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EFFECT OF GAMMA IRRADIATION ON THE QUALITY OF PROCESSED ANCHOVIES (*Engraulis encrasicolus*)

BY

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A THESIS SUBMITTED TO THE SCHOOL OF RESEARCH AND GRADUATE STUDIES, KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY, KUMASI, IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF PHILOSOPHY IN POSTHARVEST TECHNOLOGY

SEPTEMBER, 2015

DECLARATION

I hereby declare that this submission is my own work towards the award of Master of Philosophy in Postharvest Technology and that, to the best of my knowledge, it contains neither material previously published by another person nor material which has been accepted for the award of any other degree of the University, except the references to other people's work, which have been duly acknowledged.

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DEDICATION

This work is dedicated to my dearest mother Madam Agartha Boadi for her unique care and support.

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ABSTRACT

The effect of irradiation on nutritional composition, microbial load and shelf-life of anchovies (*Engraulis encrasicolus*) was assessed in this study. Irradiation doses used for the study were 2.5, 5.0, 7.5 and 10 kGy. The assessment was carried out at 3 weeks intervals for the period of 9 weeks. Samples were analysed for microbial load (total viable count, total coliform count and Staphylococcus aureus) and nutritional composition (total ash content and free fatty acids). Smoked anchovies were contaminated with TVC of 6.175 CFU/g when compared to smoked samples (6.042 CFU/g) from Keta at week 0. The sun-dried samples obtained from Chorkor at week 0, had significantly higher ($p<0.05$) TVC of 5.633 CFU/g when compared to sun-dried samples (4.490 CFU/g) from Keta. It was established on the 9th week that, there was a general decrease in microbes implying that the application of irradiation had reduced the level of TVC, TCC and SA in the anchovy samples. Samples from Chorkor were also found out to be more contaminated than those Keta and this may be as a result of the environmental condition in Chorkor. It was also established that irradiation dose as low as 2.5kGy could decontaminate processed anchovies. As regards, nutritional analysis, sun-dried samples obtained from processors (21.47%) had high amount of ash content when compared to marketers (20.80%) on the 3rd week. Smoked samples obtained from marketers (17.99%) at week 3 were not significantly different ($p>0.05$) in ash content when compared to processors (18.44%). The free fatty acids (oleic acid) levels in both smoked and sun-dried samples decreased as the storage period increased. Application of irradiation had no influence on the ash content in samples

but the free fatty acid levels reduced as the irradiation dose increased. In conclusion, irradiation to a dose of 2.5kGy is sufficient to decontaminate anchovies without significantly affecting their nutritional composition.

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LIST OF ABBREVIATIONS

FAO……Food and Agriculture Organization

PAHs……..Polycyclic aromatic hydrocarbons

GDP……Gross Domestic Product

PUFA…… ……Polyunsaturated fatty acids

IAEA…….....International Atomic Energy Agency

ECB……...……..….Ethanol chlorobenzene

CFU/g……..…..….colony forming units per gram

GSA……...….Ghana Standards Authority

APHA…….……American Public Health Association

AOAC…….American Official Association of Chemist

AOCS……American Oil Chemists' Society.

TVC…….....Total Viable Count

TCC……Total Coliform Count

SA……..*Staphylococcus aureus*

FFA……..……Free Fatty Acids

BNARI……..Biotechnology and Nuclear Agriculture Research Institute

kGy…….…….kilo gray

kg……...……..kilogram

1.0 INTRODUCTION

In Ghana and Africa as a whole, the use of advanced food processing technologies can help resolve the issues of food insecurity. These technologies should be able to address the problems of food spoilage and food borne diseases which are prevalent in the world. Improving food security include the better use of fish produced that could help reduce postharvest losses and also increase the percentage of fish used directly for human consumption (FAO, 2010).

Annually, about 10 to 12 million tons postharvest losses caused by spoilage is incurred. It is also estimated that about 20 million tons of fish in a year become wasted at sea which could possibly lead to further losses. To also contribute greater to food security, the consumption of fish and its products by humans can help make relevant use of low-value resources, rather than reducing them to fishmeal (FAO, 2010).

Anchovies are small and silvery salt water forage fish. They belong to *Engraulidae* and Anchoa family having about 145 species in 17 groups found in the Mediterranean Sea, Atlantic, Pacific and Indian Oceans. They include species of *Engraulis ringens*, European anchovy (*Engraulis encrasicolus*), *Sardina pilchardus*, *Engraulis japonicus*, and *Stolephorus commersonii* which are distinct in the world's catch and these species are harvested for consumption. About 10.5 million tons of anchovies are produced worldwide annually (Encyclopedia, 2011).

Global consumption of fish and fish products have increased in recent years due to recognition of their nutritional value (Wang *et al.*, 2003).

Preservative and or storage methods may depend on the physical control of microbes in fish. This may include the use of heat from microwave or gamma irradiation, the control of microbial activity by chemical means and also by acidification.

Traditionally, harvested anchovies are sun-dried on the roadside or at the sea shore and sometimes on raised racks in artisanal fish processing. The use of the sun in drying fish gives very little regulation over the times at which fish is dried and it may also expose fish to birds or insect attack. This may bring about physical contamination or contamination by microbes. This type of technique is totally dependent upon weather conditions.

Smoking of harvested anchovies using traditional kiln may result in the release of toxic substances such as polycyclic aromatic hydrocarbons (PAHs) and phenols. The growing concerns on the smoke from wood and inability to control smoking temperature causes poor-quality smoked fish as well as significant postharvest losses (Essumang *et al.*, 2013).

The successful use of irradiation does not only ensure how safe food is, but it also helps to extend the storability of fresh fish and meats because of how effective it is in the deactivation of pathogens without affecting the quality of the product (Mahapatra *et al.*, 2005).

The main objective of this research was, therefore, to determine the effect of irradiation on microbial load and physico-chemical properties of anchovies.

Specific objectives were to:

i. determine the microbial load of unirradiated sun-dried and smoked anchovies;

ii. determine the nutritional quality of unirradiated sun-dried and smoked

anchovies; iii. ascertain the effect of gamma irradiation (2.5, 5.0, 7.5 and 10kGy) on the microbial load and nutritional composition of sun-dried and smoked anchovies; and iv. determine the effect of gamma irradiation (2.5, 5.0, 7.5 and 10 kGy) on the shelf life of sun-dried and smoked anchovies.

2.0 LITERATURE REVIEW

2.1 GLOBAL ECONOMIC IMPORTANCE OF FISH

Fish has an economic and considerable social importance. The Food and Agriculture Organization estimates that the significance of fish sold on the international market are stated to be USD 51 billion per annum (FAO, 2001). Employment of people through fishing and aquaculture was over 36 million whiles 200 million people may rely on revenue from fish (Garcia and Newton, 1997).

According to FAO, 2007, almost 60% of the world's protein supply comes from fish and about 30% annual protein from fish is derived from 60% countries in the developing world. This implies that any shortfall in the availability of fish will one way or the other affect animal protein intake by people in different countries. Along with this for fish preservation, sorting and transporting inefficient and insufficient infrastructure available causes wastages of thousands tons of even well accepted types of fishes. Protein is a high source of nutrient in fish and its products (FAO, 1973).

Many countries now have comprehensive system of inspecting and controlling at least some aspects of fish quality. For this purpose, it is important to assess the quality of processed fish by using hurdle technology. Hurdle technology involves the application of several processing or preservation methods in small amounts that individually are insufficient for preservation, but when combined with other processing methods, becomes sufficient to preserve food for reasonably long periods. In developing countries, application of intelligent hurdle technology has proven useful for novel foods. The hurdles are eliminated or rendered harmless in the final product (FAO, 2010).

Salting, smoking, cooling and freezing are widely used methods for the preservation of fish. Cooling could not prevent spoilage but shelf-life could be prolonged through the decrease of body temperature. Cooling and freezing can also decrease the growth or multiplication of microbes in fish. During the cold or freezer storage periods, biochemical changes occur in lipids and proteins as reported by Latip *et al.,* (2013).

In 2002, the total fish production in the world was estimated to be 133.0 million tons. Aquaculture contributed 41.9 million tons to this number. World capture fisheries production amounted to 93.2 million tons, representing a slight increase of 0.4% when compared to that of 2001 (Vannuccini, 2004). In Africa, about 5 percent of the population, (about 35 million people) depend solely or partly on the fisheries sector, which is mostly artisanal fisheries, for a living (FAO, 2001). Figures for world fisheries and aquaculture production and utilization are shown in Appendix B (i) as stated by Food and Agriculture Statistics (2005).

Whole fish, fish remains or other fish by-products such as heads, tails and other offal can be used to produce fishmeal and fish oil. Though many different types of fish are used for the production of fishmeal and fish-oil, oily fish such as small pelagic, in particular anchovies, are the main groups of species utilized. In recent times, anchovies catch have experienced a series of peaks and extreme depreciation as a direct consequence of the El Niño phenomenon (FAO, 2014).

There has been a balanced increase in fish production (capture and aquaculture) since 1950, but in 1998, a sharp decrease in production was recorded. In 2003, the total world fish production (both capture and aquaculture) was 132.5 million MT (weight of fish and

shellfish at capture or harvest - freshwater, brackish water and marine species of fish, mollusks, crustaceans and other aquatic organisms) and of this 104.2 million MT were available for human consumption of which 24.4 million MT were consumed in developed countries and 79.8 million MT in developing countries (FAO, 2005).

2.1.1 World Catch of Anchovies by Countries

In terms of anchovies producing countries, Peru is evidently the largest fishing nation of anchovies in the world. The anchovy stock in the waters of Peru (and Chile) is the world's largest fishery. In 2009, Peruvian fleets caught 5.9 million tons of anchovies which correspond to 57% of the total catch of anchovies in the world. Catching a volume of 5 to 6 million tons of anchovies meant some days 150,000 tons of fish were caught which put the biomass under pressure and led to risky working conditions.

The Peruvian government introduced an individual quota system, which was supported by most companies and workers of the sector in 2009. The fishing season has been extended to 190 days and the average catches per day decreased to 30 000 tons. This was a positive measure leading to smaller number of fishing boats at bays the same fishing time and safer conditions for crew members reflected in a fewer number of accidents.

Even though the economic value of the catch is moderate compared to that of fisheries in other countries, the impact of the anchovy fishery on the economy of Peru has been enormous. Nearly all the anchovies are converted to fishmeal which is marketed in developed countries. Although Peru is taking the great majority of anchovies in the Southeast Pacific, its southern neighbor Chile has developed fish meal industry based on

the anchovies. Chile catches around 1 million tons of anchovies, being the second largest anchovies fishing country in the world and responsible for 9% of the global anchovies catch. A little less than half of the world's total supply of fish meal comes from these two countries.

China is the third largest country for anchovies catch. China's Japanese anchovy fishery started in 1990s and had dramatically expanded to 1 million tons already in 1998. At present, the total fishing volume of Japanese anchovies in China is 800 000 tons, and China is responsible for 8% of the total volume of anchovies catch in the world and 66% of Japanese anchovies catch globally. The catch by Japan of Japanese anchovies has been relatively stable since 1950 amounting to 300-350,000 tons per year.

The fourth largest fishing nation for anchovies is Turkey which harvested around 522,000 tons of European anchovy in 2009. Turkish anchovy fishery in the Black Sea has a high proportion in the total fish harvest. Anchovy had the highest catch for marine fish with 251,675 tons in 2008; this comprises of 63.6% of catch sea fish in Turkey. For example, in 2008, the total export of anchovy (fresh and frozen) was almost 878 tons and the Turkish anchovy catch in the Georgian waters was estimated at 60,968 tons between 2003 and 2009.

Refrigerated, canned, brined and fresh anchovy caught in the Black Sea was also exported to many other countries, among which countries such as USA, Germany and Holland can be listed (Abdullah and Ayse, 2010). As a traditional product for many years, anchovies retain their popularity as the most common fish caught in Turkey. Its suitability for further processing (salting, marinating, canning and processing to fish oil etc.) has led to industrialization (Harun and Gokoglu, 2014). Despite its importance, the fishing industry

suffers from relatively high amount of post-harvest losses which are estimated at 35-40% of landed weight (FAO, 1981).

Food and Agriculture Organization in 1994 estimated that post-harvest losses remain about 25% of the total world catch annually. These losses have a profound adverse impact on some fishing communities whose status and income mostly depend on postharvest activities. Such losses also have an alarming impact on the socio-economic life of these fishing communities and affect the amount of animal protein available to large segment of the population. The current demand for fish food is estimated at a little over 1 million tons per annum as against a supply of 800,000 tons per annum (West, 1989).

Despite the subsistence of our nature of capture, for instance fisheries in Nigeria, as much as 50% of post-harvest losses are recorded (Bolorunduro, 1996). According to Ndok (1982), a lot of fish are imported into Nigeria to help reduce this shortfall so as to maintain the economic demand for fish and its products. Nigeria has therefore invested much in its fish production, processing and preservation in order to meet the domestic need of the country and on the international market (Jamin and Ayinla, 2003). The larger number of fish is sold fresh for local consumption in many countries.

