UNIVERSITY OF CAPE COAST

DECONTAMINATION OF POLYCYCLIC AROMATIC HYDROCARBONS, ORGANOCHLORINE PESTICIDES AND MICROBIAL LOAD IN PROCESSED FISH IN GHANA USING GAMMA IRRADIATION: IMPLICATIONS AND PROSPECTS

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BY

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Thesis submitted to the Department of Fisheries and Aquatic Sciences of the School of Biological Sciences, University of Cape Coast, in partial fulfilment of the requirements for the award of Doctor of Philosophy Degree in Integrated Coastal Zone Management

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DECLARATION

Candidate's Declaration

I hereby declare that this thesis is the result of my own original research and that no part of it has been presented for another degree in this University or elsewhere.

Candidate's Signature: ……..…… Date: ……..…

Name: Emmanuel Kwame Gasu

Supervisors' Declaration

We hereby declare that the preparation and presentation of the thesis were supervised in accordance with the guidelines on supervision of thesis laid down by the University of Cape Coast.

Principal Supervisor's Signature: …… Date: ……

Name: Professor Edward A. Obodai

Co-Supervisor's Signature: …… Date: ……

Name: Professor Denis W. Aheto

ABSTRACT

Polycyclic Aromatic Hydrocarbons (PAHs) and Organochlorine pesticides (OCPs) are among the most hazardous class of organic chemicals in the environment. Some PAHs and OCPs have been banned or restricted in several countries. The major sources of PAHs include incomplete burning of coal, oil, wood, gas and charcoal grilled meat and fish. Major sources of OCPs include food industries, agriculture and sewage wastes. Their discharge into water bodies and food result in extremely high concentrations which ultimately cause environmental and public health concerns. Due to their high persistence, toxicity and potential to bio-accumulate, their removal from salted, smoked and sundried fish through the use of gamma irradiation is imperative. In this study the levels of seven polycyclic aromatic hydrocarbons (PAHs) and eight pesticides including three organochlorine pesticides (OCPs) residues were investigated in six (6) selected frequently consumed salted, smoked and sun dried fish collected from nine markets in three coastal Regions of Southern Ghana. Gas chromatographic-mass spectrophotometer with limit of detection of 1.0 parts per billion was employed for the analysis of PAHs. Quantification of OCPs and synthetic pyrethroids was carried out before and after irradiation by Electron capture detector (ECD) and of organophosphorus by Pulse flame photoelectric detector (PFPD) with the limit of detection of 0.01parts per million (ppm) or 10 parts per billion. The safety and quality of the fish were analysed using standard methods of Association of Analytical Chemists (AOAC), 2000. Observed mean of PAHs levels in the non-irradiated smoked fish samples ranged from 11.75 – 39.37ug/kg, ($p \geq 0.05$) with B(b)F recording the highest of 39.37ug/kg, ($p \geq 0.05$). Non-irradiated samples of sun dried fish

recorded PAHs values ranged from 5.76 – 47.68ug/kg, ($p \geq 0.05$). Gamma irradiation degradation of PAHs in sun dried fish ranged between 0.32 – 29.73ug/kg ($p \geq 0.05$) at 7.5kGy with B(b)F degraded from 39.37 to 29.73ug/kg, ($p = 0.406$) and in smoked fish, 7.54 – 22.30ug/kg ($p \geq 0.05$) at 7.5kGy. B(b)F in smoked fish degraded from 33.55 – 22.30ug/kg ($p \geq 0.05$) at 7.5kGy. Pesticides and OCPs in the non-irradiated samples ranged from 0.1 - 93mg/kg and after gamma irradiation, the values reduced ranging from non-detction to 29.73mg/kg ($p = 0.001$) in sun dried fish samples. OCPs were reduced by gamma irradiation at 7.5kGy from 0.86 – 0.007mg/kg ($p = 0.001$) in Cyfluthrin and from 0.768mg/kg to non-detection ($p = 0.001$) at 7.5kGy in Permathrin. Proximate and microbial values of smoked fish ranged as follow: Percentage weight loss (1.07±0.41), percentage moisture loss (0.02±0.02 – 9.86±0.88), percentage FFA (6.31 ± 0.00 – 8.44 ± 0.78), percentage protein loss (0.03±0.00 - 0.10±0.01), pH (6.08±0.33 – 7.38 ± 0.15), TVC logcfu/g (2.78±1.45 – 4.79 ±1.45), and MYC logcfu/g (1.90 ±0.49 – 4.14 ± 0.49). Sun dried fish recorded the following values: percentage moisture loss (0.07±0.12 – 0.31±0.12), percentage weight loss (0.05±0.02 – 0.08±0.02), percentage protein loss (0.06 ± 0.08– 0.21 ± 0.08), percentage ash (10.13 ± 2.27 – 14.66 ± 2.27), percentage FFA (2.80 ±0.71 – 4.22 ± 0.71), percentage TTA (5.33 ± 0.62 – 6.57 ± 0.62), percentage pH (6.78 ± 0.1 – 6.98 ± 0.1), TVC logcfu/g (4.80 ± 0.16 – 5.12 ± 0.16) and MYC logcfu/g (4.39 ±0.45 – 5.28 ± 0.45). On basis of the above findings, this thesis attempts to provide an assessment on the potential of using gamma irradiation as a means to improve health and safety standards of salted, smoked and sun-dried marine and freshwater fishes. Recommendations proffered included increasing the irradiation dose to

maximum of 10kGy for food in further study for the reduction of PAHs and pesticides. Protocols to regulate and enhance quality and safety of smoked and sun dried fish will add to acceptance for the export of smoked and sun dried fish from Ghana. Pesticides residue levels must be set by the Ghana's Food and Drugs Authority to monitor the activities of fish processors in consonance with international standards. These are particularly worthy for the attention of policy makers, fish processors, Fisheries Commission of Ghana, NGOs and the general promotion of food safety in the country.

KEY WORDS

Ecotoxicity

Pyrethroid Pesticides

Organochlorine pesticides

Organophsphate pesticides

Polycyclic aromatic hydrocarbons

Residue levels

Gamma irradiation decontamination

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DEDICATION

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LIST OF ACRONYMS

ADA	American Dietetic Association
AMA	American Medical Association
AOAC	Association of Analytical Chemists
ATSDR	Agency for Toxic Substances and Disease Registry
CCP	Critical Control Point
CDC	Center for Disease Control
CGIAR	Consultative Group on International Agricultural Research
CGS	Codex General Standard
Cl	Confident limit
DFID	Department for International Development
DHHS	Department of Health and Human Services
E-beam	Electron beam
EFSA	European Food Safety Authority
EPA	Environmental Protection Agency
ERLs	Environmental Risk Limit
FAO	Food and Agriculture Organization
FDA	Food and Drug Administration
FFA	Free fatty acid
FGD	Focus group discussions
GAEC	Ghana Atomic Energy Commission
GHC	Ghana Cedi
GIPC	Ghana Investment Promotion Council
IAEA	International Atomic Energy Agency

IARC	International Agency for Research on Cancer
ICGFI	International Consultative Group on Food Irradiation
ICZM	Integrated Coastal Zone Management
JECFI	Joint Expert Committee of Food Irradiation
JHS	Junior High School
Kg	kilogram
kGy	Kilo gray
MPCs	Maximum Permissible Concentrations
MeV	Mega electron volt
MOFA	Ministry of Food and Aquaculture Development
MT	Metric tonnes
NASA	National Authority of Space Administration
NGOs	Non-governmental Organizations
NRA	Nuclear Regulatory Authority
OCP	Organochlorine pesticides
OPPs	Organophosphorus pesticides
PAHs	Polycyclic aromatic hydrocarbons
PCB	Polychlorinated biphenyls
PO	Participant Observations
PUFA	Polyunsaturated fatty acids
RPI	Radiation Protection Institute
S.E	Standard error
SFMP	Sustainable Fisheries Management Programme
SHS	Senior High School
TVC	Total viable count

TMC	Total mycology count
USAID	United States Agency for International Development
USEPA	United States Environmental Protection Agency
US$	United States Dollar
UV	Ultra violet
WHO	World Health Organization

CHAPTER ONE

1.0 INTRODUCTON

Large populations of the coastal dwellers in Ghana derive their livelihood from fishing and fisheries related occupations such as fish processing and distribution. These activities ensure income and food security to both the local people, the nation at large, and for export. Traditional methods such as, salting and sun drying, smoking, and drying have been used to preserve and extend the shelf life of fisheries products. However, technologies like ionizing radiation have not been introduced into the fish processing technologies mix to extend the shelf life of fisheries products in Ghana. The irradiation of food products is a physical treatment involving direct exposure of food products to electron or electromagnetic rays, for their long time preservation and improvement of quality and safety. Salted and sundried, smoked, and dried fishes were sampled randomly from coastal markets in Southern Ghana for the current research to determine the levels of polycyclic aromatic hydrocarbon (PAHs), orgarnochlorine pesticides (OCPs) and microbial load in processed marine and freshwater fishes in Ghana.

These contaminants like the PAHs and OCPs may occur naturally in the fresh fish while some are formed during value addition processing. These sources of PAHs and OCPs vary from the air, soil and water and could be deleterious to human health. Their concentrations in air or water is relatively small compared to soil, however, due to bioaccumulation and lipid solubility, it is readily absorbed from the digestive tract of mammals and are quickly disseminated in several tissues of the body and accumulates in body fat. Several remediation methods have been tried to remove PAHs and OCPs from

the environment. However, there is little information in the use of gamma irradiation technology to degrade PAHs and OCPs from food such as salted, smoked and sundried fishes.

1.1 Background

Polycyclic aromatic hydrocarbons (PAHs) and organochlorine pesticides (OCPs) are known groups of pervasive environmental contaminants and have contributed to the increasing prevalence of non-communicable diseases such as cancers (WHO, 2010). The occurrence of PAHs in processed fish is a concern because of their carcinogenicity and mutagenicity (Palm et al., 2011; Ki-Hyun Kim et al., 2013). PAHs are generated from different sources varying from those resulting from human activities and those from natural sources such as volcanic eruptions and forest fires (Hussein & Khaled, 2014; Abdel-Shafy et al., 2016), and partial burning or pyrolysis of organic matter (Samanta et al. 2002; Taylor et al., 2014).

Humans are regularly exposed to PAHs from ambient air generated from community and industrial waste incineration, coal and wood waste burning, or vehicle exhausts, indoor air emanating from smoking or cooking of foods and also from contaminated water (Ki-Hyun Kim et al., 2013; Khalil, Albachir, & Odeh, 2016). However, incomplete combustion of organic materials is the primary source of atmospheric contamination (Boström et al., 2002; FAO, 2004; Ki-Hyun Kim et al., 2013; Khalil, Albachir, & Odeh, 2016).

Man has been exposed to PAH through the processes of body or skin contact, active smoking and passive smoking, and through food such as

smoked, grilled, fried, broiled, and roasting of meat and fish and also through drying of foods (Boström et al., 2002; Zachara et al., 2017).

Abdel-Shafy et al., (2016), reported high levels of PAHs ranging from 200µg/kg in smoked fish and meat, 130µg/kg in barbecue and 0.01 µg/kg – 1.0µg/kg in an uncooked food.

Pesticides are substances or mixture of substances that are used for preventing, destroying or controlling pests such as insects, weeds, fungi, bacteria, and are specifically called insecticides, fungicides, bactericides, herbicides or rodenticides (USEPA, 2007; Jayaraj, Megha, & Sreedev, 2016a; Mel'nikova et al., 2017). Of great importance among them are organochlorine pesticides (OCPs) and organophophorus pesticides (OPPs) which belong to a class of chlorinated hydrocarbon derivatives and are collectively known as synthetic pesticides which are used extensively globally in chemical industries and in agriculture (Jayaraj, Megha, & Sreedev, 2016b).

Several kinds of pesticides are in use depending on the purpose of application. These may be selective, non-selective, foliage or soil-applied. Selective pesticides are used to control weeds without damaging desirable crops. Non-selective Pesticides are formulated for complete removal of vegetation and they kill most plants when applied on them. Foliage Pesticides are used on leaves and other weeds above ground. These serve as contact killing and only the parts of the plant actually sprayed are affected. Soil-applied Pesticides are those applied to the ground and are recommended for pre-plant, pre-emergence and post-emergence treatments of the fields (Hayes et al., 2006).

Intense use of pesticides (OCPs, OPPs and pyrethroids) contributes to bioaccumulation in food chains (IPCS, 2004; Report et al., 2009a) posing threats to human health and the environment (Guo et al., 2008; Afful et al., 2010; FAO/WHO, 2010). Despite the threats, these pesticides are beneficial in terms of their household usage for treating wood and wood products, for the control of insects, arachnids and other pests (Fianko et al., 2011; Jayaraj et al., 2016b). They are also beneficial in agriculture such as increasing food production, food quality as well as reducing the cost of production, the price of produce and ensuring food security (Hayes and Laws, 1991; Jayaraj et al., 2016b).

OCPs and pyrethroids like their counterparts PAH, have also been identified as highly toxic and bioaccumulate in the ecosystem as well as having slow degradability (Rani, Shanker, & Jassal, 2017). Even though they have been developed with the notion to eliminate target organisms, often, non-target organisms are affected severely by their application in the environment.

Most pesticides (OCPs, OPPs, and Pyrethroids) have been banned in developed countries but the use of these contaminants has been rising in underdeveloped countries (Rani et al., 2017). The use of these pesticides for high agricultural production has increased their pollution in the air, soil and water (Jayaraj, Megha, & Sreedev, 2016b). According to the World Health Organisation study, 80% of all pesticides produced worldwide are used by developing countries (Jayaraj, Megha, & Sreedev., 2016b). OCPs' exposure have been associated with the consumption of contaminated food including meat, diary, fish and marine animals (Fitzgerald et al., 2001; Hagmar et al., 2001, Mwevura et al., 2002, Bradman et al., 2007).

Studies have shown that only a small percentage (0.3%) of pesticides applied however, goes into the target pest while 99.7% go into the environment (Rengarajan et al., 2015).

The environmental pollution of these OCPs have reached an alarming state that Darko and Acquaah, (2008) have realised pesticide residues in dairy products, meat, fish, water and sediments. Ntow (2001), reported the presence of pesticides in human blood and breast milk. Pesticides have also been detected in fruits and vegetables (Hanson et al. 2007; Hussain et al., 2002; El-Nahhal, 2004).

The application of pesticides does not only take place while the crop is growing, but is applied in post-harvest applications to assist with transportation and storage. Although washing and peeling of food produce can reduce pesticide residues, the process cannot eliminate them completely. This is because the bulk of pesticides more especially the systemic pesticides are inside the food, groundwater and the air whereas other pesticides only affect the surface and can be washed away (IUPAC, 2006).

Pesticide residue, according to USEPA, refers to the amount of a pesticide or ingredients in the mixture found in or on a raw agricultural commodity or in a processed food including fish (USEPA, 2014; USEPA, 2018). This also includes residue of degradation products of the pesticide or by plant metabolism or some other degrading process. The residue of concern may be the parent compound, a metabolite of the parent compound or both (USEPA, 2018).

The use of pesticides is restricted by international standards to ensure the circulation of only food produce that have been treated with approved

pesticides and complies with the approved Minimum Residue Levels [MRLs] (Chan, 2000, USEPA, 2018). A zero tolerance means that no amount of the pesticide chemical may remain on the raw or processed food commodity when it is offered for consumption. A zero tolerance for a pesticide chemical may be established because a safe level of the pesticide chemical in the diet has not been reliably determined. When any chemical is carcinogenic, and has a physiological effect after it shows in the diet of animals, a zero tolerance is established (Global MRL Database 2018; USEPA, 2018).

There is a stringent monitoring of pesticide residues in export crops such as cocoa beans, but such strict monitoring programmes are absent in domestic food items including fish (Musa et al., 2010; ICCO, 2011). This may have been due to a lack of relevant technical, scientific and organizational capacity to carry out effective surveillance in general.

Pesticides have been in use for decades and will continue to be used extensively in agriculture production and in household protection however, there is no effective routine monitoring of food items to check the MRL of organochlorine pesticide residues in staple foods and in smoked and sun dried fish. However, pesticide chemical residue which is superficial is normally removed through good agricultural practices (GAP) such as washing, weathering or other changes in the chemical itself, prior to the consumption of the commodity. Exposure is due to consumers coming into close contact with these pesticides during mixing, packaging, spraying on farms, lawns and during house spraying (USEPA 2005; Luo et al., 2016).

1.2 Ecotoxicity of PAH and OCP

Acute toxicity occurs in birds and in aquatic life, however higher concentrations occur in fish and shellfish than in the surrounding environment and have drastic effect on aquatic organisms (Abdel-Shafy et al., 2016).

The levels of PAH in raw foods are minimal but are high in areas close to industries and along highways vegetables along these areas have higher concentrations of about ten percent more than are found in rural areas (Phillips, 1999; Report, 2009a).

The use of OCPs and pyrethroids is beneficial to both the consumer and the farmer (Pimentel et al., 2014). Notwithstanding, ensuring crop quality improvement also ensures abundant food supply that is safe from pest and available throughout the year and that promotes good health. Indeed, food and other agricultural produce are preserved from destruction during processing, storage, transporting or marketing (Zheng et al., 2016). Although the main focus is to harm only the targeted pests, unfortunately, pesticides have become part of the food chain of humans (Eskenazi et al., 2008; Damalas & Eleftherohorinos, 2011), the environment and the people who are exposed to them.

Pesticides are toxic (IPCS, 2009) and can degrade into other chemical compounds known as derivatives. Residues and metabolites of many OCPs are stable and having long half-lives in the environment (El-Mekkawi et al., 2009). Maximum limits for residues therefore were established for selected pesticides (Boström et al., 2002). The OCPs, OPPs and carbamates are critically toxic and carcinogenic species with half-lives of about 60 years (Boada et al. 2016). Due to their high persistence, toxicity and the potential to

bioaccumulate, it is highly imperative for their complete monitoring, regulation and removal once they contaminate the ecosystem.

1.3 Health Concern of PAH and OCP

Potential exposure of PAH and OCP have adverse health effect on multiples of human organs due to wide spreading in the environment and have been associated with the risk of cancers and cardiovascular diseases (Burstyn et al., 2005; IARC, 2010; Ki-Hyun Kim et al., 2013). Earlier studies have observed associations between PAH exposure with aging indicators such as telomere length (Abdel-Shafy et al., 2016), skin aging scores (Vierkötter et al. 2010), and with global or gene-specific DNA methylation alterations though the relation between PAH exposure and DNA methylation aging is still unclear (Herbstman et al., 2012).

PAH have been classified accordingly by the International Agency for Research on Cancer with respect to the health of humans (IARC, 2010). Human exposure can be considered from occupational hazards opinion assessment such as from fire fighting and from open fireplaces (Abdel-Shafy et al., 2016). For many people such as drivers, auto-mechanics, agriculture workers, mineworkers, "galamseyers", oil refinery workers and petroleum pump and lubricant attendants, exposure may occur regularly (Ki-Hyun Kim et al., 2013).

The numerous health problems associated with organochlorine pesticides include headaches and nausea, cancer, reproductive disorder, and endocrine disruption (Whyatt et al., 2001; Mnif et al., 2011).

Endocrine disruption has been reported to produce infertility, variety of birth defects, developmental defects in offspring, including hormonal imbalances

and incomplete sexual development, impaired brain development and behavioural disorders (Sanborn et al., 2007; Zheng et al., 2016).

Children are especially sensitive to these chemicals which have been associated with the rise in autism, epilepsy and Attention-deficit hyperactivity disorder [ADHD] (Health, 2008). And in adults, they have been associated with depression, personality disorder and obsessive-compulsive disorder (Caroline, 2010; Sroubek et al., 2013). Even more challenging is the fact that pesticides can cause many types of cancer in humans (WHO, 2009b; USEPA, 2014). Earlier scientific research reported some of the most prevalent forms of cancers caused by pesticides include leukemia, non-Hodgkin's lymphoma (Luo et al. 2016), brain, bone, and breast, ovarian, prostate (Boada, 2016), testicular and liver cancers (Jayaraj et al. 2016a; Jayaraj et al. 2016b). These diseases have been reported to be especially prevalent in farmers who use pesticides (Boada, et al., 2016; Rani et al. 2017).

1.4 Removal of PAHs and OCP from the Environment

Several scientific approaches have been adopted for the removal of PAHs in the environment and these included degradation (Wang et al., 2018), biodegradation (Kim, Kweon & Cerniglia, 2010); Dandie et al., 2004; Peng et al., 2008; Fredslund et al., 2008; Haritash et al., 2009), photolysis degradation (El-Saeid et al., 2015), chemical degradation (Abdel-Shafy et al., 2013), and degradation of PAHs in crumb tyre using ultraviolet light (Taylor et al., 2014). Taylor et al., (2014) reported that UV irradiation in the presence of a catalyst (rutile TiO_2) and hydrogen peroxide were beneficial to the degradation of PAHs but Benzo(a)Pyrene which is a high molecular weight PAH was more difficult to be degraded than the lower molecular PAHs.

Nevertheless, gamma irradiation had not been tried for the removal of PAHs in sundried and smoked fish in an attempt to remove or degrade PAHs from food and stored at ambient temperature except for wheat kennel (Khalil et al., 2016).

1.5 Fish as a Major Component of Nutritious and Affordable Food for Healthy Life

The fisheries sector has been very important in the socio-economic development of Ghana and plays a major role in sustainable livelihoods and poverty reduction in several households and communities (Diei-Ouadi & Mensah, 2005). Ghana has a marine coastline of about five hundred and fifty (550) kilometres extending from Aflao in the Eastern border to Half Assini in the Western border and this together with the inland waters support the fisheries industry in Ghana (Diei-Ouadi & Mensah, 2005).

Food including fish and its constant availability, affordability and nutrition are among the basic human physiological needs considered as the main physical requirements for human survival (Kwarteng, 2015). Fish is very important cheap but quality protein source to man (FAO, 2012). Fish protein is very easy to digest and research shows that the amino acids in fish are more bioavailable than in beef, pork or chicken. In addition, fish has a balanced quantity of all of the essential amino acids, with very high Amino Acid Scores: glutamic acid (15.17g/100g - 20.76g/100g); (FAO, 2012). Large population of people rely on fish for their protein intake worldwide, and fishing is the key means of support (food and income) for millions of people around the world (FAO, 2012) particularly in Low-income food-deficit countries (LIFDCs). Fish is the most traded agricultural commodity in the

world (FAO, 2016). The diets of people have been enriched worldwide including Ghanaians by including fish in the list of options of food around the world (FAO, 2016). As a result, there was a major growth in fish utilization and it provides 60% of total animal protein need of the world and 17% of global protein requirement (FAO, 2012; Mcclanahan, Allison, & Cinner, 2013; Wang, Hu, Pan, Li, & Failler, 2016; FAO, 2016). Health benefits of fish in the diet include 36% reduction in mortality risk from coronary diseases (FAO, 2016; FAO/WHO, 2010). It has been found out that 60g of fish in a diet a day may result in 12% reduction in mortality risk of heart attack (FAO / WHO, 2010) and long-term weight loss (Badr, 2015). In addition to serving as food, it also provides by-products such as fish oil, fish manure, and fish glue for industries. It also aids in diseases control, for instance, in HIV/AIDS, by enhancing antiretroviral drugs such that people living with HIV survive up to eight years longer if they have a good and high quality protein and micronutrients in the diet regime (Kwarteng, 2015).

Despite the many health benefits of fish, frequent consumption of contaminated fish can pose considerable health risks. Although promoting the availability and the consumption of fish among the poor cannot be downplayed, fish must be healthful in terms of quality and safety to enhance healthy life. The post-harvest value addition processes must not be devoid of policies which focus on fish quality and safety. This may therefore include quality and safe fish that provides healthful food for healthy living (Haraksingh et al., 2016).

In many coastal fishing communities in Ghana, the infrastructure for post-harvest processing and preservation of fish is inadequate. This resulted in

over a 50 % of harvested fish being lost through various kinds of wastage and spoilage (FAO, 2012). Such losses have a negative impact on the socio-economics of the fishing communities and reduce the amount of animal protein available to large section of the people.

In addition, poor quality of captured fish results in about twenty (20) million metric tonnes of fish discarded into the sea every year (FAO, 2012). These constitute food insecurity and food safety issues, which have increased in recent times as the public has become concerned about the importance of fish in the diet, as well as food-borne diseases, which affect consumer safety.

Food/fish, its acquirement, processing, preservation and storage are major problems facing humanity over the years. The task that man has engaged with is to eradicate extreme hunger, to ensure environmental sustainability and ensure sustainable consumption and production patterns by halting food spoilage (FAO, IFAD, UNICEF, WFP and WHO, 2017). Food spoilage and wastage result from infestation by insects in storage (Plahar et al., 1991), contamination by physical, chemical and microbiological agents (Nyarko et al., 2013; Nwaichi & Ntorgbo, 2016), and deterioration by enzymatic reactions and high temperatures. Food safety is a significant public health concern in the world today as it has ever been (Frewer et al., 2011).

Dried fish and smoked fish are important sources of nutrition to the poor and economically disadvantaged peoples of the world including Ghana (Diei-Ouadi & Mensah, 2005).

A large number of people consume dried and smoked fish due to the characteristic flavor and taste they possess. (Aheto et al., 2017). However,

there are some disadvantages such as bacteria and insects infestation of dried and smoked fish (Plahar et al., 1991).

Generally, blowfly and their larvae frequently infest dried fishes during drying phase. While the dried and smoked fish are in the storage, beetles and mites damage them and reducing the quality (FACP, 2016). To overcome these problems, the fish processors use various insecticides and fungicides (Society, 2015). Sun dried fish contaminated with insecticides has already created a wide spectrum of health hazard in the coastal areas (Ashraf Hussain et al., 2018; Abdegadirand & Adam, 2011).

Food safety became a major concern to the public in the early 20th Century (Pires, Vieira, Perez, Wong, & Hald, 2012). The result of this concern led to the development of two technologies such as milk pasteurization and retort canning which were accepted generally as preventive measures against food borne diseases (Tauxe, 1997). However, by the beginning of the 21st century, food borne disease remains a major threat to public health because new pathogens have emerged (Mead et al., 1999). The United States have reported an estimated 76 million cases of illness due to food borne infections and 325,000 hospitalizations annually resulting in total cost of $6.7 billion in patients treatment (Hoffmann et al., 2012). The techniques used to tackle fish post-harvest problem are variable and ranging from simple sun drying and wood smoking leading to PAHs contamination in food (Abdel-Shafy & Mansour, 2016). All these processing technologies have their attended problems as food preservation is no less important than food production (Silva, Adetunde, Oluseyi, Olayinka, & Alo, 2011).

Efforts to improve food safety continued and the process control strategy of the Hazard Analysis Critical Control Point (HACCP) became the standard method for producing many foods (ICGFI, 2011). The focus is on regulating sanitation and hygiene with good manufacturing and agricultural practices, which means that food be produced under cleaner conditions.

Food irradiation may be incorporated as part of a HACCP-plan where applicable. In the overall HACCP context, irradiation is a means of reducing hazards associated with infectious parasites and microbial contamination of foods and may be used as a method of control (ICGFI, 1991 and 2011).

Besides the prevention of losses, dwindling fish stock and closed fishing seasons, demand is also growing in both the developed and the developing countries for food that are wholesome and shelf stable. In addition, some established technologies (e.g. curing, chemical preservation and fumigation), are no longer adequate with regard to their biological, chemical and physical safety, economics, and possible reduction in market quality of products so treated (Mead et al., 1999; Hoffmann et al., 2012). In the wake of growing concern about food-related illness, food irradiation has emerged to complement the traditional methods and to re-enforce safety. However, this most effective method of food preservation which is food irradiation has not been fully explored in Ghana (Gasu, et al., 2015b).

Most research works carried out in the world have only been on regional basis and mostly use irradiation in combination with cold-storage of popular fish species mostly fish fillets and fish powder used as food ingredients. Irradiation preservation and its synergistic effect on

decontamination of PAHs and OCPs from salted, smoked and sun dried fish are uncommon in the tropics including Ghana.

Irradiation is becoming one of the preferred technologies to eliminate harmful bacteria from seafood, meat and farm produce, and insect pests from fruit and stored foods (Taylor, Arvanitoyannis, Stratakos, & Mente, 2009). An increasing demand for safe food in the world is enormous and a number of horticultural growers and processors are demanding that their products be irradiated for increased safety (Taylor et al., 2009) . The term "Food Irradiation" has no practical meaning because "Food" is not really irradiated. However, specific products such as fish, spices, and other food additives which are consumed as food, are irradiated for specific purposes (Taylor et al., 2009).

1.6 Importance of Fisheries Resources and Current Challenges

Fisheries resources are important source of both macro-nutrients and micro-nutrients for humans and play a vital role in improving human nutrition and health (FAO, 2012). A very important primary source of low fat, high-quality protein and essential nutrients to humans for healthy living is fish (Alemu, 2016). It also plays very important role in terms of livelihood, food security, economic growth, poverty alleviation, and the creation of employment opportunities in coastal communities and the communities along freshwater bodies through fish production (fish capture and aquaculture), and trade (Allison & Ellis, 2001).

More than two hundred and fifty (250) million people in the world depend directly on fisheries both marine and freshwater as well as aquaculture for their livelihoods, and other several millions are engaged in the fisheries

and aquaculture value chains in terms of processing or marketing (Diei-Ouadi and Mensah, 2005; FAO, 2012). However, in recent times Ghana's fisheries sector has been battling with various challenges such as, overfishing of certain key fish stocks and the institution of fisheries "close seasons". Furthermore, pollution and the degradation of habitats that support the fisheries sector is on the ascendency (Adeyemo, 2014).

Various scientific studies on the coastal ecosystems and fisheries in Ghana have called for management interventions to address these challenges for sustainability of the sector (Minta, 2003; Program & Project, 2013; Kwarteng, 2015; Worlanyo et al., 2016). Most of these proposals focused only on increasing fish stock and building regulatory capacity of the fisheries sector. There may be the need to examine the post-harvest losses, preservation and storage of the quantity of fish that is landed each time of the season and to cater for the "close seasons" and the off seasons as well.

The dramatic and unpredictable increasing global food prices, leading to non-affordability, poses a threat that the diets of the poor may become less varied and more dependent on non-protein staples if that quantity of fish landed could not be processed and preserved for future use. Furthermore, location, seasonality, time and household socio-economic status may affect fish consumption by the poor (FAO, 2012). Constraints to increasing consumption, such as non-availability due to persistently high volume of post-harvest losses which remove significant quantities of fish from the market occur in many developing countries like Ghana (FAO, 2012). The reasons are varied and complicated but include economics such as high prices, price fluctuations, lack of infrastructure, inadequate preservation, packaging and

lack of access to credit, lack of knowledge, limited education and lack of access to modern technology (Debnath, Pandey, & Ananthan, 2014; Getu, Misganaw, & Bazezew, 2015).

There are physical losses because fresh fish, smoked, and sun dried fish cannot be stored for longer period (Getu et al., 2015). Additional losses occur during processing leading to reduction in nutritional quality, caused by damage during processing and storage (Obodai et al., 2011).

Adopting means to reduce post-harvest losses in the fisheries is a better approach to make use of the large quantities of small pelagic fish that are available for direct human consumption. Nonetheless, this will require an integrated approach, involving socio-cultural approach, changing government policies, investing in the fisheries infrastructure and research, its environmental and addressing socio-economic implications. Therefore, decisions to enhance the achievement of set goals of food safety must be intensified by encouraging the use of technology such as food irradiation to complement and enhance food and consumer safety by reducing the effect of traditional processing methods such as smoking, salting and sun drying.

There is therefore the need to renew emphasis on nutrient-rich small fish production, processing, preservation, storage and distribution, and at affordable prices to the poor. This will encourage the accessibility and full utilization of common but micronutrient-rich foods among the poor.

The extension of fish storage or shelf life, and to improve fish quality by the process of decontamination of biological (fungi and bacteria), physical (heavy metals) and organic chemical (PAHs, OCPs and PCBs) contaminants when hygiene and cold storage are disrupted and to foster acceptability among

stakeholders can be achieved by food irradiation. This integrated approach presents a challenging task in consumer protection, food safety and safety management. In addition, policies and scientific decision in addition to support tools that facilitate identification of how and where these tools and services are provided, where they are consumed, and where management decisions will affect multiple aspects of the economy, human wellbeing and the environment are required (FAO, 2012).

Possible interventions to overcome the constraints in the fisheries distribution chain are wide ranging and may involve addressing a number of hurdles to these interventions. International organizations including the International Atomic Energy Agency and the International Consultative Group on Food Irradiation, governments and civil society organizations, industry and academia would be needed. The theme of integration of efforts to reduce under nutrition spearheaded by the Food and Agriculture Organization, which enjoins the entire food system – from inputs and production, through processing, storage, transport and retailing, and consumption may contribute much more to the eradication of malnutrition by enriching the protein intake of the people (Getu et al., 2015).

International organizations such as FAO, bilateral agencies such as USAID, through Feed the Future Initiative and Department for International Development (DFID), the Consultative Group on International Agricultural Research (CGIAR) through the CGIAR Research Programs, Governments, Non-governmental Organizations (NGOs) and the private sector have all initiated programmes and interventions that provide a platform for fish to contribute to human nutrition (FAO, 2012).

Recently, however, fish supplies are failing to meet demand and there are major shortages in some critically poor countries where fish stocks are diminishing as the result of overfishing, ineffective management practices leading to post-harvest losses, processing, preservation, storage and distribution, industrial development and agricultural pollution (FAO, 2012). These may constitute risks to livelihoods and contributing to low income generation, malnutrition, disease outbreak and unemployment (FAO, 2012). The small-scale fisheries contribute immensely to livelihood, food security and income for the coastal dwellers and countries as well (FAO, 2016). However, the sector has been limited to harvesting and not much attention have been paid to preservation and storage towards fishing close and of seasons and to maintain quality and safety of capture fisheries post-capture.

By improving the productivity of fisheries and aquaculture through good post-harvest management practices to reduce losses and improve food safety, the risk to livelihood will be minimized. This is important to reduce hunger and poverty in developing countries, improve food and nutrition security, increase income and enhance livelihoods by focusing on overcoming processing and marketing barriers that reduce the availability, accessibility and affordability of nutritious and safe fish to consumers and to protect fisheries resources.

1.7 Statement of the Problem and Justification

The smoke components of the fuel (wood) give the fish specific sensory properties such as aroma, flavouring and serve as bacteriostatic agents (Salindeho, 2018). In addition to these desired sensorial properties, the smoke also contains substances that are undesirable for health. Such substances

include formaldehyde, acetone, organic acids, phenols, tar, and alcohols (Moldoveanu, Serban; 2009).

Wood smoke also contains at least 100 polycyclic aromatic hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs) among hundreds of components, and their alkylated derivatives, many of which are carcinogenic (Rengarajan et al., 2015; Abdel-Shafy and Mansour, 2016). PAHs, OCPs and PCBs are environmental contaminants, which are the result of incomplete combustion of carbon containing materials and the use of agrochemicals (Abdel-Shafy and Mansour, 2016). They are present in trace quantities in water, soils, and in air and these are due to man's daily activities such as burning of coal and from motor vehicle exhaust fumes (Shbair, 2011).

Fish and marine invertebrates may naturally contain small amounts of different PAHs absorbed from polluted waters and the environment (Alexander et al., 2008; Rengarajan et al., 2015) . Nonetheless, the highest human exposure is the presence of PAHs in smoked-food (Palm et al., 2011).

The most significant endpoint of PAH toxicity is cancer (Silva et al., 2011). Their effects on human health, other living organisms, the aquatic ecosystem, as well as soil micro biota are in many research findings (Moermond & Verbruggen, 2010; Ikenaka et al., 2014). Apart from the chemical hazards, biological hazards (spoilage and pathogenic microorganisms) also abound in smoked, salted and sun-dried fish (Nyarko et al., 2013; Nwaichi and Ntorgbo, 2016).

Traditionally fish drying is carried out directly on the ground, mats, nets, rooftops, beaches and sometimes along the roads and open spaces in homes. Sun drying of fish is an old traditional practice across the global post-

harvest processing landscape and had been generally accepted as a competent technology for preservation and storage. It has been the predominant technology for centuries throughout the developing world and played an important role in the preservation of fishery products. Nonetheless, there are problems of microbial contaminations and insect infestations during the drying process and in storage. Obodai et al. (2011) and Eze et al. (2010) observed that this then leads to losses in quantity and quality of the fish so treated during storage, transportation and marketing. Services and Service (2011) reported that microorganisms have successfully adapted to changes in food production, processing, and preservation techniques, resulting in a number of new and emerging food borne pathogens and the re-emergence of organisms that have been problematic in the past.

Oyarzabal, (2015) confirmed this in his work while studying the "Emerging and Re-emerging Infectious Diseases". The causes that play significant role in the remerging of foodborne pathogens, include those associated to the pathogen itself, the environment, food manufacture and supply, and the consumers (Table 1).

Table 1: Species of microorganisms known to be pathogenic to humans

Category	Number of infectious organisms by species
Bacteria and rickettsia	538
Helminths	287
Viruses and prion	217
Protozoa	66
Fungi	30

Adapted from Taylor et al. (2001).

1.8 Disadvantages of the use of Traditional Technology in Fish Processing

Notwithstanding its extensive use, the traditional technologies only inhibit or delay the growth and the multiplication of infectious microorganisms (Table 2) such as *Vibrio cholera*, *Escherichia coli* O157:H7, *Salmonella*, *Campylobacter*, *Listeria*, *Staphylococcus aureus*, and *Clostridium botulinum* within a very short time, which pose food safety risk to consumers (Tauxe, 2001; Thayer, 2004; Services and Service, 2011).

Since the major underlying cause of microbial food spoilage and food poisoning is the presence of microorganisms and insects in food, inactivation technique such as food irradiation is ideally preferable to inhibitory techniques as the traditional methods (Studies, 2009; Roberts, 2016). To protect public health, science must meet the challenges that result from the remarkable adaptability of food borne pathogens (Eustice, 2011).

Polycyclic aromatic hydrocarbons (PAHs) and OCPs pose serious health threat to humans and the ecosystems therefore other researchers attempted to treat, remediate and restore contaminated sites (soil, river systems, drinking and ground water, scrap tyre, paint scrapings and in wheat kennels) using ultraviolet and gamma irradiation (Taylor et al., 2014; Khalil et al., 2016). These are non-food items, however, in terms of remediation of PAHs and pesticides contamination in food items, much attention has not been paid to food items like smoked fish and sun dried fish which are the main forms in which fish is preserved in Ghana.

Table 2: Emerging infection diseases caused by bacteria and the probable factors explaining their appearance

Infection agent	Disease	Possible factors contributing to emergence
Haemophilus influenza (biotype aegyptius)	Brazilian purpuric fever	Probably new strain
Vibrio cholera	Cholera	Probably introduced from Asia to South America. Spread facilitated by reduced water chlorination
Helicobacter pylori	Gastric ulcers	Probably long widespread but just recently recognized
Escherichia coli O157:H7	Haemolytic-uremic syndrome	Mass food processing allowing point contamination of large amounts of meat
Legionella pneumophila	Legionnaires' disease	Cooling and plumbing systems
Streptococcus group A	Necrotizing skin disease	Unclear
Borrelia burgdorferi	Lyme disease	Reforestation around homes and conditions favouring the expansion of deer (secondary reservoir host)

Adapted from Morse (1995).

In Ghana, most people consume smoked and sun dried fish as their main protein source (Plahar et al., 1991). It is therefore important to monitor the various types and levels of chemical and biological contaminants in salted, smoked, and sun-dried fish. Furthermore, it is important to evaluate the effect of gamma irradiation on these foods (fish) during irradiation to reduce or eliminate these contaminants in fish products using irradiation technology.

1.9 Knowledge Gaps

The following are the key knowledge or research gaps to which this study seeks to address:

(i) Many researchers have attempted to degrade PAHs, OCPs, OPPs, and PCBs in non-food materials such as contaminated soil, wastewater and

paint scrapings, scrap tyre and polymer materials using UV and gamma irradiation (Hallab, 1968; Rani, Shanker & Jassal, 2017). No such research findings referred to irradiation degradation of PAHs, OCPs, OPPs, and PCBs in salted, smoked, or sun-dried fish in Ghana.

(ii) Numerous research findings of the efficacy of irradiation technology on the microbial decontamination of food, insect disinfestations, extension of shelf life of many categories of food and food products are available (Dessouki, Aly and Sokker, 1998; Mostafavi, 2008; Sommers, 2012). However, little information about its use for smoked, salted and sun-dried whole fish and its effect on food safety is available in Ghana.

(iii) Most research works carried out on foods were focused on the use of irradiation in combination with cold storage of fishery products such as fish fillets and fish powder which serve as food ingredients. Studies of irradiation preservation at ambient temperature are uncommon in the tropics including Ghana.

1.10 Rationale

Understanding the steps involved in processing, preservation and the distribution of fish using both quantitative and qualitative approaches is important due to their association with chronic diseases. Preservation of food items is a pre-requisite for food security.

Food borne diseases though not scientifically documented are frequently heard and are on the rise in Ghana. This study provides the opportunity to examine the problem in a wider study using a large fish samples obtained from women engaged in post-harvest management of fish in three coastal regions of Ghana.

Focus group discussions (FGDs) and Participant Observations (PO) add a unique twist to hearing and gaining insight into what women know and do about the problems confronting the fish value chain. Since information on post-harvest processing of fish is low-cut, the outcome of a FGD and OP therefore will help to better understand and address the concerns expressed by the fisher women. This will stimulate appropriate interventions that target women in the fisheries value-addition and distribution chain sector and will lead to ensure sustainable production and consumption patterns. The concern of all humans to remain healthy therefore means the protection of the environment from excessive pollution during his search for food and to foster social and economic growth.

The study seeks to identify and quantify biological and chemical contaminants in salted, smoked, and sundried fish from three Coastal Regions (Volta, Greater Accra and Central) of Ghana and nine coastal markets (Keta, Akatsi, Sogakope, Kpong, Madina, Chorkor, Mankesim, Cape Coast and Elmina) and to identify socio-economic constrains that impede the value addition and distribution chain among the markets. The study also seeks to investigate the implications and prospects of the use of food irradiation technology in the fisheries sector to enhance resilience coastal management options and ensure consumer safety in Ghana.

1.11 Hypotheses

The following hypotheses will provide in-depth guidance to the study:

(i) Ho: Smoked and sun-dried fishes in Ghana do not have PAHs, Pesticides (OCPs) and microbial contaminants.

HA: Smoked and sun-dried fishes in Ghana may have PAHs, Pesticides (OCPs) and microbial contaminants.

(ii) Ho: Gamma irradiation will be able to eliminate or reduce PAHs, OCPs, and microbial contaminants from salted, and smoked fishes.

HA: Gamma irradiation cannot eliminate or reduce PAHs, Pesticides (OCPs) in salted, smoked, and sun dried fishes.

(iii) Ho: Gamma irradiation has no significant effect on the quality, safety and shelf life of salted, smoked and sun-dried fishes stored at ambient temperature.

HA: Gamma irradiation has significant effect on the quality, safety and shelf life of smoked and sun-dried fishes stored at ambient temperature.

(iv) Ho: Consumers will accept and be willing to pay for irradiated smoked and sun-dried fish.

HA: Consumers will reject irradiated smoked and sun-dried fish.

1.12 Aim of the Study

The primary aim of the study is to assess the potential of gamma irradiation technology as a tool for decontamination of PAHs, Pesticides (OCPs) and microbial load in processed fish from selected coastal markets of Southern Ghana.

1.13 Specific Objectives

The specific objectives of the study were to:

(i) identify PAHs, Pesticides (OCPs) and microbial contaminants in six species of commonly consumed smoked and sun-dried marine and freshwater fish in the selected study areas

(ii) investigate the decontamination effect of gamma irradiation on marine and freshwater sun dried and smoked fish samples in relation to PAHs, Pesticides (OCP) and microbial load

(iii) estimate the shelf life of gamma irradiated smoked and sun-dried marine and freshwater fish over a period of 60 days

(iv) evaluate the perception, food safety practices, knowledge of packaging, and acceptance of irradiated fish through focus group discussion (FGD) and participant observation (PO)

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Overview of Global Fisheries Demand

Fish is a vital source of food for hundreds of millions of people worldwide (Kwarteng, 2015). Fish provides an important source of polyunsaturated fatty acids (PUFA), fat soluble vitamins (e.g. Vitamin A) and essential minerals, which are associated with healthy and normal growth (Hussein & Khaled, 2014; Kwarteng, 2015; State & State, 2011).

According to FAO statistics, fish accounted for about 16% of the global population's intake of animal protein and 6 % of all protein consumed (WHO, 2010). In 2008, about 81% (115 million tonnes) of estimated world fish production was used as human food with an average per capita of 17kg (FAO, 2010). In 2010, the world's consumption of fish was US$ 17.5 billion, and an estimated 4.3 billion people depended on fish globally (FAO, 2012).

Recently, global consumption of fish has increased simultaneously with the growing concern of their nutritional and therapeutic benefits. It was found that developing countries accounted for forty-nine percent (49%) of world exports by value and fifty-nine percent (59%) by volume in 2006 (FAO, 2016).

2.2. Overview of Fisheries Industry in Contemporary Africa

The seeming per capita consumption of fish in Africa is 7.6 kilogram as compared to the global average per capita of 16.9 kilogram (FAO, 2010). Africa is currently relying greatly on imports and on aquaculture for its fish supplies, as there is only limited scope for increasing fish supply from its inland and marine capture fisheries.

Africa's fish imports are dominated by small pelagic, including anchovies, herrings, mackerels, and sardines. These are also the major species fished for non-food uses such as fish meal and fish oil for use in livestock and aquaculture feed (Wang et al., 2016). In sub-Saharan African counties, twenty percent (20%) of the animal protein supply comes from fish. In Equatorial Guinea, Gambia, Ghana, and Sierra Leone (being coastal countries); fish products contribute fifty percent (50%) of total animal protein intake (FAO, 2010).

Climate change impacts and other coastal zone forcing factors tend to reduce and destabilize landings from some capture fisheries (FAO, 2012). At the same time, this influences global supplies and creates import options for Sub-Saharan Africa (FAO, 2012). For example, Ghana, once a source of fish supply to Nigeria and inland countries, now imports fish from Mali, while its trade with Nigeria appears to be limited to a low-volume but higher-value market segment (FAO, 2010). Nigeria also imports fish from all around the globe and remains an important market for intra-regional trade (FAO, 2012).

2.3. An Overview of Ghana's Fisheries Need

Ghana relies on fisheries resources from the marine, inland freshwater and coastal lagoons but marine fisheries account for over 80% of the fish consumed in the country (Fishery and Aquaculture Country Profiles. Ghana, 2016). Fish is the most traded agricultural commodities in the world and constitute a very important component in the diets of many Ghanaians and globally (FAO, 2016). This is because fish is a good source of cheap but quality protein and constitute 60% of total animal protein consumed (Aheto et al., 2017). FAO/WHO, (2010) reported that fish intake is associated with 36%

reduction in mortality risk from coronary diseases. Furthermore, 60g of fish in the diet per day leads to 12% reduction in mortality and fish consumption has been reported to be associated with long-term weight loss in the USA (Haraksingh et al., 2016).

The importance of fish cannot be downplayed. Haraksingh et al., (2012) reported 1.4 deaths that occurred in 2010 was due to diets low in seafood. Fish is also able to catalyse the uptake of micronutrients from plant-source foods in the meal.

The fisheries resources need of Ghana is over 950,000 metric tonnes of fish annually, however, she can only produce 450,000 metric tonnes and loses 30% of this through inadequate post-harvest management (MOFAD, 2015). Ghana's average per capita fish consumption in 2004 was around 20kg - 25kg, which is higher than the world's average (FID/CP/GHA FAO, 2004). Marine fisheries account for 80% of fish production in Ghana. *Sardinella* species constitute the backbone of the fishery industry in Ghana (about 49% of the total fish catch). This is declining due to dwindling fish stock, post-harvest losses and closed fishing seasons (OECD-FAO, 2015).

2.4 Fisheries Post-harvest Challenges in Ghana

Over a quarter of harvested food and fish is lost due to various kinds of wastage and spoilage (FAO, 2012). Food preservation is no less important than food production. Besides the prevention of losses, demand is also growing in both the developed and the developing countries for food that are wholesome and shelf stable (Haraksingh et al., 2016). The emerging global energy crisis has led to an evaluation of the effectiveness of traditional methods of food preservation in terms of their energy consumption.

In addition, some established technologies including curing, chemical preservation and fumigation are no longer adequate with regard to their biological safety, economics, and possible reduction in market quality of products so treated (Ihsanullah & Rashid, 2017).

Fish products are however, very perishable and subject to bacterial spoilage if left unpreserved (FID/CP/GHA FAO, 2004; Getu et al., 2015). A number of physiological and microbiological deteriorations set in, and thereby degrade the fish immediately the fish is captured and subsequently dies (Gamal et al., 2011). As fish spoils, its nutritional value decreases, the bacteria degrade the protein and produce nitrogenous compounds with noxious odour and the fish becomes highly unattractive to consumers and unsafe for consumption (Ayinsa & Maalekuu, 2013; Getu et al., 2015).

