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GHANA**

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**Microbial Profiles of *Kokonte* Chips/Pieces Produced From Three Processing Methods in
Ghana**

By

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Faculty of Biosciences, College of Science,**

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FOOD QUALITY MANAGEMENT

August 2017

presented in this thesis was done entirely by me in

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DEDICATION

I dedicate this project to the Almighty God, my Helper. To my husband, Mr. Charles Nii Korley Commodore, thank you for your immense support and encouragement and to my parents and siblings, how can I forget your love?

Love you all.



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ABSTRACT

High microbial contamination of locally processed “kokonte” chips has been a concern, as its poor quality endangers the health of consumers sometimes with heat-resistant microbial toxins. The

study assessed the extent of microbial contamination in “kokonte” chips produced from three processing methods in Ghana. Samples were purchased from “kokonte” producers in Volta Region (KCPV), cassava sellers in the Greater Accra Region (KCCA) of Ghana while Industryprocessed (IP) “kokonte” samples were processed at the CSIR - Food Research Institute, Pokuase. Moisture content, Total Viable Count (TVC), Total Coliform Count (TCC), *E. coli* Count, and identification of microorganisms were determined using standard microbial techniques. The average TVC of samples from KCPV, KCCA and IP were $\log 7.560 \pm 0.184$ cfu/g, $\log 7.440 \pm 0.573$ cfu/g, and $\log 2.038 \pm 0.409$ cfu/g, respectively. The average TCC for the samples from KCPV and KCCA were $\log 1.452 \pm 0.273$ cfu/g and $\log 1.163 \pm 0.733$ cfu/g, respectively. The IP sample did not record any coliform. No *E. coli* was identified in the IP sample but KCPV sample recorded *E. coli* count of $\log 1.140 \pm 0.180$ cfu/g while samples from KCCA recorded *E. coli* count of $\log 0.550 \pm 0.584$ cfu/g. The locally-processed cassava chips, from KCPV and KCCP, recorded mould counts above $\log 6.00$ cfu/g, however, the IP sample recorded mould count of $\log 1.200 \pm 0.281$ cfu/g. *Corynebacterium* spp., *E. coli*, *Bacillus cereus*, *Salmonella* spp., *Proteus* spp., *Streptococcus* spp., *Bacillus subtilis*, *Staphylococcus aureus*, *Enterobacter* spp., and *Bacillus licheniformis* were identified in KCPV. Mould isolates in the KCPV were *Mucor* spp., *Rhizopus* spp., *Aspergillus niger*, *Fusarium* spp. and *Cryptococcus* spp. The bacterial isolates in the KCCA samples were *Corynebacterium* spp., *E. coli*, *Bacillus cereus*, *Salmonella* spp., *Proteus* spp., *Streptococcus* spp., *Bacillus subtilis*, *Staphylococcus aureus*, *Enterobacter* spp., and *Bacillus licheniformis*. *Mucor* spp., *Rhizopus* spp., *Aspergillus niger*, *Fusarium* spp. and *Cryptococcus* spp. were the mould isolate recorded. Thus, the locallyprocessed samples were highly contaminated and may have serious health implications on consumers compared to the IP samples.

Key words:

Microbial, Contamination, “Kokonte” chips, KCPV – Kokonte Chips Processed by Kokonte Producers in Volta Region, KCCA - Kokonte Chips Processed by Cassava sellers in Greater Accra Region, IP – Industry Processed Kokonte Chips (CSIR – Food Research Institute) Moisture, Total Viable Count (TVC), Total Coliform Count (TCC), *E. coli* Count

TABLE OF CONTENTS

CONTENT	PAGE
Declaration	iii
Dedication	iv
Acknowledgement	v
Abstract	vi

Table of Contents	vii
List of Figures	ix
List of Tables	x
List of Abbreviations	xi
CHAPTER ONE	1
1.0 INTRODUCTION	1
1.1 Background to Study	1
1.2 Problem Statement	4
1.3 Justification	5
1.4 Project Objective	5
1.5 Specific Objectives.....	5
CHAPTER TWO	7
2.0 LITERATURE REVIEW.....	7
2.1 Post Harvest Loses	7
2.2 Cassava	8
2.2.1 World Production of Cassava	8
2.2.2 Cassava Production in Africa	8
2.2.3 Cassava Production in Ghana	9
2.2.4 Problems with the Storage of Cassava	9
2.2.5 Locally-Processed “Kokonte” Production	10
2.3 Equipments Used In Previous Research	14
2.4 Storage	16
2.5 Training Programs for “Kokonte” Processors	18
2.6 Microbiological Work	19
2.7 Chemical Analysis	22
CHAPTER THREE	24
3.0 MATERIALS AND METHODS	24
3.1 Materials.....	24
3.1.1 Equipment and Materials.....	24
3.1.2 Chemicals and Reagents	25
3.1.3 Sampling Area	25
3.1.4 Sampling Method.....	26
3.2 Industry - level Processing of <i>Kokonte</i> Chips.....	28
3.3 Microbiological Analysis.....	28
3.3.1 Total Viable Count (TVC).....	30
3.3.2 Total Coliform Count (TCC).....	30
3.3.3 <i>E. coli</i> Count	30