Fish after harvest become unwholesome within 12 hours at tropical temperatures. Spoilage bacteria takes over the fish as soon as it dies, and therefore, it must be processed immediately to avoid the growth of these spoilage bacteria. Fish is highly susceptible to the growth of food poisoning bacteria such as *Listeria* because of its low acid accumulation. It is important to consume it immediately after harvest or subject it to some processing techniques. Some preservation methods can cause changes to the flavor and texture of the

fish which result in a range of different products. These include cooking, lowering the moisture content and lowering the pH (FAO, 2010).

2.2 FISH PRODUCTION IN GHANA

The fisheries sector contributes about 5% of agricultural GDP and 3% to overall GDP in Ghana (DOF, 2004). Furthermore, close to 10% of the Ghanaian population are dependent on the fisheries sector (FAO, 1998). Fish is consumed by the majority of people in Ghana from the rural poor to the urban rich. With a population of about 20 million people, the average per capita fish consumption is 27 kg per annum, which is higher than the world's average of 13 kg (Nti *et al.*, 2002).

Fishing is an extremely important economic activity in Ghana. It has been estimated that the fish resources in Ghana's water bodies support the livelihoods of a total of about 2 million people which includes fishermen, fish processors (including fish canneries and cold stores), traders and boat builders. These people, together with their dependents, account for about 10% of the Ghanaian population (Onumah *et al.*, 2010).

It is historically known that artisanal fishing in Africa is dominated by fishermen in canoes and boats who one way or the other have provided fish as a nutritious source of food of high quality protein that is often cheaper than meat. With the rising cost of meat and protein foods, consumers all over the world have become increasingly interested in the consumption of fish as a source of dietary protein (Nyarko *et al*., 2011).

In many countries of Africa, the average diet contains less protein. Fish is the cheapest source of animal protein (Jamin and Ayinla, 2003). With the increase in human population, for example in Nigeria, less fish will be available per caput annually (Eyo, 1993).

The small-scale fisheries in some developing countries like Ghana, are vital because they provide a nutritious food available to a larger number of people. These small-scale fisheries provided by canoe operators represents over 80% of fish catch for consumption in the country. (*mofa.gov.gh/site/?page_id=2862*).

It is therefore important to take into account the fish catch by these canoe operators right from handling, processing, packaging and marketing of the fish and its products as shown in Appendix B (ii).

2.4 NUTRITIVE VALUE OF FISH (ANCHOVIES)

With a higher biological value of 15-23%, nutritional value in fish is generally considered high in protein due to its cheap and high quality. Interest in fish consumption increased of late due to the high content of health significant omega-3 PUFAs, particularly eicosapentaenoic acid (20:5n-3, EPA) and docosahexaenoic acid (22:6n-3, DHA) (Elvevoll and James, 2000).

Besides playing important role in cardiovascular and inflammatory diseases, anchovies are significant in the development of neuron in infants and in fat glycemic control to reduce the risk of heart attack. (Kinsella *et al.*, 1990; Mozaffarian *et al.*, 2005).

Fish provides a good source of vitamins and minerals. It may be categorized either as white, oily or shellfish. White fish such as haddock and seer contain very little fat

(usually less than 1%) whereas oily fish, such as sardines, contain between 10-25% fat (FAO, 2010). Fish are important sources for many other nutrients namely vitamins such as Vitamin A, D and E as well as iodine, calcium, selenium etc. There are abundant evidence indicating the significant of fish in brain development and learning in children, protecting vision and eye health, and protection from cardiovascular disease and some cancers.

Fish is the most important animal protein food available in the tropics that represents about 14% of all animal protein when distributed globally (Abolagba and Mello, 2008). According to Ashitey and Flake (2010), fish is a cheaper and preferred source of animal protein which contributes about 60% of animal protein intake in Ghana. Fish is also an important source iron, calcium, iodine, potassium, vitamins, poly-unsaturated fatty acids, other minerals and micronutrients (FAO, 2005).

2.5 FISH DETERIORATION

Fish is highly susceptible to deterioration by fast destruction by enzymes, high pH, high water activity, oxidation of lipids and formation of non-protein nitrogen compounds.

Preservation of fish is intended to inhibit the growth of spoilage bacteria and the metabolic changes that result in fish quality loss. Spoilage bacteria produce unpleasant odors and flavors associated with spoiled fish. An estimated amount of 90–95 percent of the total fish catch is processed into dried and smoked fish (FAO, 2010). Fish without any preservative or processing measures is exposed to a number of physiological and microbial deterioration and thereby degrade the fish (Davies and Davies, 2009). The rate of fish spoilage or deterioration is mainly owing to nature, season, catching methods, acquired micro flora,

atmospheric temperature etc. The quality of fish is a major concern to food processor and public health authorities. Hence, proper preservation of fresh fish becomes very important.

In the artisanal fishery industry in Ghana, through which most of the catch are made, anchovies are not chilled in spite of the high ambient temperatures and also, because of their smallness, gutting is not carried out. Under these conditions autolysis is accelerated in the viscera releasing bacteria and enzymes which invade the flesh (Sikorski *et al.*, 1990).

It has been estimated that in high temperatures of the tropics, for which Ghana is a part, fish deteriorate within 12-20 hours after being caught, depending on the kind and size of fish hence, a considerable proportion of the landed catch is processed to preserve most of their catch by artisanal methods (FAO, 2001). There are some reasons for deterioration of quality and spoilage; they include bacteria spoilage, autolysis, rancidity and mechanical damage (Huss, 1994).

The major challenge worldwide including Ghana, is the unhygienic environmental conditions in which fish finds itself before and after capture and before it comes to the table for consumption (Debrah *et al.*, 2011). Methods used in handling or processing fish are likely to contribute to contaminating the fish with pathogens. Of much concern in public health is the contamination of fish by fecal coliforms in contaminated waters.

In Ghana and other parts of the world, consumption of fish contaminated by pathogens have led to serious health consequences and are responsible for some of the recorded deaths.(Mensah *et al.,* 2002; Scott *et al.,* 2007). It has been discovered that the microbial flora associated with freshly harvested fish is principally a function of the environment in

which the fish are caught and not of the fish species, hence, the indigenous microbial populations of fish can vary significantly (Agbolagba and Mello, 2008).

Other studies on the microbiological quality of fish raised in contaminated water have shown that, fecal bacteria may penetrate fish flesh when fish is grown in highly contaminated water. The occurrence of pathogens such as *Staphylococcus aureus*, *Salmonella typhimurium*, *Bacillus cereus*, *Vibrio spp*. and *Clostridium botulinum* in fish has raised major concerns among researchers since they are the main causes of food borne illnesses (WHO, 1994).

Staphylococcus aureus was detected during the process of smoking and drying of eels in Alaska in 1993. It was reported after 2 to 3 days of processing that *Staphylococcus aureus* populations had increased to more than 10^5 CFU/g of the analyzed sample. Further laboratory studies indicated that the presence of fast air movement pellicles is formed on the strips. Also, bacteria grow when there is heavy deposition of smoke. Reducing air flow and eliminating pre-process drying led to smoke deposition before the formation of pellicle and enabled the product to reach levels of water-phase salt and water activity that helped to inhibit the growth of *Staphylococcus aureus* (Eklund *et al.*, 2004).

Postharvest losses in fish are represented by a clear reduction in the amount of nutrients that are possibly available to the consumer either by physical loss or nutritional loss. These factors have effect on consumer acceptability, income of fish farmers or traders and commercial value (Bostock *et al.*, 1987).

2.6 ANCHOVY PROCESSING AND PRESERVATION METHODS

As a highly perishable food, fish needs to be handled properly and some level of preservation methods is needed if it has to be kept for a longer period. This can help retain a desirable taste, quality and its nutritional composition. Preventing fish deterioration is a central problem of fish processing and this remains a fundamental concern for all other processing activities. Fish processing may also include the addition of value to produce a wide variation in products.

Keeping fish alive before processing for consumption, handling is an important technique that could be used for the preservation of fish quality. There are a number of methods that could be used to preserve fish. Some of which may include the use of ice, freezing; others on the control of water activity which include drying, salting and smoking (FAO, 2010). The common traditional preservative methods used in Ghana are depuration, freezing, smoking, sun-drying and salting (Obodai *et al.,* 2011).

Anchovies (*Engraulis encrasicolus*) are abundant in Ghana, are usually preserved by sun or smoke-drying. Because fresh fish is generally soft, it easily gets damaged; therefore, rough handling and bruising can result in its contamination. In high ambient temperatures of the tropics, fresh fish usually spoils very quickly due to heat. Unless it is subjected to some form of processing or preservation, fresh fish become unfit for human consumption normally within about a day after capture. Even after it has been processed, particularly, if traditional methods such as smoking and sun-drying are used, the fish is still subject to many forms of loss and spoilage (Abolagba and Uwagbai, 2011).

Smoking, sun-drying, salting, grilling, fermentation and frying are the major fish processing methods used in Africa. However, hot smoking methods are prevalent in West Africa for fish processing (FAO, 1998).

The shelf life of fish depends highly on ambient temperature and humidity under which the fish is processed as they dictate the rate at which chemical changes take place.

Preservation of fish by the use of smoldering wood dates back to early civilization (Clucas, 1982). The traditional or conventional methods of drying and smoking of fish results in low quality and short shelf-life products. The methods can also lead to overdrying and excessive smoking of fish causing post-harvest losses.

It must therefore be noted that in all processing operations, care is taken to avoid wastage. Value addition to processed fish product has become the order of the day as the demand for food, that requires little or no preparation before serving, is high (FAO, 2010).

2.6.1 Fish Smoking

Fish smoking is one of the traditional processing methods used to prevent or reduce postharvest losses in the fishing industry. It involves the application of heat to remove water which inhibits both bacterial and enzymatic actions (Kumolu-Johnson *et al.*, 2010) and ends up giving the product a desirable taste and odor, providing a longer shelf-life, lowering pH, imparting desirable coloration as well as accelerating the also process of spoilage (Abolagba *et al.*, 2002).

Smoking is the most widely practiced method. Anchovies is smoked using the Chorkor smoker and usually done by women. Smoking is one of the most ancient processing and

preservation technologies which have been used for centuries. The preservative effect is generally attributed to the anti-oxidant and anti-microbial properties of phenolic compounds (Essumang *et al.*, 2013).

Smoking technology is of two forms, hot and cold smoking. Cold smoking is achieved without thermal treatment usually at temperature below 30°C whereas; the commonly used method of hot smoking is carried out at thermal temperature of 70-80°C (Bykowski and Dutkiewicz, 1996) using the traditional kiln with wood burning temperature of between 300°C and 700°C usually above the 80°C of the oven's temperature (Nti *et al.*, 2002).

Levels and distribution of microbial flora in smoked fish products vary largely, depending on the quality of fish at the time of smoking, the smoking temperature and duration, the salt content and the drying time (Nickelson *et al.*, 2001). Smoking decreases the water activity in fish tissue (Sveinsdottir, 1998).

Practically, all species of fish available in the country can be smoked and it has been estimated that 70-80 percent of the domestic marine and freshwater catch is consumed in smoked form (FAO, 1992, 2001).

It was estimated in 2003 that the quantity of smoked fish from West Africa entering the United Kingdom was 500 tons per year with a retail value of £5.8 to £9.35 million (Ward, 2003).

2.6.2 Sun-Drying

Sun-drying of fish also removes water which inhibits bacterial and enzymatic actions in fish but does not add any desirable taste and odor to the end product. The length of drying depends on the type of fish, its size and the weather (Berkel *et al.*, 2004).

By tradition, small fish are dried whole while large fish are split before drying directly in the sun on the ground and sometimes on raised racks. The use of sun in drying fish usually does not allow control over the drying times and this may exposes the fish to contamination by insects attack and animal pests and dirt (UNIFEM, 1998).

During wet seasons, the drying of fish slows down and may take about seven days or beyond in some cases. Drying on the ground exposes the fish to pets and other domestic animals such as sheep, goats, pigs, lizards, etc. Drying of fish on platforms can also help give a better product.

The quality of preserved fish is therefore linked to the handling, processing and post processing procedures. During these periods, fish is susceptible to microbial attack. Microorganisms are the major cause of spoilage of most seafood products. Some microorganisms that contaminate fish are *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhimurium*, *Bacillus cereus*, *Shigella spp.*, *Clostridium botulinum* etc. (Obodai, 2011).

Limitations of this method include considerable product losses, lower fish quality because of contamination by foreign materials, insects and microorganisms as well as discoloring by ultraviolet radiation (Tiwari and Sarkar, 2007).

The processes of handling fishes are also prone to aflatoxin contamination especially in artisanal fishery due to unhygienic methods of preservation.

2.7 FOOD IRRADIATION

Food irradiation is a technology to improve food quality, to provide hygienic food and to extend its shelf life using ionizing radiation. Processed anchovies are packaged and subjected to ionizing radiation to decontaminate pathogens and extend product shelf-life. Irradiation of fishery products is the process to kill harmful bacteria and other organisms by being exposed to ionizing energy (IAEA, 2000).

Food irradiation has been used for the purposes of inhibition of sprouting, destruction of food borne insects and parasites, delay of physiological ripening and extension of shelf life or improvement of food qualities (Kim *et al.,* 2005).

This technology is also applicable for the preservation and enhancing the quality of other types of fish and its products. Ionizing radiation is used for reducing contamination in vegetables, fruits and meat products for long-term preservation. Besides sun-drying, smoking and refrigeration, ionizing radiation is the only alternative to heat processing for food preservation that has a lethal effect on micro-organisms.