Post-harvest losses of fish also occur during the fish capture through to processing and to the consumer (Getu et al., 2015). The factors causing the losses may include exposure to hash temperatures, inadequate processing and unsatisfactory preservation, poor transport network to market centres, storage and insect infestation, and further exposure during the retail-marketing process (Getu et al., 2015). These losses of fish capture could reach nearly 25 million tonnes (35 percent) of the global fish catch (FAO, 2011). Some developing countries have fish post-harvest losses reaching more than 50 percent of the landed catch and far more exceeding those of any other commodity and these losses are highest in the countries whose populations have the lowest protein intake (FAO, 2011).

Post-harvest losses in the fisheries industry as a result of spoilage due to biochemical and microbiological deterioration account for over twelve

million metric tonnes of fish shortage per year (FAO, 2012). In addition, poor quality of capture fish results in about twenty (20) million tonnes of fish discarded into the sea every year (Ayinsa and Maalekuu, 2013; Getu, Misganaw and Bazezew, 2015). These constitute food insecurity and food safety issues (FAO, 2012), which have increased in recent times as the public has become concerned about the importance of fish in the diet, food-borne diseases and consumer safety.

Despite efforts to control food spoilage and improve safety, food microbiological, physical and chemical hazards (PAHs and OCPs) still exist in food systems and the amount of illness related to pathogens in food items now is very alarming (Oyarzabal, 2015).

Post-harvest processing of fish in Ghana is mostly by smoking and sun drying and this is highly dependent on fuel wood and sunshine as the main source of energy. The fish processing industry in Ghana is largely diverse and unregulated in terms of the types of oven and wood fuels usage, and hygienic and sanitation practices (Aheto et al., 2017).

Smoked fish is a nutritious delicacy in Ghana and estimated 80% – 95% of landed fish in Ghana is smoked. About 90% - 95% of fish smoked in Ghana is locally used and only small amount is exported to other countries due to unregulated processing procedures (FOA, 2012; Kwarteng, 2015; Aheto et al., 2017). However, cold smoked fish is a product which exposed to low temperature heating and preservation and normally consumed as ready-to-eat product and possess danger of microbial contamination (Badr, 2015).

Sun drying of fish is the cheapest and simplest method of preserving fish in developing countries. Dried fish is an economically important source of

animal protein and this supplement 60% of animal protein needs of people (Services and Service, 2011; Chen et al., 2013; Farid et al., 2014).

However, dried fish is predisposed to many types of spoilage, which affect the quality and the shelf life. Getu, Misganaw and Bazezew, (2015) reported physical and organoleptic qualities of many traditional sun-dried products are unsatisfactory for human consumption. One serious problem which is of great significance in traditional sun drying and smoking of fish is damages occurring due to flies and insects infestations (Ayinsa & Maalekuu, 2013).

Foodborne diseases pose a widespread threat to human health and they are a central cause of reduced economic productivity in third world and even in advanced countries which have up-to-the-minute food processing and distribution systems (Badr, 2015). As a result, interest in food irradiation is growing because of persistently high food losses due to insect infestation, contamination, and spoilage (Taylor et al., 2009). Mounting concerns over food-borne diseases and growing international trade in food products must meet strict import and export standards of quality and quarantine. All areas in which food irradiation has been applied, demonstrated practical benefits when integrated within an established system for the safe handling and distribution of food (Arvanitoyannis (Ed.), 2010; Eustice and Bruhn, 2012). In addition, with increasingly restricted regulations or complete prohibition on the use of a number of chemical fumigants for insect and microbial control in food, irradiation is an effective alternative to protect food against insect damage and as a quarantine treatment of fresh produce (Ihsanullah & Rashid, 2017).

2.5 The Role of Gamma Irradiation Technology in Enhancing the Safety of salted, Smoked and sun-dried Fish in Ghana

Food irradiation is a processing technique that exposes food to ionizing radiation and produces a similar effect as pasteurization, cooking or other forms of heat treatment, but with less effect on appearance and texture to preserve, modify or improve the characteristics of products or materials (Bonomo, 2006; Benefits, 2015). The irradiation process involves exposing any kind of food, either in a packaged form or in bulk, to carefully controlled amounts of ionizing radiation for a specific time to achieve certain desirable objectives such as in food safety and storage (ICGFI, 2011a).

The technology is versatile, cost effective and has immense applications in the areas of postharvest management of agricultural produce, medical and dental sterilization in the form of single-use items, and household products, industrial materials modification, and environmental management (ICGFI, 1999b; Eustice, 2014). The energy absorbed by the food causes the formation of short-lived molecules known as free radicals, which kill bacteria that cause food poisoning by changing their molecular structure. The technology can also delay fruit ripening and help stop vegetables such as potatoes and onions, from sprouting by modifying and or altering the physiological processes of the tissues of the food (FAO, 2004; Mostafavi, 2008; Prakash, 2014).

The technology has increasingly become economically competitive and even in some cases superior to conventional technologies used for food and fish preservation. Combined with traditional methods of processing and preserving food, the technology of food irradiation is gaining fast and more

attention around the world (ICGFI, 2011b; Eustice, 2014). There are roughly 60 irradiation facilities being used for food processing in the world and with more under construction or at the planning stage for processing spices, grains, deboned chicken meat, beef, fruits and vegetables ICGFI, 1998). As by August 1999, over 30 countries are irradiating food for commercial purposes. It is an environmentally friendly technology and has been recommended by notable international bodies such as the Food and Agriculture Organization, World Health Organization, and the International Atomic Energy Commission (ICGFI, 1998).

The technology has been approved and accepted by over 40 countries and one hundred and fifty governments around the world following the adoption of a worldwide standard covering irradiated foods in 1983 (Ehlermann, 2016). The adoption of this global standard by the Codex Alimentarius Commission which is a joint body of the Food and Agriculture Organization of the United Nations (FAO) and the World Health Organization (WHO), responsible for issuing food standards to protect consumer health and facilitate fair practice in food trade, provided the impetus for use of the technology (Pareja, 2015; Roberts, 2016). All over the world, irradiation has been approved for the types of usage that would have a substantial impact on the presence of food borne pathogens (Ehlermann, 2016). The volume of food so treated universally has been estimated to exceed 500,000 metric tonnes (MT) annually (Mostafavi, 2008; Ihsanullah and Rashid, 2017).

2.6 Safety and benefits of Gamma Irradiation Technology

The safety and benefits of food processing by ionizing radiation has been studied at length world-over for years. This was to provide governments

and member States of the FAO, WHO, and IAEA with scientifically accurate information on issues of general interest to the public and on the safe and proper use of food irradiation technology. The Joint Expert Committee of Food Irradiation (JECFI) convened by the Food and Agriculture Organization (FAO), the World Health Organization (WHO) and the International Atomic Energy Agency (IAEA) resolved in 1980 that the irradiation of any food commodity up to an overall average dose of 10 kGy presents no toxicological hazards and requires no further testing (Thayer & Boyd, 1999; IAEA, 2009). JECFI further stated in the case of micronutrients such as vitamins losses due to irradiation treatment are comparable or lower than the conventional treatment such as heating or freezing (Mostafavi, 2008).

The Food and Drug Administration (FDA) of the USA has approved irradiation of food for limited purposes since 1963, and NASA has used irradiated food on its space missions for years as a safeguard against food borne pathogens that might affect the astronauts in orbit (Benefits, 2015).

The hot and humid climate of the Ghana may be quite favourable for the growth of several kinds of insects and microorganisms which destroy stored crops and stored fish and cause spoilage of food in storage. Once the food is irradiated, it can become prone to re-contamination unless appropriately packaged before the irradiation process (Getu et al., 2015) . Therefore, if radiation treatment is intended to control microbiological spoilage or insect infestation, pre-packaging becomes an integral part of the process and must remained packaged until it gets to the consumer (Kalyani & Manjula, 2014; Getu et al., 2015) .

2.7 Types of Irradiation Technology used for Food Preservation

The term irradiation usually excludes the exposure to non-ionizing radiation, such as infrared, visible light, microwaves from cellular phones or electromagnetic waves emitted by radio and television receivers and power supplies.

The term, furthermore, refers to a level of radiation that will serve a specific purpose, rather than radiation exposure to normal levels of background radiation. The exposure can originate from various sources, including natural sources. Irradiation technology relies on three kinds of ionizing radiation approved by the International Standard of Irradiated Foods (Benefits, 2015). These are gamma rays, electron beams and x-rays.

2.7.1 Gamma Irradiation for Food Preservation

Gamma irradiation technology uses the radiation produced by the radioactive form of Cobalt-60 or Cesium-137. These are the only radioactive isotopes approved as having the proper radiation profile and with only Cobalt-60 in use for food irradiation presently. It produces high-energy photons which has high penetration energy and can penetrate foods to a depth of several centimetres/metres in large packaging units (Kalyani & Manjula, 2014).

This type of radiation is essentially less-energetic. The Cobalt-60 (^{60}Co) emits simultaneously two photons per disintegration with energies of 1.17 MeV and 1.33 MeV). It has a low dose rate and it is highly reliable (Kalyani & Manjula, 2014; Benefits, 2015).

2.7.2 Safety of the Gamma Source and Facility

To prevent accident from the gamma ray exposure, the source has a mandatory physical protection from the outside world by a 3.0 meter thick concrete wall (Kalyani & Manjula, 2014). The gamma source is stored in a sufficient quantity of water (wet-storage), but the source is hoist from the pool in order to irradiate the target food, which could be a pallet load, for the time determined (ICGFI, 2011b).

The radiation doses applied vary depending on the types of food to be irradiated and according to the Environmental Protection Agency (USEPA) the limit is less than 10 kilo Grey for most foods. One setback of the gamma source is the need to replenish the radionuclide source when the source strength is low. This is because the half-life of Cobalt 60 (^{60}Co) is 5.3 years (Kalyani & Manjula, 2014).

2.7.3 Electron beam Irradiation Technology for Food Preservation

The Electron beam (E-beam) has a relatively limited penetration power (4cm) but a high variable dose rate with energy of 10 MeV (Bonomo, 2006; Eustice, 2014; Kalyani and Manjula, 2014; Ehlermann, 2016). E-beam irradiation requires shielding as well, but not like the concrete walls used in gamma ray irradiation (ICGFI, 1998).

The disadvantage of the E-beam is its high sensitivity to breakdown and need specialized personnel for regular maintenance. It needs regular power and cooling but the machine can be switch on and off. Unlike the gamma source, a miniature e-beam could be integrated into a production line (Bonomo, 2006; Eustice, 2014; Kalyani and Manjula, 2014; Ehlermann, 2016).

2.7.4 X-ray Irradiation Technology for Food Preservation

X-ray irradiation is a relatively new technique that combines many of the advantages of the other two methods. Like gamma ray irradiation, X-ray irradiation consists of exposing food to high-energy photons of 5 MeV with a long penetration depth (ICGFI, 1998). In this case, allowing the radiation to be controlled.

The device is a more powerful version of the X-ray machines used in medical field. The device still requires heavy shielding, although the amount of shielding required is less than that for gamma ray irradiation. Electron beams and X-ray facilities do not involve radioactive substances either (Kalyani and Manjula, 2014; Ihsanullah and Rashid, 2017).

There are three general categories of treatment by irradiation (Taylor et al., 2009) and these are:

i) "Low" doses range from zero up to 1.0 kGy and which is used to delay physiological processes, such as the ripening or sprouting of fresh fruits and vegetables, and to control insects and parasites in foods.

ii) "Medium" dose irradiation ranges from 1–10 kGy, and meant to reduce spoilage and pathogenic microorganisms in a number of foods. This is seen to improve technological properties of food, such as reducing cooking times for dehydrated vegetables, and to extend the shelf-life of many foods.

iii) "High" dose irradiation are doses greater than 10 kGy and are used for sterilization of meat, poultry, seafood, and other prepared foods in combination with mild heating. This helps to inactivate

enzymes, and disinfect certain foods or ingredients, such as spices and enzyme preparations.

2.8 Radioactivity and Irradiated Food

The concern of consumers is about the possibility of radioactivity being induced in foods treated by gamma irradiation (ICGFI, 1999a; Mostafavi, 2008; Kalyani & Manjula, 2014). For this reason, the public are however informed to look for the mandatory "Radura Logo" (Plate 1) on all foods treated by gamma irradiation. The radura logo tells which food on the shelf is irradiated and which is not irradiated.

Plate 1 : Radura Symbol (ICGFI, 1999a ; IAEA, 2002, Ehlermann, 2009)

The ''Radura''symbol is presented in green color with a plant-like structure in the center. It represents agricultural products in a closed package broken in the upper half. The upper broken gaps indicate the penetration of ionizing rays or particles. Whenever the Radura symbol is placed on the package of irradiated product, it is accompanied with the inscription "treated with ionizing radiation" or "irradiated" for consumers' information (Mostafavi, 2008).

Even though no radioactivity has been detected as a result of food irradiation, there may be background radioactivity present in foods in the

natural environment. Since radioactivity is present in soils, it is not surprising that it ends up in plants and in animals that serve as food for humans. The Brazil nut (Plate 2) has the highest radioactivity than any known food even though it has not been treated with any form of irradiation (Appendix A).

Plate 2 : Brazil nut (Anderson et al., 2005)

The Brazil nut (*Bertholletia excelsa*) is a South American tree in the family *Lecythidaceae*, and is also the name of the tree's commercially harvested edible seeds has the highest radioactivity of any food (40 – 260 Bq/kg) of $_{88}^{226}Ra$, and an equal amount of $_{88}^{228}Ra$ and $_{19}^{40}K$ (IAEA, 2002). Radioactivity occurs during the decay of an unstable nuclide to produce a different "daughter" or product called nuclide. During "transmutation", the daughter nuclide is a different element. The decaying process is accompanied by the production of one or more particles associated with "radioactivity", as shown below. The quantity of a radioactive isotope in terms of its activity or the number of decays per second is called a Becquerel (1 Bq = 1 decay or disintegration per second). High activities are often reported in curies (Ci: 1Ci = 3.7 x 10^{10} d/s). The total activity in a Brazil nut is approximately 1000

Bq/kg, as a summation of equal activities of $^{226}_{88}Ra$, $^{228}_{88}Ra$ and $^{40}_{19}K$ (IAEA, 2002).

The irradiation process using the above sources (Cobalt-60, E-beam and X-ray) does not produce radioactive substances or by-products (IAEA, 2002; Ehlermann, 2016). During the procedure of irradiation, the food is passed through the irradiation field, energy passes through the food much like a ray of light passes through a window or door. Food is irradiated with cobalt-60, which decays by beta emission to a nickel isotope. The nickel isotope decays by the gamma emission which is the radiation primarily responsible for killing microorganisms (IAEA, 2002). This energy destroys most of the bacteria and fungi spores that can cause diseases and allows the food to retain its high quality and improve its safety. This energy involved in irradiation is not strong enough to change the atoms of the food, and since the food never actually touches the radioactive source, the food does not become radioactive (IAEA, 2002). Food irradiation facilities therefore do not become radioactive and do not create radioactive waste (Bonomo, 2006; Eustice, 2014; Kalyani and Manjula, 2014; Ehlermann, 2016). Mobile irradiators have been used by scientists in search for the treatment of seasonal foods, such as fruits and vegetables, and for fish irradiation on board fishing ships.

The conclusion is that, with good public education on the benefits of irradiated foods, the public would likely receive the products without undue negative reaction to the irradiation process.

2.9 Environmental Risk Assessment of Food Irradiation Technology

The irradiation process does not produce neutrons and do not make anything around them radioactive (Bonomo, 2006; Eustice, 2014; Kalyani and

Manjula, 2014; Ehlermann, 2016). Furthermore, there are no radiation contamination of food items, water bodies, air, soil, and aquatic habitats. Nonetheless, the Nuclear Regulatory Authority (NRA) of Ghana licenses facilities that use radioactive sources in the country according to International Standard (Codex, 1984). These facilities have physical protection, tight "fail-safe" measures, extensive and well-documented safety procedures, worker training, and regular inspection and control to ensure the quality and safety of food, and the irradiation source in Ghana (Eustice, 2014). The NRA and the Radiation Protection Institute (RPI) of the Ghana Atomic Energy Commission on the other hand make sure that the relevant national and international radiological safety standards are maintained. NRA and RPI carry out inspection in order to verify that the facility is operated in accordance with the license requirements and to ensure that good irradiation and good manufacturing practices are followed (ICGFI, 1999b).

2.10 Safety of Irradiated Foods for Human Consumption

The toxicological safety and wholesomeness of foods irradiated up to specific doses is satisfactory and has no special or nutritional risk problems. All safety scientific studies have found irradiated food to be safe and wholesome. To emphasize this confidence, it has been endorsed by a multitude of organizations, including the American Medical Association, the American Dietetic Association, the Mayo Clinic, and the World Health Organization. The technique makes the food safer than other conventional processing method (ICGFI, 1999b; Mostafavi, 2008).

Food irradiation technology is currently gaining attention globally and it is more effective in combating food borne pathogens when compared with

heat or chemical treatment (Studies, 2009; Roberts, 2016; Eustice, 2018). Radiolytic species produced in irradiated foods for example benzene, is present at much lower levels than is found naturally in a variety of common foods, such as eggs or dairy products foods (Kalyani & Manjula, 2014). Numerous carcinogenicity bioassay studies have been performed which have not demonstrated any short term or long term toxicity related to the irradiation of food foods (Kalyani & Manjula, 2014). Indeed, in the RALTECH study (the largest toxicology study ever conducted on irradiation or any other food processing methods) the lowest incidence of cancer was found in the test groups that were fed the irradiated diets foods (Kalyani & Manjula, 2014). This study was initiated by US Office of the Surgeon General, and the findings reviewed by the FDA and the National Toxicology Program's Board of Scientific Counsellors agreed that the evidence did not show any carcinogenicity in irradiated foods (Kalyani & Manjula, 2014).

The veracity of the facts about food irradiation and irradiated food led one hundred and thirty (130) governments to have already approved of Codex General Standard (CGS) for irradiated foods. Jamaica and Grenada are the latest countries to announce plans to export fruit to the United States using irradiation as a mitigating process to prevent fruit fly infestation and has approved over 40 groups of irradiated foods for consumption (Eustice, 2018).

2.11 Valuation of Food Irradiation

Food irradiation facility is not a low-cost investment. Irradiation plants are expensive to acquire but once acquired the facility could provide service to a large number of users. It is also important to maximize the utilization with a high throughput for it to be cost effective (ICGFI, 1999a).

Comparative advantage for each product to be heat-treated, or irradiated, or processed in any other way need to be determined. The irradiation process is complimentary to the other traditional processes. Considering the advantages of each process against the costs of using that process and the market determines if there is a willingness to accept, and pay for, these advantages. Requirements for improved security measures at all facilities holding radioactive materials and the re-enrichment of decreasing source strength are likely to increase the costs of irradiation plants, leading to an increase in the prices of irradiated foods.

The process costs or market prices and its effect on the product price of irradiated food based on numerous cost analyses, shows that the costs of irradiation are low in comparison to the wholesale costs or market prices of some foods in some situations. It is important however, to carry out feasibility studies for each product and market (Morrison, & Roberts,1990).

Generic cost analyses taking into consideration both capital and annual operating expenditures of typical gamma processing facilities, indicate costs varying of GHC 3.12/kg for potatoes and onions, GHC 13.00/kg for poultry, fish GHC13.00/ kg, and GHC 24.00/kg for spices and dried seasonings. These numbers are contingent on realistic economies of scale. This does not however include potentially significant savings that may be realized from reduced energy costs and potentially significant marketing advantages resulting from offering improved quality or increased availability of the product.

2.12 Complementary Role of Food Irradiation Technology to Traditional Preservation Methods

Numerous advantages of the technology including reducing post-harvest losses have been cited (Ihsanullah & Rashid, 2017b). Nevertheless, not all of these losses can be prevented by food irradiation but, the technology offers unique potential that compliments other technologies to increase food supply, overall food security and Integrated Coastal Zone Management (ICZM) options by providing solution to post-harvest losses in Ghana's fishery sector, and job creation to alleviate rural poverty, reduced cold-storage and improve ambient storage of smoked and sun-dried fishery products. The huge tonnage of fish post-harvest losses in the fishery sector may reduce when the excess is processed, smoked or sun dried, appropriately packaged and irradiated to be stored for future sales.

There should therefore be a way frontward for saving the oceans' health and that of humanity and this through Integrated Coastal Zone Management, and proper landing, handling and processing of fish and the use of gamma irradiation to ensure the hygienic quality of dried fish (Ihsanullah and Rashid, 2017; Aryee, 2014). This will bring about good sanitation of the coasts and fish landing beaches to enhance good quality and safety of the fish processed for sales. Furthermore, a scientific look at the quality and safety of fish landed from the sea and processed for consumption may reveal health concerns.

2.13 Food Irradiation as a Critical Control Point in Fish Processing and Distribution Chain

Hazard analysis and critical control point (HACCP) is a system that identifies risks accompanying each food production process and decides in what way each risk can be reduced or eradicated at Critical Control Points (CCPs). HACCP depends on precise testing of food in-process for the presence of microorganisms and temperature changes that promotes the growth of those microbes (HACCP, 1994). However, testing alone cannot prevent bacterial hazards accompanying the food item from reaching the consumer. Real intervention is essential that actually kills the contaminating microorganisms and irradiation is a complementary intervention as a "kill step", or "hurdle", within HACCP plans to make food wholesome. Irradiation is combined with other "hurdles" such as good sanitation and temperature control to maximize product safety (Eustice, 2014).

Irradiation may therefore become a Critical Control Point (CCP) process in the production and long time storage of salted, sun-dried and smoked fish to meet the safety and quality standards for domestic and export markets (IAEA, 2009). However, consideration has not been given to this vital technology in the food processing sector of the economy of Ghana and in the fishery sector to reduce post-harvest losses (Gasu et al., 2015b).

The difficulties encountered with the processing of fresh fish (during glut) by smoking and drying for long time storage, marketing and for export, while not overwhelming, require a new and higher level of technical and operational complexity to overcome, such as the use of gamma (^{60}Co) irradiation in Ghana's fishery sector (IAEA, 1981 & 2009).

Irradiation is a tool that can be employed on certain foods for certain advantages. There are competitive techniques that may be employed. For example, both heat and radiation can be used to kill microorganisms in food (IAEA, 2009). However, there are technical differences between the two processes. Irradiation is a physical and cold process allowing product to be disinfected without cooking unlike the heat process which cooks the food product.

2.14 Disadvantages of Traditional Methods of Processing (salted, smoked and sun dried) Fish

Increasing human population, rapid industrialization and extensive use of modern technology, lead to discharge of toxic chemical residues into both surface and underground water bodies (FAO Newsletter, 2016). These residual chemicals (PAHs and OCPs) are absorbed by plants and animals, which are the basis of the food chain. Fish processing and storage methods also lead to fish contaminations by microorganisms resulting in human health challenges (Chanie et al., 2005; Rengarajan et al., 2015).

Polycyclic aromatic hydrocarbons (PAHs) are a group of several chemicals that are also called polynuclear aromatic hydrocarbons. They occur in nature and through human activities. PAHs are generally introduced into the eco-system naturally from forest fires and volcanoes, and from burning coal, oil, gasoline, trash, tobacco, and wood (Alexander et al., 2008). They are further generated through food preparation activities such as high-temperature cooking, grilling and smoking of foods (Dobríková & Světlíková, 2007; Moermond & Verbruggen, 2010).

These (PAHs) have half-lives of many years and are significantly harmful. Several of the PAHs, including benz(a)anthracene, benzo(a)pyrene, chrysene, benzo(b)fluoranthene, benzo(j)fluoranthene, benzo(k)fluoranthene, and indeno(1,2,3-c, d)pyrene, were identified to have caused tumors (mutagenic and tumorigenic effects) in laboratory animals when they had long periods of exposures. Some studies showed that individuals exposed for long periods to PAHs could develop cancers (Boström et al., 2002; Alexander et al., 2008). The Department of Health and Human Services (DHHS) of the United States of America, found out that Benzo(b)fluoranthene, benzo(a)pyrene, Dibenz(a,h)anthracene, Benzo(k)fluoranthene, Benzo(a)anthracene, Benzo(j)fluoranthene, and indeno(1,2,3-c,d)pyrene are known animal carcinogens (Alexander et al., 2008; Abdel-Shafy & Mansour 2016).

The International Agency for Research on Cancer (IARC) has recognized Benzo(a)anthracene and Benzo(a)pyrene as most likely carcinogenic to humans. Benzo(b)fluoranthene, Benzo(j)fluoranthene, Benzo(k)fluoranthene, and Indeno(1,2,3-c,d)pyrene are perhaps carcinogenic to humans; and Anthracene, Benzo(g,h,i)perylene, Benzo(e)pyrene, Chrysene, Fluoranthene, Fluorene, Phenanthrene, and Pyrene are not classifiable as to their carcinogenicity to humans (ATSDR, 1996).

The Environmental Protection Agency of America (USAEPA) has determined that Benzo(a)anthracene, Benzo(a)pyrene, Benzo(b)fluoranthene, Indeno(1,2,3c,d)pyrene, Benzo(k)fluoranthene, Dibenz(a,h)anthracene, and Chrysene, are possible human carcinogens and that acenaphthylene, anthracene, benzo(g,h,i)perylene, fluoranthene, fluorene, phenanthrene, and

pyrene are not classifiable as to human carcinogenicity. Acenaphthene has not been classified as a carcinogen by the DHHS, IARC, and EPA (ATSDR 1996; Boström et al., 2002).

PAHs are abundant in the environment because of combustion of fossil fuels and organic waste and some studies have shown that certain PAH metabolites interact with DNA and are genotoxic, causing malignancies and heritable genetic damage in humans. People who are heavily exposed to mixtures of PAHs as a result of their occupation occasions a substantial risk of lung, skin, or bladder cancer (Boström et al., 2002; Jayaraj, Megha, & Sreedev, 2016a).

Benzo(a)pyrene is a potent carcinogen which is generally used as an environmental indicator for PAHs. In water, PAHs concentrations are low due to their weak solubility. It was found out that the low water solubility leads to accumulation in sediments and aquatic organisms. It can also accumulate in soil and can be absorbed by plants. (ATSDR 1996; Boström et al., 2002). Due to their mutagenic and carcinogenic nature, most of them are persistent, toxic and bioaccumulate in organisms and the environment, hence their monitoring, regulation and complete removal is essential for healthy food and healthy environment (Abdel-Shafy & Mansour, 2016).

Essumang, (2010); Ledesma, Rendueles, & Díaz, (2015), observed there are variations in human exposure depending on many factors, which include smoking rates, fuel types used in cooking and pollution controls on power plants, industrial processes, and vehicles.

2.15 Sources of Human Contamination of PAH

Human exposure to PAHs comes from different sources (Figure 1). These include the air, terrestrial, aquatic systems and aquatic organisms, and fanally to man as the final target (Essumang, 2010; State & State, 2011; Khalil, Albachir, & Odeh, 2016; Zachara, Gałkowska, & Juszczak, 2017).

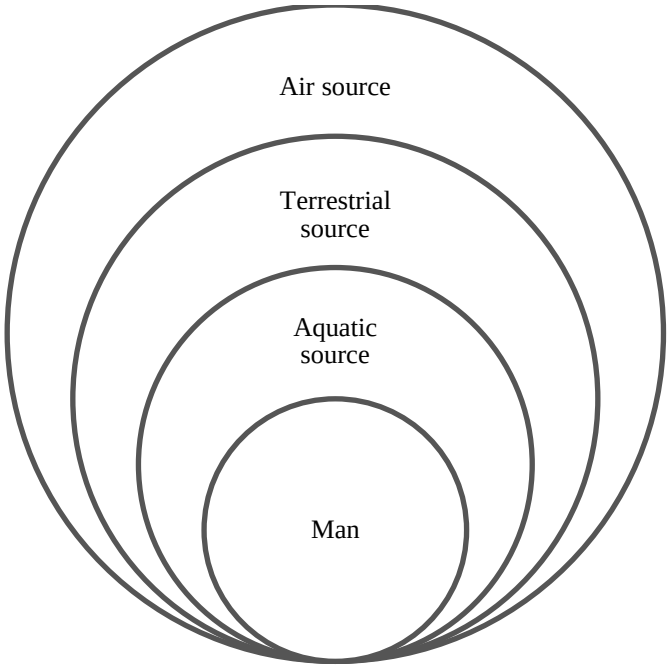


Figure 1: Conceptual Framework illustrating Ecotoxicity of PAHs and Pesticides (Adopted from: Zachara et al., 2017)

In the aquatic system, fish and shellfish absorb these contaminants which are ingested by man (Abdel-Shafy et al., 2016). In the soil, PAH are unlikely to exert toxic effect on land and soil invertebrates however, genetic mutation resulting in cancer is the end effect in living organisms (Boström et al., 2002). Scientific knowledge reported that plants absorb these PAH through roots and cuticles and bio-accumulate in their tissues (Khalil, Albachir, & Odeh, 2016). Generally, the major routes of exposure to PAHs are from food and inhaled air, while in smokers, the magnitude of exposure is the sum total

of contributions from smoking and food (Figure 1). The common entrance of PAHs into the body is through ingestion (swallowing) of grilled, charred, charcoal broiled, smoked meat and fish or foods and drinking contaminated water or milk when these are not routinely tested (Boström et al., 2002). Other routes of entrance into humans are inhalation (breathing) of cigarette smoke or secondhand smoke, vehicle exhaust fumes, fumes from asphalt, or emissions from fossil fuel fires (Alexander et al., 2008). Furthermore, direct skin contact by touching food that contains PAHs can lead to the development of skin and liver cancers. In the body, the PAHs are converted into metabolites which are passed out of the body in the urine and faeces. A few of PAHs are used in medicines and to make dyes, plastics and pesticides (ATSDR, 1996).

Several of the PAHs and some specific mixtures of PAHs are considered carcinogenic chemicals which affect human health from exposure to low levels of PAHs within the environment (Mead et al., 1999; Report, 2009b; Jayaraj, Megha and Sreedev, 2016).

With increase in human population of Ghana, less fish will be available per capita annually due to post-harvest losses (Report, 2013). These losses have an intense unfavorable impact on fishing communities whose economic standing and income often depend on post-harvest activities such as fish processing by smoking, sun drying, and marketing. Reducing these losses could increase protein availability, improve nutritional status, and eradicate the need to import fish and therefore securing food security.

2.16 Smoked Fish and PAH Contamination

Smoking is highly dependent on fuel wood and sun drying is dependent on sunshine as the main source of energy. The fish processing

industry in Ghana is largely diverse and unregulated in terms of the types of oven and wood fuel usage, and hygienic and sanitation practices. Smoking is one of the oldest ways of preservation of fish product of considerable economic importance in the world (Ayinsa & Maalekuu, 2013).

Fish processing and preservation by smoking provides an attractive characteristic of color and flavor to the fish and is largely acceptable to the populace mainly because of its sensorial characteristics (Badr, 2015). However, the Scientific Committee on Food (SCF) studied the presence and toxicity of PAHs in smoked food and reported that a number of PAHs are genotoxic carcinogens and recommended that human contacts of PAHs should be as low as reasonably achievable (Nwaichi & Ntorgbo, 2016a). The SCF identified fifteen substances as a priority that should be avoided due to their potential genotoxicity and or carcinogenicity in humans (Alexander et al., 2008; Vane et al., 2014). Based on the veracity of the report of the SCF, the Joint FAO/WHO Expert Committee on Food Additives (JECFA) performed a risk assessment on PAHs in 2005 and has also estimated Margins of Exposure (MOE) for PAHs. Based on these Margins of Exposure however, the JECFA concluded that PAHs were of low concern for human health (ATSDR, 1996; Report, 2009a).

Fish caught in Ghana are processed by the methods of drying, salting and smoking. However, smoking is the major method of fish preservation in the country (Ayinsa & Maalekuu, 2013; SFMP, 2015; Aheto et al., 2017). Through the process of fish smoking and use of the smoked products, workers and consumers are exposed to large amounts of PAHs through skin contact and from breathing smoke. They could be at risk of developing non-

communicable diseases such as cancers and cardiovascular diseases (Boström et al., 2002).

Food can be contaminated by environmental PAHs, industrial food processing methods such as heating, drying and smoking, and by home food preparation including grilling and roasting processes (Alexander et al., 2008; Gordon et al., 2013; Vane et al., 2014; Relekar, Joshi, Gore, & Kulkarni, 2014).

Damages occurring due to flies and insects are of great significance in open sun drying and this may be a serious problem in traditional sun drying and smoking of fish (Getu, Misganaw and Bazezew, 2015). Despite efforts to control food spoilage and improve safety, food microbiological and chemical hazards still exist in food systems; the amount of illness due to pathogens in food items have been reported to be very alarming (Veyrand et al., 2013). PAHs are released from various industries and from everyday activities on global scale; and due to the continued dependence on the same sources of release, PAHs will continue to be worrying hazard in foods.

PAHs found in Table 3 were identified in charred and smoked meat and fish, marine life in contaminated waters, cereals, fruits, vegetables, flour, bakery and snacks. The use of irradiation alone as a preservation technique will not solve all the problems of post-harvest fish and other food losses. Irradiation, however, can play an important role in cutting down on losses of agriculture produce and reduce the dependence on chemical preservatives and pesticides that leave residues on the food and also in the environment.

Table 3: Polycyclic aromatic hydrocarbons identified in charred and smoked fish and meat regarded as potentially genotoxic and carcinogenic to humans

No.	PAHs	No	PAHs	No	PAHs
1	Benz[a]anthracene	6	Dibenzo[ai]pyrene	11	Dibenzo[ah]anthracene
2	Benzo[a]pyrene	7	Benzo[j]fluoranthene	12	Indeno[1,2,3-cd] pyrene
3	Dibenzo[ah]pyrene	8	Cyclopenta[cd]pyrene	13	Benzo[ghi]perylene
4	Benzo[b]fluoranthene	9	Dibenzo[al]pyrene	14	Dibenzo[ae]pyrene
5	Chrysene	10	Benzo[k]fluoranthene	15	5-Methylchryzene

Source: European Scientific Committee on Food, 2002; Stołyhwo & Sikorski, 2005.

Regulations, however, have been set by the Environmental Protection Agency of the United States of America (USEPA) to protect people from the potential health effects of PAHs. This regulation stipulates that the maximum residue levels for Anthracene should not exceed 0.3 mg/kg per body weight and that of Acenaphthene should not exceed 0.06 mg/kg per body weight. The maximum residue levels for Fluoranthene was set at 0.04 mg/kg per body weight, Fluorene was set at 0.04 mg/kg per body weight, and for Pyrene at 0.03 mg/kg per body weight (ATSDR, 1996; Garcia et al., 2016). The regulations are relevant to the economy of Ghana since she exports smoked and dried fish, it is important to monitor these PAHs in smoked and sun dried fish to guarantee international acceptance and to enhance international trade.

The focus of this research is on the compounds that have been identified by USEPA to cause cancer disease in humans. These PAHs are Benz(a)anthracene, Benzo(b)fluoranthene, Benzo(a)pyrene, Chrysene, Benzo(k)fluoranthene, Benzo (g, h, i) perylene, and Indeno(1,2,3-cd) pyrene.

2.17 Stored Processed Fish and Pesticides Contamination

Smoked and sun-dried fish, and other agricultural produce are preserved from destruction during processing, storage and transporting to markets. Illegal use of pesticides to preserve fish by traders with little or no knowledge of public health policy have been reported in North-Eastern Nigeria (Musa et al., 2010). Pesticide residue levels in smoked fish were found to be above permitted tolerances and dietary intake hence could be an important process of transferring residues to humans. The presence of these residues could hamper international trade in the fisheries.

While the main focus is to harm only the targeted pests, such as insects, rodents, bacteria and fungus, pesticides have become part of the food chain of humans, the environment and the people who are exposed to them.

Pesticides are toxic and can degrade into other chemical compounds known as derivatives. Residues and metabolites of many OCPs are stable and having long half-lives in the environment (Basfar, Mohamed, & Al-saqer, 2012). Maximum limits for residues were established for only selected pesticides (Shbair, 2011; Nwaichi and Ntorgbo, 2016).

A single pesticide may be considered safe at a particular concentration level. Some foods may contain residues of several pesticides at the same time as the result of multiple exposures (American Academy of Pediatrics, 2003).

Some health problems posed by pesticides, include headaches and nausea, cancer, reproductive disorder, and endocrine disruption (Report et al., 2009a). Endocrine disruption can produce infertility and a variety of birth defects and developmental defects in offspring, including hormonal imbalances and incomplete sexual development, impaired brain development and behavioural disorders (Caroline, 2010; Sroubek et al., 2013; Boada, 2016; Jayaraj et al., 2016a; Jayaraj et al., 2016b).

Pesticides have been in use for decades and will continue to be used extensively in agriculture production and in household protection. There is, however, no effective routine monitoring of food items to check the residue level of organochlorine pesticide residues in staple food and fish. This has been due to a lack of relevant technical, scientific and organizational capacity to carry out effective surveillance in general.

There is a stringent monitoring of pesticide residues in export crops such as cocoa beans, but such strict monitoring programmes are absent in domestic food items including fish.

The application of pesticides does not only take place while the crop is growing, but is applied as a seed treatment, or in post-harvest applications to assist with transportation and storage. Although washing and peeling of food produce can reduce pesticide residues, the process cannot eliminate them because the bulk of pesticides (systemic pesticides) are inside the food, groundwater and the air, other pesticides only affect the surface and can be washed (Basfar et al., 2012).

Pesticide residue refers to the amount of a pesticide or ingredients in the mixture found in or on a raw agricultural commodity or in a processed food including fish (USEPA, 2014; USEPA, 2018). This also includes residue of degradation products of the pesticide or by plant metabolism or some other degrading process. The residue of concern may be the parent compound, a metabolite of the parent compound or both.

The use of pesticides is restricted by International Standards (Table 4) to ensure the circulation of only food produce that have been treated with approved pesticides and complies with the approved Minimum Residue Levels [MRLs] (Chan, 2000, USEPA, 2018).

Table 4: Maximum permissible residue level for some pesticides in foods and seeds

Pesticides	Allowable dietary intake mg/kg/bw/day	Pesticides	Allowable dietary intake mg/kg/bw/day
Cyfluthrin	0.02 – 0.05	Pyrethrin	0.04
Fenpropathrin	0.03	Aldrin	0.0001
Cypermethrin	0.05	Lambda-cyhalothrin	0.01
Chlorpyrifos	0.01	Fenvalerate	0.02
Deltamethrin	0.05	Malathion	0.3
Permathrin	0.05	Chlorpyrifos-methyl	0.3

Source: FAO, 2005: Plant Production and Protection (Paper 183 JMPR-2005 report).

A zero tolerance means that no amount of the pesticide chemical may remain on the raw or processed food commodity when it is offered for consumption. A zero tolerance for a pesticide chemical may be established because a safe level of the pesticide chemical in the diet has not been reliably determined.

When the chemical is carcinogenic and has an alarming physiological effect after it is present in the diet of animals, a zero tolerance is established (Global MRL Database 2018, USEPA, 2018). However, pesticide chemical residue which is superficial is normally removed through good agricultural practices (GAP) such as washing, weathering or other changes in the chemical itself, prior to the consumption of the commodity.

Exposure of consumers to these residues have been observed to occur most commonly through consumption of raw or treated food products, or coming into close contact with these pesticides during mixing, packaging,

spraying on farms, lawns and during house spraying (US EPA 2005; Luo et al., 2016).

These OCPs, being highly toxic are slow to degradation and bioaccumulation in the ecosystem (Mel'nikova et al., 2017). Although, many of the compounds that belong to Organochlorides were banned in developed countries, in Ghana the use of these agents has been rising (OECD-FAO Agricultural Outlook 2015-2024, 2015).

The extensive use of pesticides in agricultural practices and homes resulted in the discharge of pesticide residues into surface water bodies and this then contaminate fish living in such areas. The organochlorine pesticides (OCPs), organophosphorus (OPPs) and carbamates are critically toxic and carcinogenic species with half-lives of about 60 years (Boada et al., 2016).
Due to their high persistence, toxicity and their potential to bioaccumulate, it is highly imperative for their complete monitoring, regulation and removal once they contaminate the ecosystem.

Smoked and sun-dried fish are important sources of animal protein and provide nutrition to the poor and economically disadvantaged peoples of the world including Ghana. The fish processing culture in Ghana leads to the exposure of the fish to microbial, physical and chemical contamination right from the catch, through the processing, storage, transportation and final exhibition at various local markets.

2.18. Microbial Contamination of Salted, Smoked and sun Dried Fish

Foodborne pathogens of public health concerns are responsible for an increasing burden of disease globally (Appendix E). Assessing knowledge on the contribution of different sources of food and of water for disease-causing

microorganisms is essential to draw scale of preference for food safety interventions and implement appropriate control measures (Pires et al., 2012).

The impact of these disease causing microorganisms are even more alarming in the developing world where food safety and food security is on the rise. It is important to categorize the main sources of human illnesses, the available opportunities for extenuation and their associated costs.

To prioritize food safety interventions, one method to attribute human foodborne illnesses is to identify the responsible sources using microbiological approaches. In cold-smoking which results in ready-to-eat fish product, the processing temperature and the salt content is not enough to kill the microorganisms therefore cannot fully assured the absence of foodborne pathogens (Badr, 2015). This will lead to the potential for growth of foodborne pathogens in cold smoked fish or fish which is not properly smoked or sun dried. Therefore, any fish product which is not properly sun dried and well smoked is of a high risk of bacteria or fungi growth with a high fatality of approximately 30% exists (Badr, 2015). The prevention of bacterial and fungi growth would be very important in controlling foodborne diseases and safeguarding food safety and public health. Bacillus species were reported to be the most predominant bacteria in freshly smoked fish samples (Plahar et al., 1991).

The objective of this study was to identify and quantify the types of polycyclic aromatic hydrocarbons (PAHs), organochloride pesticides and microbiological contaminants in salted and sun-dried, smoked and sun dried fish sampled from nine coastal markets in Southern Ghana, and investigate the potential of gamma irradiation decontamination of these environmental

contaninants and to draw implications and prospects for Ghana's fisheries sector.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area

The study area encompassed Cape Coast, Elmina, and Mankesim markets in the Central Region; Chorkor, Kpong and Madina markets in the Greater Accra Region; and Akatsi, Keta and Sogakope markets in the Volta Region in Ghana (Figure 2). These are major market centres situated in municipalities and districts having major trunk roads passing from the national capital to other regions and neighbouring countries. These markets have vibrant fisheries activities and other produce supplied from nearby towns and fishing villages.

Figure 2: Map of Southern Ghana showing coastal markets and study sites (Source: Centre for Coastal Management, 2017).

3.2 Sampling Method

Convenience sampling method was used in selecting participants which included fish traders and processors in the selected markets. This was because the participants were readily and easily available. Typically, convenience sampling tends to be a favoured sampling technique and it is inexpensive and an easy option compared to other sampling techniques.

3.3 Fish Sample Collection

The sample collection, Focus Group Discussion and Participant Observation lasted for fourteen months (November 2016 to December 2017). Fish sampled included two sun-dried marine species of total 108 specimens each: *Engraulis encrasicolus* (*Engraulidae*; Linnaeus, 1758) and called "aborbi" in Ewe (Plate 4a), and *Selena dorsalis* (*Carangidae*; Gill, 1863) "Gbadzegbadze" in Ewe (Plate 4b) and two wood-smoked marine species *Sardinella aurita* (*Clupeidae*; Valenciennes, 1847) "Amani" in Akan (Plate 3) and *Engraulis encrasicolus*, total of 180 specimens each. Freshwater species sampled were: salted and sun-dried *Oreochromis niloticus* (*Cichlidae*; Linnaeus, 1758) "Akpatsu" in Ewe [Plate 6], (108 specimens), wood-smoked *Chrysichthys nigrodigitatus* (*Claroteidae*; Lacepède, 1803), "Blolovi" in Ewe [Plate 8], (180 specimens), *Oreochromis niloticus* [Plate 7], (180 specimens) and *Heterobranchus longifilis* (*Clariidae*; Valenciennes, 1840) called "Adeye" in Ewe [Plate 5] 180 specimens. These were obtained from the population of fishmongers in nine major markets using Stratified Random Sampling approach across three coastal zones of Ghana.

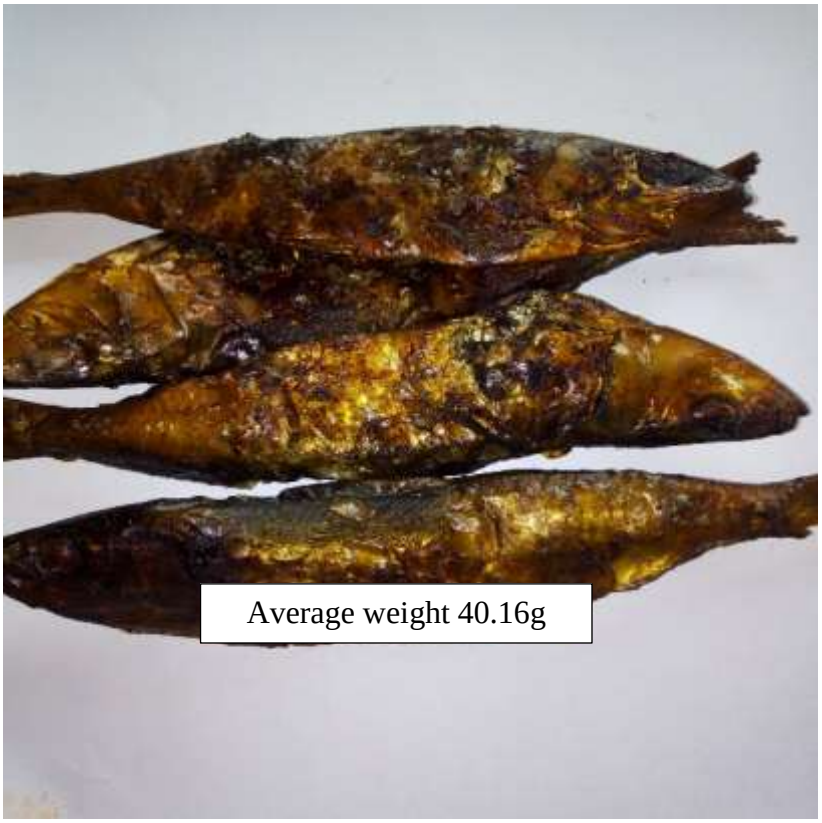


Plate 3: Smoked *Sardinella aurita* (Herring or "Amani"; Akan).

(A) anchovies ("Aborbi"; Ewe). (B) African moonfish.

Plate 4: (A) Sun dried *Engraulis encrasicolus.* (B) *Selena dorsalis* before gamma irradiation.

Plate 5: Smoked *Heterobranchus longifilis* ("Adeye" Ewe).

Plate 6: Salted and sun dried *Oreochromis niloticus* (Tilapia).

Plate 7: Smoked *Oreochromis niloticus* (Tilapia).

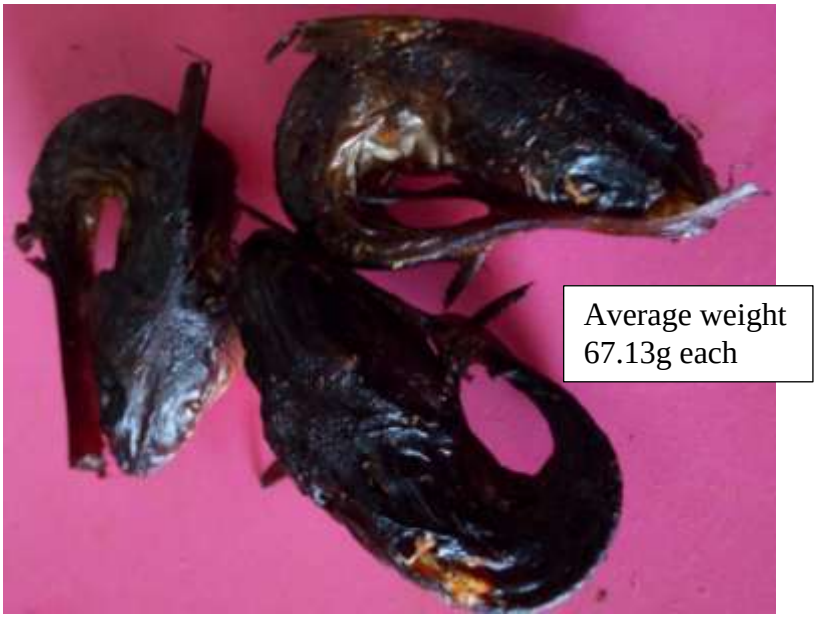


Plate 8: Smoked *Chrysichthys nigrodigitatus* ("Blolovi" Ewe).

The samples were selected randomly and packaged with polyethylene bags (Rapid-Grip^{R} 04) stacked on ice in insulated Styrofoam boxes, and then transported to the Ghana Atomic Energy Commission (GAEC) laboratory.

The research was carried out at the Radiation Technology Centre of the Ghana Atomic Energy Commission, the quality control laboratory of the Ghana Standards Authority and Animal Research Institute of the Council for Scientific and Industrial Research. Standard methods and standard operation procedures were used in all laboratory analyses.