3.3.4 Mould and Yeast Count	30
3.3.5 Isolation and identification.....	31
3.4 Physico-chemical Analysis	31
3.4.1 Moisture content determination.....	31
3.5 Statistical Tool.....	32
CHAPTER FOUR	33
4.0 RESULTS	33
4.1 Microbial Analysis	33
4.1.1. Microbial Load on Samples from Accra	33
4.1.2. Microbial Isolates from Accra.....	34
4.1.3. Microbial Load on Samples from the Volta Region	34
4.1.4. Microbial Isolates from the Volta Region	35
4.1.5. Microbial Load on Industry - Processed Samples	36
4.1.6. Microbial Isolates from Industry - Processed Samples	37
4.2. Summary of Microbial Loads on the Different Samples	37
4.3. Moisture Content Analysis	39
4.3.1. Moisture Content of Samples from the Greater Accra Region (KCCA).....	40
4.3.2. Moisture Content of Samples from the Volta Region (KCPV)	40
4.3.4 Average Moisture Contents of IP	41
4.4. Bio-Data of Respondents.....	41
4.4.1. State of Cassava Tubers Used In Processing <i>Kokonte</i>	42
4.4.2. Washing of Cassava during Processing of <i>Kokonte</i> Chips.....	43
4.4.3. The Chipping Methods used in Processing of <i>Kokonte</i> Chips.....	44
4.4.4. The Drying Methods used in Processing of <i>Kokonte</i> Chips.....	45
4.4.5. Availability of Extension Messages/Advices on Hygienic <i>Kokonte</i> Chip Processing.....	45
CHAPTER FIVE	46
5.0 DISCUSSIONS	46
CHAPTER SIX	54
6.0 CONCLUSION AND RECOMMENDATIONS.....	54
6.1 Conclusion.....	54
6.2 Recommendations	54
REFERENCES	55

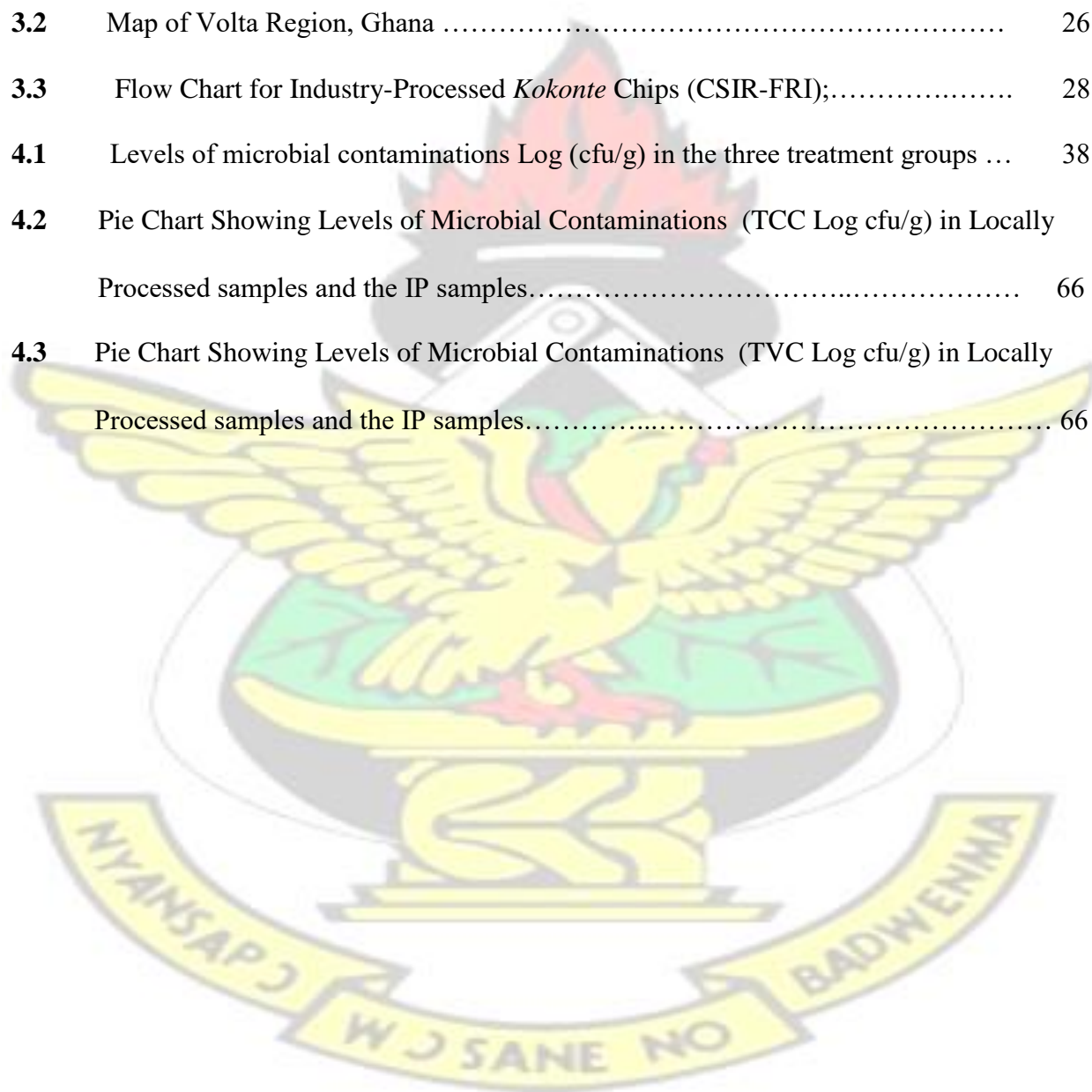
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LIST OF FIGURES

