

Impact of temperature and radiation on elimination of associated bacteria from preserved dried prawn, *Macrobrachium lamarrei*

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Abstract

The impact of different temperatures (0, 40, 60, 80 and 100° C for an hour) and radiation (0, 2.5, 5.0, 7.5 and 10.0 kGy) and combined treatment (temperature and radiation) for decontamination of bacteria from dried prawn, *Macrobrachium lamarrei* was also investigated under laboratory condition. The total viable bacterial (TVB), total staphylococcal (TS), total coliform (TC), total faecal coliform (TFC), total *Aeromonas* (TA) and total fungal (TF) count were ranged from 2.7×10^8 to 5.3×10^8 cfu/g, 2.2×10^6 to 6.0×10^6 cfu/g, 2.9×10^4 to 9.5×10^4 cfu/g, 2.8×10^3 to 8.1×10^3 cfu/g, 5.7×10^3 to 8.5×10^3 cfu/g and 1.6×10^4 to 3.8×10^4 cfu/g respectively. Seventy eight bacterial strains were isolated and identified out of which 18 (23.07 %) were *Staphylococcus aureus*, 11 (14.10 %) were *Micrococcus varians*, 8 (10.25%) were *Aeromonas hydrophila*, 5 (6.41%) were *Klebsiella ozaenae*, 7 (8.97%) were *Bacillus subtilis*, 7 (8.97%) were *Escherichia coli*, 5 (6.41%) were *Bacillus megaterium*, 7 (8.97%) were *Klebsiella edwardsii*, 6 (7.69%) were *Pseudomonas aeruginosa* and 4 (5.12%) were *Micrococcus radiodurans*. Inactivated was more favorable at 100° C for TC, TFC, TA and TF and at 5.0 kGy for TA and TF and combined treatments (40° C+2.5 kGy, 60° C+2.5 kGy and 80° C+2.5 kGy) for TFC, TA and TF respectively. TS, TC and TVB were completely eliminated in treatment 40° C+5.0 kGy, 60° C+2.5 kGy, 80° C+2.5 kGy, 40° C+7.5 kGy, 60° C+7.5 kGy and 80° C+7.5 kGy respectively. Results demonstrated that the combination treatments were more effective than single treatment for eliminating the associated microorganisms/bacteria from dried prawn.

Key words: Microorganism, dried prawn, temperature, radiation, treatment.

INTRODUCTION

Freshwater prawn is very cheap, tasty and easily available in the market. It is a rich source of animal proteins and vitamins A and D. Bangladesh is one of the prawn producing countries of the world and dried prawn has been coming up as a good exportable commodity. Bangladesh is earning a huge amount of foreign exchange through the export of dried prawn to Japan, U.S.A., Australia etc. During the fiscal year (2010-2011), 3323.7 and 36.43 crore taka of foreign currencies has been earned by exporting 47879.91 and 1200.1 metric tons shrimp and prawn and dry, salted and dehydrated fish respectively. (FSYBB, 2010-2011). So, prawn fishery products play an important role in the national economy of the country.

Dried prawn is highly nutritious, perishable and can't be kept for a long time for further consumption. Thus, in question of preservation, spoilage of fish has drawn the attention

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of people and had put effort to know the reasons of spoilage. According to Frazier & Westhoff (1988), fish is the most susceptible to autolysis, oxidation and hydrolysis of fats and microbial spoilage. Banwart (1979) reported that the microorganisms present in fish include those which are associated with the raw material and acquired during catching, handling and processing. The genera of bacteria most frequently found in fresh water fishes are: *Bacillus*, *Staphylococcus*, *Enterobacter*, *Klebsiella*, *Serratia*, *Citrobacter*, *Aeromonas*, *Escherichia*, *Micrococcus* (Jay, 1977).

Fish preservations are aimed at preventing the microbial spoilage of fish, fish products and the growth of the food borne pathogens. The microbiology of the dried prawn and its preservation by temperature, radiation and combination treatments could be considered important from the different point of view: (1) Spoilage by microbes may change the odor, texture, color and taste of the prawn ultimately causing loss of economy; (2) Pathogens may persist in the prawn and may serve as transporting agents of pathogens.

Therefore, the objectives of this study was to enumerate and detect microorganisms from preserved dried prawn and assess the suitability of temperature, radiation and combination treatments for long term preservation of dried prawn at room temperature.

MATERIALS AND METHODS

Sample collection: Dried prawn samples, *Macrobrachium lamarrei* were purchased from Savar puratan bazar fish market. All the samples were aseptically packed in pre-sterilized polyethylene bags. The polyethylene bags were sealed air tightly after collecting the samples.