3.4 Experimental Design

Total samples of 324 sun-dried fishes was used in this research based on a factorial completely randomized design (CRD) of 3x3x4x9 (3 species x 3 replicates x 4 doses x 9 markets). Additionally, total sample of smoked fishes used in this research was 540 based on a factorial CRD of 3x4x5x9 (3 replicates x 4 doses x 5 species x 9 markets).

3.5 Packaging

Combination packaging materials were used i.e. High-density polyethylene (HDPE), (Plates 10 &11) combined with Kraft paper (KP), (Plates 12 and 13) i.e. (HDPE+KP). Sub-samples were test-irradiated in similar packaging materials to determine the dose distribution rate using Ethanol chlorobenzene (ECB) dosimeter.

Plate 10: Smoked *Sardinella aurita* in HDPE 500mL Ziploc bags.

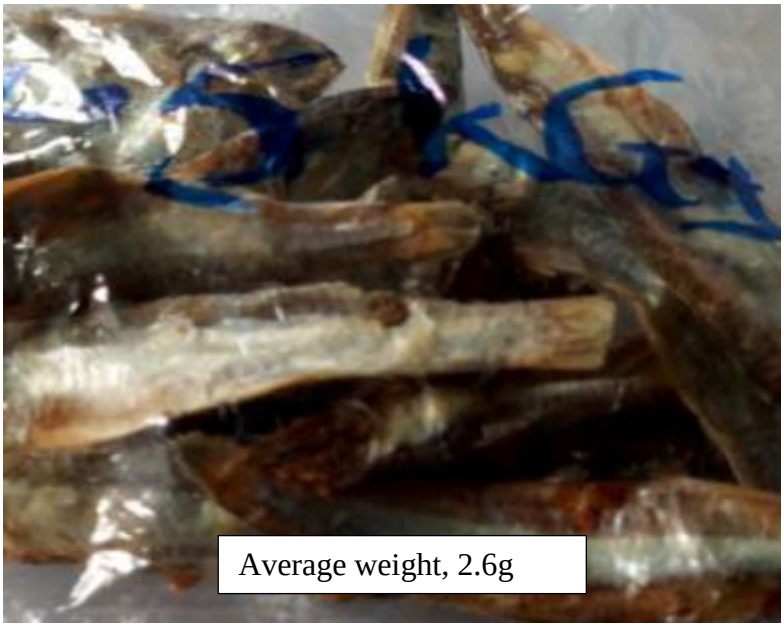


Plate 11: Sun dried *Engraulis encrasicolus* in HDPE 500mL Ziploc bags.

The samples were arranged in a lateral position and carefully wrapped in the HDPE Ziploc bags (Plates 10 and 11).

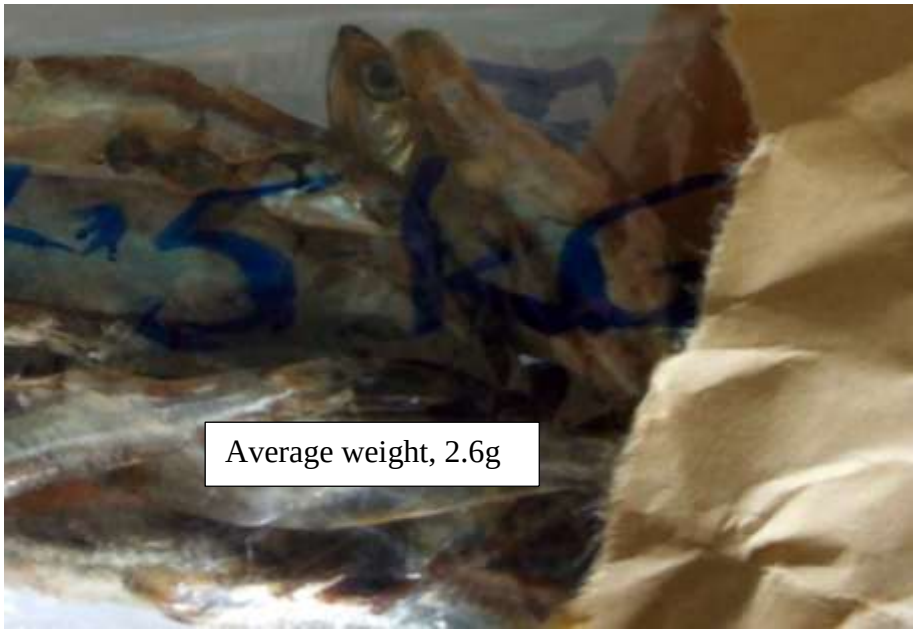


Plate 12: Sun dried *E. encrasicolus* in HDPE 500mL Ziploc bags and Kraft paper packaging.

In Plates 12 and 13, the samples that were packaged in HDPE Ziploc bags were then carefully wrapped tightly with Kraft paper. To avoid a baggy package, the Kraft paper was held together with rubber bangs.

Plate 13: Smoked *E. encrasicolus* in HDPE 500mL Ziploc bag and Kraft paper packaging.

3.6 Irradiation

Total sample of 864 were irradiated at predetermined doses: 2.5kGy, 5.0kGy and 7.5kGy (in air) using Cobalt-60 source at the Radiation Technology Center (RTC) of Ghana Atomic Energy Commission (GAEC). Samples were stored at ambient temperature (30 °C) to establish safety, quality and shelf life at days 0, 15, 30, 45 and 60 against controlled samples (0kGy).

3.7 Sample Preparation

A comparative evaluation before and after irradiation was carried out on the samples. Uniform samples were weighed using SciTech SL Chemical Balance (with maximum capacity of 5000g and a minimum of d = 0.01g) to determine the initial dry weight and the initial moisture content. Laboratory samples were freeze-dried using Freeze Dry/Shell Freeze System Labconco Freezone12, homogenized using a laboratory blender (Warring Commercial

Laboratory Blender) and then packaged in 70% ethanol sterilized 500mL Ziploc bags (Rapid-Grip^{R} 04).

Plate 9: Pre-digestion of smoked fish powder on gyration shaker.

These were stored in the freezer (Haier Thermocool). The initial pH, total titratable acidity (TTA), total ash, microbial load, free fatty acid (FFA), PAHs, OCPs and protein content were analyzed using standard methods (AOAC, 2000; Francisca, 2013). Thereafter, the samples were packaged, and test-irradiated to determine the rate of irradiation using Cobalt 60 gamma source at GAEC.

The precautions and requirements considered for sampling methods differ especially, depending on sample types and sizes in order to obtain samples that are representative for the respective fish samples. Each fish species which was to be examined was prepared separately. Larger species were subdivided into defined sub sections which were also sampled separately. Incremental samples were taken at various selected coastal markets

throughout the study area and combined into an aggregate sample. This was homogenized to represent the base material for analyses.

During the sampling and preparation of the samples, precautions were taken to avoid any changes in composition of the sample and ensure that samples do not become contaminated during sample preparation. The sampling containers were rinsed with high purity acetone or hexane before use to minimize the risk of cross contamination.

The apparatus and equipment coming into contact with the sample were of inert materials such as aluminum, glass or polished stainless steel. Materials that PAHs can adsorb onto were avoided. Such of those materials were plastics like polypropylene or polytetrafluoroethylene (PTFE). Losses of PAHs may also occur if the sample is exposed to light and high temperatures during the sample collection and storing, or PAHs react with other matrix substances during a long-term storage. Analytes were stored in brown vials in IGNIS (Model: RWN 130) with 130L storage volume capacity refrigerator kept at 4°C (Visciano et al., 2006).

3.8 Laboratory Analyses

Figure 3 outlined the evaluation of chemical hazards (PAHs) carried out before and after irradiation by Gas Chromatography and Mass Spectrometer (GC-MS) with limit of detection of 1.0 parts per billion [ppb] (AOAC, 2000).

Quantification of OCPs and synthetic pyrethroids was carried out before and after irradiation by Electron capture detector (ECD) and of organophosphorus by Pulse flame photoelectric detector (PFPD) with the limit

of detection of 0.01parts per million (ppm) or 10 parts per billion [ppb] (AOAC, 2000).

The summary of chromatographic procedure is schematically presented below:

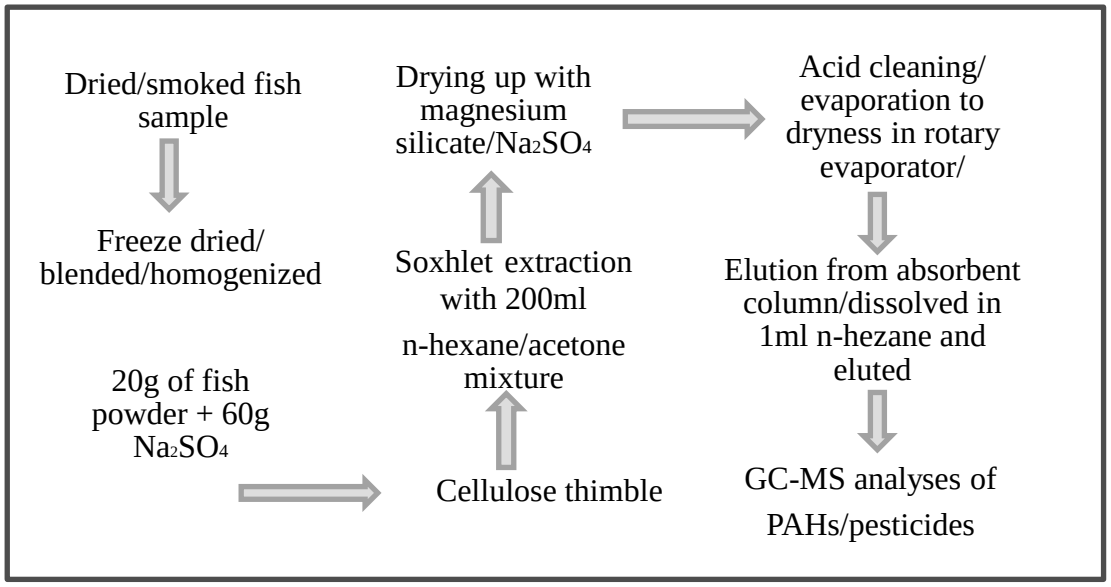


Figure 3: Flow chart of the extraction process of PAHs and Pesticides

The reagents used in the study of PAHs included the followings: n-hexane (95 % purity, Sigma-Aldrich quality), Sodium sulphate (Aldrich-Chemie quality, Germany), dichloromethane and acetone (99.5 %, BDH, England), a semi- volatile internal standard mixture with product code ISM-560, Lot number - CB-2411 containing 2000 µg/mL each of analytes of: Acenaphthene-d10, Chrysene-d12, Naphthlene- d8, Perylene-d12, and Phenanthrene-d10 in methylene chloride; p-terphenyl, product code: RAH-058, Lot number-NTO1690 with a concentration of 100 mg were all purchased from Ultra Scientific, USA.

A PAH custom standard of product code CUS-9059 and Lot number CD3298, containing 100g/mL each of analytes; naphthalene, acenaphthene, acenaphthylene, anthracene, benz(a)anthracene, benzo(a)pyrene, benzo(e) pyrene, bnzo(b)fluoranthene, benzo(j)fluoranthene, benzo(k)fluoranthene,

chrysene, benzo(g,h,i)perylene, dibenz(a,h)anthracene, fluoranthene, fluorene, indeno (1, 2, 3-c, d) pyrene, phenanthrene, pyrene, cyclopenta (c,d) pyrene and anthanthrene were purchased from Ultra Scientific, USA.

The sample preparation involved homogenization of freeze-dried fish samples, extraction of organochlorine and polycyclic aromatic hydrocarbons from the samples (with hexane-acetone) and drying them (with anhydrous sodium sulphate), pre-concentration of the extract by distillation, acid cleaning and separation of the extract on an adsorption column, final concentration (with nitrogen stripping solvent) of some chromatographic fractions.

Multiplicity of sulphuric acid treatment (applied 4 times depending on the fat content in the samples) to remove lipids from the extracts was controlled. Qualitative and quantitative analyses of the PAHs were carried out using AOAC, 2000 standard methods with Gas Chromatography-Mass Spectrometer D:\MassHunter\GCMS\1methods\PAH-methods\PAH-2019-10-14 Full Scan Parameters.M (AOAC, 2000). Qualitative and quantitative analyses of OCPs was carried out with GC, Varian CP-3800 GC-ECD with a CombiPAL Autosampler and that of OPP by GC, Varian CP-3800 GC-PFPD with a CombiPAL Autosampler all at the Ghana Standards Authority, Accra.

The operation conditions were as follows: The oven temperature was set initially at 70°C (2 minutes hold), increased to 300 °C at 25 °C/min. At 150 °C, temperature increased at a rate of 8 ºC/min to 280 ºC and then to 290 ºC at a rate of 5 ºC/min (13.133 minutes hold). Hydrogen with a purity of 99.999% and air, were as carrier gas at a constant flow of 30 and 300mL/min, respectively. The inlet temperature was maintained constant at 300 ºC with a pressure of 231.2 KPa and a total flow of 208mL/min. The detector heater was

maintained constant at 300 ºC with H_2/air flow at 35 and 300mL/min, respectively. Injection of 4mL of sample at a flow rate of 4mL/min each was performed in the split mode and the split valve was opened after 2 minutes.

The split ratio was 50:1. Identification of PAHs and OCPs in the samples was by comparison of the retention times with those in a standard solution, and quantification on the corresponding areas of the respective chromatograms. The results were statistically analysed using Minitab MPJ7777.MP Software and Excel 2013 Software. Significance was established at $p < 0.05$.

3.9 Evaluation of the quality and safety of Gamma Irradiated Salted, Smoked and sun-dried Fish

(i) Estimation of ash Content of Fish

The ash content of the fish samples was determined according to AOAC, 2007: Method 900.02 A. About 5g each of fish samples were weighed, using Mettler Toledo (Switzerland) electronic balance, in triplicates in pre-weighed quartz crucibles. The crucibles with the contents were transferred into a Muffle Furnace held at 600°C for 6 hours until all the organic materials were burnt out and the residues turned white. The crucibles were stored in desiccators with desiccant and allowed to cool considerably and later and were re-weighed. The percentage ash content was calculated as shown below (Farid et al., 2014).

$$\text{Calculation: \% ash content} = \frac{\text{weight of fish sample x100}}{\text{initial weight of fish x dry matter coefficient}} \quad (1)$$

Where dry matter coefficient = % solid/100

(ii) Estimation of pH of fish

The pH of each fish sample was determined separately according to standard methods. Ten gram (10g) of each fish sample was blended using Commercial Warring Laboratory blender 8010E (Model 38BL 40) and homogenized in 100mL of distilled water. This was centrifuged using SPECTRA Merlin centrifuge with a maximum revolution of 7000rpm for 6 minutes. Fifty millilitres (50ml) of the supernatant was measured and the pH determined according to AOAC, 2000: Method 10.028 using pH meter (Mettler Toledo 320) after the equipment had been calibrated (Farid et al., 2014).

(iii) Estimation of protein content of Fish

The protein content of the fish was determined by micro-Kjeldahl method by first digesting each sample separately with concentrated sulphuric acid according to AOAC, 2005. The steps included digestion of the samples with concentrated sulphuric acid in the presence of potassium permanganate which aided the oxidation and conversion of nitrogen into ammonium sulphate.

The digest was diluted and neutralized with sodium hydroxide after which it was distilled into boric acid with potassium iodide and potassium iodate as indicator. The liberated ammonia was determined by titrating with standard sodium thiosulphate and the results calculated as below (AOAC, 2000).

Calculation: Total Protein content

$$\%N2 = N_2 \times 5.6 \text{ (for fish)}$$

$$= \frac{(\text{titre volume x acid molarity x0.002 x 200}}{\text{weight of sample}} \quad (2)$$

(iv) Estimation of moisture content of processed Fish

Moisture was analysed using the gravimetric method (Hotbox oven with maximum temperature of 200^0C) at 130^0C for 1 hour (AOAC, 2000) About 5g of triplicate samples were accurately weighed in Petri-plates with covers over them by using Mettler Toledo (Switzerland) electronic balance.

The uncovered samples were dried in Gallenkamp Hot Box oven, (Size 1 with a maximum temperature of 200 ± 2ºC) at 130ºC for 1 hour (1 hour drying time began when oven temperature was actually 130°C). The Petri-plates were covered again after drying while still in the oven and transferred into desiccators.

After reaching room temperature, they were re-weighed using the same digital electronic balance. The percentage weight losses were reported as moisture content (with standard error) of the fish (AOAC, 2000).

Calculation:

$$\% \text{ Moisture content}$$

$$= \frac{\text{initial weight of sample} - \text{final weight of samplex100}}{\text{initial weight of sample}} \quad (3)$$

Free fatty acids, total ash, TTA and pH were analysed using standard methods

(iv) Estimation of free fatty acid (FFA) content of fish

About 5g of the homogenous sample was taken into conical flasks and 10ml of Folch Reagent (Chloroform-Methanol = 2:1) was added to the

sample, homogenized for about two minutes and kept tight in storage overnight according to the method of AOAC Method: 945.16 (8)

(Suzanne Nielsen, 2014)

Fat contents of the fish samples react with the solvent and remains in the solution. After 24 hours, the solution was filtered in another pre-weighed conical flask through a filter paper (Whatman No.42, Ashless, and 110mm).

These flasks were placed over hot water bath to dry up by evaporation and the flasks were kept in a hot oven for an hour. The flasks were re-weighed using Mettler Toledo (Switzerland) electronic balance to estimate the amount of fat contents after they had cooled (AOAC, 2000).

Calculation:

$$\% \text{ Fat (dry weight)} = \frac{\text{weight of residues x } 100}{\text{weight of sample}} \qquad (4)$$

(v) Estimation of total titratable acid (TTA) of fish

The measurement of TTA was based on the titration of organic acids, mainly free fatty acids in fish. Approximately 10g of the fish were randomly selected and blended with 100ml of distilled water using Commercial Warring Laboratory blender 8010E (Model 38BL 40) and was left to stand for about 30 minutes. This was decanted and 25mL of the supernatant was measured with a pipette into 250mL conical flask.

This aliquot was diluted with distilled water to minimize interferences from coloured and turbid samples. Three drops of 1% phenolphthalein were added and titrated with 0.1M NaOH. The sample temperature was kept at approximately 25°C during the titration. The result was reported as volume/mL of NaOH required for extracting from 1g of sample and converted

to gramme of fatty acid/10g of fish, equivalent to percentage (%) free fatty acid which represents the volume of NaOH used and multiply by 2.28 (AOAC, 2005: AOAC Method, 20.43).

(vi) Evaluation of biological hazards in fish

Ten gram of fish samples were aseptically homogenized for two to three minutes using a stomacher laboratory blender in a sterile stomacher bag containing 90mL of sterile 0.1% peptone water. This was serially diluted with fish samples and used for the microbial analysis.

Total viable bacterial count (TVBC) was determined by the standard spread plate method according to the American Public Health Association (APHA, 1996) method using spread plate technique. Nutrient agar (pH = 7.0 – 7.4) was used to determine TVBC as well as for isolation of microorganisms.

Plates were incubated at 37°C for 24 hours and the count was expressed as colony-forming unit per gram (cfu/g). Total coliform count and total fungal count were obtained in the same way using McConkey agar and Potato Dextrose Agar medium respectively (Akinjogunla, 2011; Mahin, 2011; Chaudhry, 2012).

Spoilage and pathogenic microorganism e.g. faecal coliform count were determined using phosphate-buffered saline/Violet Red Bile Agar (VRBA) at 44^0C for 48 hours. McConekey agar and Potato Dextrose agar were incubated at 37°C for 24 to 48 hours and 28°C for 5 days respectively. Microscopic examination of parasitic eggs, cysts and larvae was carried out using X100 and X60 objectives of the Light Microscope (Garcia, 2007).

(a) Isolation and identification

After overnight incubation, colonial morphology of organisms based on their physiological characteristics were studied for size, shape, outline, colour and change in medium on various media. Standard microbiological techniques including Gram staining, cellular morphology [of organisms using compound microscope magnified at x100 with oil immersion] and biochemical tests such as Motility Indole Urea (MIU) [Lioflichems.r.l. Bacteriology Products, 610236, Italy], Catalase, Triple Sugar Iron (TSI) [Oxoid, CM 0277, Hamsphire – England], Indole Methyl Red Vorges-Proskeur Citrate [IMViC] test, carbohydrates Oxidation/Fermentation (O/F) test [to detect gas and or acid production] among others were applied to isolate and identify the organisms.

(b) Culture, isolation and identification of *Salmonella*

Using the plate-out technique, subcultures were made from the Selenite F broth aseptically onto XLD agar [Oxoid, CM 469, Hamsphire – England]. Cultures were incubated at 37^{o}C for 24 – 48 hours and examined for the physiological characteristics of colonies on the media.

(c) Culture, Isolation, Identification of *Escherichia coli*

All lactose fermenting colonies on MacConkey agar were selected and aseptically subcultured onto MLGA [Oxoid, CM 1031 Hamsphire – England] to isolate and identify *E. coli*. Cultures were incubated at 45^{o}C for 24-48 hours in a bacteriological incubator.

(d) Culture, Isolation, Identification of *Staphilococcus aureus*

After the examination of the physiological characteristics of colonies on Blood Agar (BA) and the cell morphology (Gram stain) using light compound microscope at x100 magnification, all colonies showing Gram positive cocci

in clusters (halophiles) were subcultured unto Mannitol Salt Agar (MSA) [Oxoid, CM 0085, Hamsphire – England] and incubated at 37^{o}C for 18-24 hours in a bacteriological incubator.

3.10 Qualitative Study

This study involved two qualitative structured questionnaires to collect data on participants. These included Focus Group Discussion (FGD) and Participant Observation (PO).

(i) Focus group discussion (FGD)

Focus group discussion was conducted to explore perceptions and practices of women involved in fish handling from the selected coastal markets in Southern Ghana. Prior to recruiting participants, market queens in each market were contacted personally to discuss the purpose of this research to seek their interest. The market queens from each of the nine markets with the support of the researcher randomly sampled eight or ten members from their existing market associations. All prospective participants were contacted verbally in person to confirm their interest and availability for the discussion.

In defining the focus group inclusion criteria, fish traders who were more than five years old in the fish trade were sampled and fish traders who were less than five years including hawkers and floating traders in the trade were excluded in recruiting participant so that the group will be homogeneous. The team was made up of a Moderator (Student investigator) and two Assistant Moderators. A convenient venue or an available area was arranged by a member of the participating groups for this exercises in Madina, Elmina, Mankesim, Keta, Akatsi and Sogakope markets. The FGDs were conducted mainly in the Twi (Akan) language which was the preferred language by the

participants for the discussion in the Central Region and both Ga and Akan in the Greater Accra Region. In the Volta Region, Ewe language was the preferred medium used in the conversation and interaction. Each participant was made to mention her initials, the first time she responded to a question to aid voice identification during the transcription as quality control measure. Aiming to ensure order, questions on one thematic area were exhausted before moving on to the next. However, if a participant wanted to make additions to an issue already discussed she was allowed to contribute.

In all the focus groups, five thematic areas were explored: knowledge of fish safety, cost benefit analysis of the current technology, levels of concern about losses, forms of preservation, and knowledge of packaging and storage (Appendix B).

Discussions lasted an average of seventy-five minutes, ranging in length of time from about fifty-five to hundred minutes. All discussions were audio-recorded and in addition notes were taken while interactions were ongoing to capture non-verbal responses. An assessment was done after each discussion by both moderator and observers to ensure the quality of subsequent focus group discussions and the recordings then transcribed appropriately and emphasized by field notes taken during the interview.

Three cakes of toilet soap and or kitchen knives and transportation fare of GHC 20.00 were presented to each participant as motivation for involvement in the exercise.

The audio recordings were typed verbatim and translated into English by listening to all the tapes several times and comparing with the observers' notes until all the details of information were captured in the transcripts. Each

transcript consisted of mainly questions asked by the moderator with corresponding responses by each participant in the order in which the interactions took place. For the purposes of easy identification and comparison of themes, each transcript was written on numbered sheets of paper for each focus group discussion. A final transcript containing records for all focus groups was typed out.

From the transcripts, major themes used in the discussion were recorded in a codebook that was designed prior to analysis. Similar quotes were grouped systematically in a tabular form under their appropriate themes and the data was presented in figures and tables.

(ii) Participants' Observation

The study was conducted in all the nine coastal markets in the study area (Figure 3). A maximum of fifteen and a minimum of ten fish processors and traders were observed during fish handling procedures. The guiding questions (Appendix C) were solely non-verbal and included the study area, the opportunities that were available at the area for observation, the representativeness of the participants of the population at that site, and the data recording strategies (Iacono, Brown, & Holtham, 2009). Three types of participant observation were employed in this study. Every manoeuvres and processes involved in fish post harvest handling were observed and described while assuming having no knowledge of the environment or the situation. Secondly, participants were observed based on insights from focus group interviews with the same participants. And finally, different selected activities involved in fish processing, packaging and storage were observed to distinguish the differences between each activity. This offered first-hand idea

about how the processing operations were organized and prioritized with respect to how people interrelate safety to quality of fish, cultural parameters that inform their knowledge and practices in the nine markets. Data was collected on compliance or non-compliance on ten critical areas such as: environmental hygiene, layout of processing area, personal hygiene, control of fish processing operations, raw materials, fish handling procedure, water quality, storage area, packaging materials, pest control, waste management and cleaning programme, and cultural manoeuvres to check for non-verbal expression of feelings.

CHAPTER FOUR

4.0 RESULTS

4.1 Assessment of Polycyclic Aromatic Hydrocarbon (PAH) Residues in Salted, Smoked and Sun-Dried Fish from selected Coastal Markets in Southern Ghana

Eighteen (18) individual polycyclic aromatic hydrocarbons (PAHs) with varying concentrations were identified in 20g each of homogenized fish samples powder of salted and sun-dried Tilapia "Akpatogui", sun-dried or smoked anchovy ("aborbi"), African moonfish (Gbadzegbadze), and smoked Sardine "Amani", *Heterobranchus longifilis* "adeye" and *Chrysichthys nigrodigitatus* "Blolovi" fish from the coastal markets in Ghana. These were:

Naphthalene, Fluorine, Anthracene, 1-Methylnaphthalene, Benzo(a)pyrene, 2-Methylnaphthalene, Acenaphthyelene, Acenaphthene, Phenanthrene, Chrysene, Fluoranthene, Benzo(g,h,i)perylene, Pyrene, Benzo(a)anthracene, Benzo(k)fluoranthene, Benzo(b)fluoranthene, Dibenzo(a, h)anthracene, and Indenol(1,2,3,c,d)pyrene. However, the focus of this thesis was on polycyclic aromatic hydrocarbons regarded as potentially genotoxic and carcinogenic to humans. These were Chrysene, Benzo(a)anthracene [B(a)A], Benzo(k)fluoranthene [B(k)F], Benzo(a)pyrene [B(a)P], Benzo(b)fluoranthene [B(b)F], Benzo(g,h,i)perylene [B(g, h, i)P] and Indenol(1,2,3,c,d)pyrene [In(1, 2, 3, c-d)P].

4.2 Analysis of PAHs in Smoked Fish

The results of individual PAHs found in smoked fish sampled from the three coastal regions and nine markets (Tables 5 to 9, Figures 4 and 5). These tables and figures show the regions, towns and market locations of the smoked

fish species with the different PAHs distribution in smoked fish which were sampled from the selected coastal markets in Southern Ghana.

Table 5, Figures 4 and 5 presented seven (7) types of PAHs found in smoked *Sardinella aurita* sampled from markets in the Central, Greater Accra and Volta Regions of Ghana. Chrysene, and Benzo(k)fluoranthrene were not detected in smoked *S. aurita* sampled from the Volta Region.

Table 5: PAHs found in smoked *Sardinella aurita* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL	GREATER ACCRA	VOLTA
Chrysene	Cape Coast/Elmina	Chorkor/Madina	nd
Benzo(a)anthracene	nd	Madina	nd
Benzo(k)fluoranthrene	Cape Coast/Elmina	Chorkor/Madina	Akatsi/Keta
Benzo(a)pyrene	Cape Coast/Elmina	Madina	Akatsi/Keta
Benzo(g,h,i)perylene	Cape Coast/Elmina	Madina/Kpong/Tema	Akatsi/Keta
Indeno(1,2,3,c,d)pyrene	nd	Madina/Kpong	Akatsi/Keta
Benzo(b)fluoranthene	nd	Madina/Kpong	Akatsi/Keta

nd = not detected.

Benzo(a)Anthracene Benzo(b)flouranthrene and Indeno(1,2,3, c,d)pyrene were not detected in smoked *Sardinella aurita* sampled from Central Region of Ghana (Cape Coast/Elmina). Chrysene, Benzo(a)anthracene, Indeno(1,2,3,c,d)pyrene and Benzo(b)flouranthene were detected in smoked *Sardinella aurita* sampled from Greater Accra Region

(Madina/Chorkor). From the markets in the Volta Region, Benzo(a)anthracene and Chrysene were not detected in the smoked *S. aurita*.

Table 6: PAHs found in smoked *Oreochromis niloticus* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	**GREATER ACCRA**	**VOLTA REGION**
Chrysene	Cape Coast /Mankesim	Kpong/Madina	Akatsi
Benzo(a)anthracene	Cape Coast/Mankesim	Kpong/Madina	Akats
Benzo(k)fluoranthrene	Cape Coast/Mankesim	Kpong/Madina	Akatsi
Benzo(a)pyrene	Cape Coast/Mankesim	Kpong/Madina	Akatsi
Benzo(g,h,i)perylene	Cape Coast/Mankesim	Kpong/Madina	Akats
Indeno(1,2,3,c,d)pyrene	Cape Coast/Mankesim	Kpong/Madina	Akatsi
Benzo(b)fluoranthene	Cape Coast/Mankesim	Kpong/Madina	Akatsi

Table 6, Figures 5 and 6 described the kind of PAHs found in freshwater species; smoked *Oreochromis niloticus* sampled from markets in the Central, Greater Accra and Volta Regions of Ghana.

Benzo(g,h,i)perylene, Benzo(b)fluoranthene, Benzo(a)anthracene, Chrysene, Benzo(a)pyrene, Benzo(k)fluoranthrene, Indeno(1,2,3,c,d)pyrene, and were detected in all the three coastal regions and corresponding markets under study (Akatsi, Madina/Kpong/Chorkor and Cape Coast/Mankesim).

Table 7: PAHs found in smoked *Chrysichthys nigrodigitatus* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	GREATER ACCRA	VOLTA REGION
Chrysene	Mankesim	Kpong/Madina	Akatsi
Benzo(a)anthracene	Mankesim	Kpong/Madina	Akatsi
Benzo(k)fluoranthrene	Mankesim	KpongMadina	Akatsi
Benzo(a)pyrene	Mankesim	Kpong/Madina	Akatsi
Benzo(g,h,i)perylene	Mankesim	KpongMadina	Akatsi
Indeno(1,2,3,c,d)pyrene,	Mankesim	Kpong/Madina	Akatsi
Benzo(b)fluoranthene	Mankesim	KpongMadina	Akatsi

Seven (7) PAHs were detected in freshwater smoked C*hrysichthys nigrodigitatus* sampled from the three coastal regions of Ghana: Central, Greater Accra and Volta (Table 7, Figures 4 and 5).

These seven (7) PAHs were: Chrysene, Benzo(a)anthracene, Benzo(k)fluoranthrene, Benzo(g,h,i)perylene, Indeno(1,2,3,c,d)pyrene, Benzo(a)pyrene and Benzo(b)fluoranthene were detected in smoked C*hrysichthys nigrodigitatus* sampled from Mankesim, Madina, Kpong and Akatsi markets respectively (Table 7 and Figure 5).

Smoked *Heterobranchus longifilis* is one of the important freshwater fish-protein foods and is valued for its nutritional qualities in rural and urban Ghana. Study of the safety of this cherished species revealed the accumulation of seven (7) PAHs (Table 8, Figures 4 & 5). These were Chrysene,

Indeno(1,2,3,c,d)pyrene, Benzo(a)anthracene, Benzo(k)fluoranthrene, Benzo(g,h,i)perylene, Benzo(a)pyrene and Benzo(b)fluoranthene. These PAHs were found in smoked *Heterobranchus longifilis* sampled from Mankesim, Madina, Kpong and Akatsi markets.

Table 8: PAHs found in smoked *Heterobranchus longifilis* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	**GREATER ACCRA**	**VOLTA REGION**
Chrysene	Mankesim	Kpong/Madina	Akats/Sogakope
Benzo(a)anthracene	Mankesim	Kpong/Madina	Akatsi/Sogakope
Benzo(k)fluoranthrene	Mankesim	KpongMadina	Akatsi/Sogakope
Benzo(a)pyrene	Mankesim	Kpong/Madina	Akats/Sogakope
Benzo(g,h,i)perylene	Mankesim	KpongMadina	Akatsi/Sogakope
Indeno(1,2,3,c,d)pyrene	Mankesim	Kpong/Madina	Akatsi/Sogakope
Benzo(b)fluoranthene	Mankesim	KpongMadina	Akatsi/Sogakope

Table 9, Figures 4 and 5 displayed seven (7) PAHs detected in smoked *Engraulis encrasicolus* sampled from all the Coastal regions of Ghana. These were Benzo(a)anthracene, Benzo(a)pyrene, Benzo(k)fluoranthrene, Chrysene Benzo(g,h,i)perylene, Indeno(1,2,3,c,d)pyrene, and Benzo(b)fluoranthene.

Table 9: PAHs found in smoked *Engraulis encrasicolus* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	GREATER ACCRA	VOLTA REGION
Chrysene	Elmina/Mankesim	Madina	Akatsi/Keta
Benzo(a)anthracene	Elmina/Mankesim	Madina	Akatsi/Keta
Benzo(k)fluoranthrene	Elmina/Mankesim	Madina	Akatsi/Keta
Benzo(a)pyrene	Elmina/Mankesim	Madina	Akatsi/Keta
Benzo(g,h,i)perylene	Elmina/Mankesim	Madina	Akatsi/Keta
Indeno(1,2,3,c,d)pyrene	Elmina/Mankesim	Madina	Akatsi/Keta
Benzo(b)fluoranthene	Elmina/Mankesim	Madina	Akatsi/Keta

In Figure 4, the highest concentration of Chrysene occurred in smoked *O. niloticus* as 17.04 ± 2.22 µg/kg and the lowest being 8.70 ± 2.96µg/kg occurred in smoked *Heterobranchu longifilis*.

Among the smoked fish samples, B(a)A (p = 0.000) was highest in smoked *O. niloticus* 14.13 ± 2.26µg/kg, and lowest in smoked *E. encrasicolus* (1.77 ± 2.24µg/kg.

B(k)F was highest in smoked *C. nigrodigitatus* (29.05 ± 2.34µg/kg. Smoked *O. niloticus* recorded 28.28 ± 2.51µg/kg and smoked *H. longifilis* recorded 27-24 ± 2.62µg/kg and *S. aurita* recorded the lowest concentration of 1.66 ± 3.13µg/kg for B(k)F. These results were significant at 95% Cl, (p = 0.000).

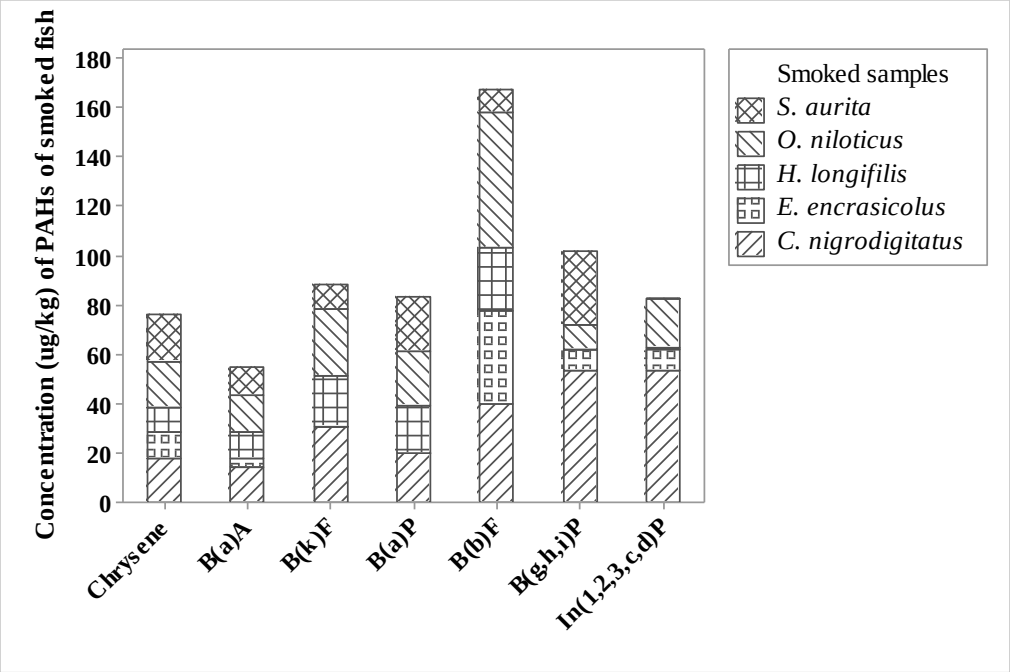


Figure 4: Mean concentrations of PAH found in smoked fishes from selected coastal Markets in Southern Ghana.

The hghest mean concentration of B(a)P was recoded in smoked *S. aurita* (17.85 ± 3.25µg/kg), *Heterobranchus longifilis* 8.51 ± 3.25µg/kg and the least value of B(a)P was recorded in smoked *E. encrasicolus* (0.97 ± 3.25µg/kg). B(b)F was highest in smoked *O. niloticus* (54.47 ± 8.80µg/kg) but lowest in smoked *E. encrasicolus* (8.72 ± 10.30µg/kg). B(k)F also recorded highest value among the smoked samples with 28.95 ± 8.10µg/kg) in *C. nigrodigitatus* and the lowest below detection level in *E. encrasicolus*.

Among the smoked fish, B(g,h,i)P was highest in *C. nigrodigitatus* (35.05 ± 6.46µg/kg) and lowest in smoked *H. longifilis* (2.97 ± 7.25µg/kg). Between the smoked samples, *C. nigrodigitatus* recorded (36.08 ± 6.29µg/kg) and *E. encrasicolus* was below detection level of B(k)F.

Figure 5 depicts the mean concentrations of PAH which contaminated smoked fishes sampled from selected markets in Sourthen Ghana. Mean concentration of Chrysene was 15.26µg/kg ± 5.64 at 95% confidence interval

and (p>0.05). B(a)A concentration was 11.01µg/kg ±15.17, (p ≥ 0.05), and B(k)F was 17.67µg/kg ± 8.87, (p ≥ 0.05). The mean concentration of B(a)P was 16.74 µg/kg ± 26.94, (p ≥ 0.05) and B(b)F was 33.55µg/kg ±26.94, (p ≥ 0.05). B(g,h,i)P was 20.45µg/kg ± 31.81, (p ≥ 0.05), and In(1,2,3,c-d)P was 16.55µg/kg ±26.48, (p ≥ 0.05). The analysis showed statistically significant differences (p = 0.006) (Appendix D) in the concentrations of the PAHs detected in smoked fish in the Ghanaian markets (Figures 5 & 6).

In Figure 5, there was a changing aspect in the occurance of PAHs in smoked fish sampled from the Coastal Regions in this research. Smoked fish sampled from the Central Region recorded the highest concentration 23.09 ± 21.5 µg/kg, Greater Accra recorded 3.60 ± 3.73 µg/kg while Volta Region recorded 3.82 ± 2.71 µg/kg µg/kg of In(1,2,3, c-d)P.

Central Region recorded 29.07 ± 1.09 µg/kg of Chrysene, Greater Accra Region 12.12 ± 2.41 µg/kg and Volta Region recorded 9.12 ± 1.33 µg/kg in smoked fish sampled. Central Region recorded 11.41 µg/kg ± 0.65, Greater Accra Region 11.41 ± 2.93 µg/kg and Volta Region recorded 10.23 ± 4.39 of B(a)A. Central Region recorded 10.23 ± 8.15 µg/kg, Greater Accra Region 12.97 ± 11.62 µg/kg and Volta Region 7.74 ± 10.27 µg/kg of B(k)F.

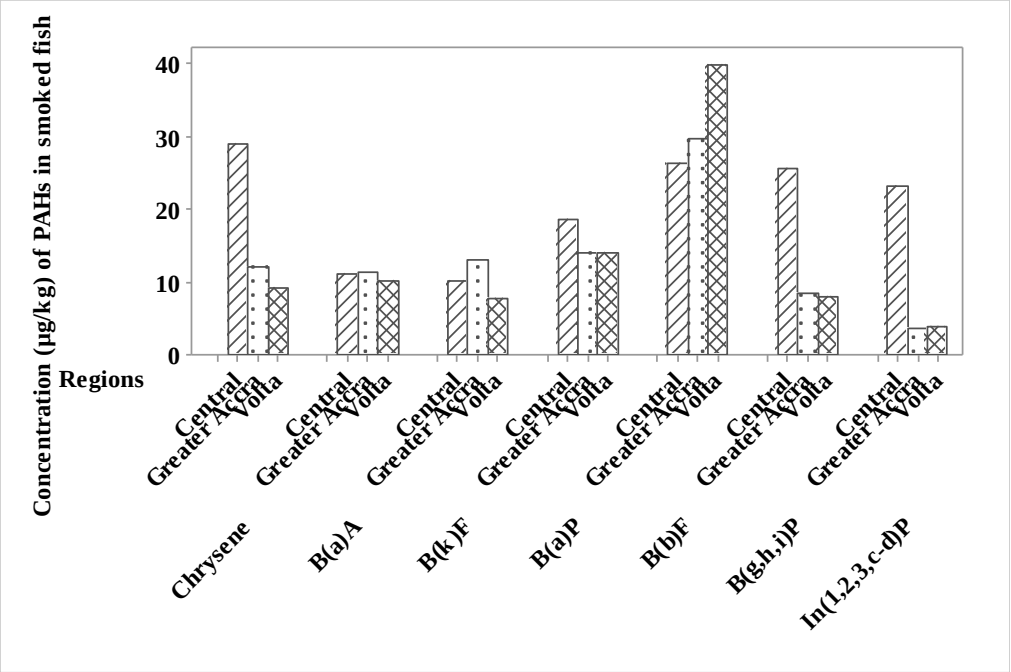


Figure 5: Regional distribution of PAHs in smoked fish sampled from selected coastal markets in Southern Ghana.

Central Region recorded the highest concentration of 18.70 ± 16.75 µg/kg B(a)P, Greater Accra Region 13.95 ± 2.85 of B(a)P and Volta Region recorded 14.1 ± 1.0. In the Greater Accra Region, B(b)F recorded 29.68 ± 15.87 µg/kg, Central Region 26.2 ± 18.8 µg/kg and the Volta Region 39.9 ± 25.2 µg/kg. The highest mean concentration of B(g, h,i)P was 25.59 ± 22.0 µg/kg in Central Region, 8.51 ± 2.92 µg/kg in the Greater Accra Region and 7.93 ± 3,6 µg/kg in Volta Region. The Central Region recorded the highest concentrations of In(1,2,3,c-d)P, B(a)P, Chrysene and B(g,h,i)P. The Greater Accra Region recorded the highest of B(a)A, B(k)F and B(b)F while Volta Region recorded the highest in only one of the seven PAHs, this was B(b)F.

The Relative Abundance Ratio of these polycyclic aromatic hydrocarbons in smoked fish PAH[(RAR)] were calculated as the quotient of the highest and lowest values for each PAHs.

The ratios were: RAR[B(a)A] = 2.69, RAR[B(b)F] = 6.86, RAR[B(g,h,i)P] = 1.80, RAR[B(k)F] = 2.03, RAR[(B(a)P] = 3.65, RAR[(Ind(1,2,3,c,d)P] = 1.0 and RAR[Chrysene] = 2.39. B(b)F scored the highest ratio of 6.86 in the smoked fish samples and B(a)P scored 3.65.

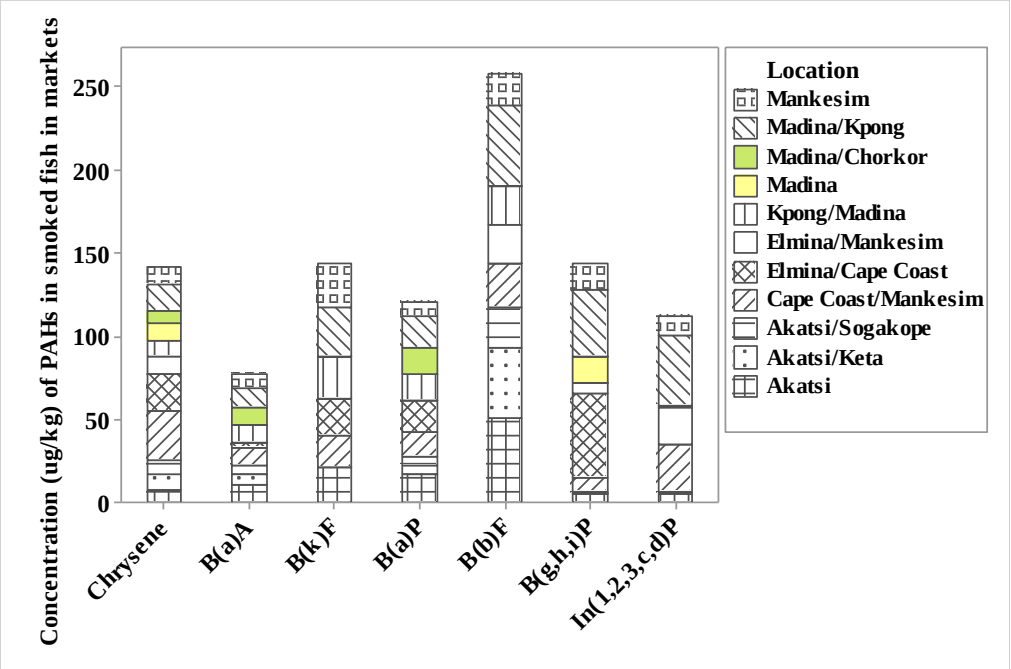


Figure 6: Occurance of polycyclic aromatic hydrocarbons in smoked fish according to markets in Southern Ghana.

Figure 6 showed the market distribution of PAHs in smoked fish sampled from the coastal regions. The aggregate subtotal concentration of Chrycene was 141.10 µg/kg: the lowest 6.88 ± 4.55 µg/kg occurred in Akatsi and the highest 29.83 ± 1.01 µg/kg in Mankesim ($p \leq 0.000$). B(a)A recorded a total of 77.52 µg/kg with lowest of 2.82 ± 5.65 µg/kg in Elmina and Cape Coast, and the highest of 11.34 ± 1.23 µg/kg in Madina and Kpong ($p \leq 0.000$). A total of 144.15 µg/kg was realized in B(k)F with lowest 19.45 ±0.57 µg/kg recorded in Cape Coast and Mankesim, and the highest 27.06 ± 9.70 µg/kg in Madina and Kpong ($p \leq 0.05$). B(a)P recorded a total of 120.88 µg/kg

with lowest of 4.99 ± 9.99 µg/kg recorded in Akatsi and Sogakofe, and the highest 19.43 ± 4.25 µg/kg in Madina and Kpong ($p \leq 0.05$). B(b)F recorded a total of 258.22 µg/kg with 19.06 ± 11.79 µg/kg being the lowest in Mankesim and 50.40 ± 17.56 µg/kg the highest in Akatsi ($p \leq 0.05$). B(g, h,i)P realized a total concentration of 141.14 ± 5.01 µg/kg being the lowest in Akatsi and Keta, the highest of 50.36 ± 25.90 µg/kg was recorded in Elmina and Cape Coast ($p \leq 0.05$). A total of 122.03 µg/kg of In(I,2,3,c-d)P was recorded with the lowest of 0.86 ± 0.42 µg/kg in Akatsi and Keta, and the highest of 42.33 ± 35.10 µg/kg recorded in Madina and Kpong ($p \leq 0.05$).

4.3 Analysis of PAHs in Sundried Fish

In Table 10, sundried *Engraulis encrasicolus* sampled from Winneba in the Central Region, Madina in the Greater Accra Region, Akatsi and Keta in the Volta Regions showed seven (7) PAHs of various kinds. Four of these PAHs were identified in the Central Region and seven each in the other regions of Ghana.

The One-way analysis of variance, (Appendix D), at 95% confidence interval (CI), showed statistically significant differences ($p \leq 0.05$) in the concentrations of the PAHs recorded in sundried *Engraulis encrasicolus*.

In Figure 7, sun-dried *Engraulis encrasicolus* recorded 10.84 ± 0.00 µg/kg of chrysene ($p \geq 0.05$) but recorded no value for B(a)A because it was below detection limit as well as B(k)F. It however, recorded 6.54 ± 11.32 µg/kg of B(a)P ($p \geq 0.05$), 26.84 ± 1.01 µg/kg of B(b)F ($p \leq 0.05$), 15.34 ± 13.28 µg/kg

of B(g, h, i)P ($p \geq 0.05$) and 15.34 ± 13.28 µg/kg of In(1, 2, 3, c – d)P ($p \geq 0.05$). The PAH with the highest concentration in sundried *Engraulis encrasicolus* was B(b)F and the least were B(a)A and B(k)F.

