heads were greater than those in hulls. No great effect of body size was found in DM and CP digestibilities. All values measured in heads + hulls were similar to the corresponding values in heads.
Table 1. Proximate composition and chitin of heads, hulls and heads + hulls of three edible shrimp species

<table>
<thead>
<tr>
<th>Parameters (%)</th>
<th>Portions</th>
<th>Black tiger</th>
<th>White leg</th>
<th>Argentine red</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Large</td>
<td>Small</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dry matter</td>
<td>Heads</td>
<td>89.6±0.2aA</td>
<td>88.4±0.3bA</td>
<td>88.3±0.2bA</td>
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<tr>
<td></td>
<td>Hulls</td>
<td>90.4±0.1ab</td>
<td>89.8±0.1bB</td>
<td>89.4±0.1bB</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>91.6±0.2ac</td>
<td>91.4±0.02c</td>
<td>91.4±0.2c</td>
</tr>
<tr>
<td>Crude protein</td>
<td>Heads</td>
<td>52.3±0.8aA</td>
<td>51.7±0.4aA</td>
<td>54.2±0.2abA</td>
</tr>
<tr>
<td></td>
<td>Hulls</td>
<td>49.0±0.3ab</td>
<td>42.2±0.7ab</td>
<td>51.3±0.3bB</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>48.9±0.1ac</td>
<td>48.8±0.3ac</td>
<td>52.5±0.2ac</td>
</tr>
<tr>
<td>Ether extract</td>
<td>Heads</td>
<td>6.4±0.2aA</td>
<td>5.1±0.6aA</td>
<td>9.7±0.5abA</td>
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<tr>
<td></td>
<td>Hulls</td>
<td>1.4±0.3ab</td>
<td>0.9±0.01bb</td>
<td>3.5±0.1bb</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>5.1±0.3ac</td>
<td>3.9±0.2bc</td>
<td>8.1±0.1bc</td>
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<tr>
<td>Crude fibre</td>
<td>Heads</td>
<td>10.8±0.03aA</td>
<td>10.7±0.1aA</td>
<td>8.5±0.2bc</td>
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<tr>
<td></td>
<td>Hulls</td>
<td>21.5±0.2ab</td>
<td>21.5±0.4ab</td>
<td>18.2±0.2ab</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>12.7±0.1ac</td>
<td>12.0±0.1bc</td>
<td>10.0±0.1bc</td>
</tr>
<tr>
<td>Crude ash</td>
<td>Heads</td>
<td>20.4±0.4aA</td>
<td>19.7±0.7aA</td>
<td>15.8±0.4bA</td>
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<tr>
<td></td>
<td>Hulls</td>
<td>27.1±0.3bA</td>
<td>23.8±0.2bB</td>
<td>19.1±0.1bc</td>
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<tr>
<td></td>
<td>Heads + hulls</td>
<td>23.1±0.2ac</td>
<td>21.6±0.3ac</td>
<td>16.3±0.3aA</td>
</tr>
<tr>
<td>Chitin</td>
<td>Heads</td>
<td>14.1±0.2aA</td>
<td>12.1±0.3aA</td>
<td>10.7±0.2bA</td>
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<tr>
<td></td>
<td>Hulls</td>
<td>24.6±0.4ab</td>
<td>22.8±0.4ab</td>
<td>19.3±0.4bB</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>16.4±0.4ac</td>
<td>14.0±0.2bc</td>
<td>12.7±0.3cC</td>
</tr>
</tbody>
</table>

1 Values are expressed on air-dry matter basis.
2 Values for each parameter represent mean±SD values with 3 observations.
  a-d Means within the same rows with different superscripts are significantly different (P<0.05).
  A-C Means within the same columns with different superscripts are significantly different (P<0.05).

Table 2. In vitro DM and CP digestibilities of heads, hulls and heads + hulls of three edible shrimp species

<table>
<thead>
<tr>
<th>Parameters (%)</th>
<th>Portions</th>
<th>Black tiger</th>
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</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Large</td>
<td>Small</td>
<td></td>
<td></td>
</tr>
<tr>
<td>DM digestibility</td>
<td>Heads</td>
<td>58.7±0.3aA</td>
<td>60.0±0.9aA</td>
<td>65.3±3.0bA</td>
</tr>
<tr>
<td></td>
<td>Hulls</td>
<td>44.0±0.1ab</td>
<td>42.7±0.4ab</td>
<td>53.3±0.4bB</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>55.9±0.1ac</td>
<td>57.3±0.2bc</td>
<td>61.5±0.5cC</td>
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<tr>
<td>CP digestibility</td>
<td>Heads</td>
<td>81.6±0.1aA</td>
<td>81.4±0.1aA</td>
<td>80.9±0.2aA</td>
</tr>
<tr>
<td></td>
<td>Hulls</td>
<td>66.8±1.0ab</td>
<td>67.4±0.6ab</td>
<td>76.1±0.2bB</td>
</tr>
<tr>
<td></td>
<td>Heads + hulls</td>
<td>80.6±0.6ac</td>
<td>79.4±0.3bc</td>
<td>79.4±0.5cC</td>
</tr>
</tbody>
</table>

1 Values are expressed on air-dry matter basis.
2 Values for each parameter represent mean±SD values with 3 observations.
  a-d Means within the same rows with different superscripts are significantly different (P<0.05).
  A-C Means within the same columns with different superscripts are significantly different (P<0.05).