Experimental design: Sufficient replicas of samples (100g each) were prepared for microbial enumeration. Among them, replicas no. 1-5 were selected for initial microbial enumeration. The replicas no. 6-10, 11-15 and 16-35 were selected for heat, radiation and combination treatments respectively. The samples of replicas 6, 7, 8, 9 and 10 were heated at 0, 40, 60, 80 and 100° C temperature for an hour respectively. Similarly replicas 11, 12, 13, 14 and 15 were exposed to 0, 2.5, 5.0, 7.5 and 10 kGy of gamma radiation respectively. For combination treatments, five replicas of samples were heated initially at 40 for an hour respectively. Then the heated samples were exposed to different doses (0, 2.5, 5.0, 7.5 and 10 kGy) of radiation. Similar procedure was followed for the samples heated at 60° C, 80° C and 100° C respectively. All of the samples were stored at room temperature and examined for microbiological qualities.

Enumeration of microorganisms: Nutrient agar, Staphylococcal agar, McConkey agar, mFc agar, Starch ampicillin agar and potato dextrose agar were used to enumerate total viable bacteria, total *Staphylococci*, total coliform, total fecal coliform, total *Aeromonas* and total fungi respectively. Spread plate technique (APHA, 1976) was followed to culture all tested microorganisms. Fecal coliform was cultured at 44.5⁰ C, whereas, all the other culture media were incubated at 37⁰ C.

Identification of the bacterial strains: All the isolated bacterial stains were identified by the methods as described in “Bergey’s Manual of Systemic Bacteriology” (Baumann & Schubert, 1984).

Statistical analysis: Univariate and bivariate statistical techniques were used for analyzing different microorganisms. The correlation analysis was used for identifying relationship among the total counts of different microorganisms.

RESULTS AND DISCUSSION

Enumeration and confirmation of microorganisms: The enumeration of microorganisms, *e.g.*, total viable bacteria, total *Staphylococci*, total coliform, total fecal coliform, total *Aeromonas* and total fungi isolated from dried prawn have been presented in the Figure 1. The lowest, highest and the average count of different bacteria indicated the similar trends of contamination. Each of the bacterial counts suggested the presence of high number of bacteria in all the tested dried prawn.

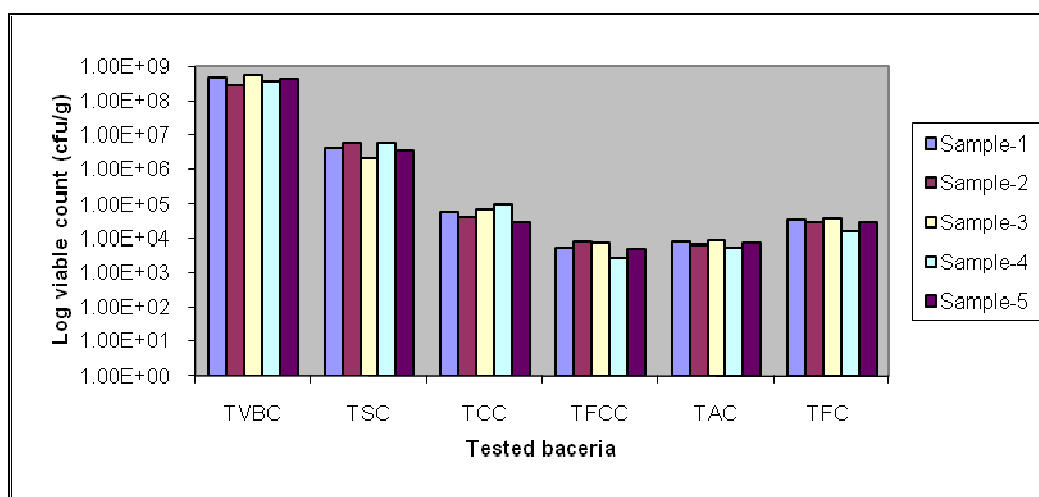


Fig. 1. Total count of different bacteria from dried prawn samples

(TVBC-total viable bacterial count, TSC-total *Staphylococcus* count, TCC-total coliform count, TFCC-total fecal coliform count, TAC-total *Aeromonas* count, TFC-total fungi count).

The highest and lowest count of total viable bacteria was 5.3×10^8 cfu/g and 2.7×10^8 cfu/g and was observed in sample-3 and in sample 2. Cho *et al.* (1992) and Ito *et al.* (1984). Khatun *et al.* (2003) also reported the variable counts in different treatments. Like total viable counts, all the other microbial studies suggested minor fluctuations of the tested microorganisms. Presence of *Staphylococcus sp* suggests that there was higher level of environmental contamination and its presence indicates the possible risks of food poisoning. Different counts of coliform bacteria may probably be due to the status of water quality used for washing (Haque, 1997). Similarly, the level of other

microorganisms e.g., faecal coliform, *Aeromonas* and fungi suggests the practice of inadequate hygienic measure, mishandling, improper storage and all unhygienic condition during sampling and processing. Contamination in dried prawn may be due to indigenous microbial contaminants, water source, processing or handling and selling condition of the prawn samples.