FIGURE

2.1.	Traditional Production of Sun-Dried Cassava Pieces	11
3.1	Map of Greater Accra, Ghana	25
3.2	Map of Volta Region, Ghana	26
3.3	Flow Chart for Industry-Processed <i>Kokonte</i> Chips (CSIR-FRI);.....	28
4.1	Levels of microbial contaminations Log (cfu/g) in the three treatment groups ...	38
4.2	Pie Chart Showing Levels of Microbial Contaminations (TCC Log cfu/g) in Locally Processed samples and the IP samples.....	66
4.3	Pie Chart Showing Levels of Microbial Contaminations (TVC Log cfu/g) in Locally Processed samples and the IP samples.....	66



LIST OF TABLES

TABLE

2.1 Limit for Biological (Microorganisms) and Mycotoxin Content In Cassava Flour.....	11
4.0.1 Microbial Counts of Samples from Accra.....	33
4.0.2 Microbial Isolates of Samples from Accra	34
4.0.3 Microbial Counts of Samples from Volta.....	35
4.0.4 Microbial Isolates of Samples from Volta	35
4.0.5 Microbial Counts in IP Samples.....	36
4.0.6 Microbial Isolates in IP Samples.....	37
4.0.7 Average Moisture Content of KCCA.....	39
4.0.8 Average Moisture Content of KCPV.....	39
4.0.9 Average Moisture Content of IP	39
4.10 State Of Cassava Tubers Used in Processing <i>Kokonte</i> Chips.....	40
4.11 Washing of Cassava During Processing of <i>Kokonte</i> Chips.....	42
4.12 Chipping Process Used in <i>Kokonte</i> Chip Production.....	43
4.13 Drying Method of Chips / Pieces	44
4.14 Availability of Extension Message on Hygienic <i>Kokonte</i> Chip Processing.....	46