Table 10: PAHs found in dried *Engraulis encrasicolus* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	GREATER ACCRA	VOLTA REGION
Chrysene	Winneba/ Mankesim	Madina/Kpong/ Tema	Akatsi
Benzo(a)anthracene	Winneba/ Mankesim	Madina/Tema/ Kpong	Akatsi
Benzo(k)fluoranthrene	nd	Madina/Kpong	Akatsi/Keta
Benzo(a)pyrene	Winneba/ Mankesim	Madina	Akatsi/Keta
Benzo(g,h,i)perylene	nd	Madina/Tema	Akatsi/Keta
Indeno(1,2,3,c,d)pyrene	nd	Madina/Tema	Akatsi/Keta
Benzo(b)fluoranthene	Winneba	Madina/Kpong	AkatsiKeta

nd: not detected.

In Table 11, the study of safety of dried *S. dorsalis* revealed four PAHs identified in the Central Region of Ghana. Seven each were identified in Greater Accra and Volta Regions of Ghana. *Selena dorsalis* is a common fish that is fried and eaten with ‘kenkey’ or “banku” in Ghana.

In Figure 7, the analysis of variance showed that *S. dosalis* contained 11.23 ± 0.00 µg/kg of chrysene, 11.03 ± 0.00 µg/kg of B(a)A, 19.65 ± 0.00 µg/kg of B(a)P and 90.28 ± 0.00 µg/kg of B(b)F ($p \leq 0.05$). However, B(k)F, B(g,h,i)P and In(1, 2, 3, c-d)P were below detection limit. The PAH with the

highest concentration in *S. dosalis* was B(b)F and this was significantly higher than the other PAHs recorded in *S. dosalis*.

Table 11: PAHs found in dried *Selena dorsalis* sampled from selected coastal markets in Southern Ghana.

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	GREATER ACCRA	VOLTA REGION
Chrysene	Mankesim/ Winneba	Kpong/Madina	Akatsi
Benzo(a)anthracene	Mankesim/ Winneba	Kpong/Madina/ Tema	Akatsi
Benzo(k)fluoranthrene	nd	nd	nd
Benzo(a)pyrene	Mankesim/ Winneba	Kpong/Madina	Akatsi
Benzo(g,h,i)perylene	nd	nd	nd
Indeno(1,2,3,c,d)pyrene	nd	nd	nd
Benzo(b)fluoranthene	Mankesim/ Winneba	Madina/Tema	Akatsi

nd = not detected.

In Table 12, four PAHs of significance were identified in salted and sundried *Oreochromis niloticus* sampled from the Central Region of Ghana. Seven each were identified in the Greater Accra and Volta Regions of Ghana. Dried salted *Oreochromis niloticus* is a cured or fermented fish product.

In Figure 7, dried salted *Oreochromis niloticus* recorded 19.62 ± 8.26 µg/kg of chrysene, 19.57 ± 7.33 µg/kg of B(a)A, 30.67 ± 0.03 µg/kg of B(k)F. It also recorded 19.77 ± 0.03 µg/kg of B(a)P, 25.92 ± 0.13 µg/kg of B(b)F, 1.95 ± 1.69 µg/kg of B(g, h, i)P and 1.95 ± 1.69 µg/kg of In(1, 2, 3, c-d)P. The PAH with the highest concentration; 30.67 ± 0.03 µg/kg, in this sample was

B(k)F, and being significantly ($p = 0.000$) different from the others in the same sample. Other PAHs with significant ($p \leq 0.05$) concentrations were B(a)A ($P = 0.004$), and B(b)F ($P = 0.000$).

Table 12: PAHs assessed in salted dried *Oreochromis niloticus* sampled from selected coastal markets in Southern Ghana

PAHs	REGIONS AND TOWNS		
	CENTRAL REGION	GREATER ACCRA	VOLTA REGION
Chrysene	Mankesim	Madina/Tema	Akatsi
Benzo(a)anthracene	Mankesim/ Winnebe	Kpong/Madina	Akatsi
Benzo(k)fluoranthrene	nd	KpongMadina	Akatsi
Benzo(a)pyrene	Mankesim/ Winneba	Kpong/Madina	Akatsi
Benzo(g,h,i)perylene	nd	KpongMadina	Akatsi
Indeno(1,2,3,c,d)pyrene	nd	Kpong/Madina	Akatsi
Benzo(b)fluoranthene	Mankesim/ Winneba	Madina/Tema	Keta/Akatsi

nd = not detected.

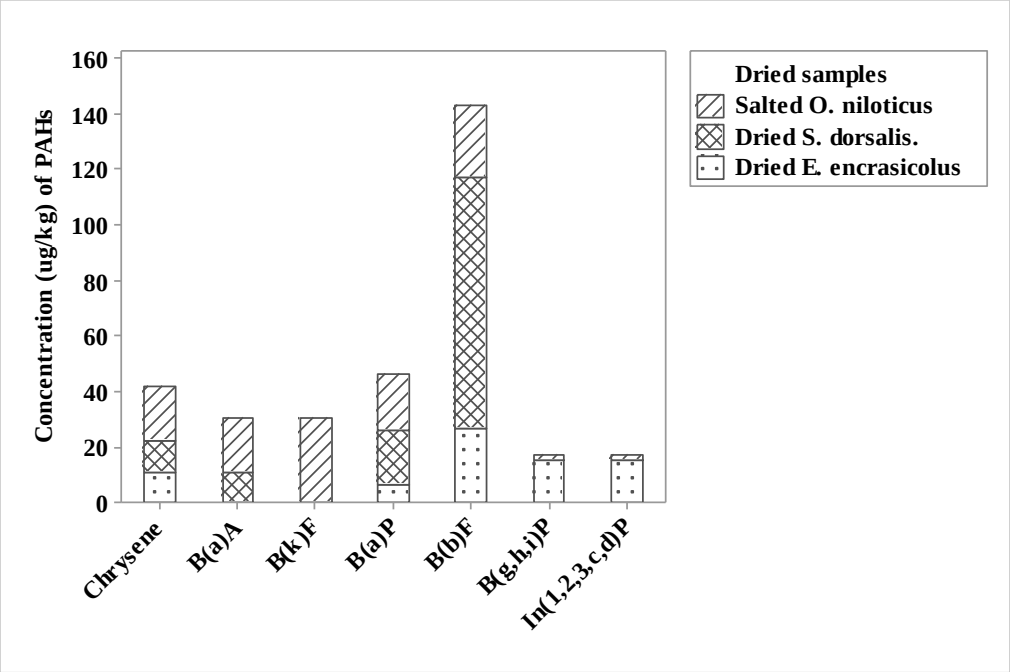


Figure 7: Mean of PAHs (µg/kg) in sundried fish sampled from selected coastal markets in Southern Ghana.

In Figure 7, the concentrations of the various PAHs differ from each other among the dried fish samples and across the markets and the regions. The stacked subtotal of concentrations of PAHs were Chrysene: 47.70 ± 4.77 µg/kg, B(a)A: 30.60 ± 4.23 µg/kg, B(k)F: 30.67 ± 0.02 µg/kg, B(a)P: 45.95 ± 6.54 µg/kg, B(b)F: 143.04 ± 0.59 µg/kg, B(g,h,i)P: 17.28 ± 7.73 µg/kg and In(1,2,3,c-d)P: 17.28 ± 7.73 µg/kg. The highest concentration of 143.04 ± 0.59 µg/kg was found in B(b)F, ($p \leq 0.05$).

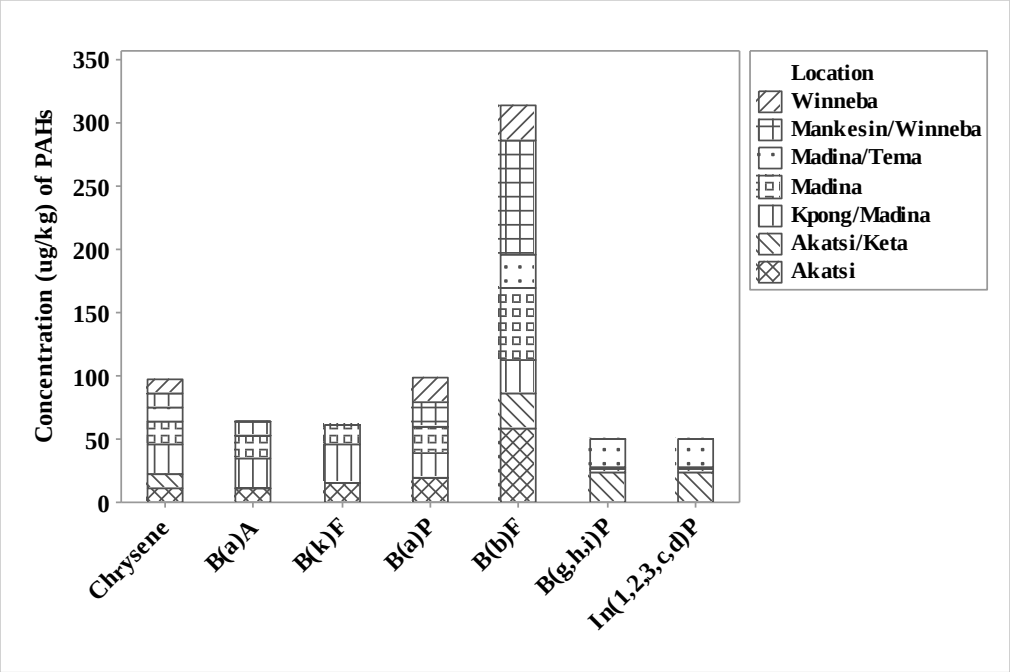


Figure 8: PAHs concentration (Aggregate subtotal) in sundried fish sampled from selected coastal markets in Southern Ghana.

Figure 8 depicts the aggregate subtotal of concentrations of PAHs by locations from where samples were taken. The subtotal of chrysene in the fish samples from all the locations was 96.62 ± 10.11 µg/kg, B(a)A was 63.31 ± 9.05 µg/kg, B(k)F recorded 61.33 ± 43.14 µg/kg and B(a)P recorded 98.43 ± 0.08 µg/kg. The others were B(b)F: 312.84 ± 91.01, B(g, h, i)P: 50.35 ± 2.12 µg/kg and In(1, 2, 3, c-d)P: 50.35 ± 2.12 µg/kg. In all the markets or locations where the fishes were sampled, the PAHs with the highest concentration was B(b)F whereas B(g, h, i)P and In(1, 2, 3, c-d)P recorded the lowest concentrations.

In Figure 8, the subtotal concentration of PAHs in the markets varied significantly. Chrysene concentration in Akatsi: 10.66 ± 0.81 µg/kg, Akatsi/Keta: 10.84 ± 0.00 µg/kg, Kpong/Madina: 24.39 ± 0.00 µg/kg, Madina: 17.81 ± 9.31 µg/kg, Madina/Tema: 10.84 ± 0.00, Mankesim/Winneba: 11.23 ±

0.00 µg/kg, and Winneba: 10.84 ± 0.00 µg/kg. The highest concentration of Chrysene (17.81 ± 9.31 µg/kg) occurred in Madina.

B(a)A concentrations in: Akatsi: 11.06 ± 0.05 µg/kg, Kpong/Madina: 23.81 ± 0.00 µg/kg, Madina: 17.41 ± 9.03 µg/kg, Mankesim/Winneba: 11.03 ± 0.00 µg/kg. The highest concentration of B(a)A (17.41 ± 9.03 µg/kg) was recorded in Madina.

B(a)P concentration in: Kpong/Madina: 19.75 ± 0.00 µg/kg, Madina: 19.72 ± 0.11 µg/kg, Akatsi: 19.70 ± 0.07 µg/kg, Winneba: 19.61 ± 0.00 µg/kg, and Mankesim/Winneba: 19.65 ± 0.00 µg/kg. The highest concentration of B(a)P (19.75 ± 0.00 µg/kg) was recorded in Kpong/Madina.

In(1, 2, 3, c-d)P and B(g, h, i)P recorded equal values of PAHs in: Kpong/Madina: 2.85 ± 0.00 µg/kg, Madina/Tema: 23.00 ± 0.00 µg/kg, Akatsi/Keta: 23.00 ± 0.00 µg/kg, and Madina: 1.50 ± 2.12 µg/kg.The highest concentration of In(1, 2, 3, c-d)P and B(g, h, i)P was 23.00 ± 0.00 µg/kg and was recorded in Madina/Tema, Akatsi/Keta, and Madina.

B(b)F concentrations in: Kpong/Madina 25.77 ± 0.00 µg/kg, Madina: 58.14 ± 45.5 µg/kg, Akatsi: 58.14 ± 45.5 µg/kg, Winneba: 27.00 ± 0.00 µg/kg, Madina/Tema 25.75 ± 0.00 µg/kg,Akatsi/Keta: 27.76 ± 0.00 µg/kg, and Mankesim/Winneba: 90.28 ± 0.00 µg/kg. The highest concentration of B(b)F was 90.28 ± 0.00 µg/kg and recorded in Mankesim and Winneba.

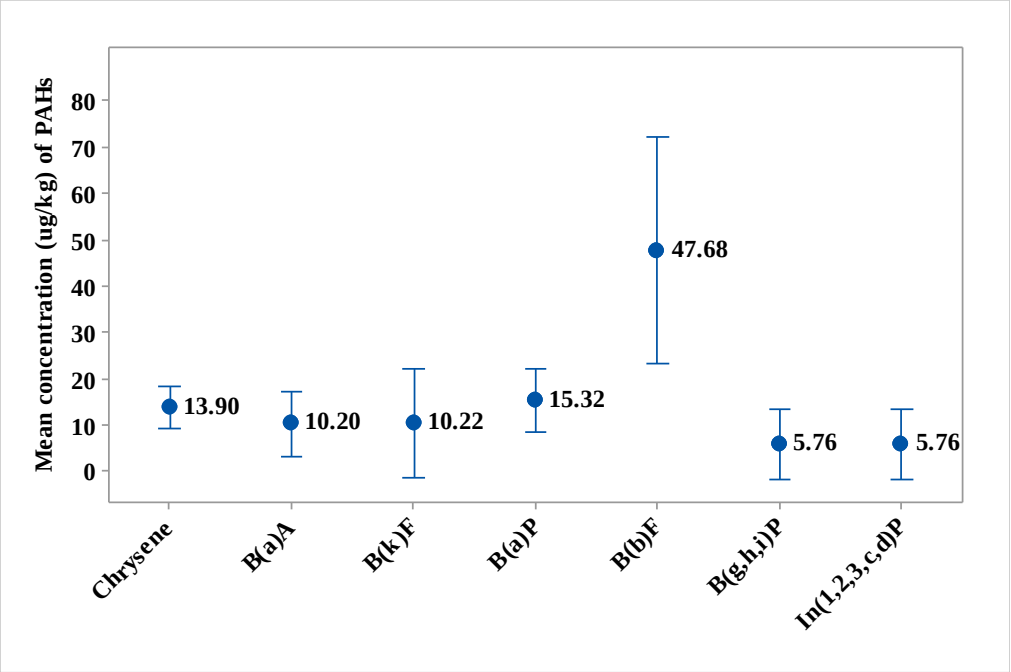


Figure 9: Mean concentration of PAHs in sundried fish samples from selected coastal markets in Southern Ghana.

Figure 9 depicts mean concentrations of PAHs in the dried samples. The PAHs occurred in varying significant ($p = 0.000$) concentrations throughout the coastal markets (Appendix D). B(b)F recorded the highest concentration of 47.7 ± 32.0 µg/kg. B(a)P:15.32 ± 8.68 µg/kg, Chrysene: 13.90 ± 5.96 µg/kg, B(k)F: 10.22 ± 15.33 µg/kg, and B(a)A: 10.20 ± 9.25 µg/kg. The rest of the PAHs such as B(g,h, i)P and In(1, 2, 3, c-d)P recorded equal concentrations of 5.76 ± 9.85 µg/kg.

4.4 Regional and Market Dynamics of PAHs Contamination in Fish

In Figure 10, the ANOVA (Appendix D) showed variable compositions and concentrations of PAHs across the regions. The highest concentration of Chrysene; 16.34 ± 7.35 µg/kg was recorded in Greater Accra Region.

Central Region recorded 11.23 ± 0.00 µg/kg and the lowest 10.72 ± 0.58 µg/kg was recorded in the Volta Region.

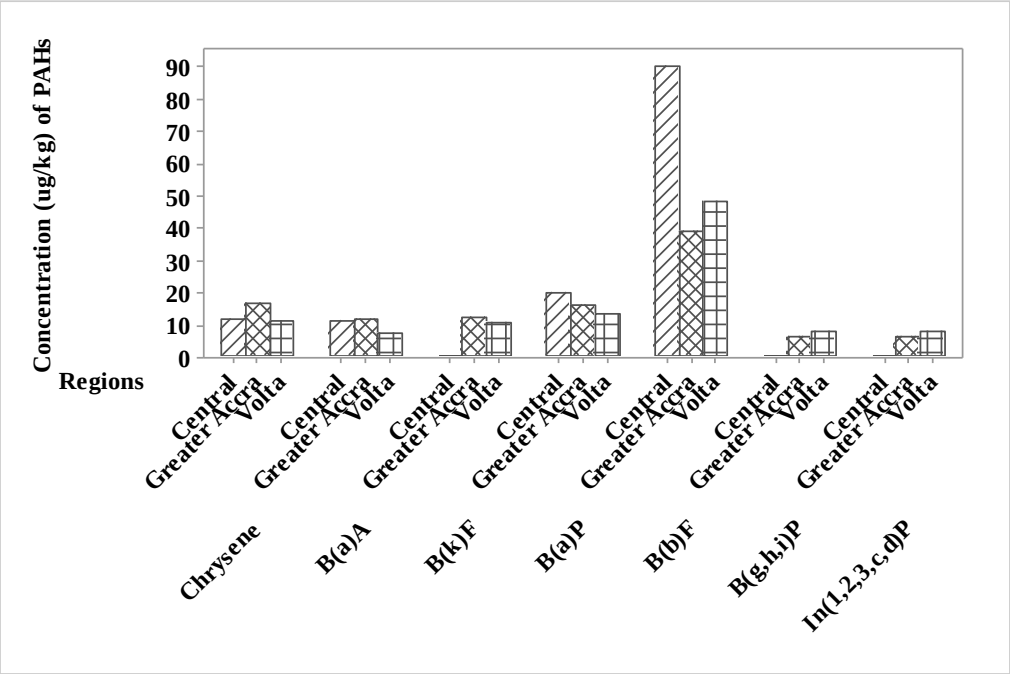


Figure 10: Regional distribution of PAHs in sundried fish sampled from coastal markets in Southern Ghana.

B(a)A varied significantly ($p \leq 0.05$) in the Greater Accra Region recording an average of 11.73 ± 11.91 µg/kg which was the highest between the regions. Central Region recorded 11.03 ± 0.00 µg/kg, while the Volta Region recorded 7.38 ± 6.39 µg/kg.

The concentration of B(k)F was below detection limit in the Central Region but recorded the highest of 12.27 ± 16.80 µg/kg in Greater Accra and 10.20 ± 17.7 µg/kg in Volta Region.

B(a)P was significantly ($p = 0.000$) high throughout the regions. These concentrations varied significantly from each other in the regions. Central Region recorded the highest of 19.65 ± 0.00 µg/kg, Greater Accra recorde 15.76 ± 8.81 µg/kg and Volta Region 13.13 ± 11.37 µg/kg. The European

Commission Regulation No 1881/2006 which replaced No 466/2001, sets maximum levels for B(a)P in muscle meat of smoked fish, smoked fishery products, smoked meat and smoked meat products at 5µg/kg to minimize the risk of cancer (Dobríková & Světlíková, 2007).

B(b)F varied from the highest, 90.28 ± 0.00µg/kg, in Central Region, 48.0 ± 36.60 µg/kg in Volta Region, and 39.0 ± 28.7 µg/kg in Greater Accra Region.

The highest concentrations of B(g, h, i)P and In(1, 2, 3 ,c-d)P of equal values 7.67 ± 13.28 µg/kg was recorded in Volta Region and 5.77 ± 9.74 µg/kg in Greater Accra Region. In Central Region, B(g, h, i)P and In(1, 2, 3 ,c-d)P were below detection levels.

These results showed that B(a)P and B(b)F were highest in the three regions with B(a)P and B(b)F more concentrated in the Central Region.

The Relative Abundance Ratio of these polycyclic aromatic hydrocarbons PAH[(RAR)] were calculated as the quotient of the highest and lowest values for each PAHs. The ratios were: RAR[B(a)A] = 2.04, RAR[B(b)F] = 15.64, RAR[B(g,h,i)P] = 1.0, RAR[B(k)F] = 2.13, RAR[(B(a)P] = 3.40, RAR[(Ind(1,2,3,c,d)P] = 1.0 and RAR[Chrysene] = 1.30. Benzo(b)Flouranthrene has the highest Relative Abundance Ratio of 15.64 and B(a)P 3.40 in sundried fish samples.

4.5 Assessment of Pesticide Residues in salted, Smoked and sun-dried Fish from Coastal Markets in Southern Ghana

Figure 11 shows the composition of pesticides which were identified in 20g of each smoked fish sampled from selected coastal markets in Southern

Ghana. Three major classes of synthetic pesticides were identified and these were organochlorines, organophosphates, and pyrethroids.

In all, thirteen different mixtures of pesticide residues were identified in 60 samples of smoked fish from the coastal markets. Only one organophosphorous pesticide (Pirimiphos-m) was identified (Figure 11).

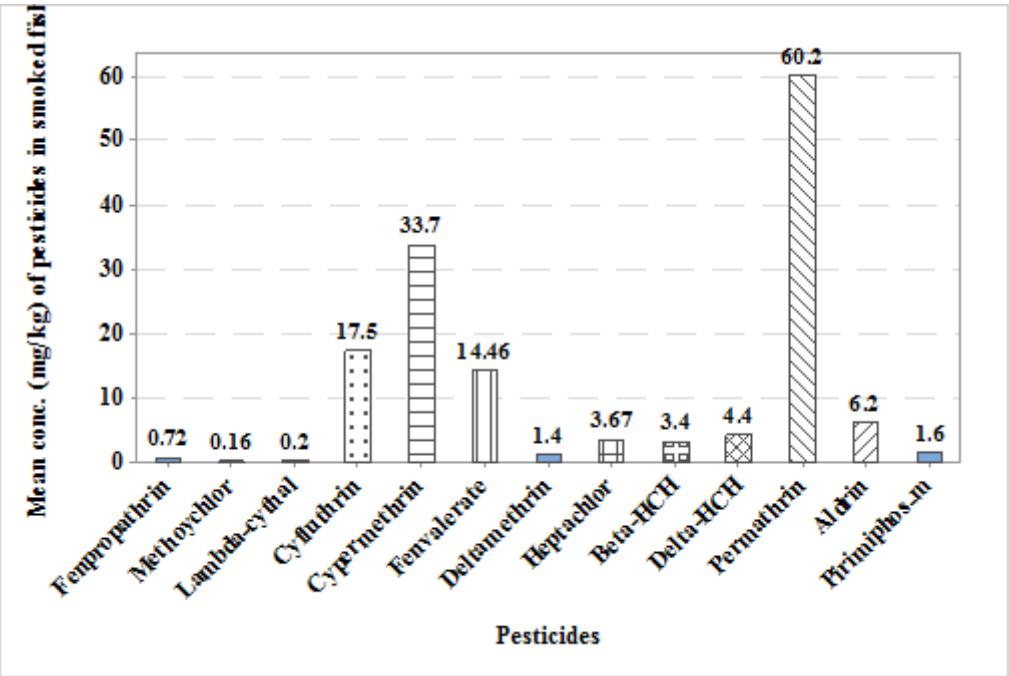


Figure 11: Sum total of pesticides identified in smoked fish sampled from selected coastal markets in Southern Ghana.

Three organochlorides (Aldrin, Delta-HCH and Heptachlor) were identified in the smoked fish samples. The remaining pesticides were pyrethroids and included Fenvelerate, Cypermethrin, Permathrin, Cyfluthrin, Deltamethoxychlor, Endosulfans, Gamma-HCH, Lambda-cythal, and Methoxychlor were present in trace concentrations.

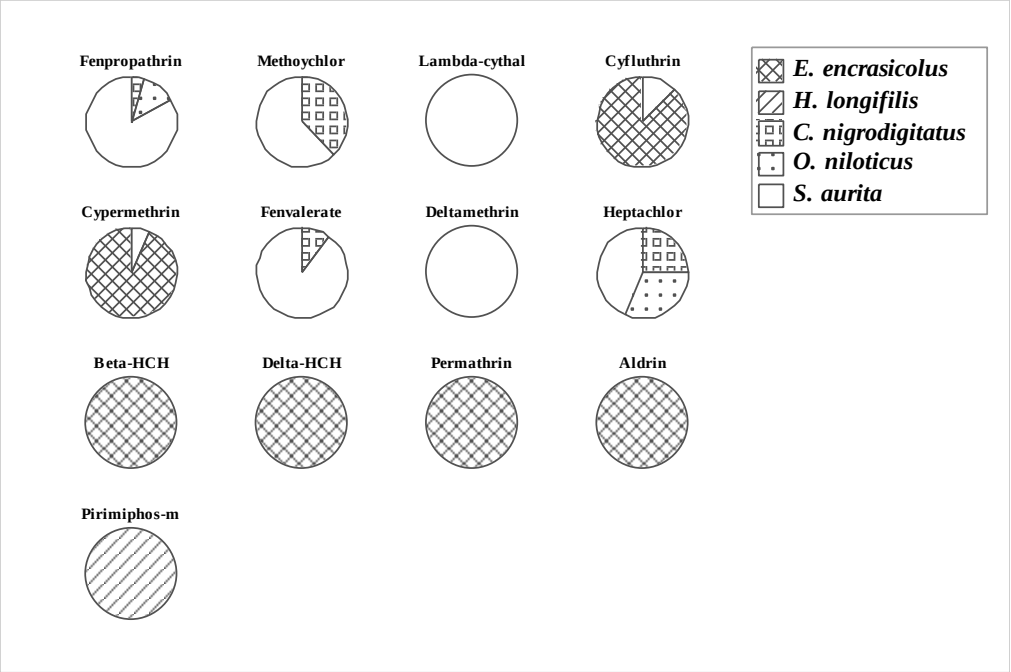


Figure 12: Types of pesticides identified in smoked fish species sampled from selected coastal markets of Southern Ghana.

Figure 12 shows six different pesticides residues in smoked *E. encrasicolus* while smoked *S. aurita* recorded seven pesticides residues. Some levels of Fenpropathrin, Fenvelerate, Heptachlor and Methoxychlor were identified in *C. nigrodigitatus*. *H. longifilis* were found to contain only Pirimiphos-m residues and similarly *O. niloticus* contained Fenpropathrin and Heptachlor residues.

Figure 13 depicts the overall concentrations of individual pesticides in 20g of each smoked fish species samples: *C. nigrodigitatus* was found to contain 0.10mg/kg of Fenpropathrin, 0.12mg/kg Methoxychlor, 2.92mg/kg Fenvelerate, and 2.24mg/kg of Heptachlor. *O. niloticus* contained 0.26mg/kg of Fenpropathrin and 3.54mg/kg of Heptachlor. *S. aurita* contained 1.80mg/kg of Fenpropathrin, 0.20mg/kg of Methoxychlor, 0.20mg/kg Lambdal-cythal, 4.20mg/kg Cyfluthrin and Cypermethrin, 26.00mg/kg of Fenvelerate, and

1.40mg/kg Deltamethrin. *E. encrasicolus* contained 30.80mg/kg of Cyfluthrin, 63.20mg/kg Cypermethrin, 3.40mg/kg Beta-HCH, 4.40mg/kg Delta-HCH, 60.20mg/kg Permathrin, and 6.2mg/kg of Aldrin. Only 1.60mg/kg of Pirimiphos-m was identified in *H. longifilis*.

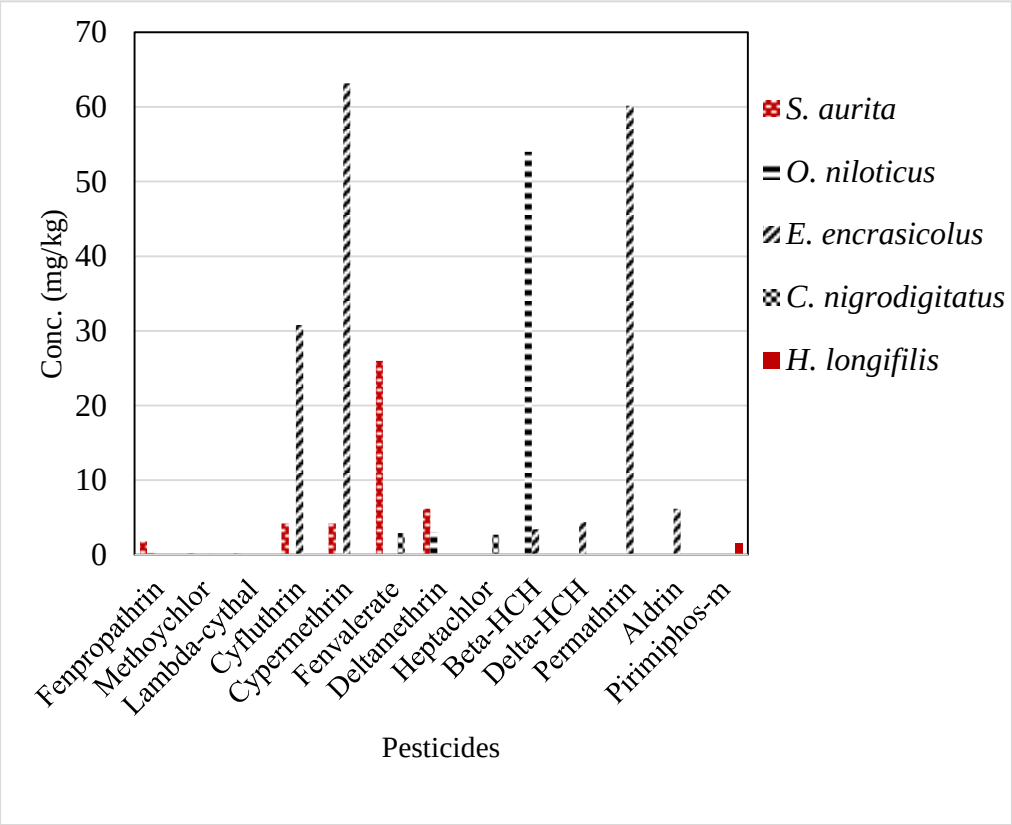


Figure 13: Concentrations of pesticides identified in smoked fish species sampled from selected markets in Southern Ghana.

Smoked *E. encrasicolus* processed in Winneba and sampled in Madina market were found to entirely contained Beta-HCH at a concentration of 3.4mg/kg of the pesticide Beta-HCH. However, Beta-HCH was not detected in smoked *C. nigrodigitatus, S. aurita, O. niloticus and H. longifilis* from the same market. Delta-HCH was detected at concentration of 4.4mg/kg in smoked *E. encrasicolus* which was processed in Winneba and sold in Madina

market. Delta-HCH was not detected in smoked *C. nigrodigitatus, S. aurita, O. niloticus and H. longifilis* from the same market.

Permathrin was detected at concentration of 60.2mg/kg in smoked *E. encrasicolus* sampled from Keta, Madina, Akatsi, and Elmina. Permathrin was however not detected in smoked *C. nigrodigitatus, S. aurita, O. niloticus and H. longifilis* sampled from the same market.

Aldrin was detected at concentration of 6.2mg/kg in smoked *Encrasicolus encrasicolus* sampled from Madina, Keta, Akatsi, Cape Coast and Elmina but not in smoked *Chrysichthys nigrodigitatus, Sardinella aurita, Oreochromis niloticus and Heterobranchus longifilis* from the same market.

Smoked *Engraulis encrasicolus* sampled from Cape Coast, Elmina, Akatsi, Chorkor and Madina markets contained Cyfluthrin residue at concentration of 30.8mg/kg ($p \leq 0.05$). Cyfluthrin residue however was not detected in smoked *C. nigrodigitatus, O. niloticus and H. longifilis* sampled from the same locations.

Smoked *S. aurita* sampled from Cape Coast, Elmina, Keta, Akatsi, Winneba, Chorkor and Madina markets contained 4.2mg/kg of Cyfluthrin residue. This value represents the mean concentration from the various markets.

Smoked *E. encrasicolus* sampled from Chorkor, Mankesim, Elmina, and Akatsi contained Cypermethrin at concentration of 63.2mg/kg. Cypermethrin however, was not detected in smoked *C. nigrodigitatus, O. niloticus and H. longifilis* sampled from the same markets. Cypermethrin residue was detected in *S. aurita* at concentration of 4.2mg/kg when they were sampled from Madina, Akatsi, Keta, Chorkor, Cape Coast, Mankesim, Elmina

and Kpong markets. The smoked *S. aurita* from Mankesim market was processed in Winneba fish market.

Smoked *H. longifilis* sampled from Mankesim, Madina, Akatsi and Kpong markets were found to contain Pirimiphos-m residue at concentration of 1.6mg/kg. This residue was absent in smoked *S. aurita, C. nigrodigitatus, O. niloticus and E. encrasicolus* sampled from the same markets.

Smoked *S. aurita* sampled from Madina, Chorkor and Akatsi contained Lambdal-cythal residue at concentration of 0.2mg/kg. This residue was however absent in smoked *E. encrasicolus*, *C. nigrodigitatus*, *O. niloticus* and *H. longifilis* sampled from the same markets.

Smoked *S. aurita* which were sampled from Madina and Chorkor markets were found to contain 1.4mg/kg of Deltamethrin residue. However, Deltamethrin residue was not detected in smoked *E encrasicolus, H. longifilis, O. niloticus* and *C. nigrodigitatus* sampled from the same locations during the study. The organophosphorus pesticide Pirimiphos-m residue was detected in smoked *H. longifilis* at a concentration of 1.6mg/kg and which were sampled from Mankesim, Madina, Akatsi and Kpong markets. Pirimiphos-m, was however, not detected in smoked *O. niloticus* and *C. nigrodigitatus, E. encrasicolus* and *S. aurita* from the same locations.

Methoxychlor residue was not detected in smoked *E. encrasicolus, H. longifilis and O. niloticus* which were sampled from Madina, Chorkor, Akatsi and Kpong markets. Methoxychlor residue at concentration of 0.12mg/kg was detected in smoked *C. nigrodigitatus* and at concentration of 0.2 mg/kg in *S. aurita* sampled from the same markets ($p \leq 0.05$).

Heptachlor residue was detected at different concentrations in different smoked fishes in this study ($p \leq 0.05$). Heptachlor occurred at concentration of 4.8mg/kg in smoked *O. aurita*, 3.5mg/kg in smoked *O. niloticus* and 2.72mg/kg in smoked *C. nigrodigitatus* sampled from Madina, Chorkor, Kpong, Akatsi, and Mankesim markets. Heptachlor however was not detected in smoked *H. longifilis* and smoked *E. encrasicolus* sampled from the same markets.

Fenvalerate residue was detected in *C. nigrodigitatus* at concentration of 2.29mg/kg, and *S. aurita* at concentration of 26.0mg/kg. Fenvalerate residue was not detected in *O. niloticus, H. longifilis and E. encrasicolus.* All these samples were from Madina, Chorkor, Kpong, Akatsi, Keta and Kpong markets.

Fenpropathrin residue was identified at concentration of 0.1mg/kg in smoked *C. nigrodigitatus*, 0.26mg/kg in smoked *O. niloticus* and 1.8mg/kg in smoked *S. aurita* sampled from Madina, Chorkor, Keta and Kpong markets. This residue was however not available in smoked *E. encrasicolus* and in smoked *H. longifilis* sampled from these markets.

Figure 14 shows eight (8) pesticide mixtures which were identified in sun dried fish sampled from the coastal markets in Southern Ghana. Among the eight, three were organochlorine base namely Aldrin, Delta-HCH and Heptachlor. Sun dried *E. encrasicolus* recorded largely Fenpropathrin, and some amount of Fenvalerate, Permathrin, Aldrin, Cyfluthrin, Cypermethrin and Delta-HCH. Salted and sun dried *O. niloticus* recorded Cyfluthrin, Cypermethrin, Delta-HCH, Aldrin and Permathrin. Sun dried *S. dorsalis* recorded Heptachlor residue. Nonetheless, these eight pesticides recorded

significantly (p = 0.002) higher mean concentrations than the rest of the pesticides in the smoked, salted and sun dried fish samples.

The overall concentration of mixture of pesticides identified in dried *E. encrasicolus* was 171.88mg/kg, in dried *Selena dorsalis* it was 3.88mg/kg, and in salted-dried *O. niloticus*, 19.4mg/kg.

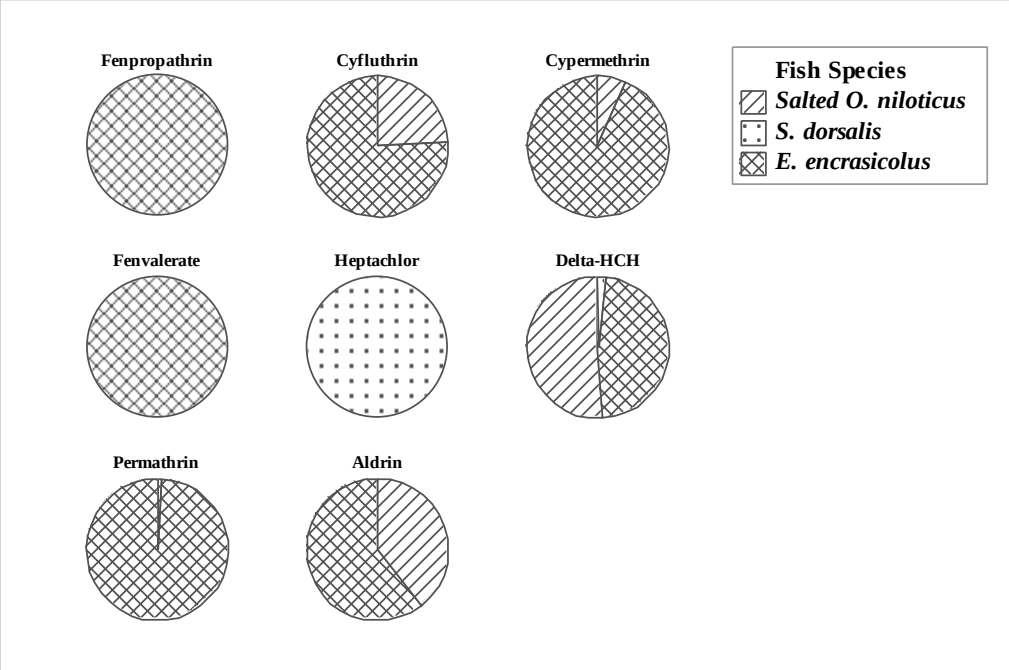


Figure 14: Composition of pesticides in sundried fish sampled from selected coastal markets in Southern Ghana.

In Figure 15, Fenpropathrin residue was detected in sundried *E. encrasicolus* at concentration of 9.2mg/kg sampled from Keta market but it was not detected in sundried *S. dorsalis* and salted and sun dried *O. niloticus* from the same market.

Cyfluthrin residue at concentration of 39.0mg/kg was detected in sundried *E. encrasicolus* and in salted and sun dried *O. niloticus* at concentration of 12.2mg/kg ($p \leq 0.05$) in samples from Madina, Cape Coast,

Kpong, Akatsi and Elmina markets. Cyfluthrin was however not detected in *S. dorsalis* sampled from the same markets.

E. encrasicolus and salted and sun dried *O. niloticus* sampled from Madina, Cape Coast, Kpong, Akatsi and Elmina markets contained a total of 14.8mg/kg and 1.0mg/kg of Cypermethrin respectively. Cypermethrin residue however was not detected in sundried *S. dorsalis* sampled from the same markets.

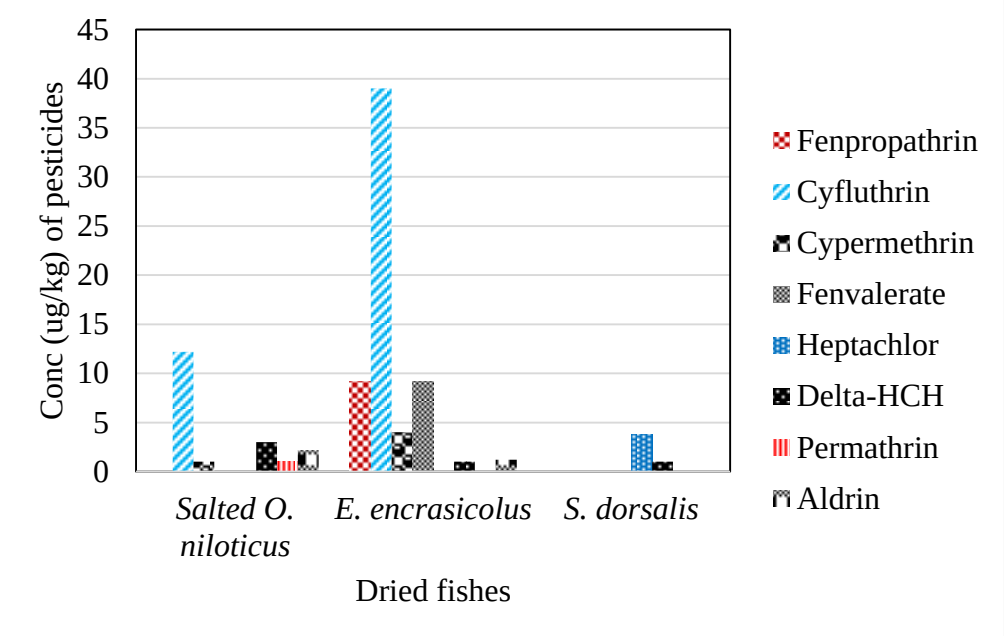


Figure 15: Pesticides identified in sundried and salted sun-dried fish species from selected markets in Southern Ghana.

Fenveralate residue was not identified in sundried *S. dorsalis* and salted and sun dried *O. niloticus* sampled from Keta market but Fenveralate was detected at concentration of 9.2mg/kg in sun dried *E. encrasicolus* sampled from the same market.

Heptachlor was detected in sundried *S. dorsalis* sampled from Mankesim market at concentration of 3.8mg/kg but this was not detected in salted and sun dried *O. niloticus* and *E. encrasicolus* from the same market.

Sun dried *S. dorsalis* recorded 0.1 mg/kg of Delta-HCH, salted and sun dried *O. niloticus* recorded 3.0mg/kg Delta-HCH residues and sun dried *E. encrasicolus* recorded 2.8mg/kg of Delta-HCH residues all sampled from Madina, Mankesim and Akatsi markets respectively.

Permethrin residue concentration in sun dried *E. encrasicolus* sampled from Keta was 9.35mg/kg and salted and sun dried *O. niloticus* was 1.0mg/kg but was not detected from sun dried *S. dorsalis* sampled from the same market.

When sun dried *E. encrasicolus* and salted and sun dried *O. niloticus* were sampled from Madina, Akatsi and Mankesim markets, Aldrin residue was not detected in *S. dorsalis* but in *E. encrasicolus* at 3.4mg/kg and salted and sun dried *O. niloticus* at 2.2mg/kg.

Figure 16 shows the pesticides including OCPs that occurred offen were Aldrin, Fenpropathrin, Delta-HCH, Fenvalerate and Heptachlor recorded between below detection limit to 0.14mg/kg. Cyfluthrin and Cypermethrin recorded between below detection limit to 0.28mg/kg, and Permathrin (0.07mg/kg – 0.42mg/kg).

The analysis of variance (Appendix D) of the samples revealed significant differences ($p = 0.001$) at $\acute{\alpha} = 0.05$ between the mean concentrations of all the pesticides/OCPs determined in the smoked, salted and sun-dried fish samples.

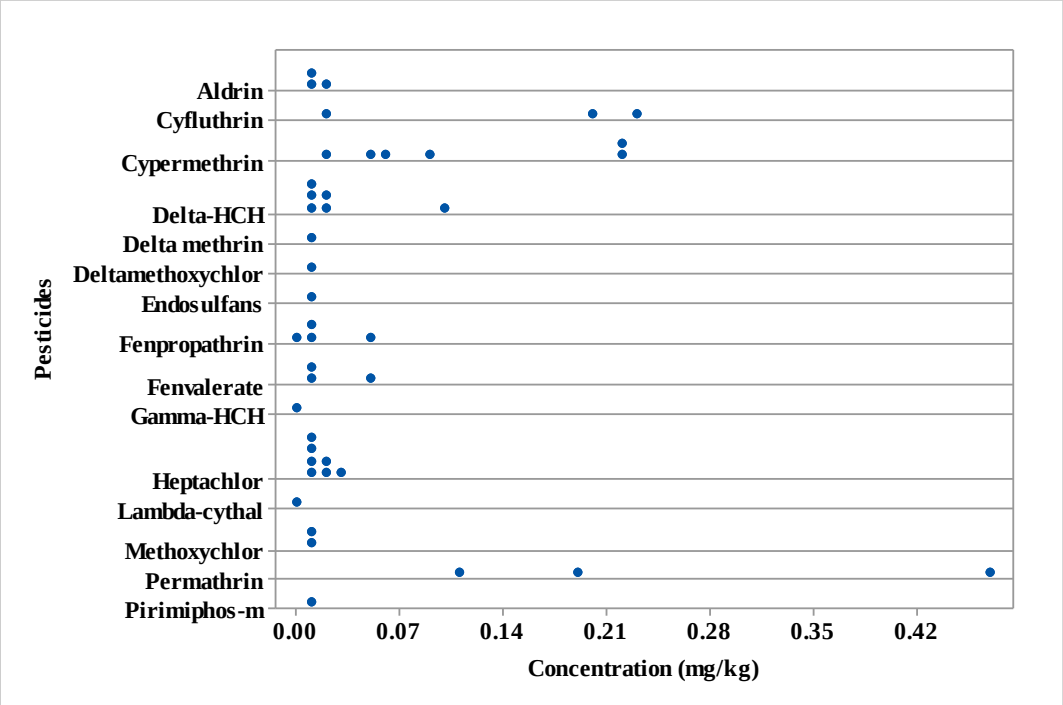


Figure 16: Dot distribution plot of pesticides in processed fish sampled from selected coastal markets in Southern Ghana.

Bonferroni Pairwise Comparisons have showed the mean concentrations of Permathrin 0.26mg/kg and Cyfluthrin 0.15mg/kg were significantly ($p \leq 0.05$) higher than Delta-HCH 0.03 mg/kg, Fenvelerate 0.03mg/kg and Cypermethrin 0.12mg/kg, which were also significantly ($p \leq 0.05$) higher than Heptachlor 0.02mg/kg, Fenpropathrin 0.02mg/kg and Aldrin 0.02 mg/kg.

These mean concentrations were significantly ($p \leq 0.05$) higher than the Maximum Residue Levels (MRL) established by FAO (2005) and by the Global MRL US EPA (2018).

4.6 Analyses of Gamma Irradiation Decontamination of PAHs, OCPs and Microbial Contaminants in Processed Fish

4.6.1 Effect of Gamma Irradiation on PAHs in Salted, Smoked and Sundried Fish from Coastal Markets of Southern Ghana

In Figure 17, the degradation of the PAHs in sundried fish were insignificant ($p \geq 0.05$) however, the individual higher irradiation doses impacted significantly ($p \leq 0.05$) on each PAHs as the doses were increased.

The study has revealed a gradual but significant ($p = 0.045$) reduction in the levels of PAHs such as Chrysene, B(a)A, B(k)F, B(a)P and B(b)F as the irradiation dose increased from 0.0kGy to 7.5kGy. This trend was observed in B(g,h,i)P when the irradiation dose was increased from 0.0kGy to 2.5kGy resulting in 49.83% degradation and in In(1,2,3,c,d)P with irradiation dose at 5.0kGy which resulted in 94.44% degradation. Gamma irradiation degradation effect on B(b)F was gradual but insignificant ($p \geq 0.05$) and resulted in 37.65% reduction from 47.68µg/kg to 29.73µg/kg when B(b)F was irradiated at 7.5kGy just as the degradation in Chrysene (51.65% reduction) at the same dose applied. There was significant ($p = 0.011$) effect of irradiation on Chrysene irradiated at 5.0kGy and when the dose was increased to 7.5kGy it caused further degradation from 10.84µg/kg to 6.72µg/kg. There were various degradation effects at each irradiation dose on the different PAHs studied in this work.