The application of radiation in the preservation of fishery products has been exploited by Nickerson *et al.,* (1954) and Proctor *et al.,* (1960). Irradiation at a dose of up to 10 kGy has been used in both animal and vegetable foods as an effective, safe and economical method of preserving food without posing any nutritional, toxicological or microbiological problems (WHO 1994). Arvanitoyannis and Stratakos (2010), reported that irradiation

doses of 2–7 kGy can reduce important food pathogens or microbes such as *Salmonella, Listeria,* and *Vibrio spp*., as well as many fish specific spoilers such as *Pseudomonaceae* and *Enterobacteriaceae* that can be significantly decreased in number. Ionizing radiation treatment of food is an effective means of slowing down pathogenic bacteria growth such as *Escherichia coli* and *Salmonella* (Olson, 1998; Thayer, 1994).

Radiation pasteurization when used does change the quality of food (IAEA, 2000). Also, food irradiation can be used to improve the microbiological safety and to extend the shelf life of foods. Though the establishment cost of food irradiation plant is very high, the operational cost is lower than most of other preservation methods (IAEA, 2000).

Eviscerated sea bass, *Dicentrarchus labrax* was irradiated at doses of 2.5 and 5 kGy at 24°C using a Cobalt-60 source. The effect of the irradiation on proximate, amino acid composition and fatty acid in cultured sea bass were then investigated. Significant differences (P<0.0.5) were found to exist between unirradiated and irradiated sea bass in terms of moisture, protein, fat, ash and carbohydrate contents. The total saturated and total mono-unsaturated fatty acid contents were 27.97-24.72% for non-irradiated sea bass respectively. The amounts of these two fay acids in irradiated samples increased to 28.1825.75% for 2.5 kGy and 29.08-28.54% for 5 kGy. Again, it was observed that the total polyunsaturated fatty acid content for irradiated samples were higher than non-irradiated samples. Aspartic acid, glutamic acid, asparagines, histidine, serine, glycine, arginine, alanine, tyrosine, cystine, methionine, lysine, hydroxyproline and proline contents for 2.5 and 5 kGy irradiated sea bass according to them were significantly different (P<0.05)

(Ozden and Erkan, 2007).

Aworh *et al.*, (2002), investigated the consequence of low dose (< 6kGy) on quality, shelf life and consumer acceptance of three traditional Nigerian meat and fish products and concluded that irradiation was able to inhibit microbial growth in 'suya' and 'kilishi' with considerable reduction in total aerobic counts, yeasts and molds and *Staphylococcus aureus*.

Prepackaging of seafood is an integral part of radiation processing primarily for avoiding post-irradiation contamination. Polypropylene bags are suitable for the process. In many instances of irradiation processes, the necessity of packaging materials is to prevent recontamination or re-infestation of the food by containing the product and essentially protecting the product from the surrounding environment.

Kombat *et al.*, (2013) carried out microbiological quality analyses on processed anchovy (*Engraulis encrasicolus*) and round sardinella (*Sardinella aurita*) collected from processing houses and local retail markets in Accra and Tema to assess their quality. Their work looked at smoked and sun-dried *Engraulis encrasicolus* and smoked *Sardinella aurita* that was randomly collected from selected processing houses and retail markets from Accra and Tema for analysis. Using the serial dilution, pour plate and spread plate methods to enumerate levels of total heterotrophic bacteria, total coliform bacteria, yeast and molds and *Bacillus cereus* colonies in the samples, they concluded that samples obtained from the retail markets recorded total heterotrophic bacteria counts ranging from $1.9 \times 10^4 - 5.9 \times 10^5$ CFU/g, while those obtained from the processing houses ranged from $1.2 \times 10^3 - 6.5 \times 10^4$ CFU/g, which were within Ghana Standards Authority accepted limits (1×10^6 CFU/g) for fish and fish products. There were counts of total coliform bacteria, yeast and molds

and *Bacillus cereus* for the samples, but it was established that they were all within the accepted limits, except for *Bacillus cereus*, which recorded counts higher than the accepted limits (1 x 10^4 CFU/g) for some samples obtained from retail markets in both Tema and Accra.

2.7.1 Fish (Anchovies) Irradiation

Anchovy is exposed to a carefully measured dose of intense ionizing radiation. This is done in a special processing room or chamber for a specified duration. The most common source of ionizing energy is Cobalt 60. These radioactive materials are contained in two sealed stainless steel tubes called "source pencils." These source pencils are placed in a rack and the entire rack is immersed in a water chamber underground when not in use. During the process, the rack is raised. Packaged food products are moved along the conveyer belt to enter the irradiation chamber where they are exposed to the rack containing source pencils. The energy in the form of gamma rays pass through the encapsulation to treat the food (IAEA, 2000).

Packaging is very important in anchovy processing to facilitate easy handling of product during storage and sale within the marketing chain. The type of packaging material to be used should be able to provide adequate protection to the packaged product from damage. This could be an economic loss of profit on fish and its products. The packaging material must be easy to use, readily available and should be able to prevent contamination with undesirable elements (FAO, 2010). The suitable packaging material used for irradiating smoked anchovies and species are polyethylene and polypropylene bags. Irradiation with

doses up to 10 kGy has been reported to have no significant effect on the physical properties of the packaging materials tested. (Maha *et al.*, 1990).

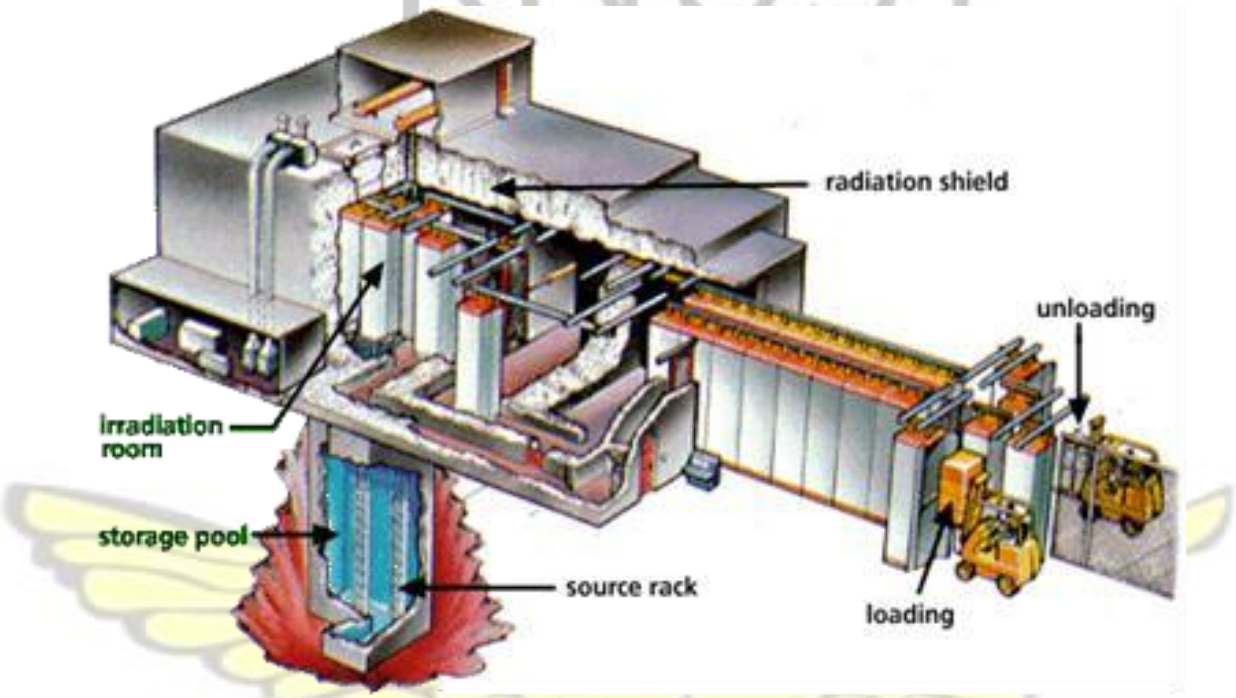

Figure 2.1. A typical gamma irradiation facility

The use of jute bags, baskets and poly sacks form the most widely used materials for packaging sun-dried and fermented fishery products for storage or during distribution and marketing. These type of packaging materials can predispose fish and its products to insects and rodent attack as well as to water or other fluids. This could render products susceptible to rapid deterioration if exposed to rain (FAO, 2010).

Research on irradiation of fish and its products indicated that the shelf life of products was enhanced by irradiation doses of ≤10 kGy (Ahmed *et al.*, 1997).

Figure 2.2. Tote boxes containing packaged anchovies ready for irradiation

3.0 MATERIALS AND METHOD

This chapter describes the processes and activities undertaken to identify the locations where anchovies were harvested, handled, processed and marketed to the final consumer and the laboratory study carried out on irradiated and unirradiated anchovies.

The microbial and nutritional analyses were carried out on samples before and after irradiation of anchovies over a period of nine weeks at three (3) weeks intervals. In the microbiological analysis, preliminary studies were carried out to investigate the presence of *Salmonella*, yeasts and molds, total viable counts, total coliform counts and *Staphylococcus aureus* in sun-dried and smoked anchovies harvested from Chorkor and Keta showed no detection of *Salmonella* and yeasts and molds in samples. Therefore, the study concentrated on the total viable count (TVC), total coliform count (TCC) and counts of *Staphylococcus aureus*.

With regard to the nutritional quality, the study emphasized the total ash content and free fatty acids (FFA) analyses of smoked and sun-dried anchovies obtained from Chorkor and Keta because the preliminary investigation carried to determine protein, ash content, free fatty acids in both unirradiated and irradiated anchovies from Chorkor and Keta found out that, the irradiation process had no effect on the protein content. Hence protein was dropped from the nutritional parameters that were further studied.

3.1 STUDY AREA DESCRIPTION

A reconnaissance survey was carried out at Chorkor, James Town, Senya Breku, Tema, Keta and Akatsi to know from the fisher folks the main fishing location of anchovies.

There were fishermen along the entire coastline of Ghana and one needed to determine where anchovies were harvested. The main survey was then targeted at the main areas of harvest of anchovies.

Laboratory analyses were done at the Radiation Technology Centre and the Food Science Department / Food Microbiology Laboratory, both at the Ghana Atomic Energy Commission in Accra.

3.2 RESEARCH PROCEDURE

The experiment was conducted in two phases; a survey and a laboratory work.

3.2.1 Survey locations

The study areas were Chorkor in the Greater Accra Region and Keta in the Volta Region. These were purposively selected as they were known to be a good source of anchovies.

Chorkor can be found in the Accra Metropolis of Ghana. It is a densely populated community found in the shorelines of Accra. The neighborhood is mainly known for fishing. Chorkor is renowned for its locally manufactured ovens called Chorkor Smokers used for smoking large quantities of fish.

Keta is the capital of Keta Municipal District in the Volta Region. It has a population of about 23, 207 and also ranked as the 61st most populous area in Ghana. Keta Lagoon is the largest lagoon in Ghana with a water area of 300 km^2. Being a Municipality with a coastline of over 60km, fish resources are in abundance as well other agricultural activities such as crop production and livestock production.

3.2.2 Questionnaire design

A structured questionnaire, both closed and open ended, was designed to obtain the requisite data. The parameters that were considered included bio-data of respondents, fishing activities, handling procedures or operation of fish, packaging and marketing of anchovies.

3.2.3 Questionnaire administration

Personal interviews together with structured questionnaires were administered to anchovy processors and marketers to obtain the requisite data. The questionnaires were administered to anchovy processors and marketers or sellers in the selected communities (Chorkor and Keta). Thirty (30) people each were selected from Chorkor and Keta; of which twenty (20) were fishermen and ten (10) were marketers or sellers. In all, sixty (60) respondents were selected from both locations. The selection of respondents was done using purposive sampling.

3.3 LABORATORY ANALYSIS

3.3.1 Sampling area

Anchovy samples were taken from the selected communities of Chorkor in the Greater Accra Region and Keta in the Volta Region.

3.3.2 Samples Collection

A total of about 1600g of anchovies were collected from both locations. From Chorkor, 200g sun-dried and 200g smoked anchovies were collected randomly from processors

(400g) and marketers (400g). Similar samples were collected from Keta for irradiation and laboratory analysis.

The smoked and sun-dried samples collected were packaged in well-labelled sterile plastic bags and transported to the Radiation Technology Centre, Ghana Atomic Energy Commission.

3.3.3 Samples Preparation

About 30g each of sun-dried and smoked anchovy samples from the two locations were weighed on an electronic balance and packaged into separate polyethylene zip lock bags in triplicates; and prepared for the study.

The first batch of the unirradiated and irradiated samples were taken to the Food Science laboratory immediately for microbial load analysis. The remaining packaged samples were stored under ambient conditions and analyzed every three (3) weeks for a period of nine (9) weeks for microbial load and nutritional analyses.

3.4 GAMMA IRRADIATION

The anchovies were packaged in polyethylene zip lock bags and irradiated at doses 2.5, 5.0, 7.5 and 10.0kGy using the category IV cobalt 60 (^{60}Co) wet storage gamma irradiation source at the Ghana Atomic Energy Commission which has a current strength of 30kCi (Bq). Non-irradiated samples were used as control.