LIST OF PLATES

PLATE

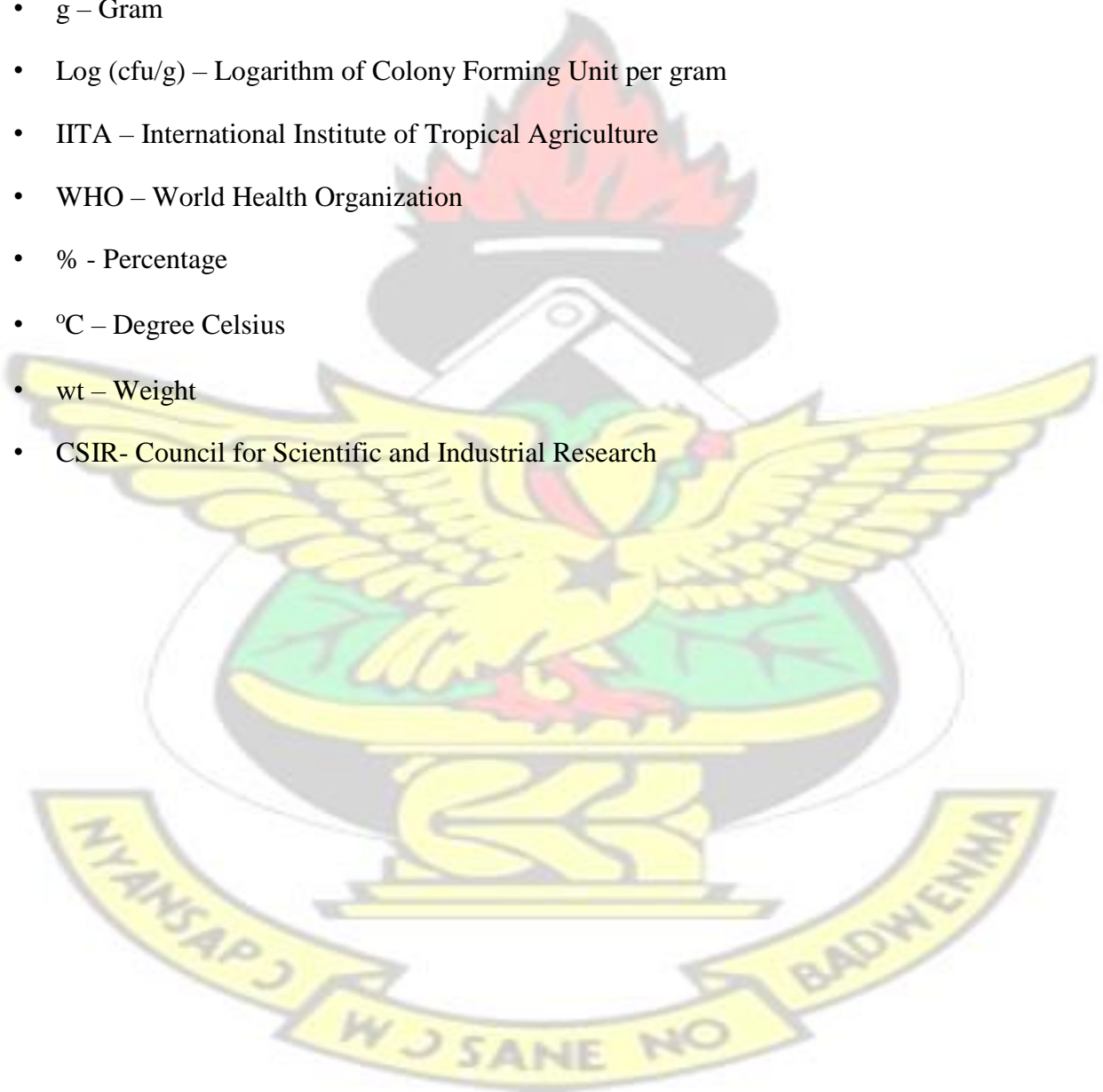
2.1. Local <i>Kokonte</i> Production	11
3.1 <i>Kokonte</i> Chips Processed by <i>Kokonte</i> Producers in the Volta Region.....	27
3.2 <i>Kokonte</i> Chips Processed by Cassava Sellers in Greater Accra Region	27

	PAGE
3.3 Processing of <i>Kokonte</i> Chips at the CSIR-Food Research Institute	28
4.1 A Representative Plate Showing <i>Salmonella</i> Spp. on XLD Agar	67
4.2 A Representative Plate Showing <i>E. coli</i> on EMB Agar	67



LIST OF ABBREVIATIONS

- IP - Industry-processed *Kokonte* Chips (CSIR-Food Research Institute)
- KCPV- *Kokonte* Chip Processed by *Kokonte* Producers in Volta Region, Ghana
- KCCA- *Kokonte* Chip Processed by Cassava Sellers in Greater Accra Region, Ghana
- FAO – Food Agricultural Organization
- g – Gram
- Log (cfu/g) – Logarithm of Colony Forming Unit per gram
- IITA – International Institute of Tropical Agriculture
- WHO – World Health Organization
- % - Percentage
- °C – Degree Celsius
- wt – Weight
- CSIR- Council for Scientific and Industrial Research



CHAPTER ONE

1.0. INTRODUCTION

1.1. BACKGROUND OF THE STUDY

The high incidence of post-harvest food losses, mainly due to inadequate food preservation technologies, affect the quality of foodstuffs in West Africa, where seasonal food shortages and diseases resulting from nutritional deficiency are a major concern (Aworh 2008). According to the FAO, food production will need to grow by 70% to feed the world population which will reach 9 billion by 2050. Further trends like increasing urban population, shift of lifestyle and diet patterns of the rising middle class in emerging economies along with climate change put considerable pressure strain on the planet's resources. Thus, integrated and innovative approaches are needed in addition to the global effort of ensuring sustainable food production and consumption (Nellemann *et al.* 2009; World Economic Forum 2009; FAO/OECD 2011; Foresight 2011; EU ERA-NET SUSFOOD 2012-2014).

In Ghana, issues of post-harvest losses are predominant, especially where locally produced crops such as cassava, yam, maize, rice, beans, and others are hardly exported leading to waste of crops especially during bumper harvest (Scudamore 2005). In order to extend the shelf-life of some of these crops and hence reduce the incidence of postharvest losses, they are processed. Methods involved in processing these indigenous foodstuffs may, however, expose them to contamination by several pathogens mainly fungi and some bacteria in addition to contamination from the farm before processing (Scudamore 2005). Some species of moulds may produce mycotoxins that intoxicate both humans and animal upon consumption (Scudamore 2005). Cassava (*Manihot esculentum* Crantz) is a shrub, 1-3 m high and reproduced through stem cuttings (Nagib Nassar 2007), is one of such perishable crops. It was introduced by the Portuguese to the tropical areas of Africa in the 1600's but found its way to Ghana before the 19th century (Jones, 1959). Cassava

ranks second to maize in terms of planted area but it is the most important crop in Ghana, in terms of quantity (Alhassan 1991).

