



**Research Article**

**MICROBIOLOGICAL QUALITY IMPROVEMENT OF DRIED FISH BY GAMMA  
IRRADIATION AND ASSESSMENT OF FOOD VALUE UPON IRRADIATION WITH RESPECT  
TO BIOCHEMICAL ASPECT**

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**Abstract:** A study was conducted to determine the effects of radiation on biochemical (moisture, protein, fat, ash, carbohydrate, calcium and phosphorus) and microbiological qualities (TVBC, TCC and TFC) of three different dried fishes (Chepa, Loitta and Chingri) collected from retail markets. Among the three non-irradiated dried fishes, the highest moisture content ( $40.85 \pm 0.42$ ) was found in Chepa compare to Loitta ( $19.48 \pm 0.40$ ) and Chingri ( $17.09 \pm 0.77$ ). Highest rate of total protein content was found in non-irradiated Chingri ( $60.39 \pm 0.75$ ) and Loitta ( $55.85 \pm 0.40$ ) compare to Chepa ( $32.67 \pm 0.06$ ). Results revealed that radiation (3 and 5 kGy) has no significant effect on biochemical composition of three dried fishes. Results of microbiology showed that total viable bacterial count (TVBC) was estimated in non-irradiated Chepa ( $5.58 \pm 0.14$  log cfu/g), Loitta ( $3.72 \pm 0.09$  log cfu/g) and Chingri ( $5.34 \pm 0.15$  log cfu/g) respectively which was significantly reduced by 1 to 3 logs at 3 kGy and 3 to 5 logs at 5 kGy gamma radiation. Maximum Total coliform count (TCC) was found in non-irradiated Chepa ( $5.28 \pm 0.02$  log cfu/g) compare to Loitta ( $3.23 \pm 0.09$ ) and Chingri ( $4.56 \pm 0.06$ ). Total fungal count (TFC) was recorded as  $3.60 \pm 0.09$  and  $3.78 \pm 0.05$  log cfu/g in non-irradiated Chepa and Chingri respectively but there was no TFC in non-irradiated Loitta. At 3 and 5 kGy, no TCC and TFC were found in all dried fishes

**Keywords:** Dry fishes, gamma radiation, microbiological and biochemical quality

**INTRODUCTION**

In Bangladesh dried fish is very demanding food stuff due to its high protein content and nutritional value. Every year Bangladesh earns foreign currency by exporting dried fishes. During the year 2008-09, Bangladesh exported 309.35 metric tons dried fish and fishery products and earned 119.9 million taka<sup>1</sup>. The quality of sun dried fishes is adversely affected by microorganisms<sup>2</sup>. Fish usually takes 5 to 7 days to dry during which it gets heavily contaminated<sup>3</sup>. Quality of fish and fishery product is strictly maintained and monitored for export to foreign countries but unfortunately, such quality control procedure is not maintained for local market. So it is very important to assess microbiological and biochemical quality of dried fishes in retail trade for guarding health and hygiene of local consumer. When considering biochemical studies, proximate composition of three commercially available marine dry fishes namely Bombay duck, Sin croaker and Ribbonfish performed a study on the traditional drying activities of commercially important marine fishes of Bangladesh and made a survey on the source of raw materials, handling, transportation, processing and marketing aspects of fish using questionnaires<sup>4,5</sup>. Major factors for poor quality of fishes in retail market are unhygienic handling and storage, lack of proper drying, physical damage, contamination with dirt and microorganism<sup>6</sup>. It was studied the load of pathogenic bacteria in different dried fishes collected from retail market and khamar of different areas of Chittagong and

Mymensingh districts of Bangladesh<sup>7</sup>. Seasonal comparative study of organoleptic, microbiological and biochemical qualities of four dried fishes was studied<sup>8</sup>.

There are some fishermen who sell low quality dry fishes in market beside the street. This low quality dried fish consumed mainly by local poor people and they have long been consumed as a traditional food. Occasionally these types of fish contain 12.50 to 40.93% moisture and are easily spoiled by the propagation of microorganisms and fly maggots. When dried fishes are spoiled by microorganisms, they become covered with molds and red spots accompanied with an unpleasant odor.

