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Original Article

Bacteriological Assessment of Value-Added Ready-to-Cook/Eat Shrimps Processed for Export from Bangladesh following the Guidelines of International Standards

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In the present investigation a total 630 black tiger and brown ready-to-cook/eat shrimp samples collected from a fish processing industry was analyzed. Total aerobic plate count (APC) was found to exceed the International Commission on Microbiological Specification for Foods (ICMSF) standard in case of 2.26% black tiger shrimp and 1.86% brown shrimp samples. *Escherichia coli* counts of all the samples were within the acceptable limit of ICMSF standard. However, considering the Council of the European Communities (EEC) standard, coliform (*E. coli*) counts of 0.96% black tiger and 0.62% brown shrimp samples exceeded the acceptable limit. Coagulase-positive *Staphylococcus aureus, Salmonella* and *Listeria monocytogenes* were not detected from any specimens. The fish processing seems to follow the hazard analysis and critical control point (HACCP) principles that should be more strictly maintained in order to keep the quality of shrimp of international standard.

Keywords: Ready-to-cook/eat shrimps, Bacteriological count, Hazard analysis and critical control point (HACCP)

Introduction

Fisheries sector is the countries second highest earner of foreign exchange next to garments manufacturing. Shrimp is the main exported fisheries products of Bangladesh[1]. About 1.2 million people directly and 10.0 million people indirectly are involved in this sector for their livelihood[2]. Although shrimp and prawns exported from Bangladesh are almost entirely in block-frozen form[3], but recently some processors establish modern equipments to produce ready-to-cook shrimp in order to fulfil the requirement of foreign buyer.

Cook shrimp is a special kind of value added ready-to-eat food and it has an international demand in the developed countries, especially in Europe, although Bangladesh start value added shrimps production from the later part of 80's[4]. Like other ready-to-eat food, cooked shrimp is considered as a serious health risk concern food for consumer due to its intended uncooked direct consumption. Therefore, cooked food and its associated microorganisms play an important role over consumer health. Among the microorganism, which associated with cook food, *Staphylococcus aureus* has a great importance because *S. aureus* is quite sensitive to microbial competition[5]. Reports available indicated that the higher the concentrations of competitive microorganism in milk the lower the rate of *S. aureus*[6]. So cooked shrimps may create great risk for *S. aureus* where the other microbial competition is very low. On the other hand, uncooked or undercooked meat often acts as a source of infection like *Escherichia coli* O157:H7, whereas ready-to-eat processed foods are potential source of listeriosis that are stored for long periods at refrigeration temperature 4°C[7]. *Salmonella* contaminated uncooked raw shrimps were also reported by several workers[8-10].

Besides these, seafood industries of Bangladesh faced a ban during the past decades by the European Union at the time of its official visit in 1997, considering that the Bangladeshi seafood is unhygienic to eat[3]. On the other hand, it was mandatory by the United States Food and Drug Administration (US FDA) that all seafood shipped to the USA from December 17, 1997 should have to be processed under hazard analysis and critical control point (HACCP) regulation[11].

Keeping the above things in view, the present study had been undertaken to analyze the ready-to-cook/eat shrimps from a leading seafood processor of Bangladesh in order to assess the microbial status from the period of April 2005 to March 2006 following the guidelines of two international food standards.

Materials and Methods

A modern shrimp processing plant was selected for microbiological assessment of ready-to-cook shrimp processing ability, which had modern shrimp cooking facilities with an approved HACCP plan. The processing plant had been regularly monitored and verified by respective competent authority.

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Total 630 shrimp samples including 310 black tiger and 320 brown shrimps were collected from the cold store of the shrimp processing plant after the completion of each day's production during the period from April 2005 to March 2006. Samples (8 oz each) were aseptically collected and transported to the laboratory according to the recommendation of the United States Food and Drug Administration (US FDA)[12].

For bacterial enumeration the US FDA recommended method[12] was followed. For the enumeration of *Listeria monocytogenes*, International Organization for Standardization (ISO) recommended method[13] was used. Aerobic plate count (APC) was performed by pour plate method using plate count agar (PCA), which was incubated at 35 ± 1°C for 48 ± 2 h. Lauryl sulphate tryptose (LST) broth was used for isolation of *Escherichia coli*. After incubation at 35 ± 1°C for 48 ± 2 h the broth tubes that showed gas production was selected and a loop-full of the broth culture was transferred to EC broth that was further incubated at 45.5 ± 0.2°C for 48 ± 2 h. Gassing tube was selected for *E. coli* enumeration using most probable number (MPN) method. For isolation and enumeration of *Salmonella* Rappaport Vassiliadis (RV), tetrathioate (TT) broth, Hektoen enteric agar (HEA), bismuth sulphite agar (BSA) and xylose lysine desoxycholate (XLD) agar media were used. Enumeration of *Staphylococcus aureus* was performed on Baird Parker agar medium. Coagulase test was performed according to standard method[12].