Discussion

In the present study, comparing with hulls, heads were rich in CP and EE, and poor in CF, crude ash and chitin in all species excepting AR. This may be explained by the fact that hulls are composed of merely exoskeleton, but heads are composed of exoskeleton and internal organs, in particular the hepatopancreas, which is considered to function as storage organ of nutrients (Vijayavel et al., 2004). Interestingly, CP level of heads was smaller than that of hulls in AR, which was due to extremely increased level of EE in heads. High level of crude ash was observed in hulls, probably due to the high levels of calcium contained in exoskeleton of shrimps (Meyers et al., 1973). Levels of CF and chitin were about two times greater in hulls than heads, and CF level changed together with chitin level.

In addition, in vitro DM and CP digestibilities were also greater in heads than in hulls, which is reasonable because the level of chitin, non-digestible amino polysaccharide, was greater in hulls than heads in all species. In this connection,
chitin digestibility in broilers has reported to be as low as about 20% (Khempaka et al., 2006b), although chitinolytic activity occurred in mucosa of the proventriculus in broilers (Koh and Iwamae, 2013). Consequently, heads are considered to be more preferable than hulls as a source for SM, and hence, heads should be used in order to generate a good nutritional quality of SM. However, use of hulls for SM is not refused as long as the numerical ratio of hull to head is maintained at 1:1, because the former is much heavier than the latter in dry weight basis (from 2.5:1 in WL to 3.0:1 in small BT). In effect, the values of proximate composition and DM and CP digestibilities in heads + hulls were closer to the corresponding values in heads than those in hulls.

As mentioned already, AR heads contained as high as 21.1% of EE. Although it is impossible to explain the reason of such high value, the reproductive cycle of shrimps should not be ignorable, because lipid concentrations in the hepatopancreas of Penaeus japonicus has been reported to increase with increasing maturity of the ovaries (Teshima and Kanazawa, 1983). EE can be used as an energy source of chickens, but excessive EE is not preferable, because of quality deterioration by oxidation. Even fishmeal, widely used animal protein source, contains 8.4–12% of EE, which is about half or less than that of AR heads. In addition, DM and CP digestibilities of AR heads were lowest among head groups. Consequently, AR waste may be less suitable for the source of SM.

WL heads contained greater level of CP and smaller levels of CF and chitin, comparing with BT heads. These observations suggest that WL heads has more improved nutritional value than BT heads. In addition, the proximate composition of WL heads was comparable to that of fishmeal containing 55% of CP, in which 56.7% of CP, 9.4% of EE, 0.6% of CF, 22.6% of crude ash are contained (National Agriculture and Food Research Organization, 2009). DM digestibility of WL heads was the greatest among the SM measured in the present study, and CP digestibility of this was similar to that of BT heads which showed the greatest value. Accordingly, WL heads may be the most nutritious SM source among shrimp wastes tested in the present study, and hence which is recognised to be a promising alternative protein source. In effect, Khempaka et al. (2011) reported that SM made of WL heads can be contained up to 15% at the expense of soybean meal without showing any negative effects on growth performance in broilers.

Effect of body size in BT was found in some parameters: increased levels of EE, crude ash and chitin were found in most portion of large BT, but the difference was less than 2%, excepting levels of crude ash in hulls and chitin in heads + hulls. In addition, there was little difference in DM and CP digestibilities in heads and hulls between body sizes. Consequently, body size of BT may not affect the nutritional value of SM greatly, but waste of small BT is rather preferable to that of large BT for a SM source.

In order to generate more improved SM, problems resulted from chitin in SM, such as decreased digestibility, should be overcome (Khempaka et al., 2006b). Unfortunately, there is no study how to decrease chitin level in SM in poultry diets to our knowledge, because of limited studies about this field, but there is an interesting paper showing that formic acid treatment can reduce chitin level in SM for shrimps (Fox et al., 1994). This treatment may be worth trying in our future study.

In conclusion, 1) the nutritional quality of SM altered depending on the shrimp species and body portion, and 2) heads of WL can be a more nutritious source of SM in poultry diets.

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