The [International Fund for Agricultural Development \(IFAD\)](#), African Union (AU) and [New Partnership for Africa's Development \(NEPAD\)](#), ranked cassava as the third most important crop in the world (IFAD, AU and NEPAD 2008) and Ghana has been ranked the fifth – largest cassava producer in the world in 2014 according to FAOSTAT (2014) with production quantity of 16,524,000 tons. Proximate analysis on the fleshy portion contains 62% moisture, 35% starch, 1% protein, 0.3% fat, 2% fiber and 1% ash (Purseglove 1991). The fresh roots contain 35 mg/100g of vitamin C, trace amount of niacin and fat soluble vitamins (Purseglove 1991).

Cassava tubers are known to have high moisture content and begin to deteriorate within 40-48 hours, a huge concern to cassava processors (IITA 1990). Fresh cassava tubers generate heat as a result of high respiration rate, leading to infection and rotting of the tubers by microbes, after three (3) to four (4) days of storage. Local farmers, thus, end up with large amounts of rotten and damaged cassava before they get to the market (Ikujenlola and Opawale 2007) while cassava sellers obtain a loss during low market turn-outs.

Food spoilage has always existed leading to the generation of different food preservation techniques which include the old technique of sun-drying of food to extend the shelf-life, as the amount of moisture in food plays a major role in food spoilage.

The original idea of cutting cassava tubers into pieces and sun-drying these pieces into sun-dried cassava pieces to extend the shelf-life of the crop was a reasonable effort by local farmers to prevent wastage of such economically important crop. These traditionally-processed sun-dried cassava chips are milled into traditional *kokonte* flour. In Ghana, traditional *kokonte* flour is produced by manually peeling the cassava tubers, cutting the tubers using heavy kitchen knife

and sun-drying the pieces on roof tops, bare ground, road sides or any available large surface for weeks to reduce the moisture content.

Different traditional methods for storing dried chips are practiced in different countries. In Ghana, traditional storage structures such as *Kanbon* (cylindrical basket about 0.2 m above the ground, built on a wooden or stone platform) and *Napogu* (cylindrical mud house covered by a thatched roof) are used for storing dried cassava chips. The quality of traditional *kokonte* was evaluated by Adu-Gyamfi and Appiah (2012), who found it to contain high Total Viable Count greater than 10^6 cfu/g and recommended the use of low dose gamma radiation to improve the hygienic quality and to extend shelf-life (Adu-Gyamfi and Appiah 2012).

In this study, market survey in Dome, Madina, Ashiaman and Accra-Tema Station markets, indicated the production of sun-dried cassava pieces in the market premises by cassava sellers. These cassava sellers in avoiding wastage of cassava due to deterioration, and in times of low market demand, have adopted a secondary occupation of producing sun-dried cassava chips from mostly partially rotten and partially fermented cassava tubers. Sometimes few of fresh cassava tubers, especially the smaller sizes may also be cut for drying. The cassava tubers are peeled, cut into pieces, washed and dried in the market premises. The cassava pieces are dried on any available large surface such as table-tops, cut-opened paper boxes laid on top of stores or kiosks, large sack-cloth on the ground and many others. The processing is done few centimeters from where they sell the fresh cassava tubers. The cassava pieces are dried for 3 to 4 weeks depending on the weather conditions and stored in baskets and sacks. These dried cassava pieces are sold to consumers who in turn mill into *kokonte* flour for consumption. In the production of traditional *kokonte* chips, the unhygienic processing steps, such as drying cut-pieces on the bare ground, roof-tops, road sides

and any available spacious area, expose the cassava pieces to microbiological contamination from sources such as dust and animal faecal matter (Stumpf 1998).

1.2. PROBLEM STATEMENT

Cassava is the most important crop in Ghana, in terms of quantity and with new technologies arising from Agricultural Research Institutions, Cassava production has been increased as tubers are ready for harvest, six (6) to seven (7) months after planting (Alhassan 1991). A research conducted by the Food Research Institute of the Council for Scientific and Industrial Research (CSIR)-Ghana, indicated that Ghana recorded a little over 14.2 million Metric tons of cassava tubers in 2011 (Ghana News Agency 2013). Wastage of cassava has, however, been a major concern of local cassava farmers as well as cassava sellers.