Figure 2 represent that among the seventy eight (78) isolated bacterial strains, 18 (23.07 %) were detected as *Staphylococcus aureus*, 11 (14.10%) as *Micrococcus varians*, 8 (10.25%) as *Aeromonas hydrophila*, 5 (6.41%) as *Klebsiella ozaenae*, 7 (8.97%) as *Bacillus subtilis*, 7 (8.97%) as *Escherichia coli*, 5 (6.41%) as *Bacillus megaterium*, 7 (8.97%) as *Klebsiella edwardsii* and 6 (7.69%) *Pseudomonas aeruginosa*, 4 (5.12%) as *Micrococcus radiodurans*. Similar taxonomical investigation was done by Islam *et al.* (2001), Amatun Nur (2001), Rashid *et al.* (1996), Anwar *et al.* (1988), Banwart (1979), Jay (1977) and Dyer (1954).

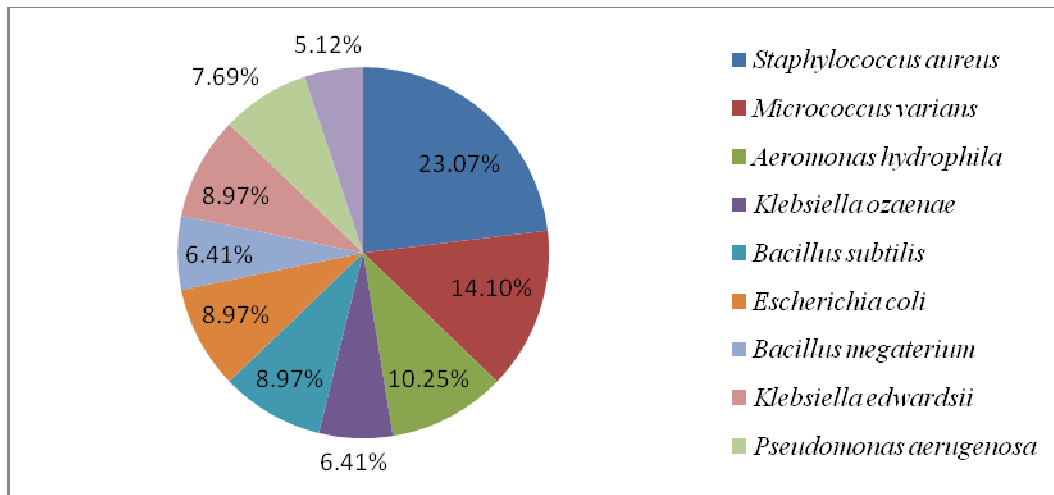


Fig. 2. Generic distribution of bacteria associated with dried prawn samples

Statistical analysis: In the present investigation, it was found positive correlation among the total counts of different microorganisms (TVBC & TVBC, TVBC & TSC, TVBC & TCC, TVBC & TFCC, TVBC & TAC, TVBC & TFC, TSC & TSC, TSC & TCC, TSC & TFCC, TSC & TAC, TSC & TFC, TCC & TCC, TCC & TFCC, TCC & TAC, TCC & TFC, TFCC & TFCC, TFCC & TAC, TFCC & TFC, TAC & TAC, TAC & TFC, TFC & TFC).

Microbial status of the non-heated and heated dried prawn: The results shown in Fig. 3 suggested that all the microbial counts decreased gradually as increased temperature in treatments. Total coliform count, total fecal coliform count, total *Aeromonas* count and total fungi count were not observe at temperature 100° C.

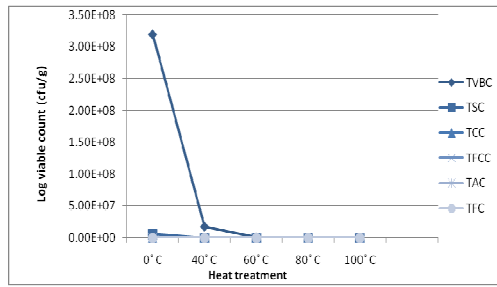


Fig. 3. Effect of temperature treatment on different bacterial counts

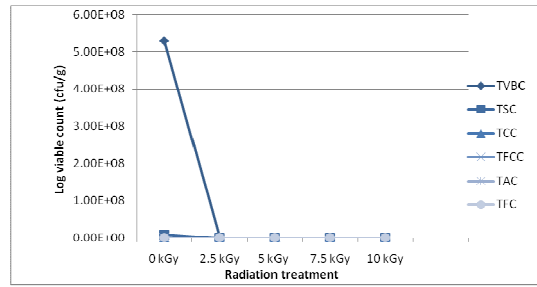


Fig. 4. Effect of radiation treatment on different bacterial counts

Microbial status of the non-irradiated and irradiated dried prawn: The data presented in the Fig. 4 indicated that the total *Aeromonas* and fungi were found to inactivate at 5.0 kGy and onwards and total viable bacteria, staphylococcal bacteria, coliform, and fecal coliform were eliminated completely at the irradiation dose of 7.5 kGy.