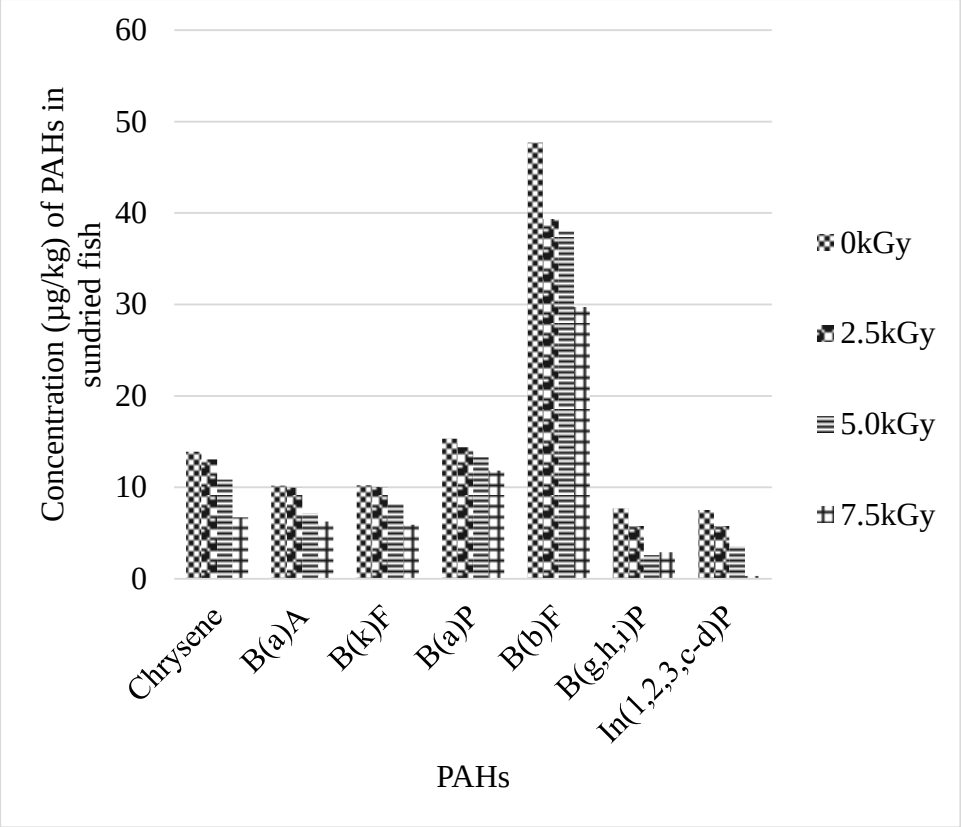


Figure 17: Effect of gamma irradiation on polycyclic aromatic hydrocarbons in sundried fish sampled from selected coastal markets in Southern Ghana. (values are mean and standard deviation of mean: N = 60).

B(a)P contamination in sundried fish was insignificantly ($p \geq 0.05$) degraded by gamma irradiation from overall mean of 15.32µg/kg in the non-irradiated (control) samples to 11.84µg/kg at 5.0kGy. B(k)F was also degraded from 10.22µg/kg to 10.05µg/kg at 2.5kGy but when the dose was increased to 7.5kGy it further degraded to 5.90µg/kg ($p \leq 0.05$). Various degrees of degradation occurred at the different doses applied.

Figure 18 depicts the analysis of smoked fish and has showed there was a gradual degradation of Chrysene, B(a)A, B(k)F, B(a)P and B(b)F at the irradiation doses applied.

The degradation of B(g,h,,i)P and In(1,2,3,c-d)P did not follow significantly well-defined trend ($p \geq 0.05$). Chrysene decreased from 15.26µg/kg at 0.0kGy to 9.66µg/kg at irradiation dose of 7.5kGy. B(a)A also decreased from 11.01µg/kg at 0.0kGy to 6.69µg/kg at 7.5kGy. B(k)F decreased from 17.67µg/kg at 0.0kGy to 13.55µg/kg at 7.5kGy ($p \leq 0.05$). B(a)P degraded from 16.74µg/kg at 0.0kGy to 9.34µg/kg at 7.5kGy. Though there were gradual degradation of the PAHs at the different irradiation doses, these degradations were not statistically significant ($p \geq 0.05$).

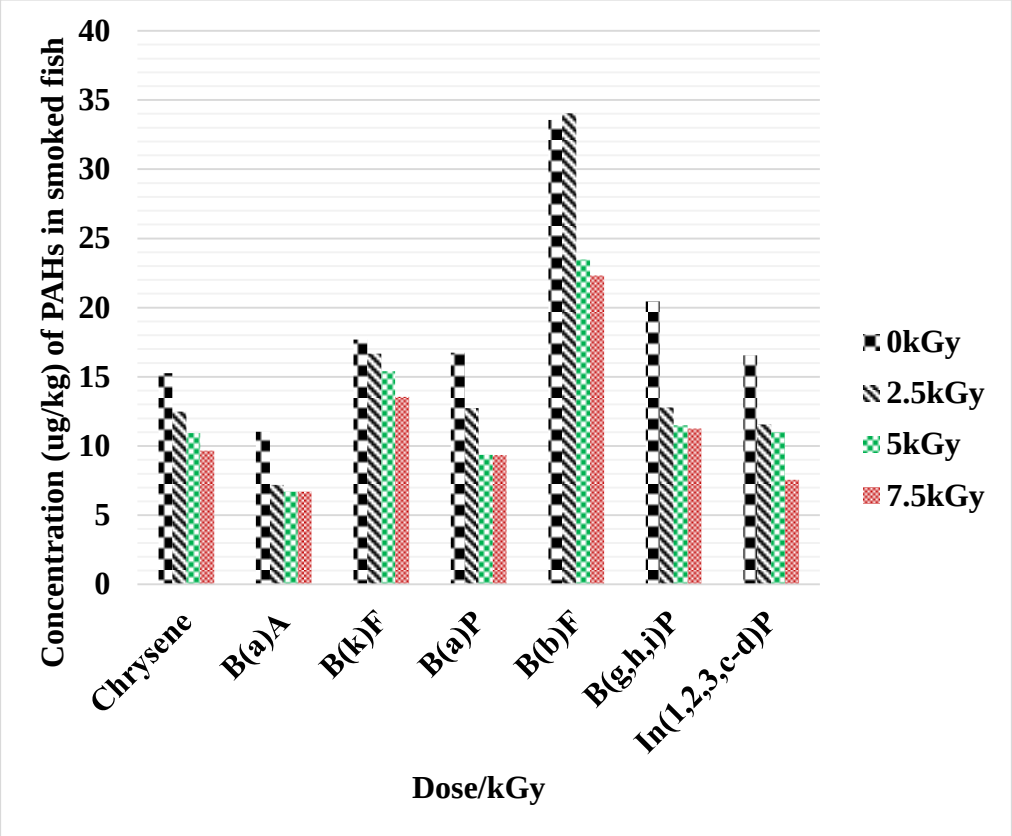


Figure 18: Effect of gamma irradiation on polycyclic aromatic hydrocarbons in smoked fish sampled from selected coastal markets in Southern Ghana. (values are means with standard deviation: N = 60).

B(b)F was degraded from 33.55µg/kg at 0.0kGy to 22.30µg/kg when the dose was 7.5kGy. When B(g,h,i)P was irradiated at 7.5kGy, the

concentration decreased to 20.45ug/kg from initial concentration of 15.59µg/kg and In(1,2,3,c-d)P decreased from 16.55µg/kg at 0.0kGy to 7.54µg/kg at 7.5kGy during the study. Though there were gradual reduction in the initial concentrations of B(a)A, B(k)F, B(a)P, B(b)F, B(g,h,i)P, and Ind(1,2,3,c-d)P, the degradation were not significant as compared with that of Chrysene in sundried fish.

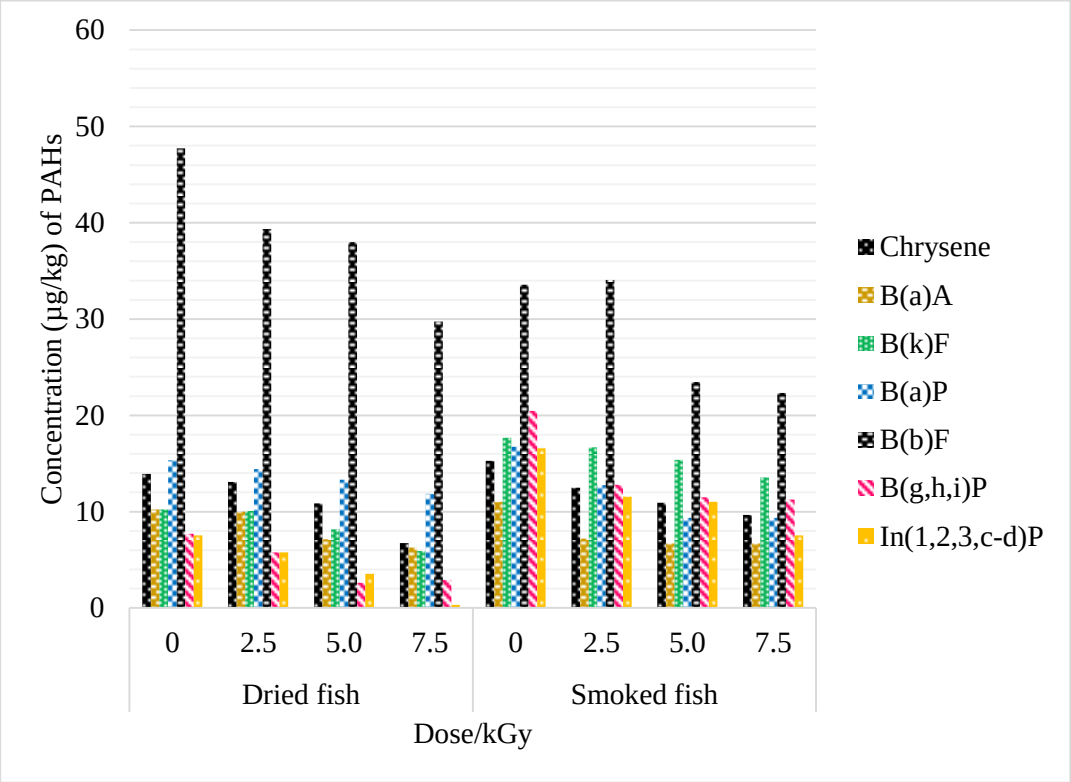


Figure 19: Comparison of the effect of gamma irradiation on PAH in smoked and sundried fish sampled from selected coastal markets in Southern Ghana (values are means with standard deviation: N = 60).

In Figure 19, gamma irradiation has a significant ($p \leq 0.05$) degradation effect on Chrysene contamination in sundried fish than in smoked fish. In sundried fish, the degradation effect was more pronounced at 2.5kGy (13.06µg/kg), 5.0kGy (10.84µg/kg) and 7.5kGy (6.72µg/kg) respectively.

Even though the degradation was gradual in B(a)A, the dose effect was not significantly different from each other just as it was in B(k)F, B(b)F, B(g,h,i)P and In(1,2,3, cd)P at 2.5kGy to 7.5kGy in all cases. Gamma irradiation degradation or decontamination at 2.5kGy, 5.0kGy and 7.5kGy was very significant ($p \leq 0.05$) with respect to B(a)P in both dried and smoked fishes.

The concentration of B(a)P in smoked fish samples was degraded from an initial mean concentration of 16.74µg/kg to 9.34µg/kg at irradiation doses of 7.5kGy.

B(b)F concentrations were high in both sundried (47.70 ± 32µg/kg) and smoked fish samples (34.04 ± 33.55µg/kg) before gamma irradiation and sundried fish (29.73 ± 7.36ug/kg) and smoked fish (22.30 ± 8.73 ug/kg) after gamma irradiation in this study.

Figure 20 and Table 13 have shown the efficiency of the irradiation method in that gamma irradiation completely degraded Deltamethrin, Pirimiphos-m, Endosulfans, Lambda-cythal and Deltamethoxychlor when they were irradiated at 2.5kGy (Appendix D).

At 5.0kGy, Aldrin (an OCP) was reduced from 0.042mg/kg to 0.014mg/kg and non-detected at 7.5kGy. Heptachlor (OCP) from 0.012mg/kg to 0.003mg/kg but non-detected at 7.5kGy. Permathrin was degraded from 0.768mg/kg to 0.01mg/kg but non-detected at 7.5kGy.

Fenvalerate was degraded from 0.074mg/kg to 0.013mg/kg but not detected at 7.5kGy and Methoxychlor from 0.019mg/kg to non-detected at 5kGy and 7.5kGy.

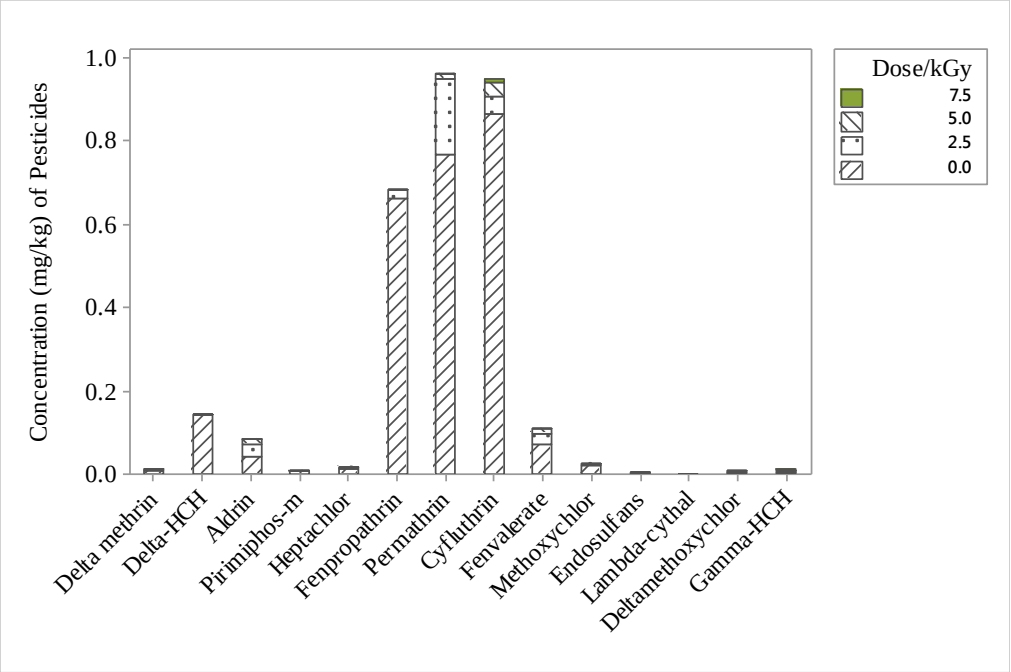


Figure 20: Effect of gamma irradiation on pesticides in processed fishes from selected markets in Sourthen Ghana.

However, two pesticides; Cyfluthrin and Gamma-HCH were not completely degraded even at 7.5kGy irradiation. At 7.5kGy, Cyfluthrin was degraded from 0.862mg/L to 0.007mg/kg being 99% degradation and Gamma-HCH from 0.004mg/kg to 0.001mg/kg being 75% degradation (Table 13).

Table 13: Gamma irradiation degradation of OCPs, OPPs and pyrethroids residues in processed fish and percentage reduction showing the performance of the irradiation method.

Pesticides	Before irradiation (0.0kGy)	After irradiation			Percentage (%) degradation
		2.5kGy	5.0kGy	7.5kGy	
Deltamethrin	0.007	0.007	nd	nd	100
Delta-HCH (OCP)	0.143	nd	nd	nd	100
Aldrin (OCP)	0.042	0.028	0.014	nd	100
Pirimiphos-m*	0.008	nd	nd	nd	100
Heptachlor (OCP)	0.012	0.003	0.003	nd	100
Fenpropathrin	0.720	0.019	nd	nd	100
Permathrin	0.768	0.180	0.010	nd	100
Cyfluthrin	0.862	0.044	0.034	0.007	99
Fenvalerate	0.074	0.023	0.013	nd	100
Methorxychlor	0.019	0.007	nd	nd	100
Endosulfans	0.005	nd	nd	nd	100
Lambda-cythal	0.001	nd	nd	nd	100
Deltamethoxy Chlor	0.005	nd	nd	nd	100
Gamma-HCH	0.004	0.003	0.003	0.001	75

* Pirimiphos-m is an OPP (Organophosphorus pesticide): nd = non detected

4.7 Estimating the Shelf Life of Gamma Irradiated Salted, Smoked and Sun-Dried Marine and Freshwater Fishes over a Period of 60 Days

The proximate composition (moisture, protein, fat and ash) and pH were determined for salted, smoked and sun dried fish species sampled from selected coastal markets in Southern Ghana to estimate the storage life. The proximate composition or bio-chemical composition and pH value of fish is an important aspect in fish processing and impact positively on both the keeping quality and the technological characteristics of the fish.

Figure 21 below depicts the levels of protein content in the smoked fish species studied.

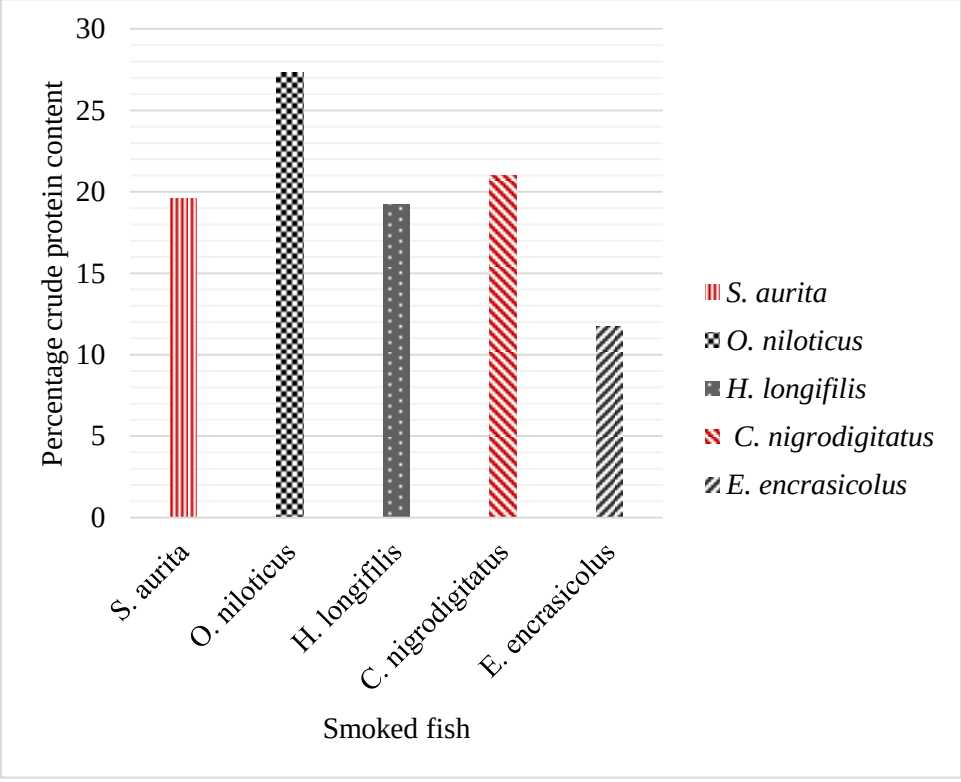


Figure 21: Initial protein contents (%) in 20g of each smoked fish samples from selected coastal markets in Southern Ghana before storage.

The findings have shown that smoked *O. niloticus* contained 27.33% of protein. *O. niloticus* have high protein content compared to smoked *C.*

nigrodigitatus (21.0%), *S. aurita* (19.6%), *H. longifilis* (19.21%) and *E. encrasiscolus* (11.7%) samples. This proved the fact that *O. niloticus* fish belongs to families of fish with high protein content (Stanby, 1982).

4.7.1 Effect of Ambient Storage Conditions on Protein Contents of Packaged and gamma Irradiated Smoked Fish from selected Coastal Markets of Southern Ghana

The analyses of the moisture content, protein, fat, ash and pH revealed a significant ($p \leq 0.05$) variations in the attributes (Appendix D). In Table 14, there was a 3% decline in protein content of *O. niloticus* from 0.14% to 0.11% between day zero and days fifteen (15) and remained constant until day fourty-five (45) of storage. There was 0.01% decrease in protein content from 0.11% on day fourty-five of storage to 0.01% on day sixty.

Table 14 shows that protein content (%) began to decline until the end of storage period on day 60. A similar trend was observed in *H. longifilis* except that between day 0 and day 15 there was a significant ($p \leq 0.05$) increase in protein content and which remained constant up till day 30. Protein content of *H. longifilis* began to decline from day 30 until day 60.

Subsequently, a decrease in protein content was observed in *C. nigrodigitatus, S. aurita and E. encrasicolus* between days 15 to the end of storage (60 days) but *S. aurita* maintained its protein content value at 0.04% to the end of storage except on day 30 when there was a minimum increase in protein content.

Table 14: Effect of storage on protein content (%) of packaged and gamma irradiated smoked fish (N = 60).

Storage (days)	Percentage protein content (w/w) in 20g of smoked fish				
	Chrysichthys nigrodigitatus	***Heterobranchus longifilis***	***Oreochromis niloticus***	***Sadinella aurita***	***Engraulis encrasicolus***
0	0.08±0.02	0.06±0.01	0.14±0.01	0.04±0.00	0.08±0.01
15	0.08±0.02	0.08±0.01	0.11±0.00	0.04±0.00	0.07±0.00
30	0.08±0.02	0.08±0.01	0.11±0.00	0.05±0.00	0.06±0.00
45	0.05±0.01	0.07±0.01	0.11±0.01	0.04±0.00	0.06±0.00
60	0.04±0.00	0.06±0.00	0.10±0.00	0.04±0.00	0.06±0.00

Protein values with standard deviation

The mean percentage protein content in smoked *O. niloticus* declined from 0.14% to 0.1%, *E. encrasicolus* and *H. longifilis* from 0.08% to 0.06% was not significant ($p \geq 0.05$).

4.7.2: Effect of Gamma Irradiation on Protein (%) Content in Smoked Fish from Selected Coastal Markets in Southern Ghana

Figure 22 revealed that protein remained fairly constant at 0.07% when *C. nigrodigitatus* was irradiated at 2.5kGy or at 5.0kGy. *H. longifilis* irradiated at 2.5kGy and 5.0kGy recorded 0.07%-0.08% protein. Furthermore, the percentage protein in irradiated *O. niloticus* at the end of irradiation remained constant at 2.5kGy and 5.0kGy. In *S. aurita*, protein decreased from 0.07% to 0.02% at the same irradiation dose however, this decrease was not significant in all the fish samples ($p \geq 0.05$). In *E. encrasicolus* the percentage protein content increased from 0.06% at 0kGy to 0.07% at 5.0kGy but the increase was not significant ($p \geq 0.05$).

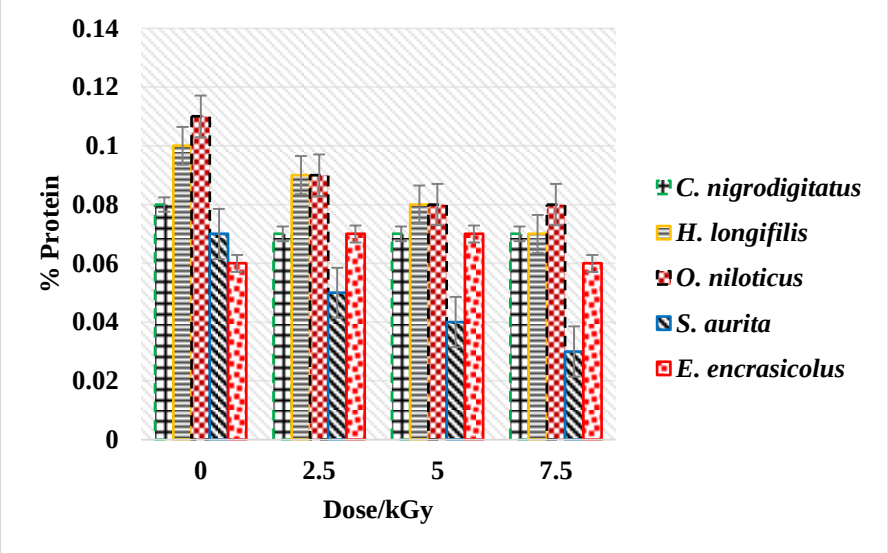


Figure 22: Effect of gamma irradiation on protein content of smoked fish from selected markets in Southern Ghana (N = 60; values with percentage errors).

4.7.3: Effect of Ambient storage on pH of Smoked Fish from selected Coastal Markets in Southern Ghana

Table 15 has shown the pH values of all the fish samples were fluctuating throughout the period of storage. The pH of the fish samples varied across the species and within the species during storage at ambient temperature.

The pH ranged from 6.08±0.33 to 7.29±0.03. The lowest pH occurred on the day of start of the storage (day zero) and the highest pH recorded was 7.38±0.15 and occurred on day 15 of storage.

Table 15: Effect of ambient storage on pH of 20g of smoked fishes (N = 60)

Storage/days	pH of smoked fish				
	Chrysichthys nigrodigitatus	***Heterobranchus longifilis***	***Oreochromis niloticus***	***Engraulis encrasicolus***	***Sadinella aurita***
0	6.68±0.15	6.64±0.20	6.72±0.21	6.86±0.05	6.08±0.33
15	7.15±0.08	7.38±0.15	7.15±0.20	6.80±0.07	6.81±0.20
30	6.89±0.08	6.93±0.09	6.83±0.22	6.91±0.06	6.92±0.15
45	7.10±0.03	7.00±0.08	6.89±0.24	6.77±0.08	7.29±0.03
60	7.05±0.00	7.16±3.47	7.36±0.00	6.92±0.00	7.24±0.00

pH values are means with standard deviation.

Within the species, *Sardinella aurita* recorded the lowest pH of 6.08±0.33 on day zero (initial pH) and rose up to above 7.29±0.03 with increasing storage days to the end of storage. *Chrysichthys nigrodigitatus* recorded pH of 6.68±0.15 to 7.15±0.08, *Heterobranchus longifilis* recorded 6.64±0.02 to 7.38±0.15, and *Oreochromis niloticus* recorded 6.72±0.21 to 7.36±0.00. *Engraulis encrasicolus* recorded a fairly constant acidic pH of 6.80±0.07 to 6.92±0.00. Across the species, pH was fairly stable within the acidic range on storage day 30 as on day zero of storage. Generally, pH increased with increased storage days signifying decay and ageing of the fish.

4.7.4: Effect of Gamma irradiation on pH of packaged Smoked Fish from selected Coastal Markets in Southern Ghana

The Figure 23 below shows how pH fluctuated with gamma irradiation of smoked fish stored at ambient temperature.

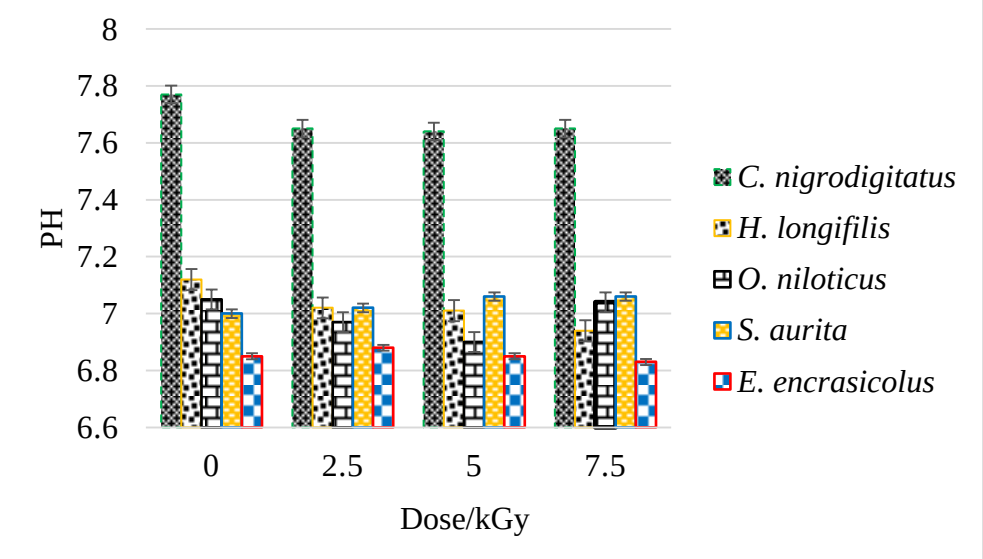


Figure 23: Effect of gamma irradiation on pH of smoked fish sampled from selected coastal markets in Southern Ghana. (values are means with standard errors: N = 60).

The pH of the fish species was not affected significantly by the irradiation doses applied as shown by the intervals in Figure 21. The pH of smoked *Chrysichthys nigrodigitatus* increased from 6.77 to 7.14 with an increased irradiation dose up to 5.0kGy ($p \geq 0.05$).

The pH decreased gradually from 7.12 at 0kGy to 6.94 at 7.5kGy in *Heterobranchus longifilis* and from 7.05 at 0kGy to 6.90 at 5.0kGy. *Sadinella aurita* had a fairly constant neutral pH of 7.0 at 0kGy and 7.06 at 5.0kGy. *Engraulis encrasicolus* recorded an increased pH from 6.85 at 0.0kGy to 6.88 at 2.5kGy. However, this value began to decrease from 6.85 at 5.0kGy to 6.83 at 7.5kGy.

4.7.5 Effect of Ambient Storage on Free fatty acid (FFA) content of packaged and Irradiated Smoked Fishes

Figure 24 has shown free fatty acid content of all the five species decreased gradually during the storage process. Free fatty acid in *S. aurita* decreased from 6.87% FFA on day zero to 6.43% FFA on days 60 of storage. Similarly, *E. encrasicolus* decreased from 7.85% FFA on day zero to 7.18% FFA on day 60. During the same time, *O. niloticus* recorded a decrease from 7.23% FFA on day zero to 5.41% FFA on day 60. *Heterobranchus longifilis* recorded 7.31% FFA on day zero but decreased to 6.77% FFA on day 60 and in *C. nigrodigitatus* FFA decreased from 8.31% FFA on day zero to 6.63% FFA on day 60.

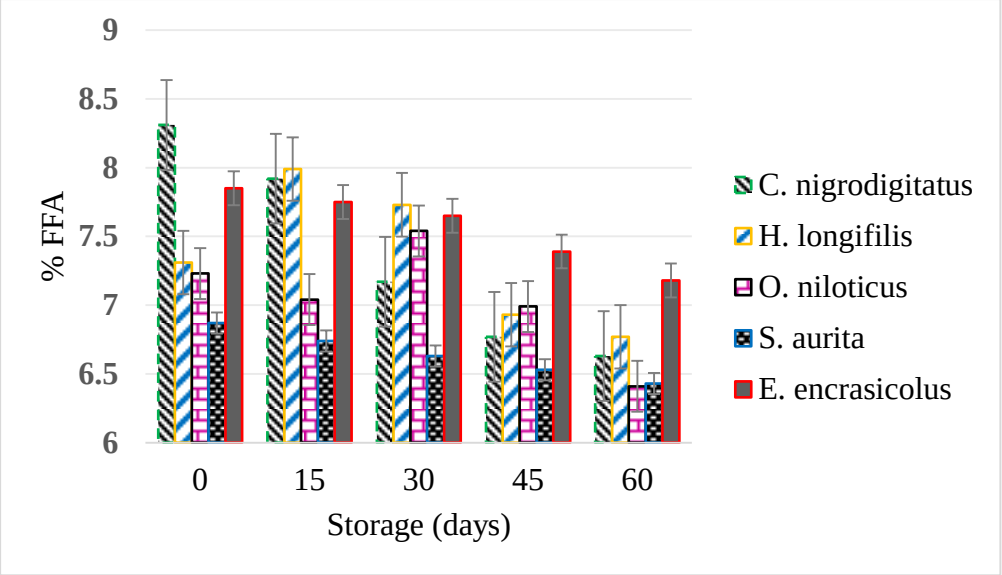


Figure 24: Effect of ambient storage on FFA content (%) in 20g of smoked fish sampled from selected coastal markets in Southern Ghana.

(values are means of FFA with standard error: N = 60).

4.7.6: Effect of Gamma Irradiation on Free Fatty Acid (FFA) content of packaged and Stored Smoked fish from selected Coastal Markets in Southern Ghana

Table 16 revealed that free fatty acid (FFA) was insignificantly ($p \geq 0.05$) affected by gamma irradiation within the dose limit applied to *C. nigrodigitatus*, *H longifilis*, *S. aurita* and *O. niloticus*. However, FFA increased in *S. aurita* with corresponding increased in irradiation dose of 2.5kGy to 7.5kGy whereas it decreased in the other species.

Table 16: Effect of gamma irradiation on FFA content (%) of smoked fishes (N = 60) sampled from selected coastal markets in Sourthen Ghana

Dose/ (kGy)	***Chrysichthys nigrodigitatus***	***Heterobranchus longifilis***	***Oreochromis niloticus***	***Sadinella aurita***	***Engraulis encrasicolus***
0	8.44±0.78	7.80±0.25	7.85±0.48	6.99±0.12	7.40±0.04
2.5	7.40±0.46	7.16±0.04	7.80±0.54	6.94±0.13	7.39±0.04
5.0	6.40±0.05	7.16±0.05	7.42±0.51	7.08±0.12	7.37±0.04
7.5	6.31±0.00	7.06±0.00	7.40±0.00	7.31±0.00	7.29±0.00

4.7.7: Effect of ambient storage on TMC/logcfu/g and TVC/logcfu/g in packaged and Gamma irradiated smoked fish from selected coastal markets in Southern Ghana

In figure 25, total mycology count (TMC) was 6.69 logcfu at the beginning of storage and the fungi population gradually decreased to 0.64 logcfu/g in all *H. longifilis* samples till the end of storage. *C. nigrodigitatus* recorded a decrease from 7.17 logcfu/g at the start of storage to 0.06 logcfu/g by the end of storage. Similar reductions occurred in *O. niloticus* (6.61 logcfu/g to 0.45 logcfu/g), *S. aurita* (5.48 logcfu/g to 1.02 logcfu/g) and *E. encrasicolus* (5.86 logcfu/g to 2.38 logcfu/g).

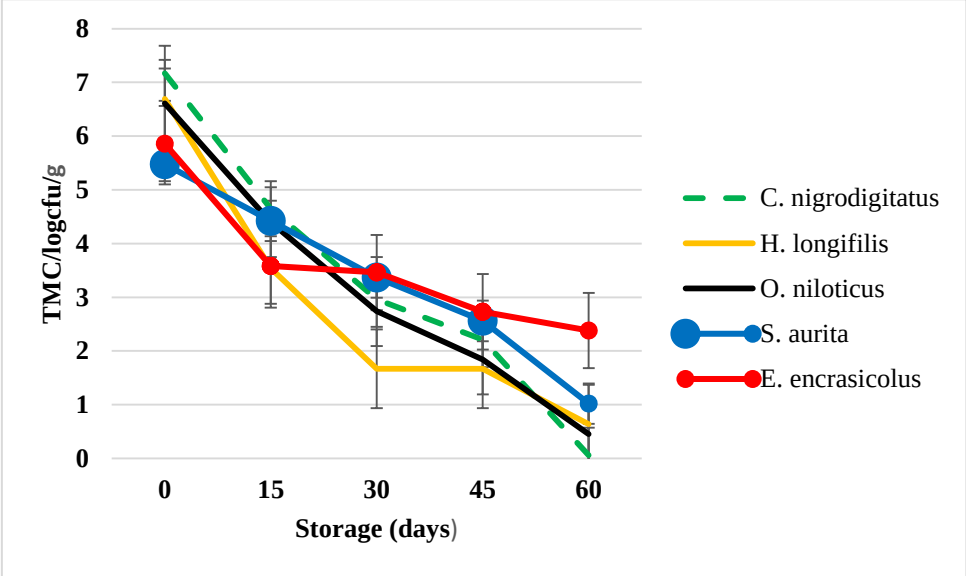


Figure 25: Effect of ambient storage on TMC/logcfu/g in packaged and irradiated smoked fish (Values are means with standard errors, N = 84).

In figure 26, total viable bacteria count (TVC) decreased from 7.08 logcfu/g before storage to 2.36 logcfu/g after storage in *O. niloticus*. The TVC for *S aurita* decreased from 4.44 logcfu/g to 1.44 logcfu/g and from 5.36 logcfu/g to 2.52 logcfu/g for *E. encrasicolu* by the end of storage. There was a

gradual decrease in the number of bacteria present in each fish sample from the beginning of storage until the end of storage. These decreases were however not significant ($p \geq 0.05$).

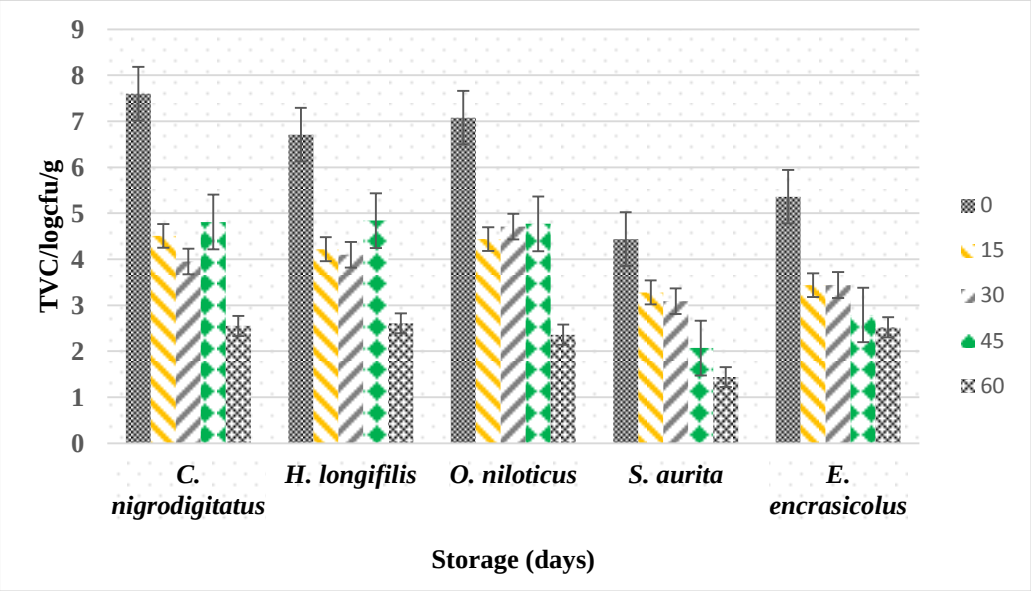


Figure 26: Effect of ambient storage on TVC/logcfu/g in smoked fish from selected markets in Southern Ghana (values are means and standard errors of mean, N = 84).

Smoked *C. nigrodigitatus* recorded a decrease in TVC from 7.60 logcfu/g to 2.55 logcfu/g and the counts in *H. longifilis* declined from 6.71 logcfu/g to 2.61 logcfu/g during the same time of storage (Figure 24). Bacteria count increased in smoked *C. nigrodigitatus*, *H. longifilis* and *O. niloticus* on day 45 of storage. *S aurita* had the lowest bacterial count on day 45 and this could be due to the packaging and irradiation. The decrease in bacterial count in *S. aurita* was consistent through out the storage period of 60 days.

4.7.8: Effect of gamma irradiation on TMC/logcfu/g and TVC/logcfu/g in packaged and gamma irradiated smoked fish from selected coastal markets in Southern Ghana

Figures 27 and 28 showed that before gamma irradiation, smoked *H. longifilis* had the highest bioburden and smoked *S. aurita* the lowest. Gamma irradiation significantly ($p \leq 0.05$) affected the contaminating microorganism in packaged and irradiated fish studied. The TVC and TMC decreased with increasing irradiation dose from 2.5kGy to 7.5kGy. The individual fish sample exhibited different sensitivity to the effect of the irradiation. *C. nigrodigitatus* recorded 7.17cfu/g (S.E. = 0.81) in the control sample and reduced to 2.87cfu/g (S.E. = 0.81) when irradiated at 7.5kGy. TVC in *H. longifilis* decreased from 7.32cfu/g (S.E. = 0.07) in the control samples to 2.78cfu/g (S.E. =0.07) at 7.5kGy irradiation dose. The bioburden of *O. niloticus* at the beginning of irradiation (0.0kGy) was 6.86cfu/g (S.E. = 0.64) and decreased to 2.61cfu/g (S.E. = 0.64) at 7.5kGy dose.

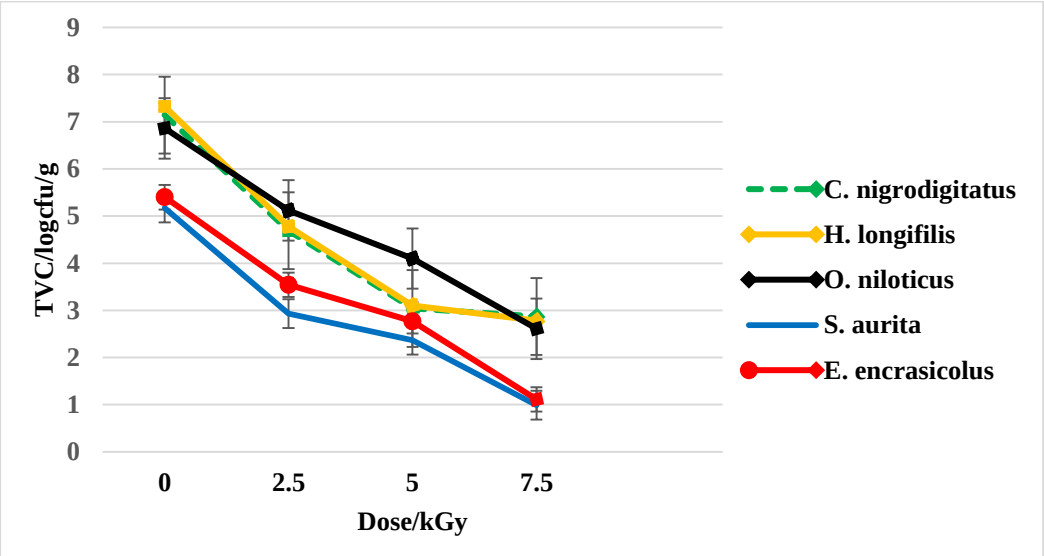


Figure 27: Effect of gamma irradiation on TVC/logcfu/g in packaged smoked fish (values are the mean with standard errors of the mean, N = 60).

The bioburden of *S. aurita* at 0.0kGy decreased from 5.17cfu/g (S.E. = 0.31) to 0.99cfu/g (S.E. = 0.31) at 7.5kGy. *E. encrasicolus* which recorded a bioburden of 5.4cfu/g (S.E. = 0.26) at 0.0kGy decreased to 1.11cfu/g (S.E. = 0.26) when the irradiation dose was increased to 7.5kGy. Irradiation up to 7.5kGy led to 4 log cycle reduction of total viable bacteria, and about 1.0 log cycle reduction of fungi in *C. nogridigitatus*. *S. aurita* recorded a lower bacteria count of 2.78 logcfu/g and *O. niloticus* recorded 4.79 logcfu/g.

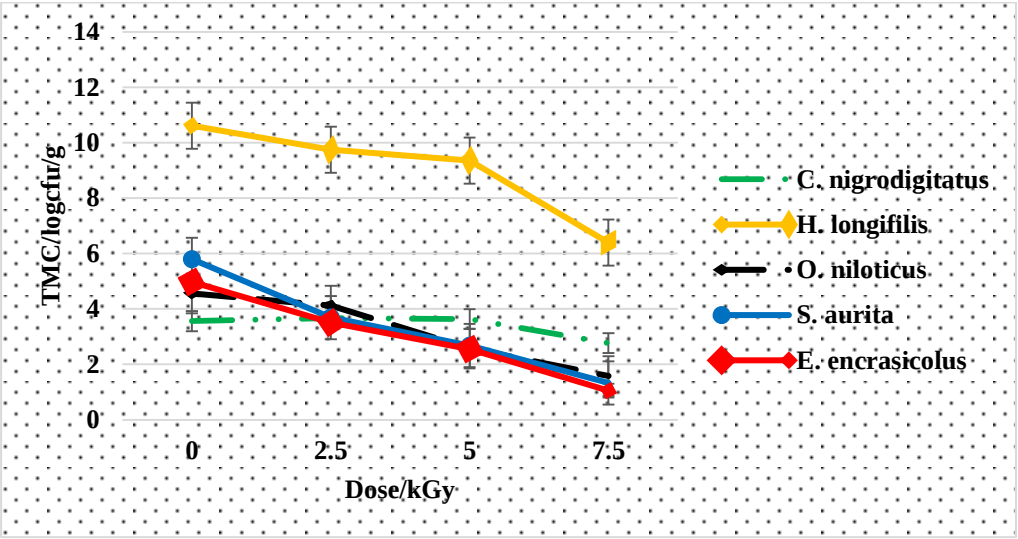


Figure 28: Effect of gamma irradiation on total fungi population in packaged smoked fish (values are the mean and standard errors, N = 60).

Figure 28 showed bioburden of fungi in these smoked fishes was highest in *H. longifilis* and lowest in *E. encrasicolus* before gamma irradiation.

The fungi bioburden count in *S. aurita* before irradiation was 5.79cfu/g (S.E. = 0.78) and 4.98cfu/g (S.E. = 0.25) in *E. encrasicolus*. Smoked *H. longifilis* recorded the highest fungi count of 10.61cfu/g (S.E. = 0.83) during storage. *O. niloticus* recorded 4.56cfu/g (S.E. = 0.71) and *C. nigrodigitatus* recorded the lowest fungi count of 3.56cfu/g (S.E. = 0.36) When *H. longifilis* was irradiated at 2.5kGy the fungi count reduced to 9.75cfu/g (S.E. = 0.83)

and to 9.35cfu/g (S.E. = 0.83) at 5.0kGy. There was a further reduction to 6.39cfu/g (S.E. = 0.83) at 7.5kGy (Figure 27). *E. encrasicolus* showed a more resilience to irradiation sensitivity of the fungi growth resulting in 1.05cfu/g (S.E. = 0.25) after irradiation at 7.5kGy.

4.7.9: Effect of ambient storage on moisture content and weight loss in packaged and gamma irradiated smoked fish from selected coastal markets in Southern Ghana.

Table 17 showed the initial weight of *C. nigrodigitatus* was 67.13g±0.04 and final weight was 66.85g±0.04, initial weight of *H. longifilis* was 61.96g±0.03 and final weight was 61.69g±0.03. The initial weight of *O. niloticus* was 86.33g±0.08 and the final weight recorded was 86.18g±0.08. *S. aurita* recorded an initial weight of 40.16g±0.08 and final weight of 40.01g±0.08.

Table 17: Effect of ambient storage on percentage weight loss of packaged and gamma irradiated smoked fishes (N = 60)

Storage (days)	Percentage weight loss in smoked fish				
	Chrysichthys nigrodigitatus	***Heterobranchus longifilis***	***Oreochromis niloticus***	***Sadinella aurita***	***Engraulis encrasicolus***
0	0.00	0.00	0.00	0.01	0.00
15	1.07±0.41	1.00±0.17	0.74±0.23	0.36±0.22	0.31±0.01
30	0.92±0.33	0.48±0.14	0.14±0.02	1.05±0.07	0.31±0.01
45	0.15±0.04	0.13±0.02	0.13±0.03	0.92±0.02	0.33±0.02
60	0.22±0.00	0.13±0.00	0.80±0.00	0.88±0.00	0.30±0.00

Percentage values with standard deviation

Engraulis encrasicolus had initial weight of 2.12g±0.05 and final weight of 2.02g±0.05. The percentage weight losses over the storage periods were not significant ($p \geq 0.05$). *C. nigrodigitatus* recorded 1.07% loss in weight after 15 days of storage but this decreased to 0.22% between days 15 to 60 of storage. *H. longifilis* recorded 1.0% loss in weight within the same period but this also reduced to 0.13% by the end of storage. During the earlier 15 days of storage, *O. niloticus* had 0.74% loss in weight and 0.08% by the 60 days of storage.

Sardinella aurita recorded 0.01% weight loss on the first day of storage and this loss increased to 1.05% until days 15($p \leq 0.05$). The percentage weight loss began to decrease (0.92%) from days 45 to (0.88%) by days 60.

Engraulis encrasicolus continued to lose weight between days zero to 45 (0.00% - 0.31%). Percentage weight loss increased up to days 45 (0.33%) but this decreased to 0.30% by days 60 (Table 17). *Sardinella aurita* had 0.15g reduction of the initial weight, *E. encrasicolus* (0.10g), *Oreochromis niloticus* 0.15g, *Heterobranchus longifilis* (0.26g) and *Chrysichthys nigrodigitatus* (0.28g). These percentage weight losses resulted in minimal reduction in the initial weights of the fish samples in this thesis.

Table 18 described the effect of ambient storage on the initial moisture content of irradiated smoked fish is shown below. The initial percentage moisture content of *Chrysichthys nigrodigitatus* was 87.37% and the final was 87.30%. *Heterobranchus longifilis* recorde an initial moisture of 87.82% and final of 87.76%. *Oreochromis niloticus* recorded 87.46% initially and 87.40% by the end of storage. *Sadinella aurita* recorded 88.67% of initial moisture and

88.57% as the final moisture content. *Engraulis encrasicolus* had an initial moisture content of 83.74% and a final moisture content of 83.59%.

