### IMPACT OF GAMMA RADIATION ON FERMENTED PRAWN (Metapenaeus cojuntus)

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### Abstract

This research focuses on the food preservation by irradiating of fermented prawn. The prawns were collected from Thanlyin market and fermented as home-made. This sample was irradiated with 3 kGy dose of Co-60 gamma source for 2:22:18 and stored in a dry place under room temperature. Nonirradiated fermented prawn sample was used for comparative study. Induced radioactivity of irradiated fermented prawn was monitored by using NaI (Tl) Scintillation Gamma Counter. Some nutritional values of nonirradiated and  $\gamma$ -irradiated samples such as water (65.34 % and 65.09 %), ash (0.08 % and 0.11 %), protein (16.00 % and 17.30 %) and fat (0.43 % and 0.52 %) were determined by analytical methods. There were no significant changes in nutritional values of  $\gamma$ -irradiated sample compared with nonirradiated sample. The shelf life of non-irradiated and  $\gamma$ -irradiated fermented prawns were studied by using microscope for seeking spoilage and pathogenic bacteria at room temperature. Shelf life of y-irradiated sample was found to be prolonged nearly two times than that of nonirradiated sample. From the monitoring of induced activity, there were no considerable activities above background. Thus radiation can be used for the shelf life extension of fermented prawn and is not harmful to health. From the observation of gamma radiation effect on microorganism and morphological tests were used to evaluate the unknown bacteria in yirradiated sample. From these investigations, the populations of Cocci and Bacilli bacteria groups in  $\gamma$ -irradiated sample were significantly decreased than that of non-irradiated sample.

Keywords: fermented prawn, irradiation, gamma radiation, shelf life extension, microorganism

### Introduction

Fermented prawn (vestals), formerly also spelled Pazun-chin, is a generic term from pungent pastes made of either shrimp or prawn in Myanmar cuisine. Pazun-chin is usually made by fermenting shrimp or prawn that is salted with rice. Pazun-chin is a main ingredient of lower Myanmar cuisine from maritime coastal province in the west and the south. It is not a main

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ingredient in traditional upper Myanmar cuisines. In the Ayeyarwaddy and Bago Regions, the majority of Pazun-chin is produced from fresh water shrimp or prawn. Pazun-chin from Bago is very well known than other regions. In this research work, home-made fermented prawn samples were prepared in such a way to have pure and reliable samples. In international literature, it was found that food irradiation can reduce risk of food-borne disease caused by microorganisms. Therefore, to have knowledge about the food preservation by concerning with microorganisms, investigation had been carried out in this work.

Food irradiation is the treatment of food by gamma rays from a cobalt-60 or caesium-137 source or electrons or X-rays from a machine source. It is analogous to other types of food processing treatments such as heat pasteurization, canning, freezing or dehydration. Irradiated foods are safer to eat and more resistant to spoilage. Irradiation destroys insects, mould, fungi and pathogens that cause food-borne illness or foods to spoil. Irradiation temporarily ionizes (electrically charging) atoms or molecules by knocking electrons out of their moulds orbits. It is stressed that the foods do not become radioactive. Irradiation extends shelf life of food in two ways. First, it can reduce spoilage bacteria and moulds than they can be grown even under refrigeration. The second way, irradiation extends shelf life by slowing the ripening process of fruits and vegetable (Charlotte, 1995).

Gamma rays with energies of 1.17 and 1.33 MeV are emitted by the cobalt-60 or energy of 0.66 MeV is emitted by caesium-137. The Co-60 is a radioactive metal that decays with a half-life of around 5.3 years. Although Cs-137 has a longer half-life of around 30.1 years, few commercial facilities use Cs-137 as a gamma ray source. Cs-137 emits gamma rays that are approximately half of the energy of those emitted by Co-60. Gamma radiation has higher penetration than electron beams. Therefore, it is suitable for treating large bulk packages of food. Gamma facilities are the majority of food irradiation worldwide. The gamma radiation can not be switched off and when not being used to treat food, must be stored in a water pool to absorb the radiation energy and product workers from exposure if they must enter the irradiation room (Mostafavi *et al.*, 2012)