Results and Discussion

The bacteriological results of the cooked black tiger peel and undeveined tail on shrimps are given in Table 1. The aerobic plate count (APC) of 97.74% samples was within the acceptable limit, while the count was within the marginally acceptable limit for the rest 2.25% samples according to the ICMSF standard[14]. ICMSF recommends three class sampling plan and microbial limit [(n = 5, c = 2, m = 5 x 10^5 and M = 10^7), where, n = No. of sample; c = No. of sample giving values between 'n' and 'M'; m = maximum recommended bacterial counts for good quality products, M = maximum recommended bacterial counts for marginally acceptable quality]. Considering the standard limit of ICMSF[17], out of 5 samples, 3 (60%) samples should be within the limit of 'm' value (5.0 x 10^5 cfu/g) and 2 (40%) samples should be within the limit of 'M' value (1.0 x 10^7 cfu/g).

All the samples showed *E. coli* count within the acceptable limit of ICMSF standard[14] (*i.e.*, 11 cfu/g) and coagulase-positive *S. aureus* was identified from any of the shrimp samples. Considering the EEC standard, *E. coli* count was within the acceptable limit (*i.e.*, 1 cfu/g) for 99.03% samples, while only 0.96% samples showed the count above the acceptable limit but the value was below the marginally acceptable limit (*i.e.*, 100 cfu/g). *Salmonella* and *Listeria monocytogenes* were not detected from any of the samples.

Table 2 represents the bacteriological results of individual quick frozen (IQF) cooked brown peel and undeveined shrimps. Aerobic plate count (APC) of 95% and 1.87% samples were within the acceptable and marginally acceptable limit respectively against ICMSF standard[14]. *E. coli* count was acceptable and no coagulase-positive *S. aureus* identified from any the samples tested. On the other hand, 99.37% samples showed acceptable results with respect to *E. coli* count against EEC standard[16], while only 0.62% samples qualified as marginally acceptable. No *Salmonella* or *L. monocytogenes* was identified from any sample.

Table 1. *Bacterial count of individual quick frozen (IQF) cooked black tiger peeled and deveined tail on shrimps*

Period	No. of sample tested	Aerobic plate count (cfu/g)		*Escherichia coli* (cfu/g)		*Coagulase-positive Staphylococcus aureus* (cfu/g)	
		Acceptable (ICMSF[a])	Marginally acceptable (ICMSF[b])	Acceptable (ICMSF[c] / EEC[d])	Marginally acceptable (ICMSF[e] / EEC[f])	Acceptable (ICMSF[g] / EEC[h])	Marginally acceptable (EEC[i])
April 2005	20	20	0	20 / 20	0 / 0	20 / 20	0
May 2005	30	30	0	30 / 30	0 / 0	30 / 30	0
June 2005	40	39	1	40 / 39	0 / 1	40 / 40	0
July 2005	40	38	2	40 / 40	0 / 0	40 / 40	0
August 2005	40	40	0	40 / 39	0 / 1	40 / 40	0
September 2005	40	38	2	40 / 39	0 / 1	40 / 40	0
October 2005	30	30	0	30 / 30	0 / 0	30 / 30	0
November 2005	30	29	1	30 / 30	0 / 0	30 / 30	0
December 2005	10	10	0	10 / 10	0 / 0	10 / 10	0
January 2006	-	-	-	- / -	- / -	- / -	-
February 2006	10	10	0	10 / 10	0 / 0	10 / 10	0
March 20006	20	19	1	20 / 20	0 / 0	20 / 20	0
Total	310	303 (97.74%)	7 (2.25%)	310 (100%) / 307 (99.03%)	0 (0.0%) 3 (0.96%)	310 (100%) / 310 (100%)	0 (0.0%)

ICMSF = International Commission on Microbiological Specification for Foods[17]; EEC = European Union Standard[16]; ICMSF[a] = 5.0 x 10^5 cfu/g; ICMSF[b] = 1.0 x 10^7 cfu/g; ICMSF[c] = 11 cfu/g; EEC[d] = 1 cfu/g; ICMSF[e] = 500 cfu/g; EEC[f] = 10 cfu/g; ICMSF[g] = <1.0 x 10^3 cfu/g; EEC[h] = 100 cfu/g; EEC[i] = 1.0 x 10^3 cfu/g.