The anchovy samples were packed into the polyethylene zip lock bags and arranged in a metal fabrication attached to the shroud covering an area of 50cm by 20cm. The shroud houses the plaque source when at the irradiation position.

Ethanol chlorobenzene (ECB) Dosimeters were put into the anchovy samples and were together subjected to gamma radiation and turned through 180°C at halfway the processing time to ensure homogenous distribution of the dose delivered under the same conditions. The ECB dosimeters were removed from the anchovies after the irradiation period and the absorbed dose was then determined using a calibrated readout instrument (High Frequency Dosimeter System, Model 2131, version 2.5, and produced by SENSOLAB LTD). The delivered doses ranged from 2.5, 5.0, 7.5 to 10 kGy with an error of ±2.3% at a dose rate of 1.43kGy/hr.

3.5 PARAMETERS ASSESSED

3.5.1 Microbial Load Analysis

The microbiological analysis was performed at the Food Microbiology Laboratory, Biotechnology and Nuclear Agriculture Research Institute (BNARI) according to the standard procedure of APHA (2000) using methods of serial dilution and pours plated to determine:

- *Salmonella*
- Yeasts and molds
- Total Viable Count
- Total Coliform Count
- Counts of *Staphylococcus aureus*

All media were prepared and sterilized according to the manufacturer's instructions. Microbiological media was prepared in appropriate quantities and sterilized by autoclaving. The autoclaved media were tempered in a water bath at 45°C to prevent media from

solidifying. It was also ensured that the water level was 1 cm above the level of the medium in the bottles.

3.5.1.1 Enumeration of *Salmonella*

Salmonella is an enteric pathogenic bacteria found in food. Ingestion of *Salmonella* may cause intestinal infection or typhoid (Environmental Health and Safety, 2012).

About 10grams of anchovies sample was aseptically weighed into sterile petri dish, macerated in a stomacher and transferred into a sterile 90ml peptone water in a 250ml conical flask to make a 1:10 dilution of the anchovy sample. Each dilution bottle was agitated to re-suspend material that may have settled out during preparation and serial diluted to 10^6. One (1) ml of each diluent was aseptically transferred into sterile welllabeled petri dishes and pour-plated using xylose lysine dexoychocolate agar (XLD) in duplicates and aerobically incubated at ±37°C (APHA, 2000).

3.5.1.2 Enumeration of Yeasts and molds

Mold is a fungi which contains multiple similar nuclei which grows in the form of hyphae of filaments. Ingestion of molds can cause respiratory problems and some allergic reactions. As a type of fungi, yeast contains only a single cell which may likely cause infection in individuals with compromised immune system (McGinnis and Tyring, 1996).

About 10grams of anchovies sample was aseptically weighed into sterile petri dish, macerated in a stomacher and transferred into a sterile 90ml peptone water in a 250ml conical flask to make a 1:10 dilution of the anchovy sample. Each dilution bottle was agitated to re-suspend material that may have settled out during preparation and serial

diluted to 10^6. One (1) ml of each diluent was aseptically transferred into sterile welllabeled petri dishes and pour-plated using oxytetracycline glucose yeast extract agar (OGYE) in duplicates and aerobically incubated at ±37°C (APHA, 2000).

3.5.1.3 Enumeration of total viable count (TVC)

Total Viable Count (TVC) gives a quantitative idea about the presence of microorganisms such as bacteria, yeast and mold in a sample. Specifically, the count actually represents the number of colony forming units (CFU) per gram (g) of the sample.

Ten (10) grams of anchovies sample was aseptically weighed into sterile petri dish, macerated in a stomacher and transferred into a sterile 90ml peptone water in a 250ml conical flask to make a 1:10 dilution of the anchovy sample. Each dilution bottle was agitated to re-suspend material that may have settled out during preparation and serial diluted to 10^6. One (1) ml of each diluent was aseptically transferred into sterile welllabeled petri dishes and pour-plated on plate count agar (PCA) in duplicates and aerobically incubated in inverted positions at 37°C (APHA, 2000).

3.5.1.4 Enumeration of total coliform count (TCC)

Total Coliform count is the most basic test for bacterial contamination of a water supply. Total coliform counts give a general indication of the sanitary condition of a water supply or environment in which fish finds itself.

About 10grams of anchovies sample was aseptically weighed into sterile petri dish, macerated in a stomacher and transferred into a sterile 90ml peptone water in a 250ml conical flask to make a 1:10 dilution of the anchovy sample. Each dilution bottle was

agitated to re-suspend material that may have settled out during preparation and serial diluted to 10^6. One (1) ml of each diluent was aseptically transferred into sterile welllabeled petri dishes and pour-plated using eosin methylene blue agar (EMBA) in duplicates and aerobically incubated at ±37°C (APHA, 2000).

3.5.1.5 Count of Staphylococcus aureus

Staphylococcus aureus is a facultative anaerobic bacterium that causes infection when ingested through food.

For enumeration of *Staphylococcus aureus*, about 10grams of anchovies sample was aseptically weighed into sterile petri dish, macerated in a stomacher and transferred into a sterile 90ml peptone water in a 250ml conical flask to make a 1:10 dilution of the anchovy sample. Each dilution bottle was agitated to re-suspend material that may have settled out during preparation and serial diluted to 10^6. One (1) ml of each diluent was aseptically transferred into sterile well-labeled petri dishes and pour-plated using bird parker agar (BP) in duplicated and aerobically incubated at 37 °C.

In each case, the media were poured not more than 15 minutes after preparation of dilutions at about 45°C on their respective petri plates. Each plate was swirled gently to mix and left for about 15 minutes to solidify. About 4ml of plate count agar, eosin methylene blue agar and bird parker agar were added to their respective plates (second layer) and left to solidify. This was to make sure that the entire sample is covered. The plates were incubated at 37°C±1 for 24 – 48hrs. The results were checked on daily basis up to 48 ± 4hrs.

Colonies were counted promptly after the incubation period using the Stuart colony counter-SC6+. Plates with 30-300 colonies or nearest to the 30-300 range were counted (including pinpoint colonies). Colonies were counted as colony forming units per gram of fish sample (CFU/g) (APHA, 2000).

3.5.2 Nutritional Analysis

Nutritional analysis was carried out at the Food Microbiology Laboratory, BNARI according to the standard procedure of AOAC (2000) International to determine the protein content, ash content and free fatty acids.

3.5.2.1 Protein determination

About 5g of sample was weighed into a digestion flask and then digested by heating it in the presence of sulfuric acid. After digestion was completed, the digestion flask was then connected to a receiving flask by a tube. Sodium hydroxide was then added to the solution in the digestion flask which converts the ammonium sulfate into ammonia gas. The ammonia gas that was formed was liberated from the solution and moves out of the digestion flask and into the receiving flask which contains an excess of boric acid. The content was then estimated by titrating the ammonium borate formed with standard hydrochloric acid using a suitable indicator to determine the end-point of the reaction (AOAC, 2000).

$$\%N = \frac{x\ \text{moles}}{1000\ \text{cm}^3} \times \frac{(v_s - v_b)\ \text{cm}^3}{m\ \text{g}} \times \frac{14\ \text{g}}{\text{moles}} \times 100$$

3.5.2.2 Total ash content

Ash content was determined in triplicate by standard procedures in (AOAC, 2000). A fairly representative sample of 2g was weighed into a crucible which has been previously dried, cooled and weighed. The crucible and its content were ignited in a muffle furnace (carbolite, CWF 1200) at 600°C for 6 hours. The crucibles were removed, cooled in a desiccator and then weighed.

Total ash content was then expressed based on weight loss.

$$\% \, Ash = \frac{weight \, of \, ash}{weight \, of \, sample} \text{ x } 100$$

Ash or mineral content is the portion of the food or any inorganic material that remains after it is burned at very high temperatures. The ash constituents include potassium, calcium, sodium and magnesium, aluminum, copper, iron, manganese or zinc, arsenic, iodine, fluorine and other trace elements. Ash content represents the total mineral content in foods which represent a small proportion of dry matter and play an important role in the physicochemical and nutritional aspect of food AOAC (2000).

3.5.2.3 Free fatty acids (FFA)

The free fatty acid values were determined in triplicate by standard procedures in (AOCS, 1997) with modifications. An amount of 1g of sample was mixed with 25ml of 95% ethanol and swirled for 5 minutes. A volume of 1m of 1% phenolphthalein solution was added as an indicator. This was allowed to stand for 5 minutes, decanted and titrated with aqueous 0.1N sodium hydroxide until pinkish color persists for 15 seconds indicating end point.

The expression as given in AOCS official method by AOCS 1997 as % free fatty acid (oleic)

$$= \frac{alkali\ volume\ (m1) \text{x}\ alkali\ normality\ (N) \text{x}\ .0282}{sample\ weight\ (g)} \text{ x } 100$$

Oleic acid is a mono-saturated fatty acid found in animal and vegetable oils Oleic acid occurs naturally in greater quantities than any other fatty acid. It is present as glycerides in most fats and oils. High concentrations of oleic acid can lower blood levels of cholesterol. It is generally believed to be good for human health (Teres *et al*., 2008).

3.5.3 Shelf-Life (Storage) Studies

Packaged samples of both unirradiated and irradiated sun-dried and smoked anchovies were stored and visually assessed every three (3) weeks for nine (9) weeks. This was done to determine the effect of the irradiation doses (2.5, 5.0, 7.5 and 10 kGy) on the anchovy samples. The samples were stored in an enclosed mesh shelf under ambient conditions (average temperature of 22°C and RH of 50%). The mesh shelf was to prevent pest damage to the fish samples.

The samples were assessed visually for color change, moldiness, insects and pest attacks. This monitoring was done every three (3) weeks before samples are taken to the laboratory for microbial and nutritional quality analyses.

Shelf life was taken as the number of days or period for a sample to be contaminated or infested. The shelf-life of a product is a critical factor in both quality and profitability, and is influenced by several factors, such as light, heat, gases intrinsic to the product and stresses on the material.

3.6 EXPERIMENTAL DESIGN

The experimental design for the study of processed anchovies was a 2×2×2×5 factorial in Completely Randomized Design (CRD) (two locations: Chorkor and Keta; source: processors and marketers; processing methods: smoked and sun-dried; and irradiation doses: control, 2.5, 5.0, 7.5 and 10 kGy).

3.7 STATISTICAL ANALYSIS

Sample analyses were conducted in triplicates for the study. Results were expressed as mean values and the differences among means of both unirradiated and irradiated samples of smoked and sun-dried Anchovies (*Engraulis encrasicolus*) obtained from Chorkor and Keta were calculated using analysis of variance (ANOVA) and statistically significant differences were reported at $P<0.05$. The Least Significant Difference (LSD) was conducted for independent sample t-test as required between two treatments. Data analyses were done with the use of GenStat software version 18.0.

4.0 RESULTS

This chapter describes the results obtained from the microbial and nutritional analyses of sun-dried and smoked anchovy samples obtained from Chorkor and Keta in the Greater Accra and Volta regions respectively.

4.1 SURVEY RESULTS

Figure 4.1 shows the percentage of fishermen and women engaged in the fishing of anchovies in both locations. From the survey results, both men and women were involved in the harvesting, handling, processing and marketing activities of anchovies. The men mostly (58%) do the fishing and harvesting while the women (42%) do the processing and marketing of the fish. However, there were few times when they had children engaged in the fishing activity. This was very common in Chorkor in the Greater Accra Region of Ghana.

Anchovies were usually harvested in the early hours of the morning with other fish using nylon seine nets attached to a canoe. Anchovies, together with other fish, are mainly harvested in these areas in August-September annually. The fishermen or fisher folks in both Chorkor and Keta usually processed anchovies into smoked or sun-dried form because of its demand by consumers.

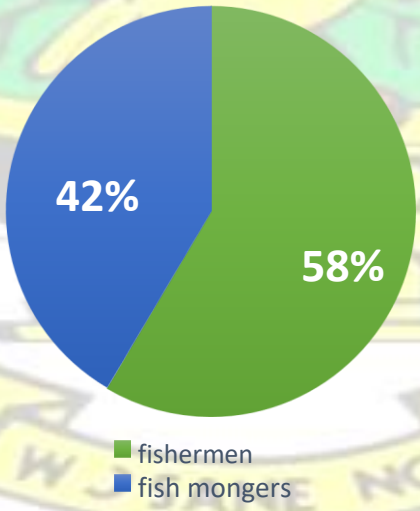

Figure 4.1. Percentage of fishermen and fish mongers engaged in fishing and handling of anchovies at both Chorkor and Keta.

4.2 LABORATORY WORK

4.2.1 Microbial Load Analysis

Table 4.1 show results of analysis of microbial load in unirradiated smoked and sun-dried anchovies obtained from both Chorkor and Keta.

Total viable count had significantly ($p<0.05$) higher contamination of samples (5.660 CFU/g) than total coliform count (3.621 CFU/g). *Staphylococcus aureus* had least contamination (2.911 CFU/g) at week 0. Sun-dried samples obtained from Chorkor at week 0, had significantly ($p<0.05$) higher total viable count (5.633 CFU/g) when compared to sun-dried samples from Keta anchovies (4.490 CFU/g); as well as smoked samples from Chorkor (6.175 CFU/g) and Keta (6.042 CFU/g).