Gamma irradiation is considered to be an alternative method of food preservation, in order to prevent food spoilage, insect infestation and to reduce microbial load, extending the shelf life of foods, while maintaining their safety and avoiding nutrients loss. As a result, the application of the radiation technique may be recommended to enhance food quality safely (Taipina *et al.*, 2013). Irradiation can be applied to liquid, solid and semi-solid foods. Gamma rays penetrate food causing DNA fragment-atom in any microbes and insects that are present. This kills the organisms or eliminates their reproductive ability. The process enhances the microbial safety and ability of the treated product. However according to the literature survey. International Agencies including IAEA, FAO and WHO stated that irradiation of any food commodity up to a dose of 10 kGy exhibits on health risk (WHO, 1981).

The effect of radiation on microorganisms in irradiated fermented prawn were also studied in this work. The dominant putrefactive bacteria, coliforms and *Staphylococcus* in fermented prawn were suggested to be decreased by the gamma radiation of 3 kGy. The shelf life of irradiated fermented prawn was prolonged nearly 2 times compared with non-irradiated fermented prawn and could be stored for 9 days at room temperature. Irradiation of fermented prawn at 3 kGy reduced the aforementioned dominant microorganism (Parachasithhisak *et al.*, 1996).

### **Materials and Methods**

#### **Preparation of Fermented Prawn**

In this work, the prawn was collected from Thanlyin Market and fermented as home-made. Prawns (400 g) were weighed, rinsed and cut of head. Then prawn body was washed with water to clear oil.100 g of cooked rice and 25 g of salt were mixed well in the separated container. After mixing these two potions thoroughly the mixture was packed with prawns in glass bottle stand in inverted position to drain liquid. The mixture became fermented prawn within 2 or 3 days (Figure 1 (a) and (b)). The prawn samples were immediately transported to the Department of Atomic Energy, Ministry of Science and Technology for radiation. The period between the starting fermentation and irradiation was approximately 12 h. Then the samples were

treated with 3 kGy doses of gamma radiation from Co-60 source (Co-60 gamma Chamber 5000) which was dose of 1.269 kGy/h (Figure 2).



Figure 1: Photographs of package of fermented prawn

### Monitoring the Induced Radioactivity of Irradiated Fermented Prawn

For study on safely consumption, the induced activity of irradiated sample was monitored by NaI (Tl) Scintillation Detector (Ludlum Model 730, 732 & 733 PC-Based Gamma Ray Spectroscopy System) at Nuclear Laboratory, Department of Chemistry, University of Yangon.



Figure 2: Photograph of Co-60 gamma chamber 5000

## Study on Shelf Life of Non-irradiated and γ-Irradiated Fermented Prawn at Room Temperature by using microscope

Firstly, the prawn samples were collected from Thanlyin Market and fermented as home-made. And then, package of fermented prawn was irradiated with 3 kGy dose of Co-60 gamma source. Non-irradiated fermented prawn was used comparative study. Non-irradiated and  $\gamma$ -irradiated samples

were kept under same condition at room temperature. The shelf life of each sample was studied daily by seeking spoilage and pathogenic bacteria by using microscope at the Fermentation Department, Pharmaceutical Research Development. The microorganisms from non-irradiated and  $\gamma$ -irradiated fermented prawn samples were extracted and cultivated on nutrient agar medium, at the Fermentation Department, Pharmaceutical Research Development as following.

A 200 mL conical flask was added by 99 mL of distilled water, four test tubes were also added by 9 mL each of distilled water, plugged them with cotton wools and label them as  $10^{-2}$ ,  $10^{-3}$ ,  $10^{-4}$ ,  $10^{-5}$  and  $10^{-6}$ , respectively. Then all of those were autoclaved at  $120^{\circ}$ C for 20 minutes.