Table 2. *Bacterial count of individual quick frozen (IQF) cooked brown peeled and deveined shrimps*

Period	No. of sample tested	Aerobic plate count (cfu/g)		*Escherichia coli* (cfu/g)		*Coagulase-positive Staphylococcus aureus* (cfu/g)	
		Acceptable (ICMSF[a])	Marginally acceptable (ICMSF[b])	Acceptable (ICMSF[c] / EEC[d])	Marginally acceptable (ICMSF[e] / EEC[f])	Acceptable (ICMSF[g] / EEC[h])	Marginally acceptable (EEC[i])
April 2005	20	20	0	20 / 20	0 / 0	20 / 20	0
May 2005	-	-	-	- / -	- / -	- / -	-
June 2005	40	40	0	40 / 39	0 / 1	40 / 40	0
July 2005	40	38	0	40 / 40	0 / 0	40 / 40	0
August 2005	50	48	2	50 / 9	0 / 1	50 / 50	0
September 2005	50	49	2	50 / 50	0 / 0	50 / 50	0
October 2005	40	40	1	50 / 40	0 / 0	40 / 40	0
November 2005	20	20	0	20 / 20	0 / 0	20 / 20	0
December 2005	-	-	-	- / -	- / -	- / -	-
January 2006	20	19	1	20 / 20	0 / 0	20 / 20	0
February 2006	10	10	0	10 / 10	0 / 0	10 / 10	0
March 20006	20	20	0	20 / 20	0 / 0	20 / 20	0
Total	320	304 (95.00%)	6 (1.87%)	320 (100%) / 318 (99.37%)	0 (0.0%) / 2 (0.62%)	320 (100%) / 320 (100%)	0 (0.0%)

ICMSF = International Commission on Microbiological Specification for Foods[17]; EEC = European Union Standard[16]; ICMSF[a] = 5.0 x 10^5 cfu/g; ICMSF[b] = 1.0 x 10^7 cfu/g; ICMSF[c] = 11 cfu/g; EEC[d] = 1 cfu/g; ICMSF[e] = 500 cfu/g; EEC[f] = 10 cfu/g; ICMSF[g] = <1.0 x 10^3 cfu/g; EEC[h] = 100 cfu/g; EEC[i] = 1.0 x 10^3 cfu/g.

In fact *S. aureus* is highly vulnerable to destruction by heat treatment and nearly all sanitizing agents[12]. Improper cooking and post cooking handling may facilitates the contamination of *S. aureus*, whereas *E. coli* is a classical indicator of faecal contamination in food[15]. Presence of *Salmonella* is also a faecal indicator, while *Listeria* excreted in animal faeces and is widely distributed in soil and water[16]. Absence of *Salmonella* and *Listeria* from any of the samples along with absence of coagulase-positive *S. aureus* from the ready-to-cook/eat shrimp samples reflects excellent personal hygiene maintained by the workers of the plant under investigated. But very negligible percentage of *E. coli* warrants the effectiveness of sanitation procedure. This finding complies with the report of Hata *et al*.[17].

International guideline for microbiological criteria in respect of many food staff has not yet been established[18]. Existing guideline of Codex Alimentarius Commission[19] for dried milk powder, food for infants and children and the criteria for certain fish and fishery product are available but no criterion yet established by the commission on cooked crustaceans. On the other hand, Council of the European Communities (EEC) recently established new regulation on microbial criteria for food[18,20] including the criterion of cooked crustaceans and has a wide acceptance in European countries. Besides these, the standard of ICMSF[14] has a worldwide acceptance, which is also cover the standard microbial criteria of cook crustaceans and hence these two standards were selected in this investigation for comparison.

No investigation carried out on ready-to-cook/eat shrimps in Bangladesh yet. Very few data are available on microbial enumeration of shrimps from neighbouring countries like India[17,21] and Pakistan[22]. In their investigations the microbial counts of raw and cook shrimps in comparison with international standard were overlooked. So the present investigation might meet this gap.

In conclusion, the most of the samples tested herein are within compliance with the two international standards except for a few cases. It could be declared that Bangladeshi ready-to-cook/eat shrimps are hygienic and safe for consumption. Our findings are in agreement with the Food and Veterinary mission of European Union, made its last visit on 8th-16th November 2005 in Bangladesh[23]. In fact, after the incidence of 1997, seafood exporters of Bangladesh prepared their establishment/facilities in all respects, and most importantly they updated their HACCP activities and providing good training facilities for their workers to meet the requirements of European Commission and along with the other international food regulating bodies.

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