There were no significant ($p>0.05$) differences in total coliform count between sun-dried (3.487 CFU/g) and smoked samples (3.645 CFU/g) obtained from Chorkor when compared to sun-dried (3.272 CFU/g) and smoked samples (3.487 CFU/g) from Keta at week 0 and also at 9th week. *Staphylococcus aureus* had least contamination in sun-dried (2.575 CFU/g) and smoked anchovy samples (3.318 CFU/g) from Chorkor; and sun-dried (2.972 CFU/g) and smoked samples (2.778 CFU/g) from Keta respectively.

There was a general decrease in microbial load with time. At the 9th week, the level of contamination had generally decreased in sun-dried samples obtained from Keta generally when compared to sun-dried samples obtained from Chorkor. Both locations had no significant ($p>0.05$) difference when anchovy samples were smoked even though some amount of contamination was recorded in the period of study (week 0, 3, 6 and 9).

Table 4.1. Microbial load of unirradiated sun-dried and smoked anchovies obtained from Keta and Chorkor

MICROBIAL LOAD (CFU/g)	PROCESSING METHODS	LOCATIONS	STORAGE PERIOD (WEEK) 0	3	6	9	MEAN
		KETA	4.490*	3.542	2.940	2.602	**3.394**
		CHORKOR	5.633	5.597	3.668	2.428	**4.332**
TOTAL VIABLE COUNT							
	SUNDRIED						
		KETA	6.042	5.117	4.285	2.265	**4.430**
		CHORKOR	6.175	6.082	3.023	2.562	**4.500**
	SMOKED						
		MEAN	**5.660**	**5.085**	**3.479**	**2.464**	
			3.272	2.620 3.412	2.590 2.938	2.047 2.202	**2.632 3.001**
			3.487	3.432	2.972	2.002	**3.122**
			4.080	3.645	3.212	2.163	**3.166**
			3.645				
		KETA					
		CHORKOR					
TOTAL COLIFORM	SUNDRIED	KETA					
		CHORKOR					

COUNT

	SMOKED					
		MEAN	**3.621**	**3.277**	**2.928**	**2.104**	
			2.972	2.310 1.985	2.547 1.767	1.605 1.620	**2.359 1.987**
			2.575	2.243	2.313	1.873	**2.302**
			2.778	2.917	2.320	1.692	**2.562**
			3.318				
Staphylococcus aureus	SUNDRIED						
		KETA					
		CHORKOR					
		KETA					
	SMOKED	CHORKOR					
		MEAN	**2.911**	**2.364**	**2.237**	**1.698**	

Lsd (5%): Microbial load = 0.1939; Processing methods = 0.1583; Storage = 0.2239;
Microbial load x Processing methods x Storage = 0.2743

*$\log_{10}$

4.2.2 Nutritional Analysis

Table 4.2 shows the percentage (%) total ash content and free fatty acids of unirradiated smoked and sun-dried anchovies obtained from both Chorkor and Keta. Sun-dried samples were not significantly different ($p>0.05$) in total ash content when compared to smoked samples. The sun-dried samples obtained from processors (21.47%) had high amount of ash content when compared to marketers (20.80%) at the 3rd week. Smoked samples obtained from marketers (17.99%) at week 3 had no significant ($p>0.05$) difference in ash content when compared to processors (18.44%).

There was no significant ($p>0.05$) difference in the ash content of unirradiated smoked and sun-dried samples obtained from both marketers and processors in Keta and Chorkor, with respect to the storage during the period (i.e. week 0, week 3, week 6 and week 9). Samples stored at the third (3rd) week (19.68%) recorded significant ($p<0.05$) higher amount of total ash content with a gradual decrease in the sixth (6th) week (15.99%) and week 9 (14.99%).

Free fatty acids (oleic acid) found in unirradiated smoked and sun-dried anchovies obtained from both Chorkor and Keta. Smoked samples (1.0975%) recorded significant ($p<0.05$) high amount of oleic acid and when compared to sun-dried samples (0.8366%). Smoked samples had significant ($p<0.05$) higher value when compared to and sun-dried samples obtained from both marketers and processors, with a slight decrease from week 3 (1.195%) to week 9 (0.7238%) and in week 6 for sun-dried samples and in week 3 (0.8037%) but an increase in oleic acid in week 9 (0.9823%) for smoked samples respectively. As storage period increases, level of oleic acid in both smoked and sundried samples decrease. Smoked samples recorded lower levels of oleic acid in week 3 (0.8037%) and in week 9 (0.9823%). Sun-dried samples recorded higher amounts of oleic

acid in week 3 (1.1915%), high in week 6 (1.1421%) and least amount in the ninth week (0.9588%). There were significant ($p<0.05$) differences during the storage duration of unirradiated smoked and sun-dried kept for a period of 9 weeks at 3 weeks interval.

Table 4.2. Nutritional Composition of unirradiated (control) sun-dried and smoked anchovies obtained from the processors and marketers.

NUTRITIONAL COMPOSITION (%)	PROCESSING METHODS	SOURCES	STORAGE ERIOD (WEEK) 3	6	9	MEAN
ASH CONTENT	SUN-DRIED	MARKETERS	20.80	15.94	13.38	**16.71**
		PROCESSORS	21.47	16.59	16.08	**18.05**
	SMOKED	MARKETERS	17.99	15.94	13.83	**15.92**
		PROCESSORS	18.44	15.48	16.68	**16.87**
		MEAN	**19.68**	**15.99**	**14.99**	
FREE FATTY ACIDS	SUN-DRIED	MARKETERS	1.1703	1.1280	0.9870	**1.0951**
		PROCESSORS	1.2126	1.1562	0.9306	**1.0998**
	SMOKED	MARKETERS	0.9024	0.8272	1.2408	**0.9901**
		PROCESSORS	0.7050	0.6204	0.7238	**0.6831**
		MEAN	**0.9976**	**0.9329**	**0.9706**	

Lsd (5%): Processing methods = 0.80; Source = 0.80; Storage period = 0.98; Processing methods x Storage = 1.13

4.2.3 Effect of Irradiation on Microbial Load and Nutritional Quality of Processed Anchovies from Chorkor and Keta

Table 4.3 and Figures 4.2 and 4.3 show the effect of gamma irradiation on the microbial load and nutritional composition of smoked and sun-dried anchovies obtained from both locations respectively.

There were differences for the various irradiation dose used. Table 4.4 at each dose, the number of micro-organisms reduced, thus reducing contamination level. There was significant ($p<0.05$) reduction in microbes from control to 5.0kGy as Total Viable Count recorded high value in smoked samples (4.502 CFU/g) and in sun-dried samples(3.393 CFU/g) at 0 kGy and least in smoked samples (1.158 CFU/g) and in sun-dried samples(1.607 CFU/g) at 5.0 kGy, respectively.

Samples exposed to dose rate of 7.5 kGy and 10 kGy had no micro-organisms. The higher the dose, the lower the contamination. Total Viable Count recorded highest microbes compared to *Staphylococcus aureus.* There was significant ($p<0.05$) difference between the microbial loads of irradiated samples from control to 10 kGy.

Sun-dried samples recorded low microbial load compared to smoked samples. *Staphylococcus aureus* recorded lower values in sun-dried samples (1.308 CFU/g) and smoked samples (1.453 CFU/g) at 2.5 kGy; and at 5.0 kGy had low values in sun-dried samples were 0.263 CFU/g and 0.554 CFU/g in smoked samples respectively. There was significant ($p<0.05$) differences in the processing methods used for the treatment and preservation of *Engraulis encrasicolus.*

Table 4.3: Gamma irradiation effect on microbial load of smoked and sun-dried anchovy samples

DOSE (kGy)	MICROBIAL LOAD (CFU/g)	PROCESSING	
		Smoked	Sun-dried
Control	Total Viable Count	4.502*	3.393
	Total Coliform Count	3.121	2.632
	Staphylococcus aureus	2.302	2.358
2.5	Total Viable Count	2.333	2.305
	Total Coliform Count	1.902	1.794
	Staphylococcus aureus	1.453	1.308
5.0	Total Viable Count	1.158	1.607
	Total Coliform Count	1.017	0.271
	Staphylococcus aureus	0.554	0.263
7.5	Total Viable Count	ND	ND
	Total Coliform Count	ND	ND
	Staphylococcus aureus	ND	ND
10.0	Total Viable Count	ND	ND
	Total Coliform Count	ND	ND
	Staphylococcus aureus	ND	ND

Lsd (5%): Dose = 0.184; Microbial load = 0.184; Processing methods = 0.150; Dose x Microbial load x Processing methods =0.319; *log $_{10}$; ND = no detection

Figures 4.2 and 4.3 show the effect of irradiation on the total ash content and free fatty acids (oleic acid) in smoked and sun-dried anchovies obtained from Chorkor and Keta. There was no significant ($p>0.05$) difference in total ash content among treatments. There was also no significant ($p>0.05$) difference in the processing methods used for the determination of total ash content and free fatty acids (oleic acid) in smoked samples

(18.04%) and sun-dried samples (18.84%). The total ash content was 18.04% and 18.84%, respectively. For oleic acid, the smoked samples had 0.69% and the sun-dried samples had 1.03%. The ash content of 18.44% recorded significant ($p<0.05$) high amount of nutritional components when compared to free fatty acids (oleic acid) (0.86%) of both smoked and sun-dried anchovies.

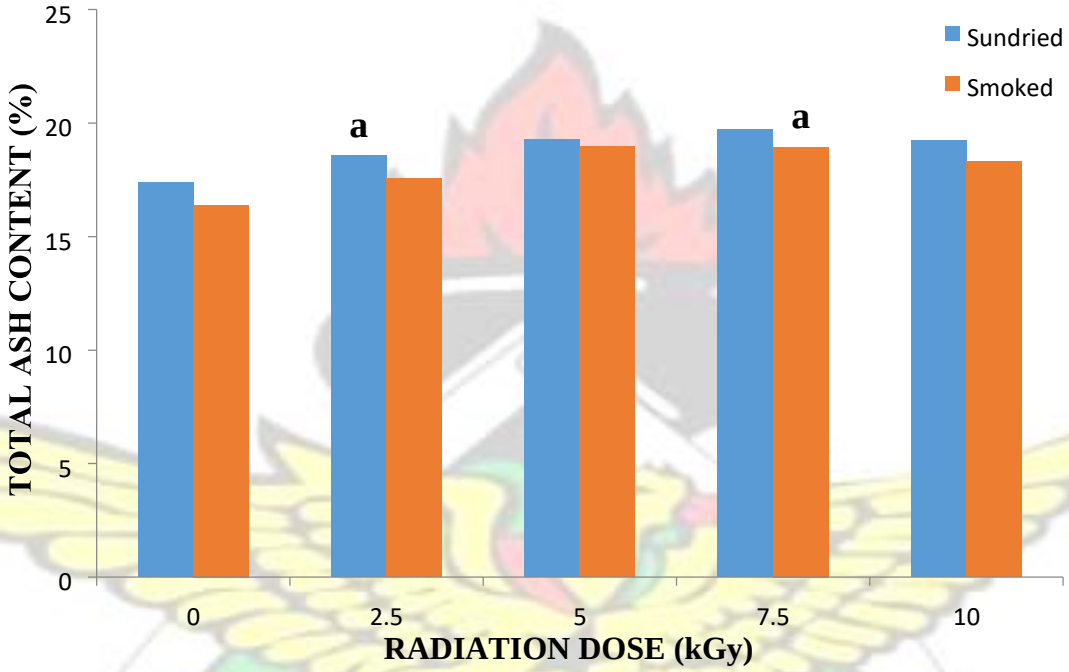

Figure 4.2. Effect of irradiation on the total ash content of sun-dried and smoked anchovies obtained from both locations.

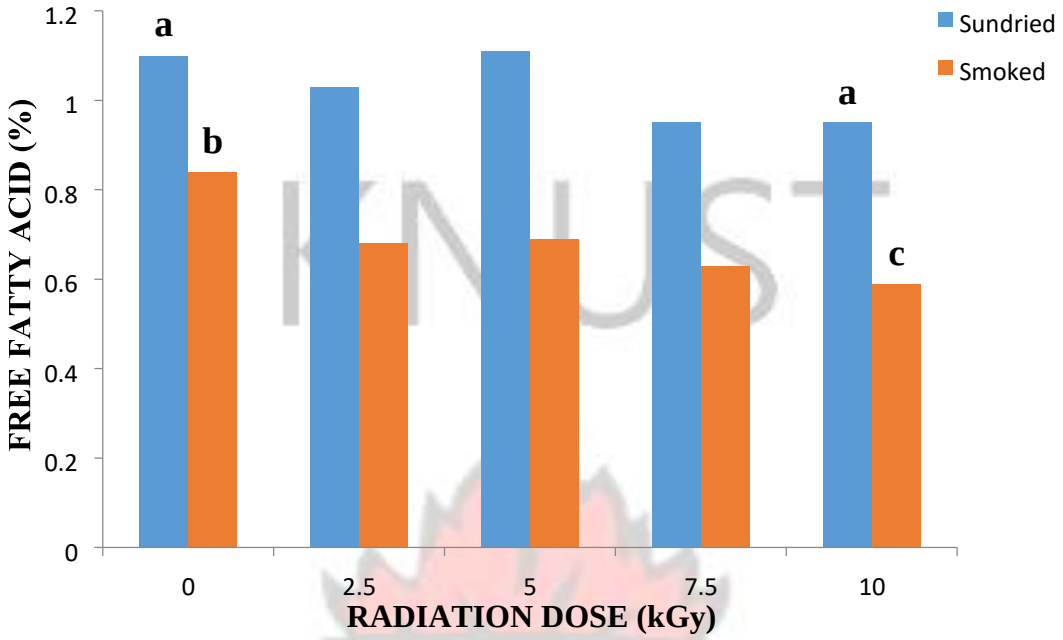

Figure 4.3. Effect of irradiation on the free fatty acids (oleic acid) on the sun-dried and smoked anchovies obtained from both locations.