Each 1 g of irradiated and non-irradiated fermented prawn sample was dissolved in 1 mL of sterile distilled water and added the mixture into autoclaved, cooled  $10^{-2}$  labeled conical flask. The flask was shaken gently for a few seconds and 1 mL of the mixture was transferred into  $10^{-3}$  autoclaved, cooled test tube. A 1 mL of mixture was transferred from  $10^{-3}$  to  $10^{-4}$  test tube and so on (Figure 3).





(a) (b) **Figure 3:** (a) Subculture of bacteria from non-irradiated and (b)  $\gamma$ -irradiated fermented prawn in test tube agar slant medium

After then, 1 mL of the prepared serial dilution was transferred to each of five nutrient agar plates and labeled them. The inoculum was spread evenly over the entire surface of the nutrient agar plate until the medium no longer appeared moist. The spreader reap the flaming and spreading for each of the remaining four plates. Then all the plates were incubated at 28°C for 24 h.

## Determination of some Nutritional Values of Non-irradiated and $\gamma$ -Irradiated Fermented Prawn

Some Nutritional Values of Non-irradiated and  $\gamma$ -Irradiated Fermented Prawn were determined by AOAC (2000) methods.

### **Results and Discussion**

#### Study on Safety Consumption of γ-irradiated Fermented Prawn

The induced activities of  $\gamma$ -irradiated sample with (3 kGy) dose was monitored by NaI (Tl) Scintillation Detector (Ludlum Model 73450, 732 & 733 PC-Based Gamma Ray Spectroscopy System) at Nuclear Chemistry Laboratory, University of Yangon. This monitoring indicated that there was no distinctive activity above the background (Table 1). According to the literature, as food is passed through the irradiation field, gamma energy passes through a window. This energy destroys most of bacteria that can cause disease, yet allow food to retain its high quality. Since the energy involved in irradiation is not strong enough to change the atoms of the food, the food cannot radioactive (Charlotte, 1995). It was found that this result was in agreement with the literature. Therefore,  $\gamma$ -irradiated fermented prawn can be handled, stored and consumed safely in the same way as non-irradiated fermented prawn.

No.	Sample	Induced activity relative to background
		(± %) (cp 300s)
1	BG 3.0	3.45

Tabl	e 1:N	Ionitoring	of Inc	luced A	Activity	γin γ-	Irradiated	Fermented	Prawn
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 $\pm$  : due to fluctuation

Note : no distinct activity above background

BG 3.0 = Gamma dose of 3.0 kGy of fermented prawn

### Effect of $\gamma$ -Irradiated on Shelf Life of Fermented Prawn at Room Temperature by Microscope

The shelf life of non-irradiated and  $\gamma$ -irradiated fermented prawn samples were assessed by microscope for seeking fermentation changes such as colour and spoilage at room temperature under same condition. According to the literature, food irradiation provides safety and extension the shelf life of fisheries products because of its high effectiveness in inactivating pathogenic and spoilage microorganism without deteriorating product quality. From the results reported in Table 2, it was found that the non-irradiated fermented prawn spoiled within 5 days and the  $\gamma$ -irradiated fermented prawn spoiled within 9 days. The shelf lives of  $\gamma$ -irradiated sample was found to be prolonged nearly two times than that of the non-irradiated sample at room temperature under same condition.

No. Sample		Shelf life at Room Temperature(days	
1	Non-irradiated	5	
2	γ-irradiated	9	

### Table 2: Determination of Shelf Life of Non-irradiated and γ-Irradiated Fermented Prawn at Room Temperature by Microscope

#### Study on Radiation Effect on Microorganism in Fermented Prawn

For the observation of effect on microorganism in samples, the bacteria from non-irradiated and  $\gamma$ -irradiated samples were extracted by using nutrient agar medium as in Figure 4 (a) and (b). From these results, it was found that non-irradiated sample occurred 2.21 x 10<sup>5</sup> CFU/mL of bacteria population and  $\gamma$ -irradiated sample occurred 4.1 x 10<sup>3</sup> CFU/mL of bacteria population after 3 days cultivated on nutrient agar medium. Thus, the population of bacteria from  $\gamma$ -irradiated sample. Among them, the eight groups bacteria that were found to be distinguished were selected for further tests. Therefore, 3 k Gy dosage of  $\gamma$ -irradiation were found to be sufficient to reduce the bacteria population of fermented prawn. Thus, the food preservation concerning with irradiation technique was found to be efficient to extend shelf life and reduce spoilage bacteria.





