JPSA
Japan Poultry Science Association

http://www.jstage.jst.go.jp/browse/jpsa
doi:10.2141/jpsa.0150128

Improvement in Nutritional Quality of Shrimp Meal with Autoclave and Chemical Treatments: an *in vitro* Study

Mustanur Rahman[1] and Katsuki Koh[2]

[1] Interdiciplineary Graduate School of Science and Technology, Shinshu University, Minamiminowa, Nagano 399−4598, Japan
[2] Faculty of Agriculture, Shinshu University, Minamiminowa, Nagano 399−4598, Japan

The present study was conducted to improve the nutritional quality of shrimp meal (SM) comprising of heads with hulls of black tiger shrimp (*Penaeus monodon*) waste by autoclaving and chemical treatments. The sun-dried SM was divided into 5 treatment groups, such as 1) control (untreated), 2) autoclaved (autoclaved at 121℃ for 10 min), 3) NaOH (treated with 3% NaOH), 4) HCl (treated with 3% HCl) and 5) formic acid (treated with 3% formic acid) groups. After treatment, they were ground to pass through 1.0 mm mesh screen and then used for analyses of chemical composition and *in vitro* dry matter (DM) and CP digestibilities. Data were subjected to one-way ANOVA and differences among treatment means ($P<0.05$) were distinguished with Tukey's test. There were no significant difference in chemical composition and *in vitro* DM and CP digestibilities between control and autoclaved groups, except ether extract level ($P<0.05$), suggesting that autoclaving affected the nutritional quality of SM little. NaOH group exhibited significantly decreased CP level and *in vitro* DM digestibility, increased crude ash (CA) level and unchanged *in vitro* CP digestibility, comparing with control group. These results suggest that NaOH treatment affected the nutritional quality of SM adversely. HCl and formic acids groups showed significantly increased CP level and *in vitro* digestibilities of DM and CP, and decreased CA level, showing that acid treatment can improve nutritional quality of SM: formic acid treatment may be more effective because of the greater values in CP level and digestibilities and decreased crude fibre level which was not observed in HCl group ($P<0.05$). The results obtained here suggest acid, especially formic acid, treatment is promising to improve the nutritional quality of SM but autoclaving and NaOH treatments.

Key words: autoclaved, chemical composition, chemical treatment, nutritional quality, shrimp meal

J. Poult. Sci., 53: 124−127, 2016

Introduction

The nutritional quality of shrimp meal (SM) has been investigated in our previous study to use this for an alternative protein source for broilers, and clarified that its nutritional quality was not good enough to be included more than 10% in a diet (Rahman and Koh, 2016), although SM quality was somewhat different depending on the portion and species (Rahman and Koh, 2014). Similar results have been reported in earlier studies (Fanimo *et al*., 1996; Gernat, 2001; Oduguwa *et al*., 2004; Khempaka *et al*., 2006a) where performances were decreased when chicken received diets containing more SM.

Several studies have been conducted to improve the nutritional quality of crustacean meals by means of physical and chemical treatments. For instance, autoclaving was applied to squilla (a stomatopod species) meal for broilers and failed to improve the nutritional quality, but assumed autoclaving temperature in their study is lower than usual one (Reddy *et al*., 1997). Alkali treatment, such as NaOH treatment, modified chemical composition of SM for broilers, so that CP and crude ash (CA) levels increased and crude fibre (CF) level decreased (Septinova *et al*., 2010). Acid treatment, such as HCl and formic acid treatments, has been reported to improve the nutritional quality of SM, but these were conducted to develop a feed ingredient not for chicken but for rats (Oduguwa *et al*., 1998) or shrimps (Fox *et al*., 1994).

In the present study, we evaluated the chemical composition and *in vitro* digestibilities of SM after receiving autoclaving or chemical treatments, and determined the most suitable treatment to improve the nutritional quality of SM as a CP source of chicken diets.