As Bangladesh produces huge amount of dried fishes and the demand of mass consumer for low cost dietary protein is fulfilled by different kinds of dried fishes, it is necessary to assure the hygienic quality of these food products with respect to both microbiological and biochemical aspects for safe consumption. With this point of view, this experiment was conducted to find out an appropriate technique for the improvement of microbiological quality of dried fishes applying gamma radiation.

**MATERIALS AND METHODS**

**Sample collection and packaging**

Three types of the dried fish Chepa (*Puntius chola*), Loitta (*Harpodon nehereus*) and Chingri (*Leander styliferus*) were purchased from the local fish market and transported

to the Food Technology Laboratory, Institute of Food and Radiation Biology, Atomic Energy Research Establishment, Savar, Dhaka.

### Irradiation

Dry fishes were selected for uniformity without any defects or mechanical injury. For each treatment 50g of sample was placed into low-density polyethylene pouches (150 $\mu$  gauge) and sealed tightly. The sealed polythene bags were labeled by indicating the name of the product and were irradiated with two selected doses of gamma radiation which were 3 and 5kGy. Irradiation was applied to the samples with a 50kCi Co<sup>60</sup> gamma source (dose rate 6.4kGy/hr) located at Institute of Food and Radiation Biology, Atomic Energy Research Establishment, Savar, Dhaka. Both treated and untreated samples were stored at room temperature (28-33°C) for biochemical and microbiological analysis.

### Biochemical analysis

The moisture content of dried fishes was determined by drying at an oven at 105°C for 5-6 hrs according to the standard method of AOAC<sup>9</sup>. Nitrogen was determined by the Micro-Kjeldahl Method according to the Ma and Zuazaga<sup>10</sup>. Total protein was calculated using the factor 6.25 X N. Calcium and Phosphorus were determined according to the method of Ranganna<sup>11</sup>. Ash was determined by drying the sample in a Muffle Furnace at 600°C for 3-5 hrs according to the method of Carpenter<sup>12</sup>. Crude fat was determined gravimetrically by AOAC<sup>13</sup>. Total carbohydrate content of foods has, for many years, been calculated by difference, rather than analyzed directly. Under this approach, the other constituents in the food (protein, fat, water, ash) are determined individually, summed and subtracted from the total weight of the food. This is referred to as total carbohydrate by difference and is calculated by the following formula:

$$100 - (\text{Weight in grams} [\text{protein} + \text{fat} + \text{water} + \text{ash}] \text{ in } 100 \text{ g of food})$$

### Determination of microbial counts

For determination of microbiological status ten grams of fish samples were aseptically homogenized for 2minutes

with a stomacher lab blender in a sterile stomacher bag containing 90ml of sterile 0.1% peptone water. Appropriately diluted fish samples were then used for microbial analysis. Total viable bacterial count (TVBC) was determined by the standard spread plate method according to the APHA using spread plate technique. Nutrient agar (pH 7.0–7.4) was used to determine TVBC as well as for isolation purposes. Plates were incubated at 37°C for 24 h and the count was expressed as colony-forming unit per gram (cfu/g). Total coliform count and total fungal count were obtained in the same way using McConkey agar and Potato Dextrose Agar medium respectively<sup>14,15,16</sup>. McConkey agar and Potato Dextrose agar were incubated at 37°C for 24 to 48 hours and 28°C for 5 days respectively.

### Statistical analysis

Statistical procedures were performed using SPSS for Microsoft version 18.0 software package (SPSS Chicago, IL) with five percent level of significance.