In order to prevent wastage of tubers due to deterioration resulting from its high moisture content (62%) (Purseglove 1991) and the generation of heat as a result of high respiration rate, leading to infection and rotting of the tubers by microbes (Ikujenlola and Opawale 2007), local dried cassava chips are processed under unhygienic conditions, milled and sieved into *kokonte* flour and consumed or sold on the market.

Microbial analysis by Ogori and Gana (2013), of cassava flour meal of retted dried balls and chunks in the Makurdi Metropolis, Nigeria, showed bacteria count ranging from 3.2×10^3 to 7.3×10^5 cfu/g and a high mould count ranging from 9.6×10^1 to 3.6×10^3 cfu/g. The significantly high bacteria and mould load obtained indicate contaminations along the processing steps. AduGyamfi and Appiah (2012), also found traditional *kokonte* to contain high Total Viable Count of $>10^6$ cfu/g and recommended the use low dose gamma radiation to improve the hygienic quality and to extend shelf-life.

1.3. JUSTIFICATION

The three “kokonte” chips processing methods used in Ghana results in different microbial profiles in the end-products due to differences in equipments and processing methods involved. The outcome of this study will bring to notice the different types of microbes that contaminate locally-processed *kokonte* chips. This would also inform the public/populace about the various types of microbes that are most common in the processed chips. Data may reveal the safety levels of processed *kokonte* chips/ flour consumed by individuals in this country. Comparing the locally-processed samples with the industry-processed samples would bring to notice if these processed chips are safe for consumption. If not, then measures would be taken to ensure that the contamination level is reduced or minimized to an acceptable level through education given to the processors.

1.4. MAIN OBJECTIVE

The main objective was to evaluate the microbial profile of *kokonte* chips produced from three processing methods in Ghana.

1.5. SPECIFIC OBJECTIVES

- To determine the moisture content of locally-processed dried *kokonte* chips and industry-processed *kokonte* chips.
- To determine the load and kinds of microbes on locally-processed dried *kokonte* chips and industry-processed *kokonte* chips.

- To compare the microbial profile of locally-processed *kokonte* chips and industryprocessed *kokonte* chips.

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CHAPTER TWO

2.0. LITERATURE REVIEW

2.1. POST HARVEST LOSSES

The Food and Agriculture Organization of the United Nations, has predicted a global food loss of about 1.3 billion tons every year (Gustavsson *et al.* 2011). It is estimated that about 1.6 million tons of food are wasted in the United Kingdom because they do not meet the retailer standards (FAO 2013) with an estimated 6.7 million tons of food going waste each year. In the case of the United States, food loss contributes to about 30% of all food produced (FAO 2013).

It has been estimated that the world's population is likely to shoot up to 10.5 billion by 2050 (Aulakh and Regmi 2013), thus, further posing concerns to the global food security issues. In order to address these concerns, it is estimated that food supply would have to be increased by about 60% (estimated at 2005 food production levels) (Alexandratos and Bruinsma 2012) in order to feed the extra mouths. Thus, it is critical to increase production while reducing food losses, however, the issue of postharvest losses is another challenge to deal with. This challenge is also significant in developing countries where food losses in the sub-Saharan Africa are estimated to be about \$4 billion per annum, estimated to feed about 48 million people (FAO 2013). Losses can be higher for perishable products accounting for as high as 50% of harvested fruits, vegetables and root crops such as cassava (Voices newsletter 2006).

In order to reduce postharvest losses as a result of raw material spoilage and hence increase food availability, most raw materials are processed into different products that are expected to extend the shelf life of the product and a typical example of such products is the local processing of fresh cassava into dried chips.

2.2. CASSAVA

2.2.1. World Production of Cassava

In 2014, 268 million tons of cassava was produced with Nigeria being the world's largest producer with 47, 406,770 tons, followed by Thailand with 30, 227,542 tons and then Brazil producing 21,484,218. (www.worldatlas.com)

2.2.2. Cassava production in Africa

In the 16th century, Portuguese traders from Brazil introduced cassava into Africa (Okigbo 1984). Currently, Africa produces about half of the world's production of cassava from Madagascar in the Southeast to Senegal and to Cape Verde in the Northwest. About 40 African countries and about 70 percent of Africa's cassava output is harvested in Nigeria, the Congo and Tanzania (FAO and IFAD 2000).

There has been a double in the cassava production in West Africa from 25.8 million tons in 1990 to 52.3 million tons in 2004 (FAO 2007). Nigeria, the leading producer of cassava globally, harvested 42.72 million tons of cassava in 2006. This harvest was 18% higher than Nigeria's cassava production in 2004. Benin also recorded an increase in cassava, however, not as much as Nigeria. The Atlantique Département recorded the highest in 2006 (934,511 tons) with Plateau following with 307,262 tons and Collines (287,864 t) according to Sanni (2009). Sierra Leone and Ghana are also producing good quantities of cassava, although not as much as that in Nigeria. Cassava is grown all over Sierra Leone with a volume of 350,000 in 2006 (Sanni 2009).