Received: August 24, 2015, Accepted: October 26, 2015
Released Online Advance Publication: November 25, 2015
Correspondence: Dr. K. Koh, Faculty of Agriculture, Shinshu University, Minamiminowa, Nagano 399−4598, Japan.
(E-mail: kkkohss@shinshu-u.ac.jp)

Materials and Methods

Preparation of Treated SM

The sun-dried SM made of heads with hulls of black tiger shrimp (*Penaeus monodon*) was obtained from a processing industry in Bangladesh. They were divided into five treatment groups: 1) control (untreated), 2) autoclaved, 3) NaOH treated, 4) HCl treated and 5) formic acid treated SM. The autoclave treatment was conducted according to Reddy *et al*. (1997) with slight modifications. Briefly, SM was autoclaved at 121℃ with 2.09 kg/cm^2 for 10 minutes and then sun-dried until achieving the moisture content of approximately 10%. The chemical (NaOH, HCl and formic acid) treatments were performed according to Septinova *et al*. (2010), Oduguwa *et al*. (1998) and Fox *et al*. (1994) with slight modifications. Briefly, about 100 g of sun-dried SM waste was suspended in 300 m*l* of 3% acids (HCl or formic acid) or alkali (NaOH) solutions at room temperature for 20 minutes. After that, they were filtered by using cheese cloth and washed with distilled water to adjust pH 7. After filtration, the solid portions were sun-dried and ground to pass through 1.0 mm mesh screen and then used as chemical treated SM. Each sample was divided into 4 aliquots for quadruplicate measurements.

Chemical Analysis

Proximate composition was analysed according to standard methods (AOAC, 1990). The chitin extraction process was done according to Ghanem *et al*. (2003), which was summarised as follows: about 1.0 g of dried SM mixed with 12.5 m*l* of 2.5 N NaOH solution, and placed in an oven at 75℃ for 6 hrs: filtrate crude chitin residue was dried at 105℃ in an oven for 1 hour. About 1.0 g of dried crude chitin was mixed with 10 m*l* of 1.7 N HCl and placed on a stir plate at room temperature for 6 hours. After filtering, the residue was washed with 95% ethanol (20 m*l*/g of crude chitin) followed by a final washing with distilled water and then dried. The dried content was weighed as chitin.

Digestibility Measurements

The *in vitro* dry matter (DM) and CP digestibilities of SM were determined according to Saunders *et al*. (1973) with slight modifications: briefly stated, about 250 mg of dried ground SM sample was suspended in 15 m*l* 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℃ for 3 hours in multi shaker (EYELA MMS-3010). After neutralisation with 0.5 N NaOH, the digesta was mixed with 7.5 m*l* of phosphate buffer at pH 8.0 containing pancreatin (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℃ 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:

$$\text{DM digestibility (\%)} = \frac{(\text{Dried sample weight} - \text{Dried residue weight})}{\text{Dried sample weight}} \times 100$$

$$\text{CP digestibility (\%)} = \frac{\text{Total N in sample} - \text{Total N in residue}}{\text{Total N in sample}} \times 100$$

Statistical Analysis

The data were analysed by one-way ANOVA. Contrasts between treatment groups means were evaluated by Tukey's test at a significance level of 5%.

Results

Chemical Composition of Treated SM (Table 1)

In control group, CP, CF, CA, ether extract (EE) and chitin accounted for 46%, 14%, 29%, 4% and 17% of dry weight of SM, respectively. These values did not change significantly by autoclave treatment, except the lower EE level ($P<0.05$). When SM was treated by NaOH, comparing with the corresponding values in control group, CP and EE levels decreased whereas CF and CA levels increased significantly but chitin level was not affected. When SM was treated by acids, such as HCl and formic acid, CP and EE levels increased significantly, comparing with the corresponding values of other treatment groups. A prominent effect was found in CA level which decreased to 40% by HCl and to 50% by formic acid treatment. Interestingly, CF and chitin levels were increased by HCl treatment and decreased by formic acid treatment, comparing with the corresponding values in other treatment groups.

Table 1. **Chemical composition of control (untreated) and treated shrimp meal[1, 2]**

Treatments	Chemical composition, %				
	Crude protein	Crude fibre	Crude ash	Ether extract	Chitin
Control	45.5±0.1^a	14.4±0.4^a	28.5±0.4^a	3.6±0.2^a	17.3±0.2^{ab}
Autoclaved SM	46.2±0.3^a	14.8±0.3^{ab}	29.6±0.4^a	2.4±0.1^b	16.6±0.2^a
NaOH treated SM	37.8±0.2^b	16.2±0.5^b	36.2±0.2^b	1.9±0.2^b	17.8±0.2^b
HCl treated SM	54.3±0.1^c	18.3±0.6^c	16.9±0.1^c	4.2±0.1^c	19.2±0.2^c
Formic acid treated SM	58.1±0.4^d	12.7±0.2^d	13.6±0.2^d	5.4±0.1^d	15.3±0.4^d

[1] Values are expressed on air-dry matter basis.
[2] Values for each parameter represent mean±SE values with 4 observations.
[a–d] Means within the same column with different superscripts are significantly different ($P<0.05$).