4.2.4 Effect of Gamma Irradiation on the Shelf-Life of Processed Anchovies Table 4.4 shows the shelf-life of irradiated smoked and sun-dried anchovies obtained from both locations when stored for a period of 9 weeks at a 3-week interval studies.

Smoked but unirradiated samples obtained from Chorkor showed signs of pest destruction at the 3rd week as well as sun-dried samples in week 5. Samples obtained from Keta at the 5th and 7th week for smoked and sun-dried samples respectively, had signs of insect pest damage.

Samples exposed to dose rates 2.5, 5.0, 7.5 and 10 kGy had no records of insect destruction within the 9weeks of storage.

Table 4.4: Gamma irradiation effect on shelf-life (weeks) of smoked and sun-dried anchovies obtained from Chorkor and Keta

DOSE (kGy)	CHORKOR		KETA		**Means**
	Smoked	Sun-dried	Smoked	Sun-dried	
Control	3	5	5	7	**5**
2.5	9	9	9	9	**9**
5.0	9	9	9	9	**9**
7.5	9	9	9	9	**9**
10	9	9	9	9	**9**
Means	**9.75**	**10.25**	**10.25**	**10.75**	

5.0 DISCUSSION

5.1 SURVEY INFORMATION

Both fishermen and fishmongers in Chorkor and Keta were involved in the fishing, handling and processing of the anchovies. Anchovies are harvested in August-September annually. This type of fish is usually marketed and consumed in smoked and sun-dried forms in Ghana. The Chorkor-smoker is usually used for smoking anchovies and can also be dried in the open to allow adequate air for complete drying. This type of fish is seasonal and therefore should be stored properly to avoid losses and also help improve food security in Africa. The application of gamma irradiation as an advanced technology can also help improve on the storability of this fish for consumption during the off-peak season.

5.2 LABORATORY ANALYSIS

5.2.1 Microbial Load Analysis

All anchovy samples collected from both processors and marketers in Chorkor and Keta recorded total viable count, total coliform count and counts of *Staphylococcus aureus*.

From table 4.2, it was observed that the unirradiated smoked anchovies from Chorkor had higher contamination with TVC (6.175 CFU/g) than TCC (3.645 CFU/g) and SA (3.318 CFU/g) being the least contaminated. The same was also observed for sun-dried samples (5.633, 3.487 and 2.575 CFU/g respectively) for TVC, TCC and SA.

These results indicate that the smoked samples were more contaminated than the sundried possibly because the sun drying produces effective and even drying of the fish than the smoked samples. The quality and freshness of fish are known to rapidly deteriorate through microbial and biochemical mechanism and therefore, with thorough drying, microbes will reduce (Al-Jasser and Al-Jasass, 2014).

The general decrease of TVC, TCC and SA in unirradiated samples when stored may be attributed to the effective packaging of the anchovies which created an environment that was not conducive for the microbes to grow or multiply. The absence of oxygen would generally decrease multiplication of microbes even though initial oxidation may lead to rancid taste and off flavor and development of many different compounds from which some have even adverse effects to human health. After storing the unirradiated smoked fish for five weeks, attack by insects was evidenced (Bari *et al.*, 2000).

On irradiating the anchovies at various radiation doses of control, 2.5, 5.0, 7.5 and 10 kGy, it was also observed from table 4.2, that contamination for smoked and sun-dried anchovies reduce with increasing radiation dose applied. For example, the contamination of smoked anchovies reduced from 4.502 CFU/g (control) to 1.158 CFU/g for TVC whiles the same trend was observed for the sun-dried samples which reduced from 3.393 CFU/g to 1.607 CFU/g when a dose of 5.0 kGy was applied. In all cases, it was found out that the contamination falls below detectable levels beyond 5.0 kGy.

However, from the present study, the results showed that contamination after being irradiated at 2.5 kGy was found out to be below the guidelines set by the Ghana Standard Authority: (Total heterotrophic bacteria count: 1 x 10^6 CFU/g; Total coliform count: 1 x 10^4 CFU/g; *Bacillus cereus* count: 1 x 10^4 CFU/g;), which implies that irradiation to a dose of 2.5kGy is enough to decontaminate the processed anchovies to meet standards set by the GSA. Mahin *et al*. (2011) stated that high radiation doses of 2.5 and 5.0 kGy reduced TVC by 3 logarithmic cycles for mola (*Amblypharyngodon mola*) at -20°C for 6 months.

5.2.2 Nutritional Quality Analysis

For the nutritional qualities of the anchovies, the investigation was to establish whether the irradiation of the anchovies after reducing microbial load in samples would affect the nutritional composition of the fish. In all cases the p-values of the test (0.999, 0.648, 0.480 and 0.775) were greater than the level of significance. This indicates that the total ash content of the irradiated samples are not significantly different ($p > 0.05$) from the ash content in unirradiated samples.

The percentage (%) total ash content of unirradiated smoked and sun-dried anchovies obtained from both locations also revealed that sun-dried samples (17.38%) indicated no significant ($p>0.05$) difference in total ash content based on the processing methods used when compared to smoked samples (16.39%). Again, similar trend was seen with samples from Chorkor and Keta and this was not affected by the processing method. This could be related to Mackerel irradiated at 1 to 45 kGy and kept in plastic bags at -22 °C. It was found out that these fish do not display any changes in amino acids, digestibility, biological values and protein use. While irradiated shrimps at 2 to 45 kGy, stored at different temperatures and level of humidity, the shrimps were found out to have lost a small amount of tryptophan (Asli, 2007).

Free fatty acid (FFA), a tertiary product of rancidity, increased during storage. The FFA is a measure of hydrolytic rancidity or the extent of lipid hydrolysis by lipase action. In most fish oils, rancidity is noticeable when the FFA (calculated as oleic acid) is found between 0.5-1.5percent (Eyo, 1993). The findings corroborate with this study in that, percentage (%) FFA amounts recorded in unirradiated smoked and sun-dried anchovies had generally undergone lipase action.

Unirradiated smoked and sun-dried samples recorded some amount of Oleic acid but reduced gradually in storage up to the ninth week. Oleic acid in smoked samples (0.69%) and sun-dried samples (1.03%) reduced after irradiation in smoked samples but increased in sun-dried samples as the storage period increases.

5.3 SHELF-LIFE STUDIES

For shelf-life studies, it was observed that samples obtained from Keta stored better than those from Chorkor. Even without irradiating, sun-dried samples stored up to a period of 7 weeks without pest damage. Irradiation doses of 2.5, 5.0, 7.5 and 10 kGy when stored for 9 weeks showed no signs of pest infestation. This indicated that, even at a dose of 2.5 kGy, the samples would be able to store at a period of 9 weeks and even beyond without insects or pest damage. Both irradiated smoked and sun-dried samples could possibly store beyond the 9 weeks duration. It could also be concluded that, 2.5 kGy dose applied to smoked and sun-dried anchovies is enough to eliminate microbes and extend the shelflife of the product.

In the light of the findings, it is not sufficient to give concern to food safety related factors alone which are usually given accentuated attentions by health personnel, the process through which the ingredients are subjected should be investigated and only the process that assured that important nutritional components are spared should be used.

Based on the present study, consumption of sun-dried anchovies offers dietary advantages for prevention of allergy associated with consumption of seafood products. Sun-dried anchovies would also find favor in the prevention of many human diseases including cancer, rheumatoid arthritis, atherosclerosis, stroke, neurodegeneration, and diabetes as concluded by Fang *et al.* (2002).

From the study, generally, both smoked and sun-dried anchovies obtained from processors and marketers in Keta are more hygienic than samples from Chorkor. This confirms the assertion of Kombat *et al.* (2013) that unhygienic environment in which fish are caught and processed have influence on the contamination level in the fish. Poor handling of anchovies found in Chorkor may also be a factor with respect to high microbial load. This unhygienic environment may be linked to the conditions under which these harvested anchovies are handled and treated before sale; the fish may be likely contaminated with pathogens. The release of fecal substances into waters is of much concern to public health. It can lead to serious health consequences.

The complex concept of fish quality consists of safety, nutritional value, availability, eating quality and product size. The most serious problem related to this product safety is the contamination with microbial pathogens. The contaminated samples were attributed to poor processing, packaging, transporting and storage conditions used by the fish mongers and marketers. The study revealed that, anchovy samples obtained from processors and marketers showed no significant difference in terms of handling and processing but some level of contamination was detected in samples.

Spoilage due to microbial activity is the main limitation of the shelf life of refrigerated fish. The spoilage development in fish is due to a combination of chemical, autolytic and microbiological changes, but the rate of spoilage could be reduced by taking into consideration different preservative measures like the use of refrigeration to extend the storage or shelf life of the fish (Oramadike *et al*., 2010).

It is known that the rate of fish spoilage depends on handling during processing, acidity level, species of fish, weather, mode of storage and temperature during transportation

(Clucas, 1982). Chemical breakdown of protein, fat and water contents contribute to quick spoilage of fish.

Since improper smoking and drying of fishes may lead to insect infestation, fungal attack, fragmentation and degradation of the product as asserted by Eyo (1993), it is important that both the artisanal fishermen and marketers adopt better methods of fish preservation. Better smoking kilns should be provided for artisanal fishermen at subsidized prices and fish product should be well stored.

In addition, continuous education of fish traders to use general good management practices and regular hygienic inspections by the standards authority is required to improve the microbial quality of processed fish in local areas.

Also, it is difficult to quantify the consumption of spoilt fish and its effect on health. Changes in chemical composition that occurs when a fish dies leads to short shelf life before processing. Smoking and sun-drying enhance flavor of the fish. However, spoilage still takes place in smoked and sun-dried fish when stored.

6.0 CONCLUSION AND RECOMMENDATION

6.1 CONCLUSION

The study was carried out to assess the microbial load and nutritional composition of unirradiated and irradiated *Engraulis encrasicolus* obtained from both processors and marketers in Chorkor and Keta. Unirradiated anchovies from both locations had some level of contamination which was above the threshold of the Ghana Standards Authority and therefore not wholesome for human consumption. The application of gamma irradiation reduced the level of contamination in the anchovies thus, making it safe for consumption

and also could be preserved for a longer period for future use. At a dose rate of 2.5kGy, the effectiveness of the irradiation was able to eliminate harmful microbes in the fish. The use of irradiation was also able to keep the fish samples for the duration of study. After the 9weeks of storage period of *Engraulis encrasicolus*, it was found out that sun-dried samples could be consumed better than the smoked one.

It has been established that irradiation of smoked and sun-dried anchovies *(Engraulis encrasicolus)* decontaminates the fish without altering or negatively affecting its nutritional qualities and hence, extends their shelf-life or storability. Irradiation is a safe and effective method of food preservation used in many countries in all over the world.

6.2 RECOMMENDATION

Based on the results of this study, it is therefore recommended that:

1. irradiation could be extended other types of fish
2. since anchovies is a seasonal fish, other seasonal type of fish could be irradiated for all year round availability.

In conclusion, irrespective of the location it is necessary to encourage the use of irradiation to provide safe fish and its products in hand with proper handling, processing and post-processing to ensure that hygienic and nutritious food reaches the final consumer.

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APPENDIX A

A questionnaire / survey designed for fisher folks in Chorkor in the Greater Accra Region and Keta in the Volta Region

QUESTIONNAIRE

A. BASIC DATA

NAME: ……

AGE: ……YEARS

GENDER: MALE☐ FEMALE☐

OCCUPATION: FISHERMAN ☐ FISHMONGER / PROCESSOR ☐

MARKETER/ SELLER ☐

HOME ADDRESS: ……

CONTACT DETAILS/ PHONE NUMBER: ……

B. FISHING ACTIVITY (To be completed by FISHERMEN only)

1. Which days do you go on fishing?

   ……

2. At what time specifically do you go for fishing? Morning☐ Afternoon☐ Evening☐
3. Which materials do you use for harvesting the fish (anchovies)?

   ……

4. Is this type of fish seasonal? YES / NO

   If YES, what is its harvesting peak season? ……

5. Are they harvested with other fishes? YES / NO

C. HANDLING & PROCESSING OPERATIONS

1. Is the fish sold immediately after harvesting? YES / NO
2. Are the fish sorted and graded after harvested? YES / NO
3. Who does the sorting and grading of the fish before sale? Fishermen☐

   Fishmongers☐
4. Do you clean the fish before selling them ashore? YES / NO
5. How is the fish processed for distant sale? .....

   .....
6. How is the fish (anchovies) dried? .....

   .....
7. How long does it takes for the fish to dry completely? .....

   .....
8. Do you incur losses during drying of the fish? YES / NO
9. How do you smoke the fish (anchovies)? .....

   .....
10. Do you incur losses during smoking of the fish? YES / NO
11. How long does the smoking process takes place? .....