2.2.3. Cassava production in Ghana

Ghana is the 6th largest cassava producer in the world in terms of value, with a Gross Domestic Product (GDP) of 22% (FAO 2005). Cassava is a staple food as well as an income generating

commodity in the country. Many cassava farmers although mostly subsistence, sell their produce to generate income for the household. Cassava ranks second to maize in terms of planted area but the most important crop, in terms of quantity in Ghana (Alhassan 1991). The Volta Region of Ghana is known to produce surplus (1350, 633.30 metric ton against regional consumption of 365,490.85 metric ton) in cassava production over the years (Larbi 2015).

2.2.4. Problems with the storage of Cassava

In many tropical countries, cassava is a staple food. Cassava tubers are known to have high moisture content of about 65%, and begin to deteriorate within 40-48 hours, a huge concern to cassava processors (IITA 1990). Local farmers end up with large amount of rotten and damaged cassava before reaching the markets as tissues of fresh cassava tubers generate heat as a result of high respiration rate, leading to infection and rotting of the tubers by microbes, after 3 to 4 days of storage (Ikujenlola and Opawale 2007).

According to the FAO (2005), Ghana loses about 50% of cassava produced along the value chain. Its perishable nature makes trading in its fresh state internationally difficult, thus intermediate products, such as dried cassava chips, High Quality Cassava and starch are internationally traded (Sanni *et al.* 2009).

Insect infestation is another major problem with storage of cassava chips/pieces. During storage of cassava chips/pieces, insect attack the chips, Anon (1991), reporting as many as twenty-one (21) insect species. Nyakunga (1982) and Hodges *et al.* (1985), mentioned *Dinoderus* spp. and *P. Truncatus* as the two most important storage pests of cassava chips. Rees (1991), also reported issues of *Dinoderus* spp. and *P. Truncatus* found attacking cassava chips. Due to insect infestation

of cassava chips during storage, about 16% weight loss of cassava chips occur (Parker and Booth 1979).

Processing cassava into dry chips often exposes them to microbial contamination through dust, secondary fermentation and insect infestation (Norris 1989). When drying period is prolonged, microbes have the favourable environment to grow, most especially moulds and this may cause health hazards to the consumer.

2.2.5. Locally-Processed *Kokonte* Production

Kokonte is pounded dried cassava chips. Depending on the drying period and method, light brown to dark colour is obtained when cooked. Modern techniques have introduced drying in ovens and the colour has since become lighter. Locally-processed *kokonte* unlike the modern laboratory-processed well packaged *kokonte*, are sun-dried on roof tops, road sides and large surfaces on the compound of households. The original motive for the production of *kokonte* was to find means of preventing wastage of fresh cassava that occurs with the abundance of cassava and little storage and processing facilities in Ghana. Traditional production of sun-dried cassava pieces/chips in Ghana is as shown in Figure 2.1 and Plate 2.1.



Plate 2.1. Local *kokonte* production
(Courtesy: McForson K. 2014)

Steps in traditional production of sun-dried cassava pieces are indicated in Fig 2.1