**Figure 4:** Photo of cultivated bacteria in non-irradiated and  $\gamma$ -irradiated fermented prawn on nutrient agar medium

# Bacteriological differentiation of non-irradiated and $\gamma$ -irradiated fermented prawn

The cultivated bacteria from non-irradiated and  $\gamma$ -irradiated samples were investigated by using optical microscope. From this microscopic examination, bacilli and cocci groups of bacteria were observed in samples and may cause spoilage of food (Figures 5 and 6).





**Figure 5:** Photo of cocci group bacteria in (a) non-irradiated and (b)  $\gamma$ - irradiated fermented prawn





Figure 6: Photo of bacilli group bacteria in (a) non-irradiated and (b)  $\gamma$ irradiated fermented prawn

### Morphology identification of bacteria by gram staining technique

The cultivated bacteria were morphologically identified by gram staining technique. According to gram staining technique, gram-positive and gram-negative cocci group bacteria and bacilli group bacteria were present in non-irradiated and  $\gamma$ -irradiated samples (Figures 7 and 8).



(b) γ-irradiated

Figure 7: Morphology of gram (+) and (-) cocci group bacteria by gram staining technique in (a) non-irradiated and (b) y-irradiated fermented prawn sample





(a) non-irradiated



(b)  $\gamma$ -irradiated

Figure 8: Morphology of gram (+) and (-) bacillii group bacteria by gram staining technique in (a) non-irradiated and (b)  $\gamma$ -irradiated fermented prawn sample

## Study on some Nutritional Values of Non-Irradiated and $\gamma\text{-Irradiated}$ Fermented Prawn

Some Nutritional values such as water contents, ash contents, fat contents and protein contents were quantitatively determined according to standard methods. The results were indicated by Table 3 and Figure 9.

No.	Nutritional parameters	Fermented Prawn			
	(%)	Non-irradiated	γ-Irradiated		
1	Water	65.34	65.09		
2	Ash	0.08	0.11		
3	Protein	16.00	17.30		
4	Fat	0.43	0.52		

Table 3: Some	Nutritional	Values	of	Non-irradiated	and	γ-Irradiated
Ferme	ented Prawn					



**Figure 9:** A bar graph of some nutritional values of non-irradiated and  $\gamma$ -irradiated fermented prawn

### Conclusion

In this work, effects of 3 kGy dose of gamma irradiation on shelf life, quality and microorganism of fermented prawn were studied. This work has found out that ;

(1) There were no distinct induced activity above background in  $\gamma$ -irradiated sample.

- (2) The shelf life of non-irradiation and  $\gamma$ -irradiated fermented prawn were 5 and 9 days, respectively.
- (3) Some nutritional values of non-irradiated and  $\gamma$ -irradiated fermented prawns with 3 kGy dose were found to be water contents (65.34 %, 65.09 %), ash contents (0.08 %, 0.11 %), protein contents of (16.00 %, 17.30 %) and fat content of (0.043 %, 0.52 %), on a dry weight basis respectively.

From overall results, 3 kGy dose of gamma irradiation do not make food radioactive. 3 kGy dose are effective in extending the storage life and do not badly change on micronutrients such as proteins and fats. The shelf life of  $\gamma$ -irradiated sample extended nearly two times than that of non-irradiated sample. 3 kGy dose of gamma irradiation reduces disease-causing bacteria (cocci and bacilli groups) in fermented prawn. Therefore food irradiation with gamma rays is a promising new food preserving technology to distribute as fresh product without harmful to eat. However, care should be taken to avoid prolong storage causing spoliation and damaging to fermented prawn.

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