## RESULT AND DISCUSSION

### Effect of gamma radiation on biochemical composition of dried fishes

The biochemical compositions of Chepa (*Puntius stigma*) were presented in Fig. 1. Each value is the mean  $\pm$  standard deviation of triplicate determination. The result showed that the moisture, protein, lipid, ash, carbohydrate, calcium and phosphorus of control Chepa dry fish were 40.93  $\pm$  0.60, 32.61  $\pm$  0.40, 12.28  $\pm$  0.45, 10.81  $\pm$  0.74, 3.37  $\pm$  0.56, 0.396  $\pm$  0.46, 0.743  $\pm$  0.92 g/100g respectively. 40.39  $\pm$  0.80 % moisture, 32.73  $\pm$  0.72 % protein, 11.90  $\pm$  0.84 % lipid, 10.93  $\pm$  0.15 % ash, 4.05  $\pm$  0.64 % carbohydrate, 0.368  $\pm$  0.32 % calcium and 0.726  $\pm$  0.28 % phosphorus were found in cheap irradiated with 3 kGy. In irradiated (5 kGy) Chepa 41.22  $\pm$  0.85 % moisture, 32.68  $\pm$  0.84 % protein, 11.78  $\pm$  0.85 % lipid, 10.90  $\pm$  0.39 % ash, 3.42  $\pm$  0.96 % carbohydrate, 0.384  $\pm$  0.25 % calcium and 0.728  $\pm$  0.64 % phosphorus were found. Biochemical analysis showed that there was no significant different between control and irradiated samples.

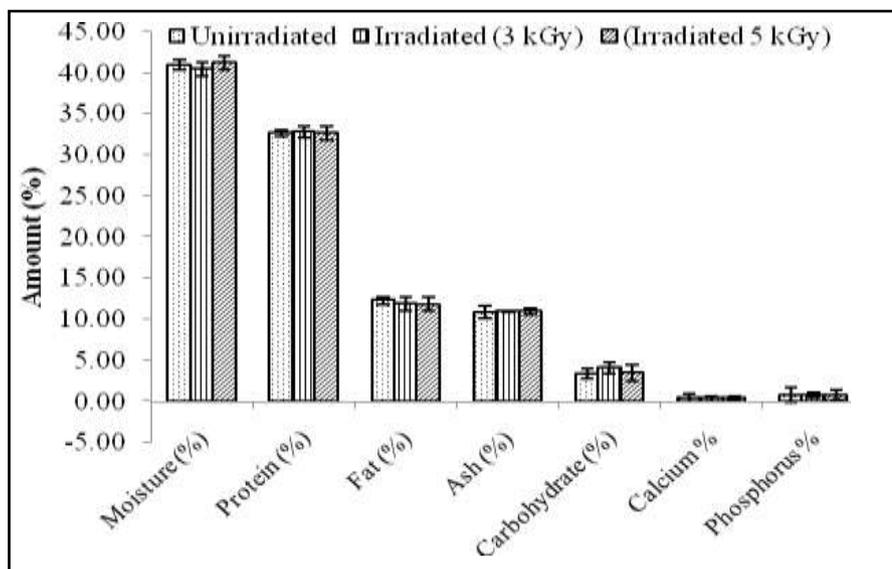


Fig.1: Biochemical composition of non-irradiated and irradiated (3 and 5kGy) of Chepa dry fish

Biochemical compositions of both un-irradiated and irradiated Loitta were presented in Fig.2.  $19.31 \pm 0.48$  % moisture,  $56.22 \pm 0.45$ % protein,  $11.10 \pm 0.10$ % lipid,  $10.08 \pm 0.20$ % ash,  $3.29 \pm 0.84$  carbohydrate,  $0.165 \pm 0.12$ %

calcium and  $0.360 \pm 0.24$  phosphorus were found in control Loitta. Irradiation (3 and 5 kGy) has no significant effect on biochemical compositions of dried Loitta.

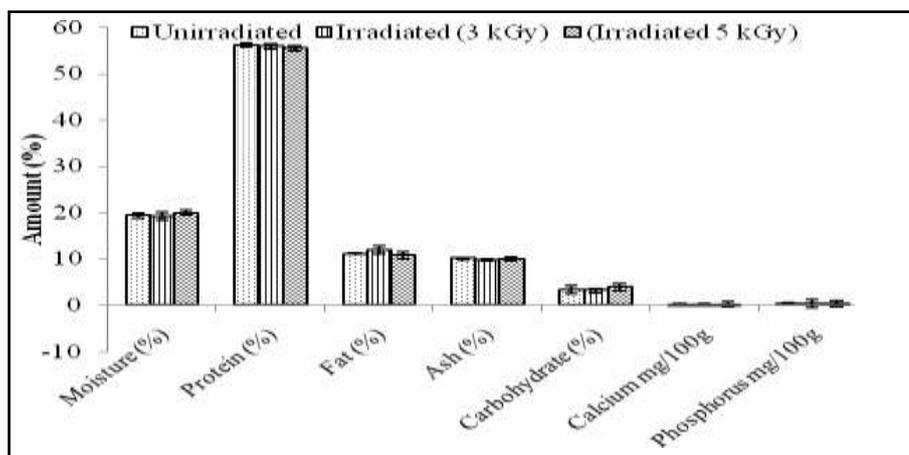


Fig.2: Biochemical composition of non-irradiated and irradiated (3 and 5kGy) of Loitta dry fish