Effect of combination treatments on microorganisms associated with dried prawn: The results presented in the Fig. 5 A, B, C and D indicated that the total viable bacterial counts were reduced gradually at the combination treatments of 40° C, 60° C, 40° C + 2.5 kGy, 60° C, + 2.5 kGy, 40° C + 5.0 kGy, 80° C, 80° C + 2.5 kGy, 100° C, 60° C + 5.0 kGy, 80° C + 5.0 kGy and 100° C + 2.5 kGy respectively. Thus no count of total viable bacteria was observed at the rest combination treatments. Similarly, total staphylococcal counts were decreased gradually at 40° C, 60° C, 40° C + 2.5 kGy, 60° C + 2.5 kGy, 80° C, 100° C and 80° C + 2.5 kGy. At the rest of combination treatments the staphylococcal count was nil. The total coliform counts were only observed at the combination treatments of 40° C, 40° C + 2.5 kGy, 60° C, 60° C + 2.5 kGy, and 80° C and also reduced gradually. Total fecal coliform count, total *Aeromonas* count and total fungal count were only assessed at all the control (40° C, 60° C and 80° C) of all combination treatments except 100° C (Control).

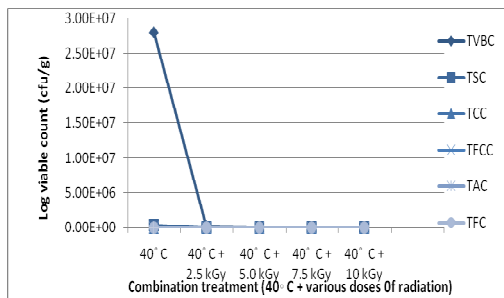


Fig. 5 A. Effect of 40° C temperature with different doses of radiation on different bacterial counts

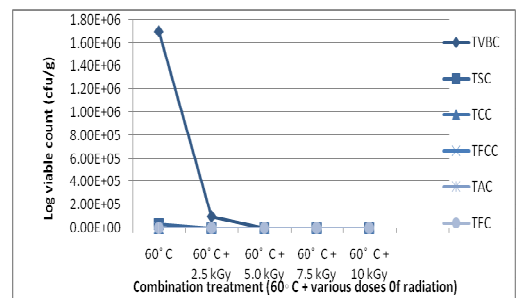


Fig.5 B. Effects of 60° C temperature with different doses of radiation on different bacterial counts

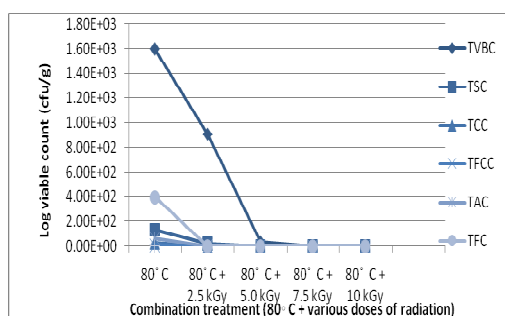


Fig. 5 C. Effect of 80° C temperature with different doses of radiation on different bacterial counts

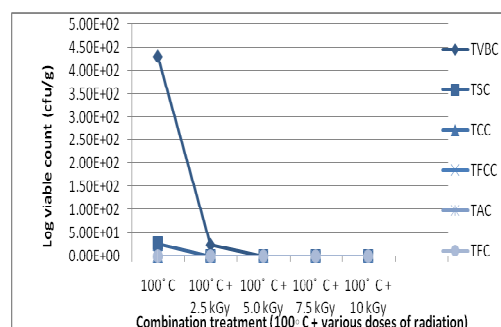


Fig.5 D. Effects of 100° C temperature with different doses of radiation on different bacterial counts

Finally, the inference is that the dried prawn samples were heavily contaminated with various microorganisms. The microbial contamination of the dried prawn samples depends upon the sources of the prawn, p^H , temperature, acidity etc. of the water, natural handling, processing, drying, time elapse between catching and marketing and water used during marketing and selling. Therefore, the control of possible contamination of dried prawn may lead to the consumption of a better quality animal protein source to the people. Accordingly, heated samples were better than that of the non-heated samples. Dried prawn with radiation could extend shelf-life of the prawn and quality of the irradiated dried prawn was better than that of non-irradiated samples. Among the three types of treatments i.e., temperature, radiation and combination treatments (temperature and radiation), combination treatments are more effective to improve the nutritional quality and it also lower the risk of food borne illness caused by microorganisms.

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