Table 18: Effect of ambient storage on moisture loss (%) of packaged and gamma irradiated smoked fish from selected coastal markets in Southern Ghana

Storage (days)	Percentage moisture loss in smoked fish				
	Chrysichthys nigrodigitatus	*Heterobranchus longifilis*	*Oreochromis niloticus*	*Sadinella aurita*	*Engraulis encrasicolus*
0	0.00±0.56	0.00±0.06	0.00±0.06	0.00±3.42	0.00±0.38
15	0.14±0.05	0.14±0.06	0.13±0.06	2.85±2.14	1.24±0.045
30	0.14±0.05	0.13±0.05	0.14±0.05	9.86±0.88	0.06±0.00
45	0.04±0.01	0.03±0.01	0.02±0.00	8.13±0.25	0.05±0.00
60	0.03±0.00	0.02±0.00	0.02±0.00	7.64±0.00	0.05±0.00

Percentage are values with standard deviation

There was significant ($p \leq 0.05$) moisture loss by *S. aurita* between day 0 of storage and day 15 and large moisture loss by day 30 (9.86%) but gradually decreased thereafter until the end of storage day 60 (Table 18).

C. nigrodigitatus continued to lose moisture from days 15 (0.14%) to (0.03%) on days 60. Moisture loss was stable between days 15 and 30.

H. longifilis lose the highest percentage moisture content during days zero to 15. However, this started to reduce from days 30 (0.13%) to days 60 (0.02%).

O. niloticus had a significant percentage weight loss between days zero to 30 (0.0% - 0.14%). This however reduced to 0.02% by days 45 and remained constant until days 60 (0.02%).

Percentage moisture loss in *S. aurita* was high between days zero (0.00%) to days 30 (9.86%) and decreased thereafter to days 45 (8.13%) and decreased further to days 60 (7.64%). Between days zero to days 15, *E. encrasicolus* lose 1.24% of moisture which began to decrease thereafter up to days 30 and remained constant until the end of 60 days of storage.

4.7.10 Effect of gamma irradiation on moisture content and weight loss in smoked fish sampled from selected coastal markets in Southern Ghana

Figure 29 showed the percentage weight loss decreased in *Chrysichthys nigrodigitatus* (0.71%-0.12%) and *Heterobranchus longifilis* (0.40%-0.12%) after irradiation at 2.5kGy-7.5kGy. When *Oreochromis niloticus* was irradiated at 2.5kGy, a very significant ($p<0.05$) percentage weight loss (0.34% – 0.07%) occurred. The percentage weight loss was constant for *Sadinella aurita* but increased (0.65%) with corresponding increase in irradiation dose at 7.5kGy. There was a gradual decrease in percentage weight loss when smoked *E. encrasicolus* was irradiated at 2.5kGy and 7.5kGy.

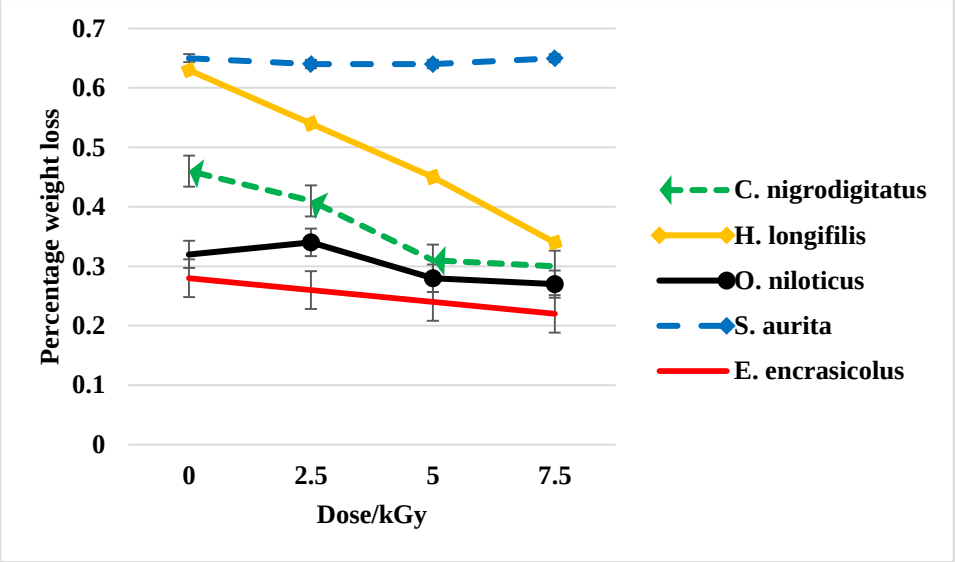


Figure 29: Effect of packaging and gamma irradiation on weight loss (%) in smoked fish sampled from selected coastal markets in Southern Ghana (values are percentages with standard errors).

Figure 30 showed percentage moisture content loss decreased with corresponding increase in irradiation dose in all the five species of smoked fish samples. This was very significant in *S. aurita* irradiated at 2.5kGy which resulted in 6.14% moisture loss and at 5.0kGy which also resulted in 5.24% moisture loss.

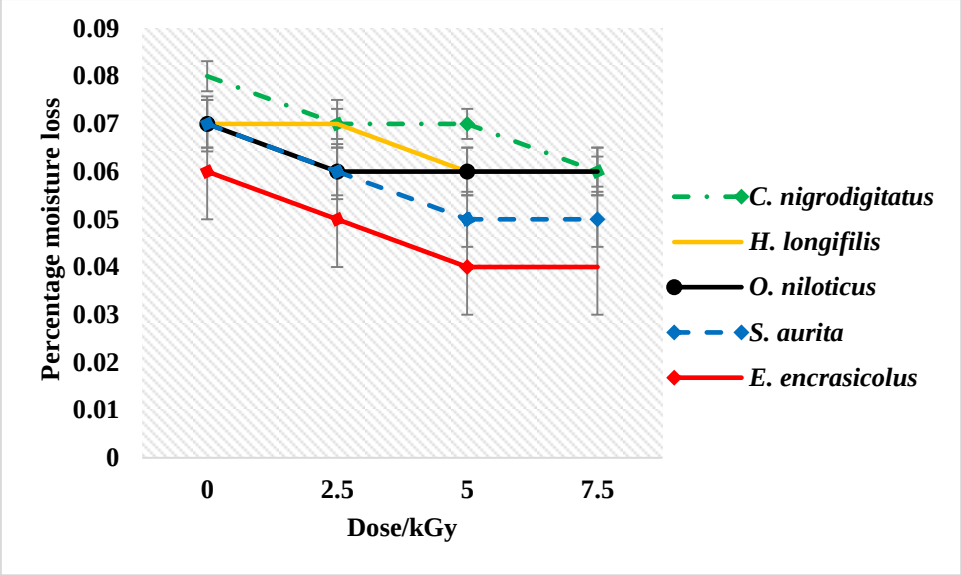


Figure 30: Effect of packaging and gamma irradiation on moisture loss (%) in smoked fish sampled from selected coastal markets in Southern Ghana (values are means with standard error).

Smoked *E. encrasicolus* recorded insignificant ($p \geq 0.05$) moisture loss of 0.08% at 2.5kGy, 0.03% at 5.0kGy and 0.89% and this resulted in significant decrease in weight loss of 0.26% at 2.5kGy and 0.24% at 5.0kGy. The effect of moisture loss has a much weight loss in the corresponding samples. In *Engraulis encrasicolus*, 0.19g of moisture loss (Figure 30) resulted in 0.49g weight loss (Figure 29).

4.7.11 Effect of gamma irradiation and ambient storage on ash content of smoked fish from selected coastal markets in Southern Ghana

In Figure 31, the initial ash content of *Chrysichthys nigrodigitatus* was 10.78% and that of *Heterobranchus longifilis* was 10.76%. *Oreochromis niloticus* had 10.85% initial ash content and *Sadinella aurita* had 17.16%. The initial ash content of Engraulis encrasicolus was 17.11%.

Percentage ash of *Chrysichthys nigrodigitatus* decreased from 12.81% on day zero to 9.26% on days 15 but increased to 11.41% on days 30. There

was a decrease in percentage ash from 11.41% on days 45 to 9.18% by days 60. *Heterobranchus longifilis* decreased in percentage ash content from 7.73% on days 30 to 6.77% by days 60.

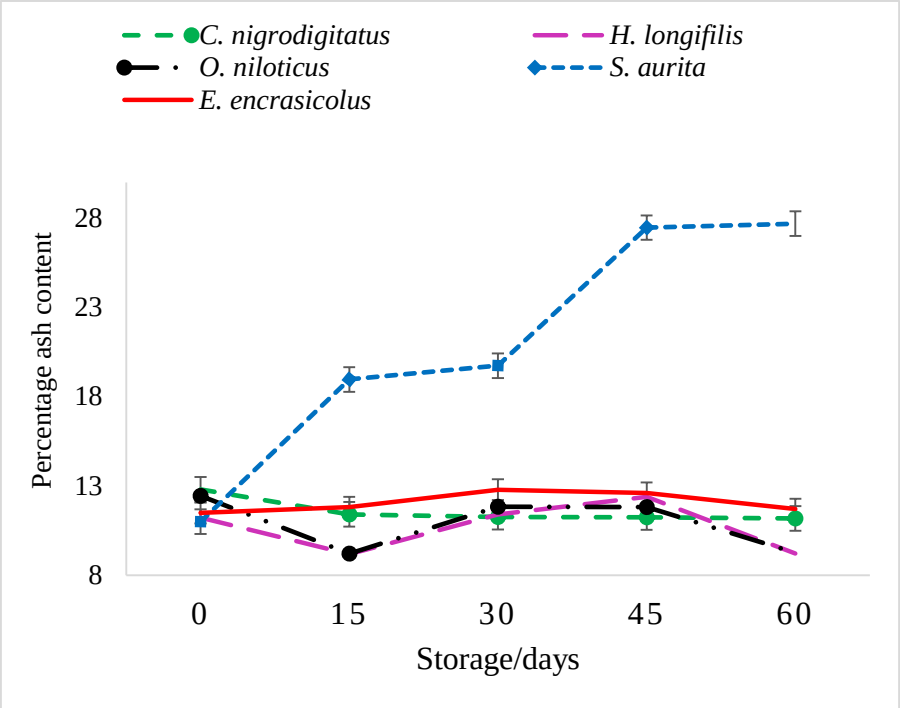


Figure 31: Effect of ambient storage on ash content (%) in packaged smoked fish sampled from selected coastal markets in Southern Ghana

(values are percentages with standard errors).

Oreochromis niloticus had a decrease in percentage ash from 12.44% on day zero to 9.25% by the end of storage on days 60. *Sadinella aurita* experienced increase in percentage ash from 11.0% on day zero to 27.69% on day 60. *Engraulis encrasicolus* also experienced increase in percentage ash from 11.49% on day zero to 11.71% on days 60.

Sadinella aurita, *Chrysichthys nigrodigitatus* and *Engraulis encrasicolus* had a steady increase in ash content throughout the storage

period. *Heterobrancus longifilis*, *Oreochromis niloticus* and *Engraulis encrasicolus* had initial decrease in ash content from day zero to day 15.

Figure 32 showed that gamma irradiation did not seemed to affect the ash content of the fish samples studied ($p \geq 0.05$).

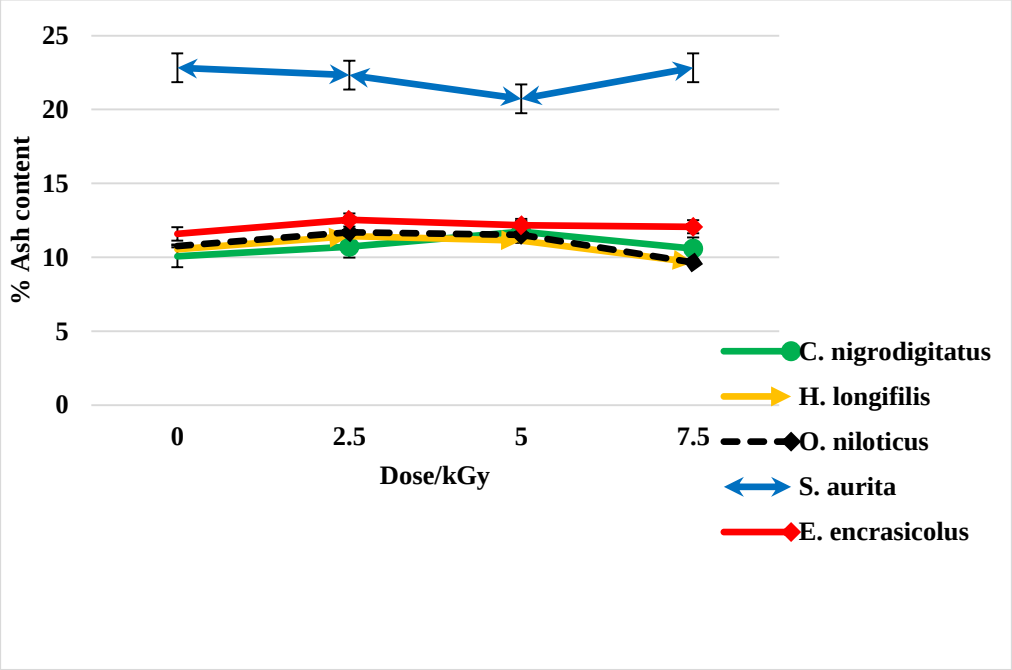


Figure 32: Effect of packaging and gamma irradiation on ash content of smoked fish sampled from selected coastal markets in Southern Ghana **(**values are percentages with standard errors).

In Figure 32, gamma irradiation at 5kGy had an ability to reduce the ash content of smoked fish; *E. encrasicolus* (12.53g – 12.07g), *C. nigrodigitatus* from 11.43g - 10.60g and O. niloticus from 10.77g - 9.65g. There was a minimal decrease of the ash content of all samples except *S. aurita* which had reduced from 22.83g – 20.70g at 5.0kGy.

4.7.12 Effect of gamma irradiation and ambient storage on total titratable acid (TTA) content of smoked fish from selected coastal markets in Southern Ghana

Figure 33 shows there was significant ($p \leq 0.05$) decrease in TTA of smoked *C. nigrodigitatus* during storage from day zero (0.36) up to days 60 (0.26). Similar trend was observed in *Heterobrancus longifilis*, *O. niloticus*, *S. aurita* and *Engraulis encrasicolus*.

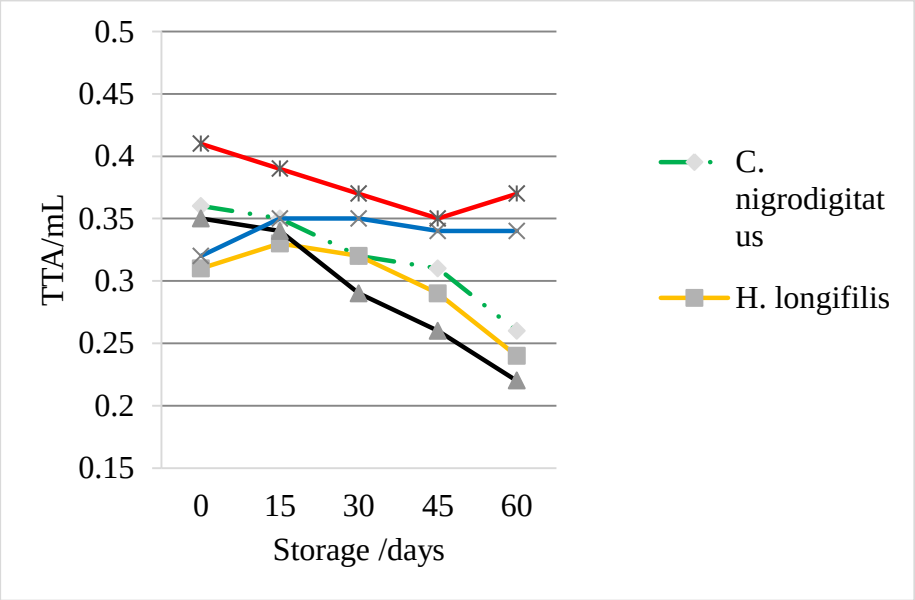


Figure 33: Effect of ambient storage on TTA in smoked fish sampled from selected coastal markets in Southern Ghana (values are with standard errors).

Engraulis encrasicolus recorded the highest TTA and *Oreocromis niloticus* recorded the lowest TTA and also had sharp decrease during the storage period.

Figure 34 shows the effect of increasing pH (decreasing acidity) on microbial growth. As pH increased from 6.97 to 7.05, TVC and TMC also increased from 4.39cfu to 4.79cfu and from 2.88cfu to2.97cfu respectively, while TTA remained fairly constant at the same time.

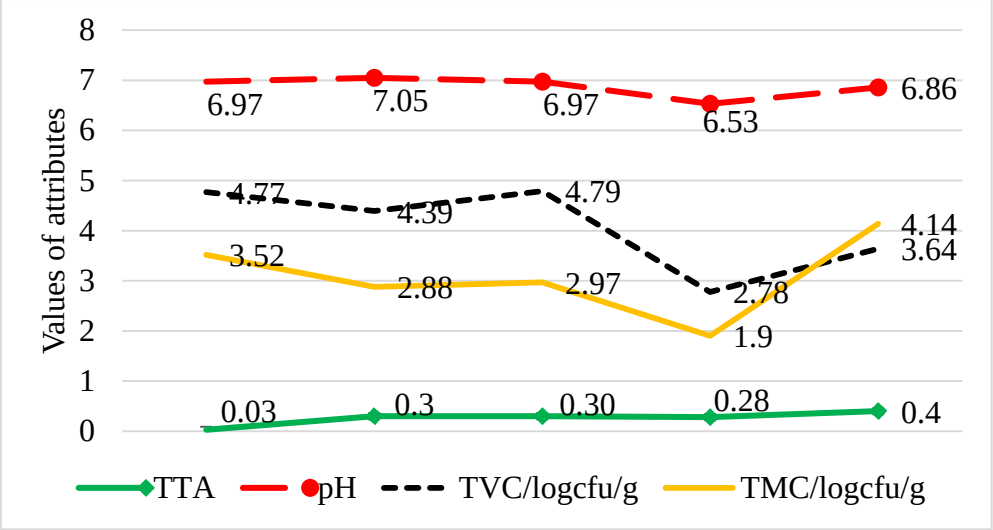


Figure 34: Comparing the effect of TTA and pH on bacteria and fungi in smoked fish sampled from selected coastal markets in Southern Ghana.

However, when pH began to decrease (acidity increased), the TVC count decreased from 4.79cfu to 2.78cfu and TMC decreased from 2.79cfu to 1.9cfu but the microbial count increased further in TVC to 3.64cfu and TMC to 4.14cfu when the acidity decreased (increase in pH value to 6.86).

Figure 34 indicates the pH rises from 6.97 to 7.05 as TTA increases slightly from 0.03 to 0.3. The pH decreased from 7.05 even when the TTA remained constant for a short time. The TTA increased to 0.4 causing the pH to rise to 6.86 during the storage period. The effect of TTA and pH is more pronounced on TMC than on TVC.

In Figure 35, the total titratable acid (TTA) of the fish were not affected significantly by the irradiation doses applied in this study. The TTA increased from 0.28 at 0kGy to 0.33 in *C. nigrodigitatus* as the irradiation dose increased at 5.0kGy.

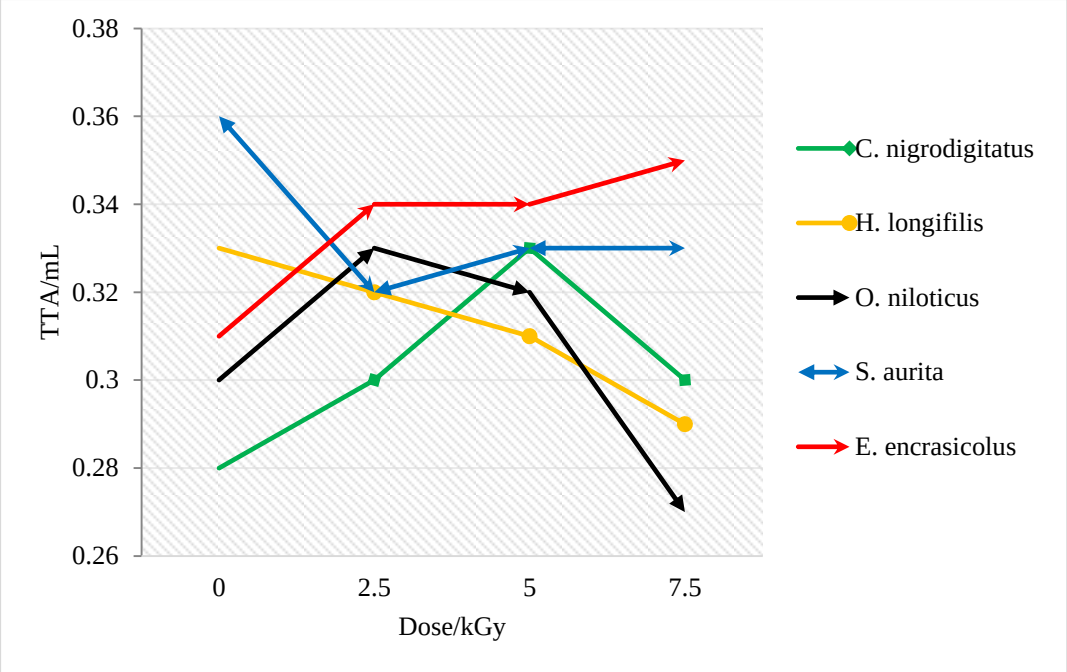


Figure 35: Effect of packaging and gamma irradiation on TTA/mL of smoked fish sampled from selected coastal markets in Southern Ghana.

In *E. encrasicolus*, TTA increased from 0.31mL at 0.0kGy to 0.35mL at 7.5kGy ($p \geq 0.05$). TTA was however stable in *S aurita* but *H. longifilis* recorded an increase in TTA from the non-irradiated 0.0kGy sample (0.03) to 0.32 at 2.5kGy. This showed that irradiation effect is insignificant ($p \geq 0.05$) on TTA of sun dried and smoked fish.

4.8 Proximate Analysis of Packaged and Stored Salted and Sun Dried Fish from selected Coastal Markets in Ghana

4.8.1 Percentage weight loss in dried fish species

The initial percentage mean weights of the samples at mean temperature of 28.8ºC and Relative Humidity (%) of 86.7 in storage area over the storage period varied from 7.438g to 63.297g. The highest was recorded in salted-dried *O. niloticus* and the lowest in *E. encrasicolus*. Figure 35 shows the percentage weight loss and the percentage moisture loss during ambient storage of the fish species. Analysis of variance showed that percentage

weight loss in the types of fish samples and the effect of storage on percentage weight loss were all statistically significant at 95% CI (p = 0.000).

The percentage weight loss in dried *E. encrasicolus* at 95% confidence interval (N = 60) was not statistically significant (p ≥ 0.405). There was however, a significant weight loss in dried *S. dorsalis* (p = 0.000).

The mean percentage weight loss at 95% CI in the fish samples (N = 60) before storage were: salted-dried *O. niloticus* (0.08%), dried *E. encrasicolus* (0.06%), and dried *S. dorsalis* (0.05% ± 0.005).

4.8.2 Percentage Weight loss During Storage

The percentage weight loss in the control dried fish samples during the beginning of storage was significant at 95% CI (p = 0.000). Subsequent storage days were significant as follows: day 15 (p = 0.004), day 30 (p = 0.000) and day 45 (p = 0.013). The control-unpackaged samples were all spoilt and discarded before day 60. Therefore, there was no result for those samples.

Bonferroni Pairwise Comparison at 95% CI has showed significant differences among the percentage weight losses over the sixty days of storage. The percentage weight loss in salted-dried *O. niloticus* was significantly different from those of dried *E. encrasicolus* and dried *S. dorsalis* (P ≤ 0.05).

4.8.3 Percentage Moisture loss in Dried Fish

The initial mean percentage moisture content of the fish species (N = 60) varied from 85.66% to 87.69% with the highest being *S. dorsalis* and the lowest was *E. encrasicolus*: dried *S. dorsalis* (87.69%), *O. niloticus* (86.44%), and E. *encrasicolus* (85.66%) respectively.

The percentage moisture loss in the dried fish species at 95% confidence interval (N = 60) before storage was not statistically significant

(p = 0.379) for *E. encrasicolus*. There was however, a significant moisture loss in dried *S. dorsalis* (p = 0.043).

4.8.4 Percentage Moisture Loss During Storage

Figure 36 shows the percentage moisture loss during ambient storage of the fish species. Analysis of variance showed that percentage moisture loss in the fish samples and the effect of storage on percentage moisture loss were all statistically significant (p = 0.000).

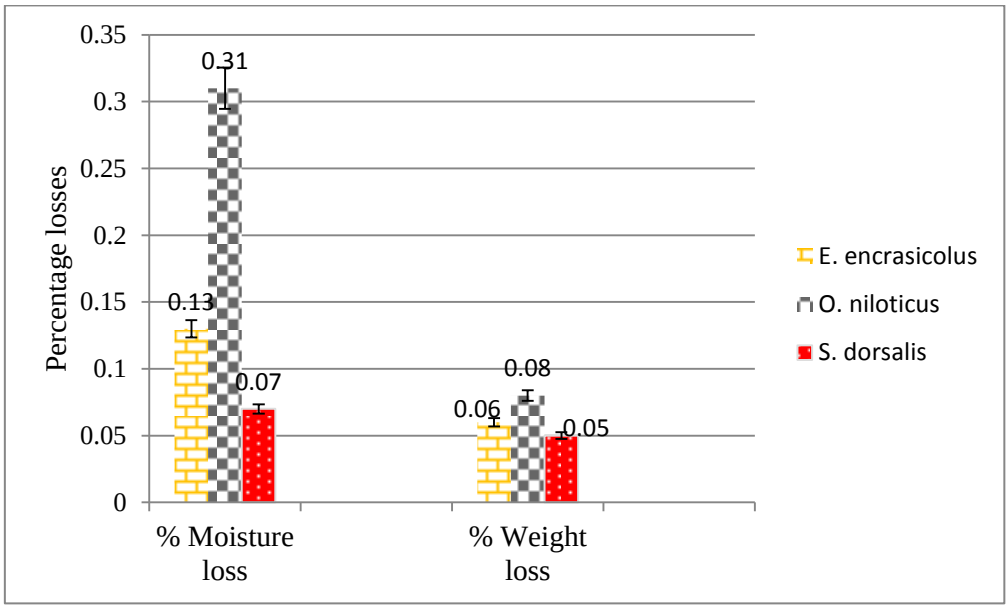


Figure 36: Percentage moisture and weight losses in three species of sundried fish sampled from selected coastal markets in Southern Ghana.

The mean percentage moisture loss at 95% CI in the fish samples (N = 60) were: salted-dried *O. niloticus* (0.31%), dried *E. encrasicolus* (0.31%), and dried *S. dorsalis* (0.07% ± 0.057).

In Figure 37, the initial percentage moisture loss by the dried and packaged fish was 0% (no transfer of moisture from product to the environment) therefore there was no loss in weight ($p \leq 0.00$) before storage. Percentage moisture loss in dried packaged fish varies from 0.12% to 0.31%

between days 15 to day 60 of storage. The lowest percentage moisture loss of 0.12% occurred on day 15 and the highest occurred on day 60.

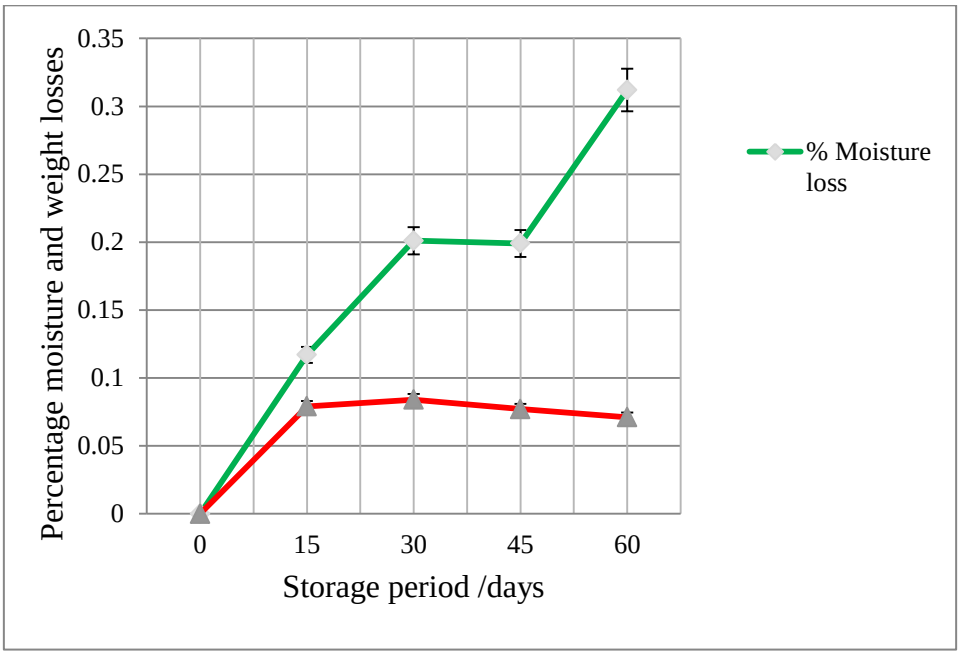


Figure 37: Effect of storage on percentage moisture and weight loss in sundried fish sampled from selected coastal markets in Southern Ghana.

Nonetheless, moisture loss between storage day zero (initial day) and day 15 was insignificant ($p \geq 0.05$) when compared with the other days of storage at 95% CI. ($p = 0.019$). There were no significant differences in the percentage moisture losses throughout the 60 days of storage ($p \geq 0.05$).

In Table 19, the microbial analysis showed no detection of coliforms in all fish sampled from the markets. The total bacteria (TVC) count/cfu/g varied from 4.80cfu/g to 5.12cfu/g with the lowest recorded in *S. dorsalis* ($p \leq 0.05$), and the highest in *O. niloticus* ($p \leq 0.05$).

Table 19: Effect of sun drying on proximate and microbiological safety of three species of fish (N = 60) from coastal markets in Southern Ghana.

Fish species	% Protein	% Ash	% FFA	% TTA	% pH	TVC(cfu/g)	MYC(cfu/g)
E.encrasicolus	0.13b	14.27a	2.91b	6.20b	6.78b	4.91a	4.75ab
Salted O niloticus	0.06a	10.13a	2.80a	6.57c	6.85a	5.12a	5.28a
S. dorsalis	0.21c	14.66b	4.22b	5.33a	6.98b	4.80a	4.39b
S.E	0.006	0.195	0.05	0.05	0.03	0.189	0.221

Mean values that do not share a common letter are significantly ($p \leq 0.05$) different.

Total fungi count (MYC) ranged from 4.39cfu/g to 5.28cfu/g recorded in *S. dorsalis* and *O. niloticus* respectively. The highest percentage ash content was recorded in *S. dorsalis* and the lowest was in salted *O. niloticus* ($p \leq 0.05$).

Variations in pH, TTA, and FFA were significant within the species ($p = 0.000$). Percentage protein content was highest in *S. dorsalis* and lowest was in salted *O. niloticus* and these however, were significant ($p = 0.000$).

In Table 20, the percentage protein varied slightly during the storage period with the highest recorded before storage (day 0) and until day 15 and remained constant until the end of storage. There was a significant difference ($p \leq 0.05$) between day 0 and day 15 of storage with respect to percentage protein loss. There were no significant differences in percentage ash, percentage TTA and reduction in percentage pH during storage ($p \geq 0.05$).

Table 20: Effect of storage on proximate and microbiological safety of sundried fish sampled from selected coastal markets in Southern Ghana.

Storage/ days	% Protein	% Ash	% FFA	% TTA	% pH	TVC (cfu/g)	MYC (cfu/g)
0	0.16a	13.08a	3.14b	5.99a	6.93a	8.20a	7.78a
15	0.15ab	13.26a	3.32ab	6.06a	6.91a	5.16b	4.94b
30	0.12b	13.22a	3.42a	6.06a	6.91a	4.42bc	4.42bc
45	0.12b	12.89a	3.35ab	6.05a	6.84a	3.77ad	3.72cd
60	0.12b	12.65a	3.33ab	6.03a	7.74a	3.18d	3.17d
S.E	0.01	0.25	0.07	0.06	0.04	0.24	0.29

Mean values that do not share a common letter are significantly ($p = 0.05$). different.

Percentage ash increased between day 0 until day 15, however, a decrease was recorded between day 30 until day 60 ($p \leq 0.05$). Total titratable acidity (percentage TTA) remained comparatively constant throughout storage just as it was recorded in percentage pH reduction.

Furthermore, there were significant variations within percentage FFA, TVCcfu/g and MYCcfu/g during storage. The least percentage FFA was recorded on day 0, and the highest on day 45 of storage ($p \leq 0.05$). Free fatty acid (FFA) increased between day 15 and day 30 and the highest was recorded by the end of day 30. However, there was a decrease from day 45 until day 60 ($p \leq 0.05$).

The highest bacteria and fungi counts were recorded on the day zero and the lowest on the day 60 of storage ($p \leq 0.05$). Before storage, total viable count (TVC/cfu/g)) was 4.8cfu/g. This increased up to 8.20cfu/g between the first day of storage (day zero) and day 15, but there after the number of counts began to decrease. Total fungi count (MYC) was 4.39cfu/g before storage. However, the number increased to 7.78cfu/g between day zero of storage and day 15 of storage. Nonetheless, both TVC and MYC recorded comparatively low counts with increasing storage and these observations were significant ($p<0.05$), (Table 20).

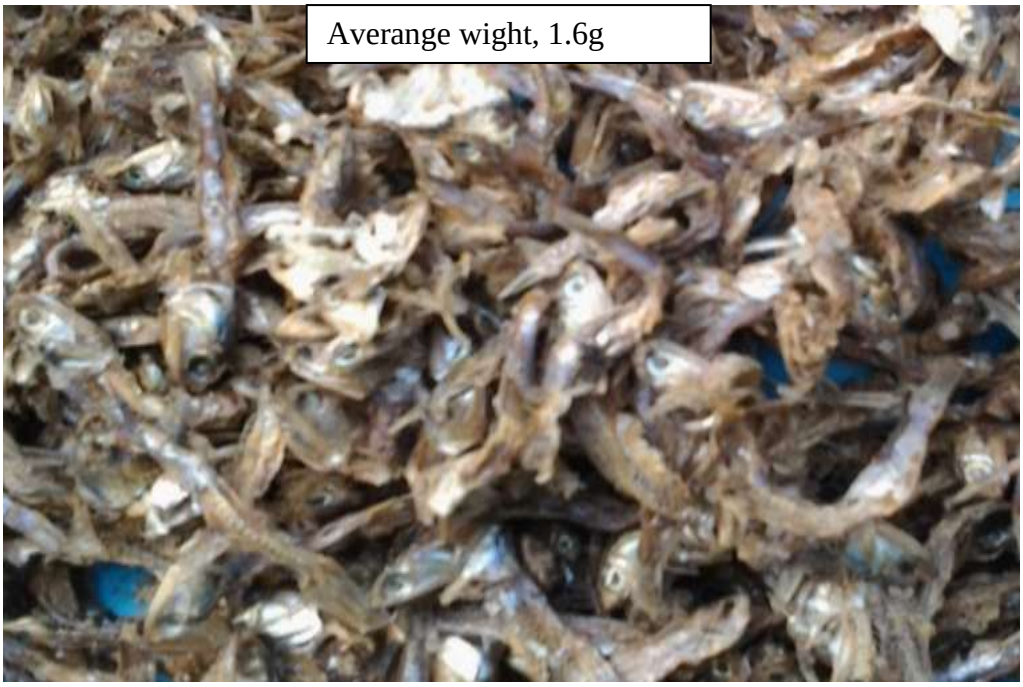


Plate 14: Non-irradiated *Engraulis encrasicolus* after storage period. This has been infested by insects during storage.

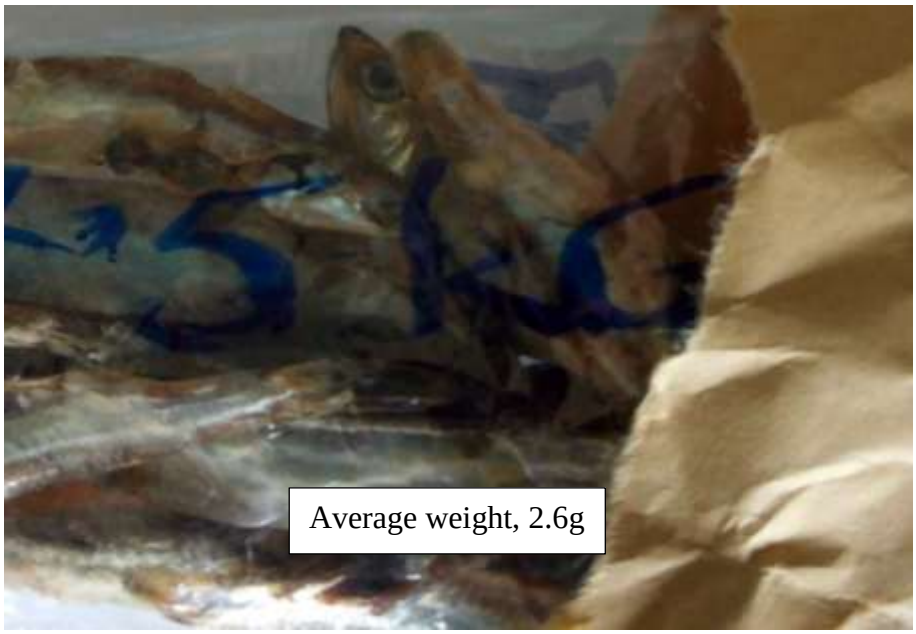


Plate 15: Irradiated *Engraulis encrasicolus* which was irradiated at 5kGy remained unaffected by insects after storage period.

Plates 14 and 15 show the differences between non-irradiated and irradiated fish products. Plate 14 shows how insect infestation negatively impact the quality of non-irradiated stored fish as compared to Plate 15 which was irradiated before storage and after storage.

Plate 16 below showed the effectiveness of irradiation technology in preserving smoked fish (*Sardinella aurita*). There was no insect infestation and deterioration of the physical appearance of the smoked fish after gamma irradiation.

Plate 16: Gamma irradiated smoked *Sardinella aurita*.

4.9 Isolation and Identification of Microorganisms and Insects from Fish Samples Before and During Storage

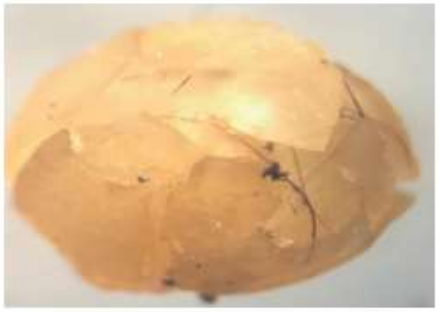
Before storage, the dominant fungi isolated and identified from all the dried fish samples had the appearance of creamy-white, yellow or orange coloured colonies. These were identified as *Aspergillus niger, Aspergillus flavus, Aspergillu formigatus, Pennicillium and Rhizopus spp.*

The bacteria species identified in the dried fish were *Bacillus cereus, Staphylococcus aureus, Bacillus mycoides, Bacillus lichmiformis, and Micrococcus, Pseudomonas* and *Streptococcus* species. *Micrococcus* and *Pseudomonas* were dominant in dried *O. niloticus*.

Two major insect infestations were identified that caused damage to the smoked and sun-dried fish products but mostly in the dried fish. These were *Dermestes maculatus* and *Musca domestica.*

The predominant insect species was *Dermestes maculatus* commonly named: hide beetle, and this infested and damaged the stored smoked and sun-dried fish in this study. The stages of insects identified in non-irradiated and stored smoked and sun dried fish taken with X100 HP of the Light Microscope are shown in Plate 17. The damages come from the insects' life cycle stages such as larvae or maggots of *Dermestes maculatus* and of *Musca domestica* during the early stages of drying of the fish.

Plate 17 shows the egg (17 A), larvae (16 B) and adult beetle (17 D) of *Dermestes maculatus* (magnification = X100 High Power of Light Microscope), which infested the stored fish. Mainly adult females lay their eggs in the gills and belly of the fish and after hatching, the young larvae then fed vigorously on the fish leaving behind powdery residues. Most of the damages in the dry fish were caused by the larval stage (Plate 17 B). These insects contaminated the fish with their faeces which appeared dark brown and chewy.



A: Egg

B: Larva

C: Pupa

D: Adult

Plate 17: Stages of development of *Dermestes maculatus* (Magnification: X100)

4.10 Evaluation of perception, food safety practices, knowledge of packaging, and acceptance of irradiated fish through focus group discussion (FGD) and participant observation (PO)

4.10.1 Background characteristics and outcome of FGD and PO study

This section summarizes the outcome of FGD and PO conducted as part of this study. Each FGD and PO was made up of an average of ten women (total sample of 80 women) with younger, middle-age and older women in a heterogeneous trade. This is because women play a key role along the fish value chain.

The participants had an average minimum age of 25 ± 3.53 years and a maximum average age of 64.29±10.61 years. The ages of the women in Akatsi ranged between 39 – 60 ± 21.21 years, Keta 30 – 75±31.82 years,

Madina 18 – 70 ±36.77 years, Chorkor 15 – 65 ±35.36 years, Mankesim 35 – 70 ±24.75 years, Elmina 45 – 65±35.36 years and Kpong 33-45 ±7.01 years.

The educational status of these participants who engaged in the fisheries post harvest industries was also assessed and each group was heterogeneous in terms of education. For example, out of the eighty participants, 5% (4) attended SHS, 38.75% (31) attended JHS and 56.25% (45) had no formal education. Between the FGs, only 3 (20%) participants in Keta had no formal education, Akatsi 6 (50%) participants had no formal education, Madina 7 (58.33%), Kpong 7 (70%), Chorkor 7 (70%) and Elmina 7 (63.635%) participants had no formal education. Mankesim recorded the highest number of 8 (80%) participants who had no formal education. The most experienced participant had been in the trade for 65 years and the least experienced was in the trade for 5 years. Looking at the age limit, the oldest age limit was up to 75 years, and falling within this age limit, it implies she started the fish trade at the age of 10 which could have revealed child labour practice in the fisheries. This implies that the women engaged in the fish value chain have an in-depth experience of fish postharvest handling.

(i) Knowledge of fish safety and description of the nature of spoiled fish

Across many of the focus groups, participants reported abysmal knowledge of food safety and uncertain feelings about fish spoilage. In expressing their perceptions about what safe fish is, the participant either attempted to define what the condition is or expressed opinions about how fish becomes unsafe for food. As noted by a respondent who participated in group one (1):

"I can see it getting spoiled because it is soft and smells and worms coming out of it. Sometimes I see something growing on it....." That growth could probably be fungi *(*Group 1). *"Sometimes the belly looks different…., the gills and eyes look green in colour and I look at the texture also which is soft and that is different from how it normally looks"* (Group 2), *If it is "aborbi" it turns red in colour.* One participant in Group three (3) said *"if the fish is the big type and it gets spoilt, the fish is used for fermented produce called "momoni" or lafufui fotsi (Ewe).*

(ii) Source of knowledge and training

The participants gave an account of how they acquired the knowledge of fish post-harvest handling and fish safety:

"*It was a family business",* one participant said. *"I grew to meet it and I have been doing this work since my early days of life", I learned it from my grandparents."* said another participant, *"I grew up to know it because I live in the coastal area and also from experience". "There was no employment except the fish trade which I grew up to meet"* (Group 1, 2, 3).

(iii) Identifiable features of spoiled fish

In narrating their skills on how fish going bad can be identified, the women attempted to explain their knowledge.

"When the fish is processed late after been caught, (Group 1), *it appears dark in colour, it becomes soft and houseflies come around it and it changes appearance……the bigger fish cannot be easily processed and stored for longer time….. four (4) days at best except continuous smoking …. This makes it costly on the market* (Group 1, 2 and 3). *"Houseflies infestation and itching on the tongue, and bad odour from the mouth of fish"* (Groups 2, 4, 5 and 6).

(iv) Causes of fish spoilage

The women also described the causes of fish spoilage and post-harvest losses. Trying to narrate their experiences, the participants said ……*"some fish spoiled after they are landed before reaching the shore, late processing ….and inadequate smoking, bad weather conditions for fish drying and improper storage facilities…., are some of our setbacks"* (all groups).

(v) Profitability of the current technology

"We do not have a better alternative to the current technology. What we have now is adequate for our needs but if there is any other way please teach us….(all groups)".

(vi) Benefits of either smoked or sundried fish trade

All the groups described their trade in terms of profit and loss as follows: *"We prefer the dried fish trade to smoked fish trade because it fetches more profit and involves less cost to process". " You don't need to buy fuel wood often" Eh!, for smoked fish I have to smoke it often so that it will not spoil"* One other participant said *"continuous smoking leads to the fish becoming small and light weight so that customers don't like light smoked fish.*

(vii) Profitability of the fish trade

The participants also admitted that *"all businesses come with profit and loss" "How for do I have no other job so I am just managing this for living" "I can hence hold to it and I can advise more people to engage in fish processing because no matter what, they can get some for food and sell the rest to cater for their children". "When fish is being dried and the rains come, the loss in terms of money is high… the weather is the determining factor when one*

wants to dry the fish"…. "the fish trade is very profitable (all groups) but it is plagued with many challenges".

"I lose a lot of money through my fish spoiling" I sometimes lose about two hundred Ghana Cedis (GHC200.00) at a time due to bad weather especially when I just dried the fish and then the rain comes" "I see insects and flies. They eat the fish leaving only bones" "When I see insects, then I spray them with mosquito spray or I light mosquito coil and place it under the basket".

(viii) Possible causes of fish spoilage

The groups in the three Coastal Regions unanimously responded: *I think that the "method of processing could be the cause of spoilage and losses:….the main means of fish processing is drying and smoking and that was what we have been practising for years… frying is just for a short period and this also goes rancid i.e. it tastes unusual to the tongue after few days."…… and from the way the fish is packaged before they bring them to the us the fishmongers …. storage and preservation, we sometimes use paper and baskets so the air and water cannot worry the fish, ……we also use pest and insect killers to prevent cockroaches and other insects from coming to eat the fish".*

(ix) Levels of concern of economic losses

In conveying their concerns with regard to financial losses, the groups stated that…."*we are very concern about the losses we incur because we work with bank loans. …. The cost of processing is high in terms of firewood, and transportation, and we do not have cold stores, we cannot pay back the money we took from money lenders and the bank……our time and personal money invested is lost and we cannot continue the business any longer so our children stay home because we cannot pay their school fees".*

(x) Worries of inflow (import) of fish from neighbouring countries

Expressing their worries about the losses in the community, the groups across the regions said *"concerns about the losses are very common with all of us"… the reason is that we do not get ready markets for our fish, others bring fish from Cote D'Ivoire, Gambia, Cameroon, Sierra Leone and Togo so when we are not able to sell ours. They go bad and we lose money".*

(xi) Losses due to the current technology

We incur losses due to the processing and sometimes preservation methods,….our packaging and the way we store the finished product also make us to lose money…. by the time we open the stored fish, it has already spoiled,…pests also disturb us".

(xii) Traceability and feedback

Traceability and feedback is a key to improving marketing relationships with clienteles and when the women's concerns were heard the researcher put the question which the participants responded as *"we do give feedback to our sources of acquisition of salted, smoked and dried fish products……"Yes*! *We do because the losses can be high sometimes….and they sometimes add up to the quantity purchased or they give a monetary reduction.*

(xiii) Preservation processes available to the women

When the women were asked the methods of preservation they employ in their processes, the responses were diverse: …. *"we the women have to buy the fish at high price so by the time we finish smoking and drying the price goes higher again….because the price is high, the customers do not buy often so the remaining go bad and waste..* "*The cost of preservation is high therefore we process on time, we smoke or dry the fish as soon as we come*

back from the shore or from the riverside"….the price of the fish is very high because of "no fish in the sea"… the men go to sea and come back with no fish or little fish".

(xiv) Suggestions to address the problems

To the question as to what they think can be done to address the problem of preservation, the participants seem to know little or oblivion. *….we have no idea so if the government can come to our aid…. (All Groups)".*

(xv) Willingness to use new technology if education is given on its benefits

To the question about knowledge of food irradiation, *the participant responded "we do not know of food irradiation" (All Groups). "….if it is going to help us to increase profit then we can use the food irradiation as you have said to us"….."If it will not cost us too much we will use it to keep our fish for a longer time and increase our livelihood (All Groups)".*

(xvi) Willingness to pay a little more for the use of irradiation facility

"*…if it is the new way, definitely it will cost high initially so we will try it" …..the prices are already high and difficult to sell, so any new preservation method that will cause more increase in prices will bring about low sales …… if the new method will increase the preservation and we can store the fish for longer time then we will be able to embrace it ……and if it will increase our profit in the long run we will use it but we cannot give any margin of increase to add to the selling price"* (All Groups).

(xvii) Knowledge of packaging and packaging materials currently available

The participants expressed their current knowledge of packaging of smoked and sun dried fish. *"..... We pack the fish in rubber or cement paper and then in old unused clothes and we put them in baskets" "....sometimes we leave the smoked fish on the smoking oven and cover them with either cement paper or jute sacks...."* (All the groups). *.....we also continue to dry the already dried fish to avoid spoilage and to decontaminate the fish of insects.*

(xviii) Forms of storage currently employed

Probing the knowledge of packaging and storage, the women emphasized: "*we have hired a room where we keep the fish in the market or at home,....We also can keep them on the oven for as long as it is not sent to the market* (Group 4)*...we have to keep smoking them every two to three day to keep the fish safe,..... We place them on lorry tires or on cement blocks and planks".*

(xix) Problems encountered during storage

We encounter flies and rodents some times,... the room has no windows so we spray the room with insect killer and sometimes we place lighted mosquito coils in the room to keep houseflies away,.....we also sprinkle cockroach and mice killers in the room,....some of us also use kerosene or we burn Nim tree leaves".