Table 2. ***In vitro*** **DM and CP digestibilities of control (untreated) and treated shrimp meal[1, 2]**

Treatments	DM digestibility, %	CP digestibility, %
Control	44.3±0.3[a]	73.9±0.4[a]
Autoclaved SM	43.9±0.3[a]	73.8±0.3[a]
NaOH treated SM	39.6±0.4[b]	72.8±0.3[a]
HCl treated SM	48.1±0.5[c]	76.2±0.5[b]
Formic acid treated SM	51.6±0.3[d]	79.1±0.3[c]

[1] Values are expressed on air-dry matter basis.
[2] Values for each parameter represent mean±SE values with 4 observations.
[a–d] Means within the same column with different superscripts are significantly different ($P<0.05$).

Digestibility of Treated SM (Table 2)

DM and CP digestibilities in control group were about 44% and 74%, respectively. Autoclave treatment did not improve these values and NaOH treatment rather deteriorated DM digestibility. In contrast, acid treatments improved DM and CP digestibilities and the magnitudes of improvements were greater in formic acid treatment.

Discussion

The obtained results revealed that autoclaving failed to improve the nutritional quality of SM. Similar observation was found in squilla meal reported by Reddy *et al*. (1997). Our autoclaving condition (at 121℃ for 10 min) seemed to be more severe than their condition (at 1.09 kg/cm^2 for 5 min), because according to Antoine equation (Thomson, 1946) of temperature in an autoclave increased until 102.05 ℃ at a pressure of 1.09 kg/cm^2. This may suggest that this level of autoclaving is not effective to alter the chemical composition of crustacean meals. In this connection, autoclaving can improve pepsin digestibility of feather meal significantly, even though there was no significant impact on the chemical composition (Kim and Patterson, 2000). This makes us expect an improved *in vitro* digestibility of autoclaved SM in the present study, but the fact was different: not only DM but also CP digestibilities did not show improved values. Consequently, autoclaving may not be suitable to improve the nutritional quality of SM.

NaOH treated SM had lower CP level and higher CA level than other treatment groups ($P<0.05$), which was contrary to the results of Septinova *et al*. (2010) who found increased CP, CA levels and decreased CF level in NaOH treated SM. Thus, it is quite difficult to discuss this inconsistency, because there was no difference in treatment condition between them. Our results exhibited decreased DM and CP digestibilities and it may assume that CP retention should be low in chickens, although Septinova *et al*. (2010) found increased protein retention in broilers given diet containing NaOH treated SM. In this connection, it has been reported that NaOH treated feather meal showed decreased protein and increased ash levels, although *in vitro* protein digestibility increased (Papadopoulos *et al*., 1985) and no significant difference in CP level (Steiner *et al*., 1983). Therefore, further investigation is necessary to confirm the effect of NaOH treatment on nutritional quality of SM.

Beneficial effect was obtained in two acid treatment experiments: significantly higher CP and lower CA levels were observed in both acid treated SM, comparing with the corresponding values of other treatment groups. This may be results of leaching the minerals, such as calcium, in exoskeleton (No *et al*., 1989; Fox *et al*., 1994; Oduguwa *et al*., 1998), and accordingly, relative content of CP increased. In this context, increased CF and chitin levels in HCl treated SM can also be explained. However, it is very interesting that formic acid treatment decreased CF and chitin levels, suggesting that chitin, main source of CF and possible factor to decrease digestibility (Austin *et al*., 1981; Fanimo *et al*., 2006; Khempaka *et al*., 2006b), was leached from SM by formic acid treatment. In this regard, Win and Stevens (2001) reported that formic acid treatment results in a weakening of the crystal structure and able to dissolve the shrimp chitin.

Treating the SM with acid solution resulted in increased DM and CP digestibilities relative to other treatments ($P<0.05$). Similarly, it has been reported that apparent protein digestibility increased in rats (Oduguwa *et al*., 1998) and protein retention increased in broilers (Septinova *et al*., 2010) when HCl treated SM was included in diet. Moreover, significantly greater digestibilities observed in formic acid treated SM, may be because of the higher CP level and lower CF, CA and chitin levels in formic acid treated SM. The positive response of formic acid treated SM was also observed by Fox *et al*. (1994) who reported an improvement in growth and survival rate in marine shrimps (*Penaeus monodon*) fed diet containing formic acid treated SM.

In conclusion, the obtained results revealed that formic acid treatment showed improved nutritional quality of SM over autoclaving, NaOH and HCl treatments, and suggest that formic acid treated SM can be used as a potential source of protein in chicken diets.