Fig. 3 Showed that the moisture, protein, lipid, ash, carbohydrate, calcium and phosphorus of non-irradiated dried Chingri (*Leander styliferus*) were  $17.09 \pm 0.77$ %,  $60.00 \pm 0.78$  % ,  $8.89 \pm 0.51$  % ,  $10.22 \pm 0.25$  % ,  $3.80 \pm 0.92$ % ,

$0.410 \pm 0.22$  % and  $0.792 \pm 0.02$  % respectively. No significant effect was found on biochemical composition analysis in dried Chingri when treated with 3 and 5 kGy gamma radiation.

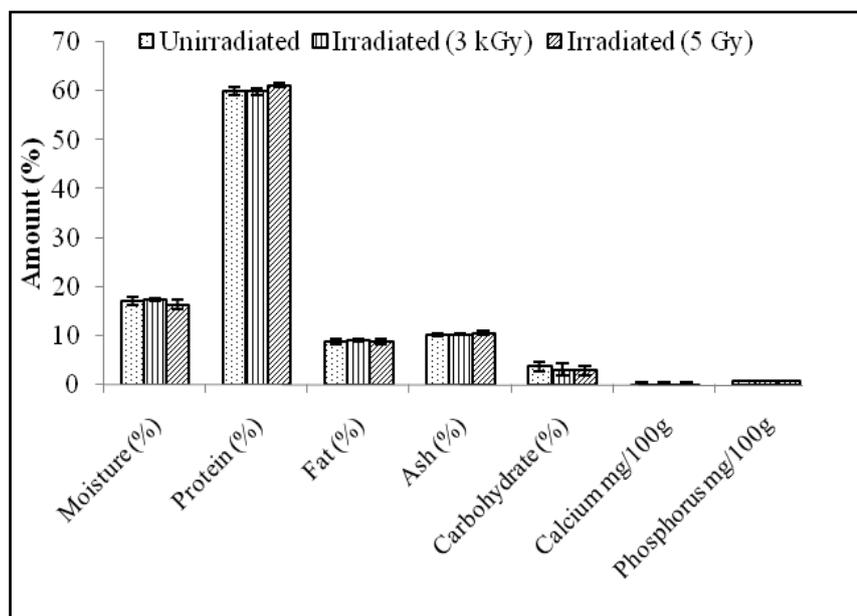


Fig. 3: Biochemical composition of non-irradiated and irradiated (3 and 5kGy) of Chingri dry fish.

Among the three dried fishes highest moisture content was found in dried Chepa. It was found that moisture content varied from 39.62 to 46.89% in Chepa obtained from producer, wholesaler and retailer and highest moisture content was found in the product obtained from retailer compare to the product from producer<sup>17</sup>. Highest moisture content is probably due to continuous adding of moisture in the products from the environment in the marketing chain. There is also a possibility to absorb moisture during storage

since in most of the places Chepa stored in bamboo made basket or clay vat under open air<sup>18</sup>.

Current results revealed that Chingri and Loitta showed highest value of protein content compare to Chepa. It was also found the lowest value of protein content in retailer dried Chepa compare to Chepa from producer and wholesaler<sup>17</sup>.

**Microbiological load in non-irradiated and irradiated dried fish**

The average TVBC of the non-irradiated three types of dried fish (Chepa, Loitta and Chingri) were  $5.58 \pm 0.14$ ,  $3.72 \pm 0.09$  and  $5.34 \pm 0.15$  log cfu/g respectively (Table-1).