(xx) Willingness to re-package

The women in all the groups responded *"Yes!"* to willingness to re-packaging and adding that *"if there are new ways of packaging and storage we will like to know"* (All Groups).

4.10.2 Evidence of complaince

Ten participants in each market were assessed for compliance or non-compliance on ten critical areas such as: environmental hygiene, layout of processing area, personal hygiene, control of fish processing operations, fish handling procedure, water quality, storage, pest control, waste management and cleaning programme (Appendix C).

4.11.1 Site isolation of fish processing areas

In Figure 38, 10 participants observed in the Keta Municipality were scored for 1 representing 4.5%. These had fresh fish cleaning locations separated from fish smoking and fish drying sites. Only 21% of participants observed throughout the study were scored for very good site isolation of working area. Other participants observed in Akatsi, Keta, Madina, Kpong, and Sogakope all have good sites isolation with a score of 2 (47.7%) for their fish processing activities in response to good environmental hygiene. Elmina, Mankesim, Chorkor and part of Akatsi scored 3 (47.7%) for bad site isolation.

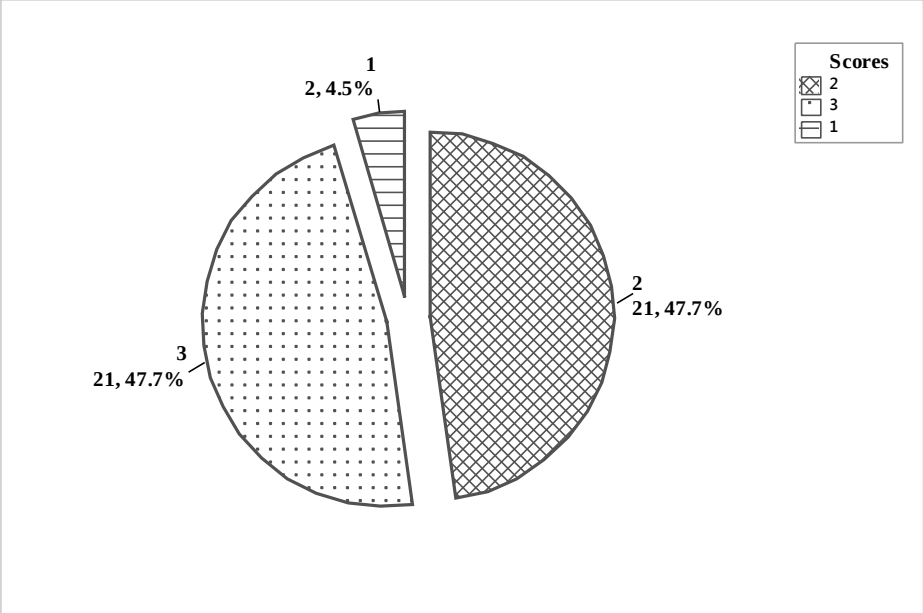


Figure 38: Scores for Site isolation of fish processing areas among fish participants in selected coastal markets of Southern Ghana (Legend: 1 = very good; 2 = good; 3 = bad).

4.11.2 Physical protection of processing site

In Figure 39, about 81% of all participants from Akatsi, Madina, Chorkor Mankesim, Elmina, Kpong and part of Sogakope had no physical protection for fish processing facilities. Only 6.3% had scored very good (1) physical protection for the processing sites and 12.5% was scored for good (2) physical protection sites.

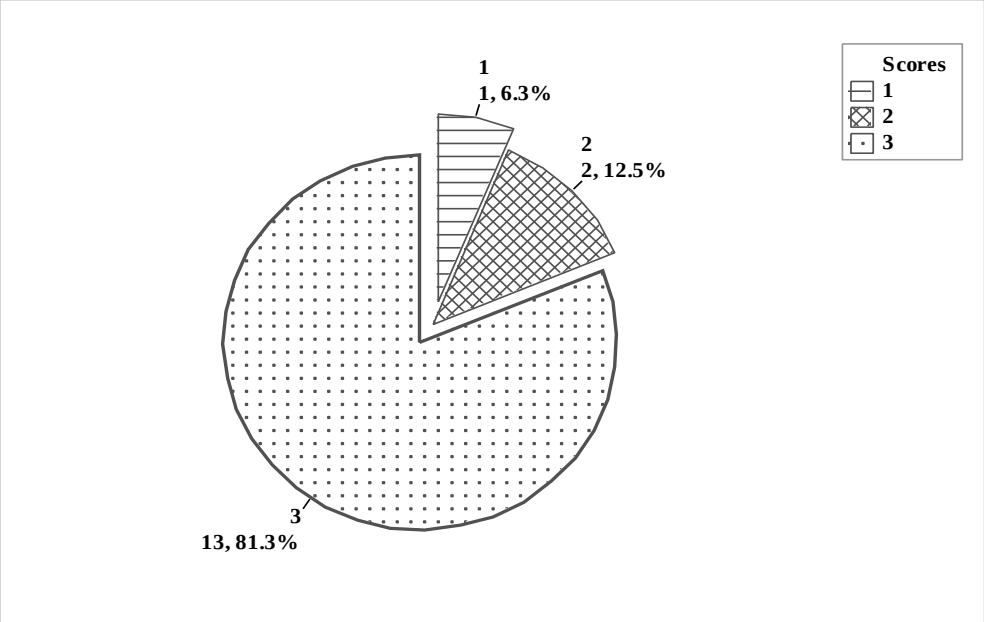


Figure 39: Scores for Physical protection of fish processing areas among fish participants in selected coastal markets of Southern Ghana (Legend: 1 = very good; 2 = good; 3 = bad).

4.11.3 Clean location for fish processing

Figure 40 indicated that participants in Akatsi, Keta and Madina were scored 2 constituting 31% for good clean location for used for processing as compared to participants in Chorkor, Kpong, Sogakope, Mankesim and Elmina who were scored 3 constituting 68.8% for having unclean (bad) locations.

Accessibility to drains for easy cleaning of the processing area was scored 1 for very good making up only 6.3% in Keta but in Madina, Mankesim and Elmina participants were scored 2 (37.5%) for good access to cleaning the processing locations.

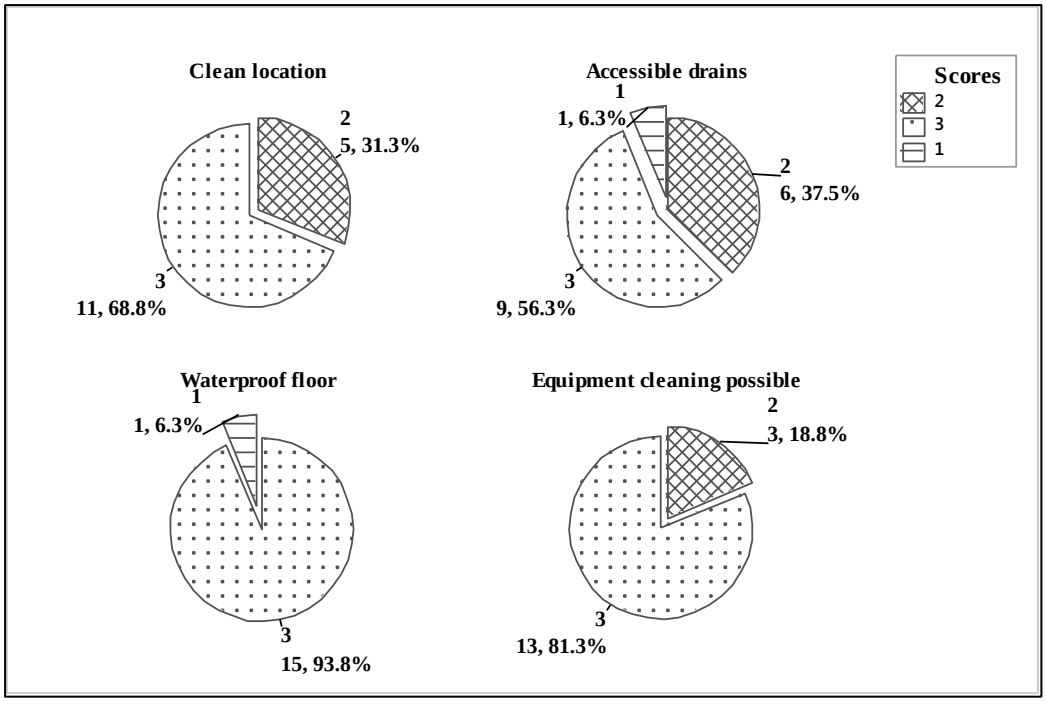


Figure 40: Scores for clean location, accessible drains, equipment cleaning and regular cleaning programme in processing areas (*Legend: 1 = very good; 2 = good; 3 = bad*).

The rest of the participants did not meet up to the accessibility test therefore were scored 3 (56%) for bad access to drains (Figure 40).

Some areas in the Keta Municipality have waterproof floor for fish processing and were scored 1 constituting only 6.3% for very good compliance. Other areas observed were scored 3 (93.8%) for not having waterproof floor therefore designated bad areas.

Areas having equipment cleaning programme were scored 2 (18.8%) for very good at Kpong, Chorkor and Keta which suggested compliance but the rest of the markets were scored 3 (81.3%) for bad practices and non-compliance to environmental hygiene (Figure 40).

4.11.4 Water quality management for fish processing

4.11.4.1 Presence of portable water

In Figure 41, few operations observed, 75% of the participants did not use portable water and were awarded score of 3 signifying bad practices or non-compliance. Markets such as Akatsi, Keta, and Madina where only 25% of the participants use portable water were assigned score of 2 denoting good or compliance.

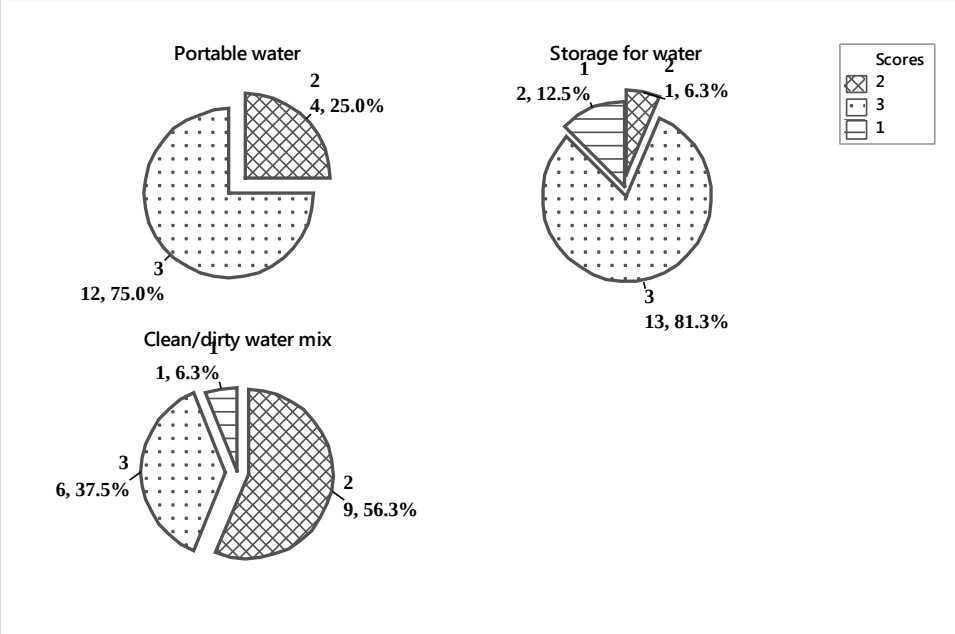


Figure 41: Scores for Clean water system for fish processing areas among participants in selected coastal markets of Southern Ghana (*Legend: 1 = very good; 2 = good; 3 = bad).*

4.11.4.2 Presence of water storage facilities at the processing site

In Figure 41, only 6.3% of participants in Keta and Madina were awarded scores of 1 for very good water storage facilities and 12.5% of the participants were scored 2 indicating good water storage compliance. The rest of the participants 81.3% observed in Chorkor, Kpong, Mankesim and Akatsi markets were awarded scores of 3 signifying bad water storage compliance.

4.11.4.3 Separation of clean water from unclean water for fish processing

In Figure 41, when participants in Akatsi and Madina were observed for the separation of clean water from dirty water, they were awarded scores of 1 (6.3%) and 2 (56.3%) for compliance. Other markets including Kpong, Sogakope, Mankesim and Elmina were assigned the score of 3 (37.5%) signifying non-compliance.

4.11.5 Regular waste removal from site

Figure 42 showed that waste marterials were regularly removal from the fish processing areas in Elmina, Mankesim and Sogakope markets and were assigned the score of 2 (75%) as good. In the rest of the markets the score of 3 (25%) was assigned as bad practices of waste management.

4.11.6 Possibility of pest infestation

Figure 42, most stores observed were possibly free from pests in both the processing and storage areas and therefore were assigned the score of 2 (25%). There was a high possibility (75%) of pest infestation depicted by scores of 3 in all the markets observed.

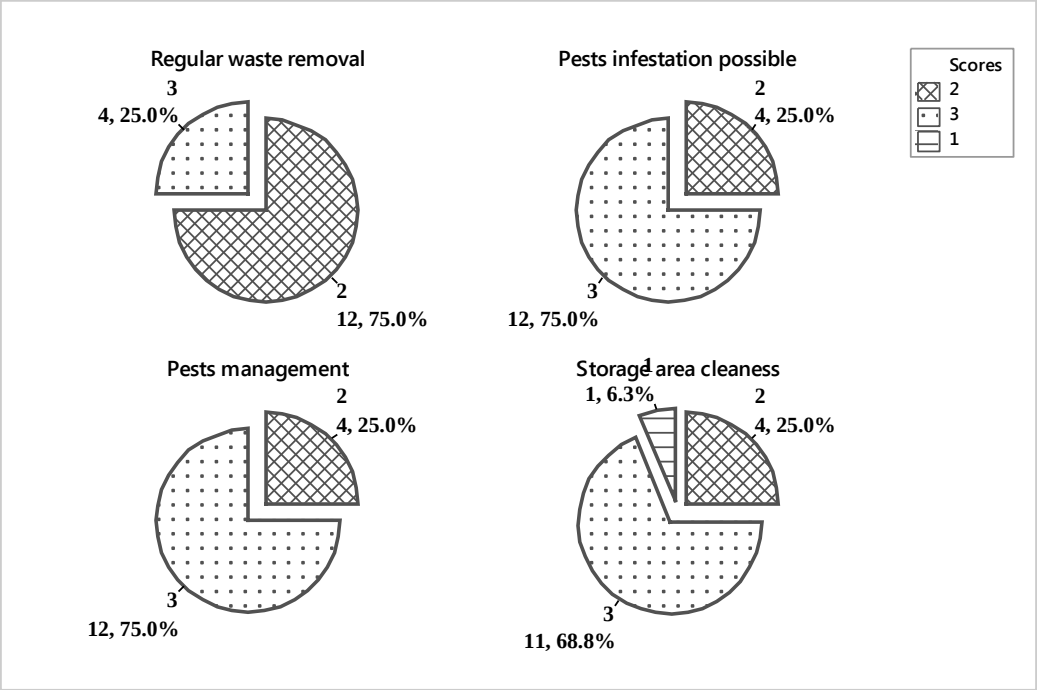


Figure 42: Scores of pests' management in storage areas in selected coastal markets in Southern Ghana (*Legend: 1 = very good; 2 = good; 3 = bad).*

4.11.7 Pest management

Bad pest management was scored for 3 (75%) in some markets among the traders observed and scores of 2 assigned for good (25%) pest management in other markets. Signs of the presence of pests in some markets were high with a score of 3 signifying bad pest's management in the markets (Figure 42).

4.11.8 Clean storage area

In Figure 42, participants were scored 1 representing 6.3% in Keta for clean storage area for fish. Those in Madina, Sogakope and Elmina were all awarded the score of 2 (25%) for good and clean storage areas. In all other markets 68.8% of the participants were scored 3 for having bad and non-compliance to clean store areas.

Plate 18: Landing of *E. encrasicolus* (Aborbi) on the Keta beach

Fish post harvest losses begin at fish landing since fish is landed directly on the beach sand. The landed fish may begin to deteriorate under the hot weather condition conducive for fast microbe multiplication. In Plate 18, Aborbi is landed at Keta beach in heaps ready for sale.

Plate 19: Preparing fresh fish for smoking

In Plate 19 fish is often processed on the floor which constitute the basis for contamination. The wash water is not changed often and several rounds of

washing of fish leads to recontamination of fish before either drying or smoking.

CHAPTER FIVE

5.0 DISCUSSION

Polycyclic aromatic hydrocarbons (PAHs), pesticides (OCP, OPPs and Pyrethroids) and microbial contaminants have been studied in samples of six species of commonly consumed sun-dried and smoked marine and freshwater fish in Ghana. This study was carried out using standard AOAC methods and Standard Operation Procedures attested to in consumer and food quality and safety studies (Yeung and Morris, 1986; Andrada, 2014; Walker, 2017; Horita *et al.*, 2018), and food irradiation technology (Farkas, 2006; Studies, 2009; Ehlermann, 2016). Secondly, a subjective analysis comprising of focus group discussion and participant observation was carried out to ascertain the post-harvest risk factors that plague the fishery industry in Ghana.

5.1 PAH in the Ghanaian sun dried and smoked fish

This study revealed high concentrations of PAHs: 11.01 µg/kg for B(a)A to 33.55 µg/kg for B(b)F, all in smoked fish and 5.76 µg/kg for B(g, h, i)P to 47.68 µg/kg for B(b)F, all in sun dried fish in the Ghanaian markets. The high concentrations of PAHs in the fish samples were above the maximum limits (MLs) or maximum permissible concentrations (MPCs) of 0.9µg/kg for Chrysene: 0.07µg/kg for fresh water and 0.0007µg/kg for marine water (Moermond/Verbruggen, 2010); MPC for B(a)A is 0.1µg/mg for freshwater; and 0.01ug/kg for marine water; B(a)P has been set at 5µg/kg (Debrikova and Svetlikovi, 2007). These high concentrations realized above may be the result of uncontrolled processing such as smoking and re-smoking of fish and also drying on the ground and by the road sides where dried fish

gets secondary contamination from the soil and exhaust fumes from vehicular traffics (Alexander et al., 2008; Vane et al., 2014).

There were similarities in the PAHs observed in the different fish species from the regions and this confirmed the dynamics of the smoked and sun dried fish trade across the markets and also revealed the cultural practices in the fisheries post-harvest processes.

The MPs of Chrysene (0.9 µg/kg) is lower than it was found in smoked and sundried fish analysed in this study. The high concentration of Chrysene (15.26 µg/kg) in this study could have resulted from human activities such as incineration of waste materials such as polythene sheets and incomplete combustion of wood for smoking fish and the indiscriminate waste burning. The concentrations were higher than the Maximum Acceptable Concentration for water and the ecosystem (MACwater,eco); 0.07µg/kg for freshwater and 0.007µg/kg for marine water (Moermond & Verbruggen, 2010). This therefore revealed the state of pollution of the Ghanaian smoked and sun-dried fish. Most rivers from which fish is captured flow through farming areas which become sinks for smokes from vegetation fires, fossil fuels, industrial activities including illegal mining and illegal fishing effects. Vehicle washing along water bodies, open dumps, and solid wastes such as rubber bags and tyres and E-waste burning, all of which currently occur in the vicinity of most of these water bodies could lead to increased concentration of Chrysene as well as other PAHs as was observed in other research works (Final, 2007; Alexander et al., 2008; State & State, 2011). These authors found out that the major sources of PAHs in the biosphere are human utilization of petroleum products and incomplete combustion of fossil fuels, biofuels or other forms of

organic matter, which far exceed natural sources. As a result, the PAH concentrations in sediments increase at points that are near emission sources, especially near urban and industrial areas that often have several point sources of release such as smoke houses (Abdel-Shafy et al., 2016).

This study also identified Benzo(a)anthracene in all fish samples and showed that the mean concentrations in each fish sample were higher (14.13 µg/kg) than the MACwater, eco: 0.1µg/kg for freshwater and 0.01µg/kg for marine water. The mean concentrations of

Benzo(a)anthracene in this research were also higher (14.13 µg/kg) than that set by the EPA of the USA, called maximum contaminant level (MCL): 0.0002 mg/kg, to protect and prevent potential health problems. However, other studies reported by Veyrand et al. (2013); Rengarajan et al. (2015) and confirmed by Abdel-Shafy et al. (2016) showed that, different PAH sources depend on the specific region or local sources examined and on the analytical methods applied including number of PAH analytes. This is why the lists of PAHs assigned to sources differ from those in this study (Peter & Oliver, 2013).

The maximum acceptable concentrations for Benzo(k)fluorathrene MACwater, eco = 0.1µg/kg for freshwater and 0.01µg/kg for marine water however, the concentrations found in smoked and sun dried fish in the study were higher than the MAC and could pose a possible health hazard to consumers.

The high concentrations of B(a)P and the other PAHs realized in dried fish could not only be due to its presence in the water, but also due to drying of fish along roads and in open compounds. Vane et al., (2014), realized PAH

in soils studied in the United Kingdom and Essumang (2010), also observed some levels of PAH in roadside soils in Ghana. This could have accounted for the high concentrations of PAH 47.63 µg/kg realised in sun-dried fish. In most markets in Ghana, fish and other fish products are displayed in bowls placed on the ground or are displayed on table tops without protection from dust and this could contribute to air-borne PAHs contaminating fish.

The concentration of B(a)P in smoked fish in this work was much higher (14.08µg/kg) than the MACwater,eco (5.0 µg/kg) than MACwater, eco.

The concentration of Benzo(b)fluoranthene realized in the current research was in excess of the MACwater, eco for freshwater and for marine water because each PAH occurs in the mixture of other PAH. Acute toxicity was reported seldom in humans, fish, or wildlife, because of exposure to low levels of a single PAH compound such as this one. Although there are no human data that specifically link exposure to Benzo(b)fluoranthene to human cancers, Benzo(b)fluoranthene is a component of mixtures that have been associated with human cancer. The rejection of smoked fish exported from Ghana was due to the high concentrations of PAHs specifically B(a)P which is reported as carcinogenic. These include coal tar, soot, coke oven emissions and cigarette smoke Alexander et al., 2008; Abdel-Shafy & Mansour 2016).

The mean concentrations (5.76 µg/kg – 20.45 µg/kg) of Benzo(g, h, i)P and (5.76 µg/kg - 16.55 µg/kg) of Indeno(1,2,3,c,d)P in this study were also above the MACwater,eco for freshwater and also for marine water (0.01 µg/kg). Palm et al. (2011), observed higher mean concentrations (28.5 µg/kg – 406.37 µg/kg) of PAHs in smoked fish samples from Winneba, Madina, Chorkor and Ada markets compared to those found in this study. The high

concentrations (5.76 µg/kg – 20.45 µg/kg) was suggestive of deposition of more smoke from previous smouldering processes that occurred in the smoking chamber.

The incomplete burning of the previous PAH residues may have led to the formation of additional higher molecular weight PAH and, subsequently increased the PAH concentration in the samples (Palm et al., 2011; Silva et al., 2011; Vane et al., 2014). Earlier researchers had found out that the concentration of PAHs in smoked marine fish ranged from 46.5µg/kg to 124µg/kg and B(a)P was 0.7µg/kg (Moermond & Verbruggen, 2010). The results (5.76 µg/kg – 20.45 µg/kg) of the present study are comparable to the results found by these researchers therefore the concentration of PAH in smoked and sun-dried fish on the Ghanaian markets are above the recommended values for human safe consumption.

5.2 OCPs in Ghanaian sun dried and smoked fish

A number of organochlorine pesticides (OCPs) have been identified in samples of six species of smoked and sun-dried marine and freshwater fish from selected coastal markets in Southern Ghana. The results of the analyses of salted, smoked, and sundried fish in this study (Aldrin: 15.2 mg/kg/bw/day; Cyfluthrin: 30.8 mg/kg/bw/day) showed that the concentrations of the individual OCPs realised were above (Aldrin: 0.0001mg/kg/bw/day; Cyfluthrin: 0.02 – 0.05 mg/kg/bw/day) the requirements for maximum permissible concentration for water and the ecosystem (MPCwater, eco). This observation could probably be the result of increased pollution of the environment through extensive use of OCPs for the purpose of high agricultural production (Jayaraj et al., 2016a). This finding has further

underscored the fact that there is probably unregulated use and disposal of OCPs that are potentially hazardous to human health (Moermond & Verbruggen, 2010; Fianko et al., 2011). These authors attributed their observations to unregulated application of OCPs and other pesticides on farms in the vicinities of water bodies and also in fishing activities. The presence of OCPs in processed fish therefore renders the sun dried and smoked fish products from Ghana unhealthy and may not meet the export standard.

The concentrations of Permathrin (60.2 mg/kg/bw/day), Cyfluthrin (30.8 mg/kg/bw/day) and Cypermethrin (63.2 mg/kg/bw/day) residue were high in smoked *Engraulis encrasicolus* as well as in sun dried *Engraulis encrasicolus*. This raises safety concern of both sun dried and smoked *Engraulis encrasicolus*. These fish products contained more than one OCP residue and bio-accumulate in them as was reported by WHO (2009). These findings reported the possibility of encountering one or more pesticide residues in one food item. The contamination of processed *Engraulis encrasicolus* with three OCPs in this study agrees with the findings of Jayaraj et al., (2016) and Abdel-Shafy et al., (2016) when they reported pesticides bio-accumulation in water-bodies and in aquatic organisms.

The concentration of these OCPs in the fish samples such as the smoked, sun dried and salted and sun-dried fish in this study exceeded the WHO's GV (World Health Organization Guideline value) and the Australia and New Zealand's HV (Health Value) limit of 0.03µg/kg for Heptachlor and Aldrin in water and the environment (Hamilton, Ambrus, Dieterle, Felsot, & Harris, 2003). Although these residues occurred at lower concentrations in some of the fish sampled than found in other studies (Tashk & Ka, 2015), they

may bio-accumulate to higher levels in human beings who consume these fish (Rani et al., 2017). Bio-accumulation leads to health problems in animals including humans because OCPs are lipophilic and aid the bio-accumulation in tissues of organisms (Ka, 2015).

The consumption of smaller fish such as sun dried *Engraulis encrasicolus* and smoked *Engraulis encrasicolus* which can be eaten as a whole would provide readily available calcium and micronutrients such as iodine, selenium, zinc, iron, phosphorus, potassium and vitamins like vitamin A, vitamin D, and vitamin B-complexes (Ross et al., 2007). However, the concentration of the OCPs in these smaller fishes may be contributing to health harzards in the consumers. Large number of the poor coastal dwellers would therefore tend to consuming smaller sun dried and smoked fish resulting in an increased health risk from the indiscriminate use of toxic pesticides such as OCPs by the fish processors to preserve and store their produce (Allison & Ellis, 2001).

The high concentrations (3.0 mg/kg/bw/day – 154.68 mg/kg/bw/day) of OCPs recorded in these fish samples during this study could be the results of their use to prevent insect infestation during storage. This was also evident from the focus group discussions and participants' observation organized during this study. Earlier researchers have reported high concentrations of various pesticides in several media including water, foodstuffs and human breast milk (America Academy of Pediatrics, 2003; Jayaraj et al. 2016a).

Others similarly reported persistence of organochlorine compounds to have varied from moderate concentration with half-life of approximately 60 days to high concentration with half-life up to 10–15 years (Jayaraj et al.,

2016a). This suggests that even after a long time storage of salted, smoked and sun dried fish, organochlorine pesticide residues could persist and could have adverse effect on consumers. Various types of pesticide residues have been found to remain in the environment for longer periods and bio-accumulate in plants and animals, which might have led to contamination of food including fish consumed by humans (Rani et al., 2017).

In Bangladesh, studies on the conservation of dried fish showed that a mixture of organochlorine insecticides (DDT & Heptachlor) was used to protect dried fish from bacterial infestation (Huda, Dawlatana, Rahman, & Akter, 2009). This is confirmed by this study in Ghanaian sun dried and smoked fish.

The acceptable daily intake of pesticides for man, (mg/kg/body weight/day), were reported for some pesticides as Permathrin 0.05mg/kg/bodyweight/day, Fenpropathrin 0.03mg/kg/bodyweight/day (Ólafsdóttir, 2012); Aldrin 0.0001mg/kg/bodyweight/day (WHO/FAO, 2005), Fenvelerate 0.02mg/kg/bodyweight/day (Ólafsdóttir, 2012); Cyfluthrin 0.05mg/kg/bodyweight/day (USEPA, 2014); Heptachlor 0.0005mg/kg/body weight/day and for Methoxychlor 0.1 mg/kg/bodyweight/day (Boada et al., 2016). The consumption of any food product with pesticides concentrations higher than the daily allowable concentration constitute health hazard to consumers as in this study. The values (3.0 mg/kg/bw/day – 154.68 mg/kg/bw/day) in the present study therefore raise health concerns compared with the USA cancer risk of 0.0008 mg/kg for Heptachlor (Hamilton et al., 2003).

The most prevalent pesticides in the smoked and sundried fish in Ghana may have been ingested by fish from the polluted water bodies and sediments due to illegal fishing methods such as the use of chemicals in fishing and unwholesome preservation and storage methods as revealed by the focus group findings in the present study.

In the European Union, the minimum specified requirement is 0.0001mg/kg for each pesticide in drinking water intended for human consumption and a maximum of 0.0005mg/kg for total pesticides, except for Aldrin, Dieldrin, Heptachlor and Heptachlor epoxide, which are each limited to maximum levels of 0.00003mg/kg (Hamilton et al.,2003). Levels exceeding the maximum established limits have been reported in this study however, maximum limits for residues have been established only for certain pesticides (USEPA, 2014).

The concentrations of Permathrin (60.20 mg/kg), Delta-HCH (4.40 mg/kg) Fenvelerate (26.0 mg/kg), Heptachlor (3.54 mg/kg) and Aldrin (6.20mg/kg) in the smoked, salted and sundried fish exceeded the WHO's (World Health Organization) Guideline value (GV) and the Australia and New Zealand's Health Value (HV) limit of 0.00003 mg/kg for Heptachlor and Aldrin in water and the environment (Hamilton et al, 2003).

5.3 The decontamination effect of gamma irradiation on sun dried and smoked fish in relation to polycyclic aromatic hydrocarbons (PAHs)

It was evident from the results in this study that gamma irradiation has the potential to decrease the contamination levels of PAHs in the salted, smoked and sun dried fish samples with the increase of the absorbed dose applied and at a lower dose rate. This observation has been reported by

Lbpine et al. (1991) when they reported that the degradation extent of organic substances increases with increasing irradiation dose.

Aubin et al. (2011) observed in their work that B(b)F occurred at a higher concentration than all other PAHs in a given medium. In this study, B(b)F recorded the highest concentration before gamma irradiation which suggested the prevalence of this PAH in the environment. B(b)F was seen to be the most suitable PAH as a potential alternative for the environmentally labile B(a)P as an index of exposure to PAHs in the environment (Aubin et al., 2011).

This observation was comparable to that observed in the removal of organic pollutants in wastewater by another researcher group (Basfar et al., 2012). However, the irradiation doses used in wastewater decontamination were higher than that which is allowed for use on food for human consumption. The International Atomic Energy Agency (IAEA) recommended that any food material irradiated up to a maximum dose of 10.0 kGy is safe for human consumption and does not require any further toxicological tests (Arvanitoyannis & Tserkezou, 2007). In this study, the degradation of PAHs in smoked and sun dried fish increased with increase absorbed dose as also reported by Meguenni et al. (2011) that a 99.96% degradation of Naphthalene was achieved when the irradiation dose was increased from 6.0 kGy to 30.0 kGy. In the work of Khalil, Albachir, & Odeh (2016), the results demonstrated a reverse relationship between PAHs concentrations and gamma irradiation doses as well. They also reported PAHs concentrations decreased when the gamma irradiation dose increased as also reported in the current study and the total of PAHs concentration decreased by 35% at 1.0 kGy while

the reduction exceeded 70% for doses higher than 5.0 kGy. Khalil and colleagues found out PAHs reduction in irradiated wheat grain samples versus irradiation dose was linear/exponential depending on the structure of PAHs compounds. The results in this current study could be of great importance when gamma irradiation is used to complement the various ovens currently used for the elimination or reduction of PAHs from sun dried and smoked fish.

5.4 The decontamination effect of gamma irradiation on sun dried and smoked fish in relation to organochlorine pesticides (OCPs)

The total Organochlorine pesticides (OCPs: Cyfluthrin, Aldrin and Heptachlor) content in the different fish samples varied from 0.001 µg/kg to 0.862 µg/kg before gamma irradiation (Table 13). OCPs degradation has been shown to be feasible in this study through the use of gamma irradiation. Furthermore, the degradation level increased with increasing irradiation dose (Lbpine et al., 1991). OCP concentration in the fish samples after gamma irradiation ranged from non-detection (0.00 µg/kg) to 0.007 µg/kg. Studies carried out by some group of researchers found out that irradiation of OCPs resulted in their degradation in solution (Mel'nikova et al., 2017). Lbpine et al. (1991) observed irradiation degradation products have been identified in organic solvents, but very few studies of this type have been performed on irradiated food signifying a knowledge gaps. The reports showed the degradation of Aldrin at 5.0 kGy from 1.0 µg/kg to 4.9 x 10^{8}mol L^{-1}/kGy. Lbpine et al. (1991) concluded that the degradation of OCPs upon irradiation is linearly proportional to the irradiation dose applied. Irradiation degradation of OCP residues is depended on factors such as the physical state of the substrate, the irradiation dose rate applied and the concentration of the residue.

In this study OCPs' concentration was a factor determining the irradiation efficiency. Considering the maximum radiation dose of 10 kGy recommended by the Food and Agriculture Organization of the United Nations (FAO) for food processing, the complete degradation of some OCPs in the study which used up to 7.5 kGy irradiation dose could not degrade some OCPs due to their high initial concentrations as seen in Cyfluthrin and Gamma HCH. Nevertheless, some OCP residues which were identified in the non-irradiated samples in this study were completely degraded to 0.00µg/kg from their initial concentrations after gamma irradiation at 5.0 kGy and 7.5 kGy (Table 13). This could be due to their initial low concentrations as seen in Gamma HCH, Lambda-cythal, Heptachlor, Deltamethrine and Delta HCH in the study. This showed the effect of increasing radiation dose on the degradation of these OCPs as was also reported by Lbpine et al. (1991) that any pesticide degradation increases with increasing irradiation dose. Furthermore, it was observed that pesticides absorbed on dry solids such as dried fish, or frozen food, degradation is very low than that in non-frozen matrix or solution. Toxicologically, there are very few data about this aspect which does not give a conclusive fact about irradiation degradation of OCPs in food (Lbpine et al., 1991).

5.5 Effect of gamma irradiation on quality and safety of salted, smoked and sun dried fish

Chemical modifications by major and minor radiolysis effects of ionizing radiation depend on the absorbed dose, dose rate and irradiation source type, presence or absence of oxygen and temperature (Mostafavi, 2008). By and large, nutrient contents of food micronutrients mainly water-soluble and fat-

soluble vitamins, and macronutrients as carbohydrates, proteins, and lipids are not affected when irradiated at 10 kGy except when the dose is above 10 kGy (Bahraini, Salari, Sari, Fayazi, & Behgar, 2017).

The effect of ionizing radiation at maximum dose of 7.5 kGy on the quality and safety of salted, smoked and sun dried fish in coastal markets of Southern Ghana were assessed and found that it could be a tool for enhancing value-chain development, reduce postharvest losses and give the fishers improved technology for fish preservation.

Traditional processing of fish by salting, sun drying and smoking in combination with packaging and irradiation controlled the microbial population better (Tarhouni, Miloud and Mtar, 2015) under low moisture and neutral pH than the non-irradiated samples. This was due to the reduced moisture content which is an index of perishability of food products (Tarhouni et al., 2015). Thus low moisture content and fairly neutral pH realized in this study may have been the reason for low microbial load during storage.

Bacterial population reduction to 2.55 logcfu/g and fungi population reduction to 0.06 logcfu/g at low dose irradiation of 1.0 kGy - 5 kGy in this study confirmed the findings by Tarhouni, Miloud and Mtar (2015). Since gamma irradiation inactivated the microorganism in food, the microorganisms were not exposed to adequate period to deteriorate the quality of tissue lipids and proteins. The low total viable count of 2.55 logcfu was due to the irradiation effect on the spoilage microorganisms that could break down the protein component of the smoked fish (Mostafavi, 2008). The heating process associated with sun drying, hot smoking and re-smoking in combination with gamma irradiation could also reduce the moisture content (Akuamoa,

Odamtten, Kortei, & Yildiz, 2018), and therefore the water activity of the microbes and in that way limiting microbial growth (Diei-Ouadi & Mensah, 2005). Nonetheless, the rate of microbial spoilage depends upon the number of microorganisms present on the fish and the temperature at which the fish is kept (Diei-Ouadi & Mensah, 2005). However, the effect of gamma irradiation on the microbes could be the reason for the total elimination of some disease causing species. The presence of microbes on the fish after smoking and drying, and high moisture content may have resulted in the spoilage of the unirradiated fish samples.

Total protein content of smoked fish was not affected by either irradiation or the period of storage and to have a longer storage life, high protein value is required. In this study, since gamma irradiation did not negatively affect protein content and thereby not significantly reduced its level, oxidation and rancidity was likely reduced. Ayinsa & Maalekuu (2013) reported similar observation when they studied non-irradiated smoked red fish and found out that protein nitrogen was not reduced during drying and smoking but that the process probably concentrated crude protein component of the fish. However, in salted

and sun dried *O. niloticus* the protein content reduced during storage probably due to the salt effect as also observed by Ayinsa et al. (2013). Free fatty acid content in salted and sun dried *O. niloticus* also reduced probably due to tissue breakdown and sun drying effect similar to other findings (Ayinsa et al., 2013). A similar trend was also observed in soybean seed by Botanicae et al. (2016) that total protein content was not affected by irradiation. However, the fish protein denatured at higher temperatures of smoking and re-smoking

resulting in the fish obtaining a "cooked" appearance as in hot-smoked products such as those found in this study, irradiation may not affect the macronutrients (Mostafavi, 2008).

The pH of smoked fish samples remains stable ranging from 6.53 to 7.05 during storage in this study. The pH of a fish is a reliable indicator of the degree of freshness or spoilage. The pH of fresh-water fish is almost in the neutral range (Farid et al., 2014). According to other researchers, as the fish decomposes the pH increases (Farid et al., 2014). The fairly stable pH within the neutral range as realized in this study may indicated the preservation of the quality of the salted, sun dried, smoked and packaged fish samples and the inhibition of bacterial growth in this study (Tarhouni, Miloud and Mtar, 2015).

It has been reported that harmful food-poisoning bacteria are not found on fish in their natural aquatic environment. When fish comes into contact with dirty unclean surfaces after fish capture, poor handling during value addition potentially dangerous food-poisoning bacteria infestation resulted in spoilage (Tauxe, 1997; Report et al., 2009a; Badr, 2015). This situation was observed during the participant observation section in this study. *Sardinella aurita* had the lowest bacteria count of 2.78 logcfu/g when the pH was within the neutral region and *O. niloticus* the highest of 4.79 logcfu/g and these high levels of bioburden may have been reached through unhygienic hand (Figure 33). This was evident in the present research with *C. nigrodigitatus* having the highest bioburden of both total bacterial (7.60 logcfu/g) and fungi counts (7.17 logcfu/g) before gamma irradiation (Figures 24 & 25).

The lowering of the water activity also reduced the pH and this was also observed by Adenike (2014). There was therefore a synergy between the

pH and the moisture content in limiting bacterial growth and therefore preventing spoilage. A similar relation was explained by Prakash (2014), that heat resistance of microorganisms was likely related to pH of foods/fish.

Weight losses occurred in all the smoked and packaged stored fish samples in this study especially during the first 15 days of storage. Smoked *Sadinella aurita* may maintain a fairly constant weight for a longer time up to 60 days of storage when packaged and irradiated. Reduction in weight in storage of similar foods have been observed by several workers (Arvanitoyannis & Tserkezou, 2007; Taylor et al., 2009). Weight losses decreased with higher irradiation doses implying that irradiation would not significantly affect the initial weight of smoked fish and the fish products can be sold by weight. This shows that irradiation can preserve the weight of processed fish for long time storage and to enhance livelihood through trade by fisher women.

Percentage moisture loss was low (0.05% - 0.08%) when smoked and packaged *S. aurita* was irradiated at a minimum dose of 2.5kGy, and this preserves the corresponding body weight which will then appeal to customers who buy the fish by weights. Though this moisture loss was significant, the corresponding weight of the fish was not affected (Reza et al., 2015). Free fatty acid in *Engraulis encrasicolus* increased with increasing irradiation but this did not affect free fatty acid in the rest of the smoked fish samples analyzed.

There was significant reduction in both bacteria and fungi in all the smoked fish species at the limit of the irradiation dose applied in this study as also observed by Prakash (Prakash, 2014). The synergistic effect of hot-

smoking and irradiation contributed to the reduction of microbes (Taylor et al., 2009; Tarhouni, Miloud and Mtar, 2015). The fairly constant retention of protein suggested the ineffectiveness of bacterial degradation of the protein hence the fish maintained its freshness during storage (Getu et al., 2015). The reduction in the moisture content indicated the ability of gamma irradiation to preserve the quality of the smoked and packaged stored fish as also reported by (Arvanitoyannis and Tserkezou, 2007; Reza et al., 2015).

Protein therefore was not significantly affected by the irradiation doses used in this study being low and medium doses which caused less chemical reactions and induced only a small breakdown of food proteins into lower molecular weight proteins and amino acid (Mostafavi, 2008; Akuamoa *et al.*, 2018).

5.6 Effect of gamma irradiation on shelf life of salted and sun dried fish in Ghana

The effects of storage on proximate composition of salted and sun dried *Oreochromis niloticus*, sun dried *Engraulis encrasicolus* and *Selena dorsalis* with respect to shelf life were appraised.

The significance of the high percentage moisture content at the beginning of storage (day zero) denotes the inefficiency of the drying process across the markets. This might be due to the initial high moisture content of the dried fish on the markets as found out by other researchers (Adenike, 2014; Farid et al., 2014). The stable moisture content of the dried fish samples during the 60 days of storage could be due to the packaging and in that way conserving the integrity of the fish and protecting the quality and consequently the health of the consumer (Coles, McDowell, & Kirwan, 2012).

Packaging and storage in addition to the stable moisture content therefore could maintain an appreciable weight that could appeal to customers even after 60 days of preservation and storage of sundried fish. Due to the minimal moisture loss during the 60 days of storage, the packaging may have provided protection from external influences such as chemical, biological and physical agents. This therefore may ensure efficiency of the dried fish supply chain distribution and offers opportunities for cost reduction and benefit communication, package protection and performance evaluation through the retail processes (FAO, 2010; Prakash, 2014). Even though there was reduction in fungi and total viable counts in all the three dried fish samples when stored for 60 days, the reduction was still above the permissible level of $<10^2$ logcfu by Ghana Standards Authority for some microbial species.

Generally, *Salmonella* and *E. coli* O157 should not be present in any food product. The presence of *E. coli* characterizes faecal contamination and should also not be present but very low levels may be acceptable, e.g., <10 cfu/g. *Staphylococcus aureus* though detected (<20cfu/g) will not produce enterotoxins until the population exceeds 10^4cfu/g. However, the reduction in microbial contaminants in the fish samples could be attributed to the effective packaging to avoid recontamination or reinfestation of the fish products coupled with the irradiation process. Gamma irradiation of *Engraulis encrasicolus* may have created an environment that was not conducive for the microbes to multiply and grow. The use of gamma irradiation as HACCP plan may eliminate the regular microbial testing for verification and phytosanitary certification for the export market to enhance the acceptability of Ghana's dried fish export. *Bacillus cereus* can form spores which are able to resist heat

and survive the cooking temperature and can either grow in the presence or absence of oxygen but cannot survive irradiation. The optimal growth temperature for *B. cereus* is between 30 °C and 37 °C which is the storage temperature in this study. This favours the growth of *B. cereus* however the effect of the irradiation may have eliminated them during storage.

At temperatures below 10°C, *B. cereus* is unable to grow and produce toxin that causes vomiting. Therefore, gamma irradiation and controlled storage temperature of food may be important to prevent foodborne disease caused by this bacterium (Deeley, 2002).

The reduction in moisture during storage and the artificial barrier created by packaging reduced the multiplication of microbes to the minimum level as was also observed by other researchers (Reza et al., 2015). *Escherichia coli* is a commonly used faecal indicator organism. Substantial number of *Escherichia coli* in food suggests a general lack of cleanliness in handling or processing, packaging and improper storage. However, the absence of *Escherichia coli* in this samples reflected the hygienic quality of the irradiated fishes. Where *Escherichia coli* occurred it should not exceed 20cfu/g.

Micrococcus and *Pseudomonas* are freshwater genera and were dominant in samples of freshwater *O. niloticus*. The identification of these in the unirradiated salted and dried *O. niloticus* showed that the salting and drying processes could not totally eliminate them from the fish (Reza et al., 2015). In the unirradiated fish samples fungi count was higher in salted-dried *O. niloticus* than in dried *E. encrasicolus* or dried *S. dorsalis* because some fungi can tolerate high salt concentrations. The presence of *Aspergillus* and

Pennicillium genera in these dried fishes is an indication of mycotoxins production, which can lead to food poisoning in dried fish.

The constancy of the protein value during storage showed that sundried fish is a good source of protein and in no doubt the loss of moisture led to increase in dry matter and this could be because of storage stability (Ayinsa & Maalekuu, 2013).

The pH and TTA also remained relatively constant during the storage period making it prohibitive to the rapid multiplication of these spoilage microorganisms. The pH of a medium determines the survival of microbes in the medium hence the pH of the fishes under study was in the acidic phase of pH = 6.84 from the first day until pH = 6.93 at day 45 and then it entered the neutral phase pH = 7.74 by day 60 of storage (Table 20).

It was noted that proximate composition of dried fish varied with the species, body size and the environment of their storage. This was also observed by Reza and his colleagues when they studied *Laubuka dadiburjori* at room temperature (Reza et al., 2015). From the discussion it may be evident that dried fish samples studied in the current work could be packaged, irradiated and stored at ambient temperature for about 60 days without loss of quality.

5.7 Effect of gamma irradiation on shelf life of smoked fish in Ghana

The decrease in percentage weight loss suggested that smoked *Chrysichthys nigrodigitatus*, *Sardinella aurita*, *Heterobranchus longifilis*, *Oreochromis niloticus* and *Engraulis encrasicolus* could be packaged, irradiated and stored for about 60 days at ambient temperature of 28 ºC without significant weight loss. This could likely be the result of less moisture

loss which could most likely be due to the sealed polyethylene bags (0.04mm thick) combined with Kraft paper packaging and the effect of irradiation before storage Prakash, 2014). This might be due also to effective smoking (Plahar et al., 1991), that might have lowered the moisture content of the fish and to the change in the temperature of the environment to normal room temperature. The low moisture content probably ensured extended shelf life which was also reported by Plahar et al., (1991). The low moisture content prior to storage could be the factor of storability and shelf life stability.

The increase in percentage moisture loss in *Sardinella aurita* could be due to initial high moisture content due to insufficient smoking processes or freshly smoked sample at the time of sampling. This resulted in high mouldiness indicated in total fungi counts of 5.48 logcfu/g in the unirradiated samples (ICGFI, 1999b). This however, decreased with increased in storage days and the effect of irradiation. Other researches indicated that the shelf life of unirradiated smoked *Sardinella aurita* varied between 4 days and 40 days according to the moisture content when stored in sealed polyethylene bags with desiccant (Plahar et al., 1991). Plahar et al., (1991) indicated that storage of smoked fish in polyethylene bags at ambient temperature was ineffective and that inclusion of desiccant only delayed total decomposition of the fish just beyond about 30 days. However, the application of gamma irradiation could be the difference in the current results.

Plahar et al., (1991) have reported the "Effect of Storage Conditions on the Quality of Smoke-dried Herring". In this report, the highest microbial counts were observed in samples stored in polyethylene bags. However, in the current study there was initial high microbial count nevertheless the effect of

gamma irradiation and a combined packaging of Polyethylene-Kraft paper resulted with the microbes likely deactivated and were unable to multiply and grow. The package integrity resulted in the reduction of the number and or activity of all organisms (including their spores) of food spoilage and public health significance achieving shelf-stability without refrigeration (IAEA, 2003). As long as the package integrity was not affected (broken or pieced), the smoked or sundried fish were subsequently stored independent of the conditions of storage as found in this study.

The decrease in ash content of *C. nigrodigitatus* suggested it had less mineral content. Ash is the inorganic residue that remains after the organic matter has been burnt off and depicts the nutrient content of the fish. This was found insignificant ($p \geq 0.05$) in the fish sampled (Francisca, 2013).