.....
12. How long can the anchovies stay after processing prior to consumption? .....

    .....
13. Which other means do you process the fish into? .....

    .....
14. Are you given any form of education on the handling and processing of fish? YES / NO

15. If YES, how often do you get such education and by whom? ……..

……..

D. PACKAGING & MARKETING OPERATIONS

1. Which packaging material is used for transporting the anchovies to the market?

   Baskets☐ Boxes☐ Polythene bags☐ Sacks☐

   Other (*specify*)……

2. Which packaging material is commonly used for selling the processed anchovies on the

   market? Metal cans☐ Paper☐ Polythene bags☐

   Other (*specify*)……

3. How is the processed fish stored? ……..

   ……

4. What is the average demand for dried anchovies on the local market? High☐

   Moderate☐ Low☐

5. Do you export the anchovies to other countries? YES/ NO

6. What is the demand of anchovies on the international market? High ☐ Moderate ☐

   Low ☐

7. Do consumers/ buyers complain on the fish sold to them? YES / NO
8. If YES, what are the complains?

……

APPENDIX B (i)

Table 2.1: World fisheries and aquaculture production and utilization

PRODUCTION (Million tons)	**2006**	**2007**	**2008**	**2009**	**2010**	**2011**
Capture						
Inland	9.8	10.0	10.2	10.4	11.2	11.5
Marine	80.2	80.4	79.5	79.2	77.4	78.9
Total Capture	90.0	90.3	89.7	89.6	88.6	90.4
Aquaculture						
Inland	31.3	33.4	36.0	38.1	41.7	44.3
Marine	16.0	16.6	16.9	17.6	18.1	19.3
Total Aquaculture	47.3	49.9	52.9	55.7	59.9	63.6
Total World Fisheries	**137.3**	**140.2**	**142.6**	**145.3**	**148.5**	**154.0**
UTILIZATION						
Human consumption	114.3	117.3	119.7	123.6	128.3	130.8

Non-food uses	23.0	23.0	22.9	21.8	20.2	23.2
Population (billions)	6.6	6.7	6.7	6.8	6.9	7.0
Per capita food fish supply (kg)	17.4	17.6	17.8	18.1	18.6	18.8

Source: The State of World Fisheries and Aquaculture 2012. Data for 2011 are provisional estimates.

APPENDIX B (ii)

Table 2.2: Fish Production Figures for Marine, Inland and Aquaculture Sectors (20052009)

1	**MARINE**	**2005**	**2006**	**2007**	**2008**	**2009**
	Canoes	218,871.9	231,680.6	187,088.1	254,133.5	226,755.3
	Inshore Vessels	7,591.3	9,877.2	10,008.7	6,140.3	12,047.7
	Industrial Trawlers	12,494.0	17,419.1	19,892.8	18,289.3	20,836.7
	Paired Trawlers	1,163.5	1,090.4	1,217.9	1,181.1	0.0
	Shrimp Vessels	443.0	299.4	143.0	123.7	0.0
	Tuna Vessels	82,225.9	63,252.4	72,355.0	64,093.9	66,470.0
	TOTAL MFP	322,789.6	323,619.1	290,705.5	343,961.8	326,109.7
2	**INLAND**					
	Volta Lake	74,500	74,500	74,500	74,500	74,500
	Rivers & Dams	7,000	7,000	7,000	7,000	5,826

	Reservoirs & Ponds	1,154	1,668	3,256.7	5,595.7	1,377.4
	TOTAL IFP	82,654	83,168	84,757	87,095.7	81,703.4
3	**TOTAL DOM. CATCH**	405,443.5	406,786.6	375,462.2	431,057.5	407,813.1
4	**EXPORTS**					
	Tuna	59,892.2	43,340.7	54,989.8	48,070.4	41,211.4
	Fish	2,519.1	1,943.5	1,932.4	8,107.1	30,389.5
	Shrimp	36.4	62.6	25.7	23.7	0.0
	TOTAL EXPORTS	62,447.7	45,346.8	56,947.9	56,201.2	71,600.9
5	**FISH IMPORTS**	166,003.1	165,559.7	212,945.4	191,656.50	182,400.00
6	**TUNA SOLD LOCALLY**	22,333.7	19,911.7	69,769.3	16,023.50	25,258.60
7	**FISH SUPPLY /CONSUMPTION**	508,998.9	526,999.5	531,459.7	566,512.8	518,612.2
8	**POPULATION (M)**	21.6	22.1	22.7	23.3	23.9
9	**REQUIREMENTS (MT)**	862,400	884,800	907,600	931,198	956,000
10	**% ACHIEVED**	59.0	59.6	58.6	60.8	54.2
11	**PER CAPITA CONSUMPT'N (KG)**	23.6	23.8	23.4	24.3	21.7

Source- ***mofa.gov.gh/site/?page_id=2862***

Table 2.3: Production Figures for Imports, Exports and Consumption of Fish (2009-2011)

MARINE	**2009**	**2010**	**2011**
Canoes	226,755.3	213,000.00	209,200.28
Inshore Vessels	12,047.7	9,823.30	9575.99
Industrial Trawlers	20,836.7	18,859.30	21,596.90
Paired Trawlers	0.0	0.0	0
Shrimp Vessels	0.0	0	0
Tuna Vessels	66,470.0	77,875.50	86771.6
TOTAL MFP	326,109.7	319,558.1	327,144.8
INLAND			
Volta Lake	74,500.0	83,127	95,353.30
Ponds, Cages & Pens	5,826.0	10,200	19,091.97
Reservoirs, Dugouts & Dams	1,377.4	0	0
TOTAL IFP	81,703.4	93,327	114,445
TOTAL DOM. CATCH	407,813.1	412,884.7	441,590.0
EXPORTS			
Tuna	41,211.4	46,725.30	
Fish	30,389.5	15,724.70	
Shrimp	0.0	0	0
TOTAL EXPORTS	71,600.9	62,450.0	44,144.80

FISH IMPORTS	182,400.0	199,798.40	191,428.90
TUNA SOLD LOCALLY	25,258.6	31,150.20	
FISH SUPPLY /CONSUMPTION	518,612.2	550,233.1	588,874.1
POPULATION (M)	23.9	24.2	24.8
REQUIREMENTS (MT)	956,000.0	968,000.0	992,000.0
% ACHIEVED	54.2	56.8	59.4
PER CAPITA CONSUMPT'N (KG)	21.7	22.7	23.7

Source- ***http://mofa.gov.gh/site/?page_id=2862***

APPENDIX C

Results obtained from Chorkor

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Microbial_load	2	876.8	438.4	3.01	0.050
Processing_methods	1	156.1	156.1	1.07	0.301
Microbial_load.Processing_methods	2	270.5	135.2	0.93	0.396
Residual	426	62091.4	145.8		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Microbial_load		Processing_methods		
rep.	144	216	72	426	426	426
l.s.d.		2.797		2.283		3.955

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Source	1	72.3	72.3	0.49	0.483
Microbial_load	2	876.8	438.4	2.99	0.052
Processing_methods	1	86.3	86.3	0.59	0.444
Source.Microbial_load	2	86.7	43.4	0.30	0.744
Microbial_load.Processing_methods	2	189.3	94.7	0.64	0.525
Residual	423	62083.4	146.8		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Source	Microbial_load	Processing_methods	Source	Microbial_load			
rep.	unequal	144	216	unequal	d.f.	423	423	423	423
l.s.d.					5.613	min.rep		
	2.646	2.806	2.806	4.583	max-min			
					3.241	max.rep		

Table	Microbial_load	Processing_methods
rep.	72	l.s.d. 4.437
d.f.	423	

Except when comparing means with the same level(s) of

Microbial_load 4.861 d.f. 423

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Microbial_load	2	876.8	438.4	3.01	0.050
Processing_methods	1	156.1	156.1	1.07	0.301
Microbial_load.Processing_methods	2	270.5	135.2	0.93	0.396
Residual	426	62091.4	145.8		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Microbial_load	Processing_methods	Microbial_load	Processing_methods
rep.	144	216	72	
d.f.	426	426	426	
l.s.d.	2.797	2.283	3.955	

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	700.1	350.0	2.42	0.091
Microbial_load	2	876.8	438.4	3.03	0.050
Processing_methods	1	156.1	156.1	1.08	0.300
Dose_kGy.Microbial_load	4	527.9	132.0	0.91	0.457
Dose_kGy.Processing_methods	2	307.5	153.8	1.06	0.347
Microbial_load.Processing_methods	2	270.5	135.2	0.93	0.394
Dose_kGy.Microbial_load.Processing_methods	4	581.2	145.3	1.00	0.406
Residual	414	59974.6	144.9		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Dose_kGy Microbial_load Processing_methods		Dose_kGy Microbial_load	
rep.	144	144	216	48
d.f.	414	414	414	414
l.s.d.	2.788	2.788	2.277	4.829

Table Dose_kGy Microbial_load Processing_methods

	Dose_kGy Processing_methods	Processing_methods Microbial_load	Processing_methods
rep.	72	72	24
d.f.	414	414	414
l.s.d.	3.943	3.943	6.830

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Source	1	72.3	72.3	0.49	0.485
Dose_kGy	2	700.1	350.0	2.36	0.095
Microbial_load	2	876.8	438.4	2.96	0.053
Processing_methods	1	86.3	86.3	0.58	0.446
Source.Dose_kGy	2	99.1	49.6	0.33	0.716
Source.Microbial_load	2	86.7	43.4	0.29	0.746
Dose_kGy.Microbial_load	4	527.9	132.0	0.89	0.469

Dose_kGy.Processing_methods

	2	209.0	104.5	0.71	0.494
Microbial_load.Processing_methods	2	189.3	94.7	0.64	0.528
Source.Dose_kGy.Microbial_load	4	187.7	46.9	0.32	0.867
Dose_kGy.Microbial_load.Processing_methods	4	394.8	98.7	0.67	0.615
Residual	405	59964.7	148.1		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Source	Dose_kGy	Microbial_load	Processing_methods			
rep.	unequal	144	144	216	d.f.	405	405	405	405
l.s.d.	2.658	2.819	2.819	2.819					

Table Source Source Dose_kGy Dose_kGy Dose_kGy Microbial_load Microbial_load Processing_methods

rep.	unequal	unequal	48	72
l.s.d.	5.638	5.638		min.rep
d.f.	405	405		
l.s.d.	4.603	4.603	4.883	4.457 max-min
d.f.	405	405	405	405
l.s.d.	3.255	3.255		max.rep
d.f.	405	405		

Except when comparing means with the same level(s) of

Dose_kGy	4.883	d.f.	405

Table Microbial_load Source Dose_kGy Processing_methods

Dose_kGy Microbial_load

Microbial_load
Processing_methods

rep.	72	unequal	24	
l.s.d.		9.765		min.rep
d.f.		405		
l.s.d.	4.457	7.973	7.720	max-min
d.f.	405	405	405	
l.s.d.		5.638		max.rep
d.f.		405		

Except when comparing means with the same level(s) of

Microbial_load	4.883	
d.f.	405	
Dose_kGy.Microbial_load		8.457
d.f.		405

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	700.1	350.0	2.42	0.091
Microbial_load	2	876.8	438.4	3.03	0.050
Processing_methods	1	156.1	156.1	1.08	0.300
Dose_kGy.Microbial_load	4	527.9	132.0	0.91	0.457
Dose_kGy.Processing_methods	2	307.5	153.8	1.06	0.347
Microbial_load.Processing_methods	2	270.5	135.2	0.93	0.394
Dose_kGy.Microbial_load.Processing_methods	4	581.2	145.3	1.00	0.406

Residual	414	59974.6	144.9	
Total	431	63394.8		

Least significant differences of means (5% level)

Table Dose_kGy Microbial_load Processing_methods Dose_kGy Microbial_load

rep.	144	144	216	48
d.f.	414	414	414	414
l.s.d.	2.788	2.788	2.277	4.829

Table Dose_kGy Microbial_load Dose_kGy Processing_methods Processing_methods Microbial_load Processing_methods

rep.	72	72	24
l.s.d.	3.943	3.943	6.830
d.f.	414	414	414

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	700.1	350.0	2.40	0.092
Storage_period	3	485.9	162.0	1.11	0.344
Processing_methods	1	156.1	156.1	1.07	0.301
Dose_kGy.Storage_period	6	1045.0	174.2	1.20	0.308
Dose_kGy.Processing_methods	2	307.5	153.8	1.06	0.349

Storage_period.Processing_methods	3	361.8	120.6	0.83	0.479
Dose_kGy.Storage_period.Processing_methods	6	895.6	149.3	1.02	0.409
Residual	408	59442.8	145.7		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Dose_kGy	Storage_period	Processing_methods	Dose_kGy Storage_period
rep.	144	108	216	36
d.f.	408	408	408	408
l.s.d.	2.796	3.229	2.283	5.593

Table	Dose_kGy	Storage_period	Dose_kGy Processing_methods	Processing_methods Storage_period	Processing_methods
rep.	72	54	18		
d.f.	408	408	408		
l.s.d.	3.955	4.566	7.909		

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Source	1	72.3	72.3	0.48	0.488
Dose_kGy	2	700.1	350.0	2.33	0.098