References

Association of Official Analytical Chemists. Official Method of Analysis. 15th edition. Association of Analytical Chemists, Washington, DC. 1990.

Austin PR, Brine CJ, Castle JE and Zikakis JP. Chitin: New facets of research. Science, 212: 749−753. 1981.

Fanimo AO, Mudama E, Umukoro TO and Oduguwa OO. Substitution of shrimp waste meal for fish meal in broiler chicken rations. Tropical Agriculture (Trinidad), 73: 201−205. 1996.

Fanimo AO, Susenbeth A and Sudekum KH. Protein utilisation, lysine bioavailability and nutrient digestibility of shrimp meal in growing pigs. Animal Feed Science and Technology, 129: 196−209. 2006.

Fox CJ, Blow P, Brown JH and Watson I. The effect of various processing methods on the physical and biochemical properties of shrimp head meals and their utilization by juvenile *Penaeus monodon* Fab. Aquaculture, 122: 209−226. 1994.

Gernat AG. The effect of using different levels of shrimp meal in laying hen diets. Poultry Science, 80: 633−636. 2001.

Ghanem A, Ghaly AE and Chaulk M. Effect of shrimp processing procedures on the quality and quantity of extracted chitin from the shells of northern shrimp *Pandalus borealis*. Journal of Aquatic Food Product Technology, 12: 63−79. 2003.

Khempaka S, Koh K and Karasawa Y. Effect of shrimp meal on growth performance and digestibility in growing broilers. Journal of Poultry Science, 43: 250−254. 2006a.

Khempaka S, Mochizuki M, Koh K and Karasawa Y. Effect of chitin in shrimp meal on growth performance and digestibility in growing broilers. Journal of Poultry Science, 43: 339−343. 2006b.

Kim WK and Patterson PH. Nutritional value of enzyme- or sodium hydroxide-treated feathers from dead hens. Poultry Science, 79: 528−534. 2000.

No KH, Meyers SP and Lee SK. Isolation and characterization of chitin from craw shell waste. Journal of Agricultural and Food Chemistry, 37: 575−579. 1989.

Oduguwa OO, Fanimo AO, Iyayi EA, Kalejaiye OO and Oyekola OA. Preliminary studies on the effects of different processing methods on the nutritive value of shrimp waste meal. Nigerian Journal of Animal Production, 25: 139−144. 1998.

Oduguwa OO, Fanimo AO, Olayemi VO and Oteri N. The feeding value of sun-dried shrimp waste-meal based diets for starter and finisher broilers. Archivos de Zootecnia, 53: 87−90. 2004.

Papadopoulos MC, El-Boushy AR and Roodbeen AE. The effect of varying autoclaving conditions and added sodium hydroxide on amino acid content and nitrogen characteristics of feather meal. Journal of the Science of Food and Agriculture, 36: 1219−1226. 1985.

Rahman M and Koh K. Nutritional quality and *in vitro* digestibility of shrimp meal made of heads and hulls of black tiger (*Penaeus monodon*), white leg (*Litopenaeus vannamei*) and argentine red (*Pleoticus muelleri*) Shrimps. Journal of Poultry Science, 51: 411−415. 2014.

Rahman M and Koh K. Effect of shrimp meal made of heads of black tiger (*Penaeus monodon*) and white leg (*Litopenaeus vannamei*) shrimps on growth performance in broilers. Journal of Poultry Science, 53: 149−152. 2016.

Reddy, VR, Reddy VR and Qudratullah S. Utilisation of squilla meal (a novel animal protein source) by broilers. British Poultry Science, 38: 263−269. 1997.

Saunders RM, Connor MA, Booth AN, Bickoff EM and Kohler GO. Measurement of digestibility of alfalfa protein concentrates by *in vivo* and *in vitro* methods. Journal of Nutrition, 103: 530−535. 1973.

Septinova D, Kurtini T and Tantalo S. Evaluation the usage of treated shrimp waste as protein source in broiler diet. Journal of Animal Production, 12: 1−5. 2010.

Steiner RJ, Kellems RO and Church DC. Feather and hair meals for ruminants. IV. Effects of chemical treatments of feathers and processing time on digestibility. Journal of Animal Science, 57: 495−502. 1983.

Thomson GW. The Antoine equation for vapor-pressure data. Chemical reviews, 38: 1−39. 1946.

Win NN and Stevens WF. Shrimp chitin as substrate for fungal chitin deacetylase. Applied Microbiology and Biotechnology, 57: 334−341. 2001.