In the current study, index of perishability parameters such as total titratable acidity, pH, moisture content, and storage temperature likely exerted intensive selective pressures on the original fish bio-burden as was also observed by Ayinsa & Maalekuu, (2013). Thermophiles grow best at high temperatures, in the range of 45ºC to 70ºC but due to the high smoking temperature of 50ºC – 70ºC and regular smoking coupled with gamma irradiation effect, the mesophiles and psychrophiles could not likely survive to the detriment of the stored fish samples. The heat treatment (smoking) likely resulted in destruction of some microorganisms and also likely inactivated the enzymes in food which may cause spoilage microorganisms to proliferate (Sala et al., 1995).

The physical and organoleptic qualities of many traditionally processed sun-dried products are of low value and unhealthy for human consumption as

was reported by (Getu, Misganaw and Bazezew, 2015; FACP, 2016). They asserted insect infestation is the main problems in sun dried fish leading to both losses in quantity and quality as realised in this study. Salted and sun-dried, and sun dried fishes in Ghana are inclined to many types of spoilage including, blackening and discoloration of these fish products which affect the quality and storability of such fish products. There were damages due to insects that laid their eggs in the belly of the fish during early stages of drying. This could be the result of drying on the ground and home compounds since some of these insects lay their eggs in the soil as well, fish dried on the ground also easily gets infected with fly larvae that stay in the soil (Obodai et al., 2011). The insect infestations are of great economic significance in unprotected sun-drying, excessive re-smoking and this is a serious problem identified in this study.

5.8. Fishmongers' perception of food safety regarding preservation and storage based on focus group dialogue

The findings discussed in this study were based on the collective experiences of the participants and highlighted the complex and dynamic nature of post-harvest processing of fisheries resources specific to salted, smoked and sun dried fish. In the development of services and systems that are responsive to the views and values of these diverse groups across the value chain landscape, beliefs and social practices were highlighted. The fishmongers believed that achieving important progress in addressing issue of food safety within the value chain, multiparty efforts of all stakeholders will be required particularly the governance structures and based on this, interventions may be selected to increase participation and bring about

competitiveness and quality of the value chain sector. Currently, the knowledge of gamma irradiation of food among Ghanians has not been fully known due to lack of education (Gasu et al., 2015a).

The major theme of the knowledge of fish spoilage discussed summarized a range of positive as well as negative food safety experiences by the women. As conjuntured by Adenike who studied the different tradional methods of fish processing indicated that the fish processed by traditional methods were safe for consumption throughout the period of storage (Adenike, 2014). Many of the negative experiences discussed exist within the context of parallel themes in the broader practices and processes know-how. Nevertheless, the experiences discussed in this study represented an important call to action by stakeholders in the fisheries post harvest industry to institute educational programmes for the women on how to improve on the traditional methods. Needed are exceptional and innovative interventions intended to improve attention to address the worries in the fisheries value addition industries in the context of food safety framework by providing the needed infrastructure for storage of smoked and sundried fishery products.

In spite of the concerns of the women engaged in fisheries post-harvest processing, the majority also know what works for them while others are in the fish business for subsistence living. This was well illustrated by the range of recommendations for improving what they already know about fish spoilage, preservation and storage, and they gave different experiences and characteristics of spoiled fish. The women depend on the odour, colour and firmness or floppiness of the fish as the first criterion for estimating the freshness or spoilage of the fresh fish before processing. This same experience

and criterion of the use of odour was expressed by women participants in Ethiopia (Chanie et al., 2005). This revealed the basic knowledge of food safety the women have in practicing their trade. The women also attributed fish spoilage to late arrival of the fishermen from sea and the rivers and also to inadequate transportation network as also observed by other authors (Alemu, 2016). This resulted in the fish going bad indicative by the odour, colour and firmness before they start the drying and smoking process. It became evident that when the fish spoiled they are not thrown away to avoid debts but rather they are processed by salting and drying to produce dried fish product or fermented or cured as "mormorni". This is evident by Aheto et al. (2017) that 3% of landed fish in Ghana is processed by salting while the remainder is sun dried or fermented.

The sources of their knowledge gave the dynamics of handed down tradition from family members and from the community of the fish trade. In this, the diverse knowledge expressed by the women showed the need to improve upon the current process and disseminate a new and improved technology to enhance food safety.

Most of the women reiterated their setbacks encountered in practicing their trade to lack of modern technology to preserve the fish from spoilage, no training, no financial support to buy storage equipment for improving on their trade (Alemu, 2016). Most of the women across the markets expressed their worries of being excluded in decision making concerning their trade.

The level of formal education being low among the fishmongers they became less visible to be consulted about the management of their work. They

expressed their inability to know from the onset what policies affect them except that the decision is pushed down their throat.

In terms of profitability of the sector, there were many challenges identified such as the inability to pay off bank loans and credits from money lenders as a result of losses after sales. Most of the women could not access loan facilities and therefore could not expand their trades which they operate on small scales. They however preferred the dry fish trade to the smoked fish since the dried fish trade does not require buying of fuel wood and re-smoking. They expressed their worries about fish price fluctuations on the markets since they buy the fish at a higher price from the fishermen and process them incurring more cost against various competing prices (Ayinsa et al., 2013).

They expressed worries about packaging materials and the indiscriminate use of pesticides to protect the stored fish from insects' infestation and rodents attack in storage. The lack of knowledge on the harmful effect of the use of all kinds of pesticides calls for education and training.

Across the markets the women desired knowledge on knew technologies to assist in the fish processing and favourable marking avenues in terms of smoking, drying, packaging and storage. They however have no knowledge of food irradiation (Gasu et al., 2015b), but expressed desire to know and use the technology with little cost (Sapp & Downing-matibag, 2009; Eustice & Bruhn, 2012).

During the participant observation (PO), it became clear that spoiled fish are used for fermented products. There was no stringent adherence to

hygiene at the processing areas, storage rooms and even transportation to markets. There were no water proof floor and fish were poured on the bare ground both at the shore and the smoking houses in most of the markets surveyed (Wang et al., 2016).

Portable water systems and water storage facilities were very poor and fish were processed on bare ground overlaid with polyethylene sheets as a result, the raw materials were not separated far from the cleaning materials. Insects and pest infestation was predominant in some of the markets and smokehouses.

Quality checks on raw materials was not stringently adhered to for safe and quality fish selection that could be dried or smoked. However, few identifiable characteristics of spoiled fish include dull eyes, redden belly, blacken gills, and flaccid body were observed.

Equipment cleaning and regular cleaning programmes of the premises were absent in most of the markets selected in this study. Clean and dirty water could not be identified because once the women began to wash the fish, the same dirty water was used for several washings.

There were no waiting rooms for most processing homes and as a result there were no code of conduct for visitors. There was no dress code for attendants therefore they dressed in jewelleries, bungles and chains, and without head nets during fish washing and packing for sales. Most participants observed did not wear hand gloves and also did not have medical certification to handle food. The use of cold storage was not a norm with these fish handlers therefore the use of plastic boxes was also scarcely observed.

Another setback was the absence of storage facility and lack of new methods of preservation. The only method they knew was salting, smoking or sun drying of the fish and these activities are largely unregulated by scientific protocols and complete absence of food safety regulations in the markets. The absence of storage facilities for sun dried and smoked make the women to engaged in re-smoking and re-drying of the fish regularly to keep the fish from growing mouldy. Various oven types and wood fuels are employed to generate the smoke needed for the continuous re-smoking processes. The re-smoking could be the reason of increase in the PAH concentration of the fish and makes the fish have a burnt appearance. This process adds up to the poor quality of sun dried and smoked fish on the Ghanaian market.

CHAPTER SIX

6.0. SUMMARY OF FINDINGS, CONCLUSIONS AND RECOMMENDATIONS

6.1 Summary of Findings

The traditional methods of fish processing and preservation in Ghana comprise of ancient technology such as smoking, drying, frying, salting and curing/fermenting. One of these or a combination of these methods are used to process and preserve fish. The Women who predominate the post-harvest industry, however, have no effective means of processing, preserving and storing fish. According to Aheto et al., (2017), about 95% of fish processed in Ghana is by smoking. Further 3% of processed fish is by salting while the remainder is sun dried, fried or cured/fermented *mormorni*. These methods especially sun drying as observed by Obodai et al., (2011), predispose the processed fish to microbiological contamination. In addition, chemical (PAHs and OCPs), and physical (heavy metals and plastics) hazards that are of public health concern cannot be overlooked if food safety goals should be attained.

The activities of fish processors are dependent on knowledge handed down and largely unregulated by scientific protocols and there are no food safety regulations in the market places that could withstand the stringent regulations of food safety authorities to meet export standards.

Various types of oven and wood fuels are employed to generate the smoke needed for fish processing and sun drying depends on sunny weather. These methods of preservation result in re-smoking and continuous sun drying hence the quality and safety of salted, smoked and sun dried fish vary from market to market and region to region along the coast of Ghana.

The current research has revealed high concentrations of PAHs, OCPs and microbial contaminants in salted, smoked and sun dried fish on the Ghanaian markets.

There is scarcity of research data in the Ghanaian context, which has used packaging and gamma irradiation technology to decontaminate smoked and sun dried fish of PAHs, OCPs and biological pathogens.

In this study, the contaminants associated with fish smoking and sun drying (PAHs, OCPs and microbial bio-burden) were found to be above the recommended maximum residue levels (MRLs) and the daily allowable intake of PAHs and OCPs. These limiting values were established in the European Commission Regulation (EU) 2015/1125), WHO, Australia and New Zealand daily allowable intake per body weight.

Gamma irradiation has been found to significantly reduce the concentrations of PAHs in smoked and sun dried fish. Gamma irradiation have also been found to reduce the concentrations of OCPs contaminations in sun dried fish. Most OCPs were completely degraded while others were reduced to acceptable limits. At an absorbed dose of 7.5kGy, Deltamethrin, Delta HCH, Aldrin, Pirimiphos-m, Heptachlor, Fenpropathrin, Permathrin, Fenvalerate, Methoxchlor, Endosulfans, Lambdacythal and Deltamethoxychlor were completely degraded and removed from the fish. However, Cyfluthrin and Gamma HCH were reduced within the daily allowable intake or maximum residue levels (MRLs).

Gamma irradiation within the limits of the doses used in this research had no significant effect on the protein content, moisture loss and the pH of sun dried and smoked fish. The gamma irradiation however had little effect on

weight loss, the ash content and the TTA of the sun dried and smoked fish. The bacteria count (TVC) and fungi count (TMC) were significantly reduced by the irradiation process.

Storage had great effect on the free fatty acid and protein contents, weight loss, pH, TVC and TMC but not on PAH and OCP of the sun dreid and smoked fish that were studied in this research.

In the subjective measurements which included focus group discussion and participant observation, most of the participants were women and engaged in the smoking and sun drying activities of fish but this varied across socio-demographic factors. Focus group discussion revealed the problems limiting the fish value addition industry and impacting on the livelihood of coastal dwellers especially, women who seemed to dominate the enterprise.

The oldest woman fishmonger was between 70 – 75 years old and the youngest was between 33 - 35 years in the Regions. This suggest that value-addition and distribution chain in fisheries form a major livelihood to many women for years.

There were improper storage facilities and unhygienic preservation of processed fish. Smoked and sun dried fish were packaged in wooden baskets and covered with old discarded clothes or jute sacks and polythene sheets. There was smoking and re-smoking, and continuous sun drying to maintain good appearance and to avoid the growth of moulds. Insect infestation was very high in smoked and sun dried fish trade and this resulted in high financial losses as was expressed through focus group discussion.

Most participants have no knowledge of packaging and food irradiation. They used insecticides to keep flies away from the stored fish which were stacked on cement blocks or wooden blocks with lit mosquito coils under them or insecticides sprayed in the store rooms and around market stalls where these fishes were stored and displayed for sales. This could be health hazard to consummers of these fish products.

6.2. Conclusion

This thesis has investigated polycyclic aromatic hydrocarbons, orgarnochlorine pesticides and microbial load of salted, smoked and sundried fishes from coastal markets in Southern Ghana.

High concentrations of polycyclic aromatic hydrocarbon were found in smoked and sun dried fish from the Ghanaian markets. This high levels of PAHs may be dependent on accumulation through the environment or the procession methods. PAHs were not only identified in smoked fish but also in sundried fish in this study as result of the practice of drying fish on the ground by the roadside and at the beaches.

Drying of fishes is predisposed to several types of damage which can affect the quality and shelf life and this is a serious problem in traditional drying. Physical and organoleptic qualities of many traditional sundried products are unacceptable for human consumption. Damages to smoked and sun dried fish was found to be due to lack of adequate preservation and packaging, high moisture content and insect infestation during storage.

This research has investigated pesticides residues in salted, smoked and sundried fishes from coastal markets in Southern Ghana. High concentrations of pesticides were found in smoked and sun dried fish from the

Ghanaian markets. This high levels may be dependent on accumulation through the environment or the processing and preservation methods for storage. The results therefore revealed food safety concerns and unregulated practices currently in the Ghanaian fishery value chain. Developing protocols to regulate and enhance quality and safety of smoked and sun dried fish will enhance acceptance of smoked and sun dried fish from Ghana by countries who have pesticides residue regulatory standards to enhance food safety and protect the citizens.

The thesis documents the potential of gamma irradiation as a means to improve the safety standards of processed fish and extended shelf life due to the irradiation's effectiveness in decontaminating PAHs and inactivating pathogenic and spoilage microorganisms.

The adoption of gamma irradiation as a complementary technology to traditional preservation technology to decontaminate PAHs, OCPs and biological contaminants in smoked and sun-dried fish would also produce synergistic effect of prolonging the shelf life of smoked and sun dried fish for storage at ambient temperature. It has proved to be more effective and costs less at the long-run compared to the traditional technologies using the smoking oven alone in terms of shelf life and quality. Gamma irradiation has also shown in this work as an effective processing method for improving the nutritional value of smoked and sun dried fish. However, consumer knowledge, acceptability and government policies toward the acceptance of its commercialization remains a challenge.

6.3. Implications and Prospects of Irradiation Technology in the Fishery Industry of Ghana

This research is a fundamental to: promote Integrated Coastal Zone Management, reduce poverty in fishing communities by adding value to fish post-harvest efforts in Ghana, support supplementary livelihoods within fisheries value chain and reduce conflicts in fisheries as result of improved fisheries economy, enhance value-chain development and reduce postharvest losses in fisheries.

Gamma irradiation could maintain food quality and address food safety and food security problems without significantly affecting the food's sensory or nutritional attributes. In this way providing nutrition to the people hence ending hunger, achieving food security and improving nutrition and sustainable fisheries for Ghana.

The extension of shelf life of processed fish and fish products may result in improved income of the fish traders and the fishermen as well. This may lead to preservation and storage of bumper harvest against closed and off seasons providing continuous business and income to the stakeholders. This will therefore reduce poverty among the stakeholders therefore ending poverty in all forms everywhere for all.

The use of irradiation will reduce the negative impact of smoke on the women who smoke fish. This will ensure healthy lives and promote wellbeing for all at all ages and to sustain the environment and answering climate change challenges. It is a safe technology and may be used on sundried and smoked fish in Ghana to avoid excessive use of wood fuel which depletes the

mangroves and coastal vegetations when in search for wood fuel for smoking fish.

Currently, irradiation treatment is used on a wide variety of food and non-food products and have a far-reaching implication for Ghana's fishery industry to reduced risk of food-borne diseases caused by micro-organisms such as *Campylobacter, Salmonella, E. coli* and *Listeria* especially in meat, poultry and fish. This may be beneficial to the fisheries sector of Ghana and to aid coastal management policies such as clean coastland and beaches.

The use of gamma irradiation will result in less need for pesticides usage, less need for some food additives, such as preservatives and antioxidants, lower risk of importing or exporting insect pests hidden inside dried fish products from other countries, and to reduce the need for toxic chemical treatments, such as those used to kill bacteria found in some spices.

For the export of sundried and smoked fish, food irradiation technology may serve as phytosanitary certification to avoid emigrating pests and insects to the importing countries. This may position Ghana with comparative advantage over other countries in terms of salted, smoked and sundried fish export. It will promote international trade and improve the economy and further enhance the livelihood of fisher folks. Salted, smoked and sundried fish may then be processed with gamma irradiation and stored for a longer time for future use and for export.

This technology may give the closed and off seasons a better meaning when the fishermen know that they can preserve and store their stock for a longer period and still make profit while preserving the life of the ocean and allowing the fisheries stock to replenished. The food irradiation technology

has a far more fetching implication to enhance the Integrated Coastal Zone Management Programme than the initial investment in establishing an irradiation source.

Policy makers and coastal and fisheries managers would find solace in food irradiation to address poverty alleviation, increase fish protein availability, nutritious and affordable food and healthy life for all. The current overall Government policy framework for the fisheries sector of Ghana is devoid of some scientific information on the use of modern technology for preservation of sun dried and smoked fish which when considered may enhance buffer stocking for the close and off season. Policies on the use of food irradiation to increase food safety and food security will create new jobs in the fisheries and value chain distribution sector. The surest way is to adopt food irradiation as a complementary option.

The prospects are enormous because there is already a Gamma Irradiator situated at GAEC manned by a well trained scientists and technologies by the International Atomic Energy Agency and International and National Universities. There is also the Radiation Protection Authority and the Biosafety Authority to regulate and licence the activities of people ready to use the irradiator for commercial purposes.

6.4: Recommendations

This thesis has contributed to the growing body of evidence that smoked and sun-dried fish have the potential to be preserved for a longer time by gamma irradiation and the following recommendations made:

(i) Government needs to formulate policies to include the use of food irradiation into the agricultural sector.

(ii) Mobile irradiators may be installed at fishing harbours and smokehouses for the purpose of irradiating smoked and sundried fishes to reduce post harvest losses.

(iii) Public education on the benefits of food irradiation must be carried out to inform the population that gamma irradiated foods cause no side effects. This can eventually be translated into the population's knowledge of the safety of irradiated food and provide the policy to enhance the process of food irradiation and decontamination.

6.5 Further studies may be carried out into:

(i) An assessment of the composition and properties of radiation metabolites in salted sundried, smoked and dried fishes in Ghanaian markets.

(ii) Investigating ways to improve the processes of decontamination by adopting packaging to prevent the probable deterioration of smoked and sun-dried fish to avoid cross contamination.

(iii) Developing protocols to regulate and enhance quality and safety of smoked and sun dried fish to curb unregulated practices currently in the Ghanaian fishery value chain.

(iv) Packaging technology for fish preservation and storage should be employed to enhance consumer protection and safety.

(v) Freshly sun dried and freshly smoked fish to be analysed for determining the effective moisture content for storage at ambient temperature.

(vi) Effect of an increased dose up to 10kGy be evaluated for complete decontamination of polycyclic aromatic hydrocarbons and orgarnochlorine pesticides in processed fish.

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8.0 APPENDICES

APPENDIX A

Common nuclides in food and in human body:

Natural Radioactivity in Food [5]		
Food	$^{40}_{19}$K Bq/kg	$^{226}_{88}$Ra Bq/kg
Banana	130	.04
Brazil Nuts	210	37-260
Carrot	126	0.02-.07
White Potatoes	126	.04-0.1
Beer	14	--
Red Meat	111	0.02
Lima Bean	172	0.07-0.2
Drinking Water	---	0-0.006

Natural Radioactivity in a 70 kg human			
Nuclide	Total Mass	Total Activity	Daily Intake
$^{238}_{92}$U	90 µg	30 pCi (1.1 Bq)	1.9 µg
$^{232}_{90}$Th	30 µg	3 pCi (0.11 Bq)	3.0 µg
$^{40}_{19}$K	17 mg	120 nCi (4.4 kBq)	0.39 mg
$^{226}_{88}$Ra	31 pg	30 pCi (1.1 Bq)	2.3 pg
$^{14}_{6}$C	22 ng	0.1 µCi (3.7 kBq)	1.8 ng
$^{3}_{1}$H	0.06 pg	0.6 nCi (23 Bq)	0.003 pg
$^{210}_{84}$Po	0.2 pg	1 nCi (37 Bq)	~0.6 fg

APPENDIX B

Focus Group Discussion for Analysis of Irradiation Technology of Smoked and Sun Dried Fish in Ghana

Evaluation of perceptions, practices and local traditional knowledge of coastal communities about fish safety and fish irradiation technology

Defining the group

Eight participants in each focus group will be selected according to Eliot & Associates (2005) from each of the six markets in the study sites.

Inclusion criteria

Fish traders who are more than five years old in the fish trade will be sampled for the discussion

Exclusion criteria

Fish traders who are less than five years in the trade, hawkers and floating traders will be excluded in recruiting participant so that the group will be homogeneous.

Recruiting Participants

Market queens in each market will be contacted personally to discuss the purpose of this research to seek her interest and after which she will randomly nominate eight members from their existing association. This will constitute members of the same group and of the same job titles. All recruits will be contacted verbally to confirm their interest and availability.

Conducting the focus group

The team will be made up of a Moderator (Student investigator) and two Assistant Moderators. The proceedings will be recorded using an audio recorder. The Assistant Moderators will be taking notes on the discussions.

Venue

A convenient room or an available area will be sort for this exercise in each market.

Incentives

Three cakes of toilet soap and transportation fare of GHS 20.00 will be presented to each participant as "thank you" incentives.

1. Knowledge on fish safety

1.1 Can you describe how fish looks when it is safe and unsafe for consumption?

(Probe: differences between safe and unsafe fish by looking at the fish)

1.2 What is the source of your knowledge?

(Probe: how you came by the source i.e. by effort, accidentally or by experience)

1.3 How can you tell if fish is going bad and not safe for consumption?

(Probe: appearance, odour, softness, maggots etc)

1.4 What causes fish spoilage and post-harvest/post-capture losses?

(Probe: among the most relevant causes to my research)

2. Cost benefit Analysis of Current Technology

2.1 What is the benefit of the fish trade currently?

(Probe: what is/was the profit margin?)

2.2 How much in monetary value do you lose when your fish spoils?

(Probe: the worthiness of the fish trade)

2.3 Is the loss due to processing/preservation/storage? (Probe: do you engage in the above all by yourself)?

Based on information from 10 record books (from market queens) per market determine

(i) Seasonal income, (ii) annual income, (iii) seasonal expenditure, (iv) annual expenditure,

(v) Profitability

Months	Community concerned: _____									
	1	2	3	4	5	6	7	8	9	10
Jan										
Feb										
Mar										
Apr										
May										
Jun										
Jul										
Aug										
Sep										
Oct										
Nov										
Dec										
TOTAL INCOME										
TOTAL EXPENDITURE										
PROFIT										

3. Levels of concern of losses

3.1 Are you concerned about the losses? If yes, why?

(Probe: both positive and negative responses)

3.2 Are the losses a problem in your community among the fish traders?

(Probe: what are the reasons for the losses?)

3.3 Do you think the losses are due to the method of processing?

(Probe: method of processing)

3.4 Do you give feedback to your source of acquisition of dried and smoked fish?

(Probe: what responses do you receive from your sources?)

4. Forms of preservation

4.1 What forms of preservation are available to you?

(Probe: knowledge of preservation technology / merits and demerits of technology)

4.2 What do you think can be done to address the problem of preservation?

(Probe: education etc)

4.3 Do you know about fish irradiation preservation?

(Probe: same as X-ray and Scan, then background radiation, sun rays etc)

4.5 What is the source of your knowledge?

(Probe: where did you hear this from?)

4.6 Would you be willing to use it if you are educated on it?

(Probe: it is a clean and a safe method of destroying spoilage microbes that cause diseases)

4.7 Irradiation can extend the shelf life of your stored fish; will you like to use this technology?

(Probe: marginal increase in cost)

4.8 Will you be willing to pay extra for irradiated fish to enable you increase your profit margin? If yes, how much would you be willing to pay by way of margin?

(Probe: how much)

5. Knowledge of packaging and storage

5.1 Do you have any form of packaging prior to storage?

(Probe: kinds of packaging available to you)

5.2 Do you have any form of storage for your fish until the fish is sold?

(Probe: storage area, well ventilated, on the ground, on raised stack)

5.3 What problems do you encounter when you store your fish?

(Probe: insects, maggots, moulds, change in colour, mice, etc)

5.4 Will you wish to re-package your fish before sales if you have extra knowledge? (Probe: suggest packaging materials)

APPENDIX C

Participant observation

This is to design checklist for fish safety auditing. Variables will be scored for as

"Very good = 1, Good =2, Bad = 3"

Area	Requirements	Comp-liance	Non-comp-liance	Observation
1.Environmental hygiene	Is the location of production clean and free from obvious contamination?			**1** ☐ **2** ☐ **3** ☐
	Is dump isolated from site of operation and protected			**1** ☐ **2** ☐ **3** ☐
	Is the premises protected with fence to keep away animals?			**1** ☐ **2** ☐ **3** ☐
2. Layout of processing area	Is there a reception area separating visitors from operatives in the fish processing fscility?			**1** ☐ **2** ☐ **3** ☐

	Is the floor waterproof to withstand constant washing?			**1** ☐ **2** ☐ **3** ☐
	Are drainages assessable for cleaning regularly?			**1** ☐ **2** ☐ **3** ☐
3 Personal hygiene	Do staff have medical permit to work in fish processing facility?			**1** ☐ **2** ☐ **3** ☐
	Do operatives wear jewelries, bungles and wrest watches?			**1** ☐ **2** ☐ **3** ☐
	Observing for chewing of gum, eating and smoking of cigarettes in the processing area.			**1** ☐ **2** ☐ **3** ☐
	Observing for long/short fingernails and false nails			**1** ☐ **2** ☐ **3** ☐
	Observing for the wearing of hand gloves and hair covers			**1** ☐ **2** ☐ **3** ☐
	Do visitors adhere to the laid down hygiene requirements?			**1** ☐ **2** ☐ **3** ☐

4.Control of fish processing operations	Is clean water and dirty water separated from each other?			**1** ☐ **2** ☐ **3** ☐
	Are smoked, dried and fresh fish kept close to each other?			**1** ☐ **2** ☐ **3** ☐
	Are quality checks performed on in-coming raw fish to ensure quality adherence?			**1** ☐ **2** ☐ **3** ☐
5.Handling procedure	How is sorting of fish done (in ice salt water, bowl of water, running water?)			**1** ☐ **2** ☐ **3** ☐
	Is there a possibility of contamination and cross-contamination?			**1** ☐ **2** ☐ **3** ☐
	Can all equipment and containers be adequately cleaned, disinfected, and to avoid cross-contamination?			**1** ☐ **2** ☐ **3** ☐
6.Water quality	Is the source of water portable or acceptable? What is the source of water in the			**1** ☐ **2** ☐ **3** ☐

	facility?			
	Are there water storage tanks			**1** ☐ **2** ☐ **3** ☐
	Are the storage tanks easy to clean?			**1** ☐ **2** ☐ **3** ☐
7.Storage	Is the storage area cool and clean?			**1** ☐ **2** ☐ **3** ☐
	If ice is used, what is the source of the ice?			**1** ☐ **2** ☐ **3** ☐
	Are clean plastics boxes used to store the ice			**1** ☐ **2** ☐ **3** ☐
	Is the storage area well ventilated?			**1** ☐ **2** ☐ **3** ☐
	Is the storage area protected from pests			**1** ☐ **2** ☐ **3** ☐
	Are cleaning agents, and chemicals, brooms, brushes and dusters stored in the			**1** ☐ **2** ☐ **3** ☐

	same area as raw, smoked or dried fish?			
8.Pest control	Is there any sign of pest activity			**1** ☐ **2** ☐ **3** ☐
	Is the facility kept in a hygienic condition to avoid pest harbourage?			**1** ☐ **2** ☐ **3** ☐
	Are there methods of pest elimination?			**1** ☐ **2** ☐ **3** ☐
9.Waste management	Is there a waste disposal system for both liquid and solid wastes in the facility?			**1** ☐ **2** ☐ **3** ☐
	Is the waste removed regularly and in a timely manner?			**1** ☐ **2** ☐ **3** ☐
10.Cleaning programme	Are all equipment and utensils clean regularly?			**1** ☐ **2** ☐ **3** ☐

APPENDIX D

(i) Statistical analysis of PAHs in Smoked Fish

General Linear Model: Chrysene versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	986.5	123.32	2.93	0.006***
Error	86	3616.0	42.05		
Total	94	4602.5			

General Linear Model: Chrysene versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	529.5	176.50	3.94	0.011***
Error	91	4073.0	44.76		
Total	94	4602.5			

General Linear Model: B(a)A versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	2075	259.37	14.96	0.000***
Error	76	1318	17.34		
Total	84	3393			

General Linear Model: B (a)A versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	244.9	81.62	2.10	0.107
Error	81	3147.6	38.86		
Total	84	3392.5			

General Linear Model: B (k)F versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	13301	1662.60	29.87	0.000***
Error	84	4676	55.66		
Total	92	17977			

General Linear Model: B (k)F versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	386.0	128.7	0.65	0.584
Error	89	17590.5	197.6		
Total	92	17976.5			

General Linear Model: B (a)P versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	3392	423.94	11.08	0.000***
Error	87	3328	38.25		
Total	95	6719			

General Linear Model: B (a)P versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	529.7	176.56	2.62	0.055*
Error	92	6189.4	67.28		
Total	95	6719.1			

General Linear Model: B(b)F versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	26097	3262.1	8.53	0.000***
Error	83	31747	382.5		
Total	91	57844			

General Linear Model: B(b)F versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	4683	1560.9	2.58	0.058*
Error	88	53161	604.1		
Total	91	57844			

General Linear Model: B(g,h,i)P versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	12480	1559.9	5.92	0.000***
Error	85	22387	263.4		
Total	93	34867			

General Linear Model: B(g,h,i)P versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	726.4	242.1	0.64	0.592
Error	90	34140.5	379.3		
Total	93	34866.8			

General Linear Model: In (1,2,3,c,d)P versus Samples

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Samples	8	11708	1463.5	6.19	0.000***
Error	83	19619	236.4		
Total	91	31326			

General Linear Model: In(1,2,3,c,d)P versus Dose/kGy

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Dose/kGy	3	710.5	236.8	0.68	0.566
Error	88	30616.0	347.9		
Total	91	31326.5			

(i) Market dynamics

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Regions	2	694.37	347.183	118.79	0.000
Error	6	17.54	2.923		
Total	8	711.90			

Statistical Analysis of PAHs in sun dried fish

General Linear Model: B(a)P versus Location

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	6	603.311	100.00%	603.311	100.552	12076.45	0.000***
Error	2	0.017	0.00%	0.017	0.008		
Total	8	603.328	100.00%				

General Linear Model: B(g,h,i)P versus Location

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	6	772.229	99.42%	772.229	128.705	57.20	0.017**
Error	2	4.500	0.58%	4.500	2.250		
Total	8	776.729	100.00%				

General Linear Model: In(1,2,3,c,d)P versus Location

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	6	772.229	99.42%	772.229	128.705	57.20	0.017**
Error	2	4.500	0.58%	4.500	2.250		
Total	8	776.729	100.00%				

General Linear Model: B(a)A versus Dried samples

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Dried samples	3	577.5	84.30%	577.5	192.50	8.95	0.019**
Error	5	107.6	15.70%	107.6	21.52		
Total	8	685.1	100.00%				

General Linear Model: B(k)F versus Dried samples

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Dried samples	3	1881.07	100.00%	1881.07	627.023	2045209.11	0.000***
Error	5	0.00	0.00%	0.00	0.000		
Total	8	1881.07	100.00%				

General Linear Model: B(a)P versus Dried samples

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Dried samples	3	603.326	100.00%	603.326	201.109	560095.80	0.000***
Error	5	0.002	0.00%	0.002	0.000		
Total	8	603.328	100.00%				

General Linear Model: B(b)F versus Dried samples

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Dried samples	3	8167.06	99.97%	8167.06	2722.35	6645.63	0.000***
Error	5	2.05	0.03%	2.05	0.41		
Total	8	8169.11	100.00				

General Linear Model: B(g,h,i)P versus Dried samples

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Dried samples	3	771.021	99.27%	771.021	257.007	225.14	0.000***
Error	5	5.708	0.73%	5.708	1.142		
Total	8	776.729	100.00%				

General Linear Model: In(1,2,3,c,d)P versus Dried samples

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Dried samples	3	771.021	99.27%	771.021	257.007	225.14	0.000***
Error	5	5.708	0.73%	5.708	1.142		
Total	8	776.729	100.00%				

Analysis of Variance of OCPs Appendix 4.4 &4.5

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Factor	12	4690	390.8	1.32	0.354
Error	8	2365	295.6		
Total	20	7055			

(ii) Statistical analysis of the effect of gamma irradiation on pesticides in fish

General Linear Model: 0kGy versus Pesticides

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Pesticides	7	0.1887	0.026963	5.14	0.001***
Error	27	0.1416	0.005244		
Total	34	0.3303			

General Linear Model: 2.5kGy versus Pesticides

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Pesticides	7	0.011832	0.001690	5.49	0.001***
Error	27	0.008309	0.000308		
Total	34	0.020141			

General Linear Model: 5.0kGy versus Pesticides

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Pesticides	7	0.000364	0.000052	1.63	0.168
Error	27	0.000860	0.000032		
Total	34	0.001224			

General Linear Model: 7.5kGy versus Pesticides

Analysis of Variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Pesticides	7	0.000016	0.000002	1.76	0.136
Error	27	0.000035	0.000001		
Total	34	0.000050			

a. Statistical analysis of proximate of smoked fish

General Linear Model: Initial weight/gm versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	892.3	5.45%	892.3	446.1	1.81	0.175
Dose/kGy	3	1819.2	11.11%	1819.2	606.4	2.45	0.074
Storage/days	4	1311.9	8.01%	1311.9	328.0	1.33	0.273
Error	50	12354.3	75.43%	12354.3	247.1		
Total	59	16377.7	100.00%				

General Linear Model: Final weight/gm versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	889.6	5.45%	889.6	444.8	1.81	0.174
Dose/kGy	3	1834.5	11.24%	1834.5	611.5	2.49	0.071
Storage/days	4	1326.0	8.12%	1326.0	331.5	1.35	0.264
Error	50	12273.6	75.19%	12273.6	245.5		
Total	59	16323.8	100.00%				

General Linear Model: % weight loss versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.00317	0.04%	0.00317	0.00158	0.03	0.975
Dose/kGy	3	0.87284	10.66%	0.87284	0.29095	4.65	0.006***
Storage/days	4	4.18769	51.14%	4.18769	1.04692	16.75	0.000***
Error	50	3.12535	38.17%	3.12535	0.06251		
Total	59	8.18905	100.00%				

General Linear Model: Initial moisture versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	3.976	1.31%	3.976	1.988	0.48	0.624
Dose/kGy	3	27.345	9.03%	27.345	9.115	2.18	0.102
Storage/days	4	62.448	20.62%	62.448	15.612	3.73	0.010**
Error	50	209.144	69.04%	209.144	4.183		
Total	59	302.913	100.00%				

General Linear Model: Final moisture versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	4.041	1.35%	4.04	12.020	0.48	0.619
Dose/kGy	3	27.217	9.06%	27.217	9.072	2.17	0.103
Storage/days	4	60.401	20.11%	60.401	15.100	3.62	0.011**
Error	50	208.651	69.48%	208.651	4.173		
Total	59	300.310	100.00%				

General Linear Model: % Moisture loss versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.000769	0.32%	0.000769	0.000384	2.81	0.070
Dose/kGy	3	0.000902	0.38%	0.000902	0.000301	2.19	0.100
Storage/days	4	0.230454	96.43%	0.230454	0.057614	420.54	0.000***
Error	50	0.006850	2.87%	0.006850	0.000137		
Total	59	0.238975	100.00%				

General Linear Model: % Ash versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.537	0.23%	0.537	0.2686	0.18	0.837
Dose/kGy	3	38.860	16.82%	38.860	12.9532	8.62	0.000***
Storage/days	4	116.429	50.40%	116.429	29.1073	19.36	0.000***
Error	50	75.168	32.54%	75.168	1.5034		
Total	59	230.994	100.00%				

General Linear Model: TTA versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.000421	0.23%	0.000421	0.000211	0.18	0.837
Dose/kGy	3	0.030466	16.82%	0.030466	0.010155	8.62	0.000***
Storage/days	4	0.091281	50.40%	0.091281	0.022820	19.36	0.000***
Error	50	0.058932	32.54%	0.058932	0.001179		
Total	59	0.181099	100.00%				

General Linear Model: pH versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.0936	0.64%	0.0936	0.04680	0.21	0.811
Dose/kGy	3	0.2296	1.57%	0.2296	0.07654	0.34	0.794
Storage/days	4	3.1725	21.67%	3.1725	0.79314	3.56	0.012*
Error	50	11.1466	76.13%	11.1466	0.22293		
Total	59	14.6424	100.00%				

General Linear Model: %FFA versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.705	0.09%	0.705	0.3527	0.03	0.969
Dose/kGy	3	48.516	6.44%	48.516	16.1720	1.46	0.237
Storage/days	4	149.392	19.83%	149.392	37.3480	3.37	0.016*
Error	50	554.819	73.64%	554.819	11.0964		
Total	59	753.432	100.00%				

General Linear Model: TVC/logcfu versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	1.342	0.35%	0.293	0.1464	0.05	0.950
Dose/kGy	3	147.581	38.06%	106.876	35.6253	12.50	0.000***
Storage/days	4	113.480	29.26%	113.480	28.3699	9.96	0.000***
Error	44	125.381	32.33%	125.381	2.8496		
Total	53	387.782	100.00%				

General Linear Model: MYC/logcfu versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	4.205	0.89%	4.205	2.103	0.78	0.463
Dose/kGy	3	111.379	23.61%	71.105	23.702	8.83	0.000***
Storage/days	4	238.096	50.47%	238.096	59.524	22.19	0.000***
Error	44	118.055	25.03%	118.055	2.683		
Total	53	471.734	100.00%				

General Linear Model: Final % Protein versus Location, Dose/kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Location	2	0.01563	4.02%	0.01563	0.007813	1.25	0.296
Dose/kGy	3	0.01010	2.60%	0.01010	0.003367	0.54	0.659
Storage/days	4	0.04912	12.65%	0.04912	0.012281	1.96	0.115
Error	50	0.31350	80.73%	0.31350	0.006270		
Total	59	0.38835	100.00%				

General Linear Model: Initial weight/gm versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	1.5089	9.33%	1.50894	0.25149	11.90	0.000***
Dose kGy	3	6.0810	37.61%	6.08100	2.02700	95.94	0.000***
Storage/days	4	0.0000	0.00%	0.00000	0.00000	0.00	1.000
Error	406	8.5780	53.06%	8.57798	0.02113		
Lack-of-Fit	126	4.6164	28.55%	4.61640	0.03664	2.59	0.000
Pure Error	280	3.9616	24.50%	3.96159	0.01415		
Total	419	16.1679	100.00%				

General Linear Model: Final weight/gm versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	1.5273	9.38%	1.52730	0.25455	11.91	0.000
Dose kGy	3	6.0710	37.28%	6.07104	2.02368	94.68	0.000
Storage/days	4	0.0084	0.05%	0.00845	0.00211	0.10	0.983
Error	406	8.6774	53.29%	8.67740	0.02137		
Lack-of-Fit	126	4.6361	28.47%	4.63614	0.03679	2.55	0.000
Pure Error	280	4.0413	24.82%	4.04126	0.01443		
Total	419	16.2842	100.00%				

General Linear Model: % weight loss versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	0.2136	1.87%	0.2136	0.03560	3.24	0.004***
Dose kGy	3	0.2254	1.97%	0.2254	0.07513	6.84	0.000***
Storage/days	4	6.5330	57.15%	6.5330	1.63326	148.72	0.000***
Error	406	4.4586	39.01%	4.4586	0.01098		
Lack-of-Fit	126	1.5512	13.57%	1.5512	0.01231	1.19	0.125
Pure Error	280	2.9074	25.44%	2.9074	0.01038		
Total	419	11.4307	100.00%				

General Linear Model: Initial Moisture versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	8391.0	36.77%	8391.0	1398.50	50.48	0.000***
Dose kGy	3	3.0	0.01%	3.0	0.99	0.04	0.991
Storage/days	4	3177.9	13.93%	3177.9	794.47	28.68	0.000***
Error	406	11247.8	49.29%	11247.8	27.70		
Lack-of-Fit	126	10728.2	47.01%	10728.2	85.14	45.88	0.000
Pure Error	280	519.6	2.28%	519.6	1.86		
Total	419	22819.7	100.00%				

General Linear Model: Final Moisture versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	8594.9	29.57%	8594.9	1432.48	34.92	0.000***
Dose kGy	3	54.9	0.19%	54.9	18.30	0.45	0.720
Storage/days	4	3756.6	12.93%	3756.6	939.16	22.89	0.000***
Error	406	16656.3	57.31%	16656.3	41.03		
Lack-of-Fit	126	12285.8	42.27%	12285.8	97.51	6.25	0.000
Pure Error	280	4370.5	15.04%	4370.5	15.61		
Total	419	29062.7	100.00%				

General Linear Model: %Moisture loss versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	112.59	1.38%	112.59	18.76	0.96	0.449
Dose kGy	3	52.34	0.64%	52.34	17.45	0.90	0.443
Storage/days	4	97.55	1.19%	97.55	24.39	1.25	0.288
Error	406	7902.94	96.79%	7902.94	19.47		
Lack-of-Fit	126	2453.94	30.05%	2453.94	19.48	1.00	0.490
Pure Error	280	5449.00	66.73%	5449.00	19.46		
Total	419	8165.42	100.00%				

General Linear Model: % Ash versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	608.47	36.73%	608.47	101.411	46.30	0.000***
Dose kGy	3	47.07	2.84%	47.07	15.692	7.16	0.000***
Storage/days	4	111.64	6.74%	111.64	27.910	12.74	0.000***
Error	406	889.20	53.68%	889.20	2.190		
Lack-of-Fit	126	602.95	36.40%	602.95	4.785	4.68	0.000
Pure Error	280	286.25	17.28%	286.25	1.022		
Total	419	1656.38	100.00%				

General Linear Model: TTA versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	1.6934	26.53%	1.6934	0.282231	37.47	0.000***
Dose kGy	3	0.1082	1.69%	0.1082	0.036052	4.79	0.003***
Storage/days	4	1.5228	23.86%	1.5228	0.380708	50.54	0.000***
Error	406	3.0584	47.92%	3.0584	0.007533		
Lack-of-Fit	126	2.9028	45.48%	2.9028	0.023038	41.43	0.000
Pure Error	280	0.1557	2.44%	0.1557	0.000556		
Total	419	6.3828	100.00%				

General Linear Model: pH versus Markets, Dose kGy, Storage/days

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	3.5952	21.90%	3.5952	0.59920	21.56	0.000***
Dose kGy	3	0.1036	0.63%	0.1036	0.03453	1.24	0.294
Storage/days	4	1.4312	8.72%	1.4312	0.35780	12.87	0.000***
Error	406	11.2833	68.75%	11.2833	0.02779		
Lack-of-Fit	126	7.7904	47.46%	7.7904	0.06183	4.96	0.000
Pure Error	280	3.4929	21.28%	3.4929	0.01247		
Total	419	16.4133	100.00%				

General Linear Model: %FFA versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	5.904	1.98%	5.904	0.9839	1.63	0.137
Dose kGy	3	22.532	7.55%	22.532	7.5106	12.45	0.000***
Storage/days	4	25.102	8.41%	25.102	6.2756	10.40	0.000***
Error	406	244.943	82.06%	244.943	0.6033		
Lack-of-Fit	126	198.633	66.55%	198.633	1.5765	9.53	0.000
Pure Error	280	46.309	15.52%	46.309	0.1654		
Total	419	298.481	100.00%				

General Linear Model: % Protien versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	0.19424	13.87%	0.19424	0.032373	11.21	0.000***
Dose kGy	3	0.01204	0.86%	0.01204	0.004012	1.39	0.246
Storage/days	4	0.02142	1.53%	0.02142	0.005356	1.85	0.118
Error	406	1.17292	83.74%	1.17292	0.002889		
Lack-of-Fit	126	0.75099	53.62%	0.75099	0.005960	3.96	0.000
Pure Error	280	0.42193	30.12%	0.42193	0.001507		
Total	419	1.40061	100.00%				

(iv) General Linear Model: TVC/logcfu versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	60.29	3.28%	59.98	9.997	5.52	0.000***
Dose kGy	3	678.60	36.96%	614.42	204.808	113.12	0.000***
Storage/days	4	398.37	21.70%	398.37	99.592	55.01	0.000***
Error	386	698.85	38.06%	698.85	1.810		
Lack-of-Fit	120	642.05	34.97%	642.05	5.350	25.06	0.000
Pure Error	266	56.80	3.09%	56.80	0.214		
Total	399	1836.11	100.00%				

General Linear Model: MYC/logcfu versus Markets, Dose kGy, Storage/days

Analysis of Variance

Source	DF	Seq SS	Contribution	Adj SS	Adj MS	F-Value	P-Value
Markets	6	189.47	7.19%	199.55	33.259	14.27	0.000***
Dose kGy	3	936.25	35.53%	865.96	288.655	123.85	0.000***
Storage/days	4	597.88	22.69%	597.88	149.470	64.13	0.000***
Error	391	911.30	34.59%	911.30	2.331		
Lack-of-Fit	122	847.95	32.18%	847.95	6.950	29.51	0.000
Pure Error	269	63.36	2.40%	63.36	0.236		
Total	404	2634.90	100.00%				

Analysis of Variance of Pesticides in smoked fishes

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Factor	12	4690	390.8	1.32	0.354
Error	8	2365	295.6		
Total	20	7055			

Analysis of Variance of Pesticides: Dose/kGy

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Factor	13	0.4180	0.03216	1.07	0.407
Error	42	1.2588	0.02997		
Total	55	1.6768			

APPENDIX E

(v) Common Foodborne Pathogens This appendix provides general information on the common foodborne pathogens included in this set of guidelines for reference.

Common foodborne pathogens	Infective dose*	Incubation period**	Associated foods**
Bacillus cereus x	Greater than 106 organisms per gram in food is indicative of a potential human health hazard; the number of organisms most often associated with human illness is 105 to 108 ; however, the pathogenicity arises from preformed toxin	Emetic intoxication (Preformed heat stable toxin in food): Usually 1 – 6 hours Diarrhoeal (Toxin producedinside body): Usually 10 – 16 hours	Meat, stews, gravies and improperly refrigerated cooked and fried rice
Campylobacter spp.	About 104 organisms in general; however, in trials, as few as 500 organisms led to disease in some individuals	Usually 2 – 5 days (C. jejuni)	Raw and undercooked poultry

Clostridium perfringens x	Greater than 106 organisms or greater than 106 spores per gram of food; toxin production in the digestive tract is associated with sporulation	Range from 6-24 hours; usually 10-12 hours	Meat, poultry and gravies
Escherichia coli O157 (and other Shiga toxin-producing E. coli (STEC))	As low as 10 organisms (E. coli O157:H7)	Range from 2 – 10 days; usually 3 - 4 days	Raw or undercooked ground meat products, fruits and vegetables
Listeria monocytogenes	Undetermined; less than 103 organisms may cause disease in susceptible individuals	Range from 3 – 70 days; 3 weeks on average	Ready-to-eat food with long shelf lives under refrigeration e.g. soft cheese and cold cuts
Salmonella spp.	Typhoid fever: less than 103 organisms	Range from 7 – 21days	Food, water or beverages contaminated with faeces and urine of infected people e.g. shellfish (particularly oysters), raw fruits and vegetables and unpasteurised

			milk and dairy products
Shigella spp.	Less than 1µg preformed heat stable toxin ; greater than 105 organisms per gram in food is needed to produce this toxin level	Range from 30 minutes to 8 hours; usually 2 – 4 hours	Any food contaminated by food handlers with skin infection or nasal carriers, especially those food involving manual handling and no reheating afterwards e.g. sandwiches, cakes and pastries
Vibrio parahaemolyticus	Risk assessment states that the median infective dose is 108 organisms; however, evidence from an outbreak suggests an infectious dose >1,000-fold less than that	Usually 12 – 24 hours	Seafood, salted food e.g. salted vegetables and smoked knuckles or other food cross-contaminated by seafood
Vibrio cholerae	About 106 organisms	Range from a few hours to 5 days, usually 2 – 3 days	Contaminated fish and shellfish

* Source: U.S. FDA Bad Bug Book, Foodborne Pathogenic Microorganisms and Natural Toxins Handbook, 2nd edition. (Accessed 4 March 2014).

** Sources: (1) Diagnosis and Management of Foodborne Illnesses – A Primer for Physicians and Other Health Care Professionals: Foodborne Illnesses Table: Bacterial Agents. AMA/CDC/FDA/US Department of Agriculture, February 2004. And (2) Communicable Diseases of Centre for Health Protection. (Accessed 4 March 2018). x Spore-forming

PUBLICATIONS

Manuscript drafts at consideration stage

i. Title: Assessment of organochlorine pesticide residues in smoked, salted and sun-dried fish in selected coastal markets in Ghana

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ii. Title: Assessment of polycyclic aromatic hydrocarbon (PAH) residues in smoked, salted and sun-dried fish in selected coastal markets in southern Ghana

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