Storage_period	3	485.9	162.0	1.08	0.358
Processing_methods	1	86.3	86.3	0.57	0.449
Source.Dose_kGy	2	99.1	49.6	0.33	0.719
Source.Storage_period	3	123.8	41.3	0.27	0.844
Dose_kGy.Storage_period	6	1045.0	174.2	1.16	0.327
Dose_kGy.Processing_methods	2	209.0	104.5	0.70	0.499
Storage_period.Processing_methods	3	239.3	79.8	0.53	0.661
Source.Dose_kGy.Storage_period	6	284.0	47.3	0.32	0.929
Dose_kGy.Storage_period.Processing_methods	6	616.2	102.7	0.68	0.662
Residual	396	59433.8	150.1		
Total	431	63394.8			

Least significant differences of means (5% level)

Table	Source	Dose_kGy	Storage_period	Processing_methods
rep.	unequal	144	108	216
d.f.	396	396	396	396
l.s.d.	2.676	2.838	3.278	2.838

Table	Source	Source	Dose_kGy	Dose_kGy
	Dose_kGy	Storage_period		
	Storage_period		Processing_methods	
rep.	unequal	unequal	36	72
l.s.d.	5.677	6.555		min.rep
d.f.	396	396		
l.s.d.	4.635	5.352	5.677	4.488 max-min
d.f.	396	396	396	396
l.s.d.	3.278	3.785		max.rep
d.f.	396	396		

Except when comparing means with the same level(s) of Dose_kGy 4.916 d.f. 396

Table Storage_period Source Dose_kGy

Processing_methods
Dose_kGy Storage_period

Storage_period
Processing_methods rep. 54 unequal 18

l.s.d.		11.354		min.rep
d.f.		396		
l.s.d.	5.182	9.270	8.976	max-min
d.f.	396	396	396	
l.s.d.		6.555		max.rep
d.f.		396		

Except when comparing means with the same level(s) of

Storage_period	5.677	d.f.	396
Dose_kGy.Storage_period		9.833	
d.f.	396		

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	700.1	350.0	2.40	0.092
Storage_period	3	485.9	162.0	1.11	0.344
Processing_methods	1	156.1	156.1	1.07	0.301
Dose_kGy.Storage_period	6	1045.0	174.2	1.20	0.308
Dose_kGy.Processing_methods	2	307.5	153.8	1.06	0.349
Storage_period.Processing_methods	3	361.8	120.6	0.83	0.479
Dose_kGy.Storage_period.Processing_methods	6	895.6	149.3	1.02	0.409

Residual	408	59442.8	145.7	
Total	431	63394.8		

Least significant differences of means (5% level)

Table	Dose_kGy	Storage_period	Processing_methods	Dose_kGy Storage_period
rep.	144	108	216	36
d.f.	408	408	408	408
l.s.d.	2.796	3.229	2.283	5.593

Table	Dose_kGy	Storage_period	Dose_kGy Processing_methods	Processing_methods Storage_period	Processing_methods
rep.	72	54	18		
l.s.d.	3.955	4.566	7.909		
d.f.	408	408	408		

Results obtained from Keta

Analysis of variance Variate:

CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.

Microbial_load	2	102.544	51.272	33.57	<.001
Processing_methods	1	7.758	7.758	5.08	0.025
Microbial_load.Processing_methods	2	1.934	0.967	0.63	0.531
Residual	426	650.573	1.527		
Total	431	762.808			

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Microbial_load	2	102.544	51.272	33.57	<.001
Processing_methods	1	7.758	7.758	5.08	0.025
Microbial_load.Processing_methods	2	1.934	0.967	0.63	0.531
Residual	426	650.573	1.527		
Total	431	762.808			

Least significant differences of means (5% level)

Table	Microbial_load	Processing_methods	Microbial_load	Processing_methods
rep.	144	216	72	
d.f.	426	426	426	
l.s.d.	0.2863	0.2337	0.4048	

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Source	1	2.328	2.328	1.56	0.213
Microbial_load	2	102.544	51.272	34.32	<.001
Processing_methods	1	20.161	20.161	13.50	<.001
Source.Microbial_load	2	0.984	0.492	0.33	0.720

Microbial_load.Processing_methods	2	4.935	2.467	1.65	0.193
Residual	423	631.855	1.494		
Total	431	762.808			

Least significant differences of means (5% level)

Table	Source Microbial_load Processing_methods				Source Microbial_load					
rep.	unequal	144	216	unequal	d.f.	423	423	423	423	
l.s.d.					0.5662	min.rep				
	0.2669	0.2831	0.2831	0.4623	max-min					
				0.3269	max.rep					

Table	Microbial_load	Processing_methods	
rep.	72	l.s.d.	0.4476
d.f.	423		

Except when comparing means with the same level(s) of

Microbial_load	0.4904	d.f.	423

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Microbial_load	2	102.544	51.272	33.57	<.001
Processing_methods	1	7.758	7.758	5.08	0.025
Microbial_load.Processing_methods	2	1.934	0.967	0.63	0.531
Residual	426	650.573	1.527		

Total	431	762.808

Least significant differences of means (5% level)

Table	Dose_kGy Microbial_load	Processing_methods Dose_kGy	Microbial_load	
rep.	144	144	216	48
d.f.	414	414	414	414
l.s.d.	0.1842	0.1842	0.1504	0.3191

Table	Dose_kGy Microbial_load	Dose_kGy Processing_methods	Processing_methods Microbial_load	Processing_methods
rep.	72	72	24	
d.f.	414	414	414	
l.s.d.	0.2605	0.2605	0.4513	

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Source	1	2.3282	2.3282	3.91	0.049
Dose_kGy	2	361.8780	180.9390	303.84	<.001
Microbial_load	2	102.5444	51.2722	86.10	<.001
Processing_methods	1	20.1612	20.1612	33.86	<.001

Source.Dose_kGy	2	0.9617	0.4808	0.81	0.447
Source.Microbial_load	2	0.9841	0.4921	0.83	0.438
Dose_kGy.Microbial_load	4	8.3947	2.0987	3.52	0.008
Dose_kGy.Processing_methods	2	3.0599	1.5300	2.57	0.078
Microbial_load.Processing_methods	2	4.9349	2.4675	4.14	0.017
Source.Dose_kGy.Microbial_load	4	6.2790	1.5697	2.64	0.034
Dose_kGy.Microbial_load.Processing_methods	4	10.1041	2.5260	4.24	0.002
Residual	405	241.1779	0.5955		
Total	431	762.8080			

Least significant differences of means (5% level)

Table	Source	Dose_kGy	Microbial_load		Processing_methods			
rep.	unequal	144	144	216	d.f.	405	405	405	405
l.s.d.	0.1686	0.1788	0.1788	0.1788	Table	Source	Source	Dose_kGy	Dose_kGy
		Dose_kGy	Microbial_load			Microbial_load			
		Processing_methods	rep.	unequal	unequal	48	72		
l.s.d.		0.3576	0.3576				min.rep		
d.f.		405	405						
l.s.d.		0.2919	0.2919	0.3097	0.2827	max-min			
d.f.		405	405	405	405				
l.s.d.		0.2064	0.2064			max.rep			
d.f.		405	405						

Except when comparing means with the same level(s) of

Dose_kGy	0.3097	d.f.		405

Table Microbial_load Source Dose_kGy

Processing_methods
Dose_kGy Microbial_load

Microbial_load
Processing_methods rep. 72 unequal 24

l.s.d.		0.6193		min.rep
d.f.		405		
l.s.d.	0.2827	0.5057	0.4896	max-min
d.f.	405	405	405	
l.s.d.		0.3576		max.rep
d.f.		405		

Except when comparing means with the same level(s) of

Microbial_load	0.3097	d.f.	405

Dose_kGy.Microbial_load	0.5363		
d.f.	405		

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	361.8780	180.9390	286.11	<.001
Microbial_load	2	102.5444	51.2722	81.07	<.001
Processing_methods	1	7.7575	7.7575	12.27	<.001
Dose_kGy.Microbial_load	4	8.3947	2.0987	3.32	0.011
Dose_kGy.Processing_methods	2	3.4543	1.7271	2.73	0.066
Microbial_load.Processing_methods					

	2	1.9336	0.9668	1.53	0.218
Dose_kGy.Microbial_load.Processing_methods	4	15.0245	3.7561	5.94	<.001
Residual	414	261.8210	0.6324		
Total	431	762.8080			

Least significant differences of means (5% level)

Table	Dose_kGy	Microbial_load	Processing_methods	Dose_kGy Microbial_load
rep.	144	144	216	48
d.f.	414	414	414	414
l.s.d.	0.1842	0.1842	0.1504	0.3191

Table	Dose_kGy	Microbial_load	Dose_kGy Processing_methods	Processing_methods Microbial_load	Processing_methods
rep.	72	72	24		
l.s.d.	0.2605	0.2605	0.4513		
d.f.	414	414	414		

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	361.8780	180.9390	301.24	<.001
Storage_period	3	116.1514	38.7171	64.46	<.001
Processing_methods	1	7.7575	7.7575	12.92	<.001
Dose_kGy.Storage_period	6	11.9255	1.9876	3.31	0.003

Dose_kGy.Processing_methods	2	3.4543	1.7271	2.88	0.058
Storage_period.Processing_methods	3	13.7104	4.5701	7.61	<.001
Dose_kGy.Storage_period.Processing_methods	6	2.8634	0.4772	0.79	0.575
Residual	408	245.0675	0.6007		
Total	431	762.8080			

Least significant differences of means (5% level)

Table Dose_kGy Storage_period Processing_methods Dose_kGy Storage_period

rep.	144	108	216	36
d.f.	408	408	408	408
l.s.d.	0.1795	0.2073	0.1466	0.3591

Table Dose_kGy Storage_period Dose_kGy Processing_methods Processing_methods Storage_period Processing_methods

rep.	72	54	18
d.f.	408	408	408
l.s.d.	0.2539	0.2932	0.5078

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Source	1	2.3282	2.3282	4.10	0.043
Dose_kGy	2	361.8780	180.9390	318.91	<.001
Storage_period	3	116.1514	38.7171	68.24	<.001
Processing_methods	1	20.1612	20.1612	35.53	<.001
Source.Dose_kGy	2	0.9617	0.4808	0.85	0.429

Source.Storage_period	3	2.7262	0.9087	1.60	0.188
Dose_kGy.Storage_period	6	11.9255	1.9876	3.50	0.002
Dose_kGy.Processing_methods	2	3.0599	1.5300	2.70	0.069
Storage_period.Processing_methods	3	12.1453	4.0484	7.14	<.001
Source.Dose_kGy.Storage_period	6	2.8347	0.4725	0.83	0.545
Dose_kGy.Storage_period.Processing_methods	6	3.9564	0.6594	1.16	0.326
Residual	396	224.6796	0.5674		
Total	431	762.8080			

Least significant differences of means (5% level)

Table	Source	Dose_kGy	Storage_period	Processing_methods
rep.	unequal	144	108	216
d.f.	396	396	396	396
l.s.d.	0.1645	0.1745	0.2015	0.1745

Table	Source	Source	Dose_kGy	Dose_kGy
	Dose_kGy	Storage_period		
	Storage_period		Processing_methods	
rep.	unequal	unequal	36	72
l.s.d.	0.3490	0.4030		min.rep
d.f.	396	396		
l.s.d.	0.2850	0.3291	0.3490	0.2759 max-min
d.f.	396	396	396	396
l.s.d.	0.2015	0.2327		max.rep
d.f.	396	396		

Except when comparing means with the same level(s) of

Dose_kGy	0.3023	d.f.	396

Table Storage_period Source Dose_kGy

Processing_methods
Dose_kGy Storage_period

Storage_period
Processing_methods rep. 54 unequal 18

l.s.d.		0.6981		min.rep
d.f.		396		
l.s.d.	0.3186	0.5700	0.5519	max-min
d.f.	396	396	396	
l.s.d.		0.4030		max.rep
d.f.		396		

Except when comparing means with the same level(s) of

Storage_period	0.3490		
d.f.	396		
Dose_kGy.Storage_period		0.6046	
d.f.		396	

Analysis of variance

Variate: CFU_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Dose_kGy	2	361.8780	180.9390	301.24	<.001
Storage_period	3	116.1514	38.7171	64.46	<.001
Processing_methods	1	7.7575	7.7575	12.92	<.001
Dose_kGy.Storage_period	6	11.9255	1.9876	3.31	0.003

Dose_kGy.Processing_methods

	2	3.4543	1.7271	2.88	0.058
Storage_period.Processing_methods	3	13.7104	4.5701	7.61	<.001
Dose_kGy.Storage_period.Processing_methods	6	2.8634	0.4772	0.79	0.575
Residual	408	245.0675	0.6007		
Total	431	762.8080			

Least significant differences of means (5% level)

Table Dose_kGy Storage_period Processing_methods Dose_kGy

Storage_period

rep.	144	108	216	36
d.f.	408	408	408	408
l.s.d.	0.1795	0.2073	0.1466	0.3591

Table Dose_kGy Storage_period Dose_kGy

Processing_methods Processing_methods Storage_period

Processing_methods		rep.	72	54
18	l.s.d.	0.2539	0.2932	0.5078
d.f.		408	408	408

APPENDIX D

Plate 1. Sun-dried anchovies

Plate 2: Smoked anchovies

Plate 3. Packaged anchovies obtained from Chorkor and Keta ready for storage

Plate 4. Packaged anchovies under storage