These associated microorganisms indicated the presence of indigenous microorganisms and further contamination during harvesting, processing and selling of these dried fishes. Previous reports presented the abundance of total aerobic bacteria at 3- 4 logs at different dried fishes<sup>19,20</sup>.

**Table-1: Microbiological load in non-irradiated and irradiated dried fishes**

Name of the fishes	Treatments	Microbial parameter (Mean±SD)		
		TVBC	TCC	TFC
<b>Chepa</b>	Un-irradiated	$5.58 \pm 0.14$	$5.28 \pm 0.02$	$3.60 \pm 0.09$
	Irradiated (3 kGy)	$2.02 \pm 0.05$	Nil	Nil
	Irradiated (5 kGy)	Nil	Nil	Nil
<b>Loitta</b>	Un-irradiated	$3.72 \pm 0.09$	$3.23 \pm 0.09$	Nil
	Irradiated (3 kGy)	Nil	Nil	Nil
	Irradiated (5 kGy)	Nil	Nil	Nil
<b>Chingri</b>	Un-irradiated	$5.34 \pm 0.15$	$4.56 \pm 0.06$	$3.78 \pm 0.05$
	Irradiated (3 kGy)	$2.84 \pm 0.12$	Nil	Nil
	Irradiated (5 kGy)	$2.78 \pm 0.01$	Nil	Nil

SD= Standard deviation, Values are mean of 3 replicates.

The average TCC of the untreated fish samples were  $5.28 \pm 0.02$ ,  $3.23 \pm 0.09$  and  $4.56 \pm 0.06$  log cfu/ g respectively (Table-1). Coliforms might be present due to the inadequate hygienic measures, inappropriate handling, improper storage and all unhygienic conditions of the shops from where dried fishes were collected<sup>21</sup>. Two log cfu/ g of coliforms was reported previously<sup>22</sup>. Some food borne illness, in dry fishes are mainly due to chemical agent histamine, also known as histamine poisoning. Coliforms are responsible for the production of histamine in dried fishes<sup>23,24</sup>.

TFC of the untreated dry fish samples were  $3.60 \pm 0.09$ , Nil and  $3.78 \pm 0.05$  log cfu/ g respectively (Table-1). Previous report showed the presence of fungi in dried fish at 2 log cfu/g concentration<sup>25</sup>. One of the fungi, *Aspergillus flavus* is responsible for the production of aflatoxin which causes food borne intoxication and might lead to serious health hazards. *Aspergillus* is the main genus which is commonly involved in the production of mycotoxin<sup>26</sup>.

The observed results indicated that dry fishes are prone to contamination by pathogenic microorganisms. As fishes to be dried are usually handled with bared hands and drying process is carried out in the open atmosphere, this unhygienic practice might cause the contamination of dried fishes posing a risk of food borne diseases. Ordinary processing cannot eliminate the risk of avoiding microbial contamination. The level of microorganisms associated with these foods can be decreased by irradiation that depends upon the absorbed dose of radiation<sup>22</sup>.

To reduce/ eliminate microorganisms dried fish samples viz. Chepa, Loitta and Chingri were irradiated at different doses of gamma radiation. In case of TVBC, the counts were reduced by 1.0 to 3.0 logs and 3.0 to 5.0 logs at 3.0 and 5.0 kGy, respectively. Reduction of TVBC at similar rate at the response to irradiation was also reported by<sup>25</sup>. At 3.0 kGy irradiation dose TVBC of three different dried fishes (Chapa, Loitta and Chingri) were  $2.02 \pm 0.05$ , Nil,  $2.84 \pm 0.12$  log cfu/g respectively. At 5.0 kGy, TVBC of these

three samples were Nil, Nil,  $2.78 \pm 0.01$ , log cfu/g, respectively. TFC and TCC were below detection limit after irradiation at 3.0 and 5.0 kGy in all the three samples (Table-1).

**CONCLUSION**

The results suggest that gamma irradiation can be effectively applied for ensuring the hygienic quality of dried fishes. The availability of dried fishes with satisfactory hygienic quality is highly desirable and to achieve this quality of these products, a hygienic process adhering good manufacturing practice (GMP) is suggested to be maintained in all of the steps of the process including harvesting, washing, drying and packaging.

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