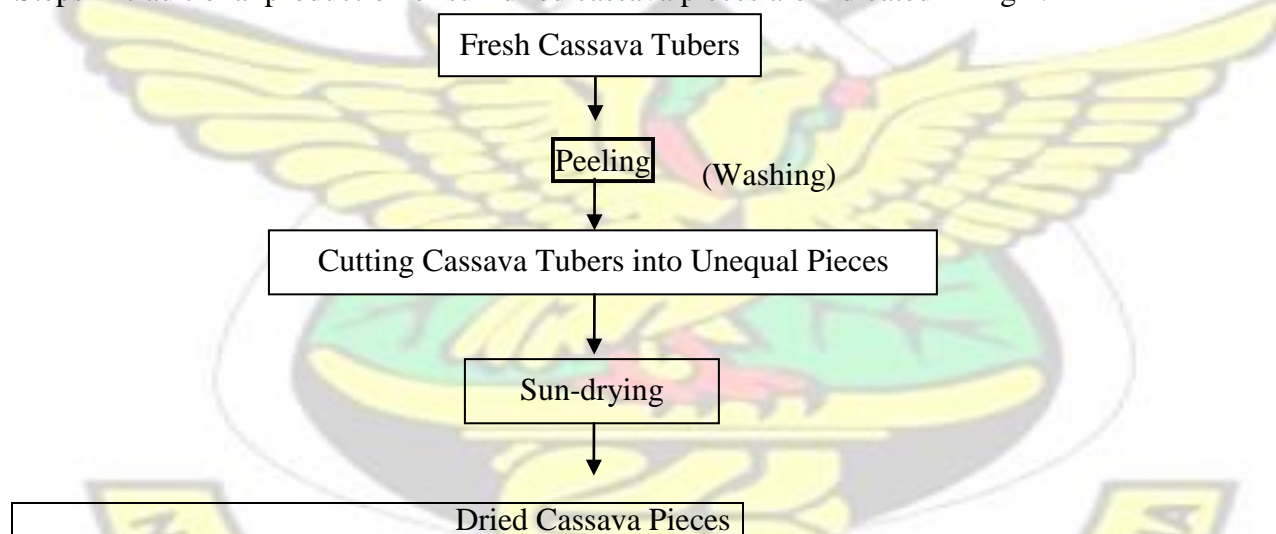


Figure 2.1: Traditional Production of Sun-Dried Cassava Pieces

Codex has defined edible cassava flour (*kokonte*) as the product prepared from dried cassava pieces or paste by pounding, grinding or milling process, followed by sifting to separate the fibre from the flour (Codex Alimentarius Commission 1989).

The Codex standard for edible cassava flour (*kokonte*) is that;

- Edible cassava flour shall be safe and suitable for human consumption.
- Edible cassava flour shall be free from abnormal flavours, odours, and living insects.
- Edible cassava flour shall be free from filth (impurities of animal origin, including dead insects) in amounts which may represent a hazard to human health.
- Edible cassava flour shall have Moisture content of 13% m/m max.

The rudimentary methods used in the production of traditional *kokonte*, however, result in contaminations along the processing stages. There is the need to trace the sources of contamination in relation to the microorganisms identified in the *kokonte* products.

The focus of the literature review is to relate the reports on the various traditional processing stages of the traditional *kokonte* production to the microbiological load and as well as compare the Total Viable Count, Total Coliform Count, Mould and Yeast Count of previous works. The samples used for the microbiological analysis in this research were traditionally produced by cutting the cassava tubers into unequal pieces, sun drying to reduce moisture content of the cassava pieces as shown in Plate 2.1. Nicol (1991) reported that sun-drying is the common method practiced by local cassava farmers in drying fresh cassava pieces to safe moisture content to prevent spoilage. This method is weather dependent and causes problems in rainy seasons. The long-term pattern of weather in the regions in Ghana that are involved in *kokonte* production such as the Volta, Northern, Southern and Eastern part of Ghana differs. On the southern half of the eastern border of Ghana is Volta Region, with a tropical climate, characterized by moderate temperatures between 21 and 32 °Celsius (70–90 °F), for most part of the year. There are two rainfall regimes in the year; that is, March to July and mid-August to October with the maximum average annual rainfall figure of 2,103 mm with 1,168 mm, being the minimum (VRRC 2013). In the Northern Region, however, the mean annual rainfall is approximately 1100 mm and occurs over 95 rainy days. Small rains are

experienced in March/April to a maximum in August. It then declines till it stops completely in mid-November when the dry Saharan wind brings in the harmattan season. Dry wind blows from the northeast lowering humidity and leads to hot days. This enables the farmers to dry harvested cassava roots naturally (Runge-Metzger and Diehl 1993).

In the southern part of Ghana, the effects of harmattan occur in January but highest temperatures occur in March and lowest in August. Dried cassava pieces are used solely for food preparation in Southern Ghana. Sun-drying is difficult when the weather temperature is low and quality of the cassava pieces is affected due to insufficient drying. For good storage, the moisture content should be 12% or below to avoid growth of microorganisms.

Thus, climate changes are an associated factor to the quality production of dried cassava pieces. The quality of dried cassava pieces processed by traditional methods is often poor as longer drying period (> 2 weeks) is required due to the large sizes of the cassava pieces they produce. The drying surfaces used in the production of traditionally-processed sun-dried cassava pieces contaminate cassava chips during the drying period. Market survey indicates the availability of improved high quality *kokonte* products on the market, especially the CSIR-FRI *kokonte* products. CSIR-FRI practice Hazard/Quality Analysis Critical Control Point (HACCP) System to ensure food safety. Pre-requisite programmes such as Good Manufacturing Practices (GMPs) and Good Hygiene Practices (GHPs) are ensured in their processing. There is also mechanized labour leading to increased production as well as hygienic processing, and all these practices contribute to the production of high quality *kokonte* flour and dried cassava pieces.

2.3. Equipments Used In Previous Research

Literature survey shows the efforts of research and funding agencies in promoting mechanization of labour and improved technologies in the traditional *kokonte* production as well as ensuring quality of the traditional *kokonte* products.

Improvements have been made in the area of manufacturing *kokonte* processing equipments to ensure mechanization of labour and enhance human capacity to bring about increased production. Despite some impressive technological achievement, there are still daunting challenges. The traditional *kokonte* processors do not have the working capital to purchase and maintain the equipments; they therefore opt for small-scale production which involves low level of technology. The high costs of most of the cassava processing machinery constitute a major hindrance to acquisition of the readily available machinery. Cooperative bodies are sometimes formed to buy the machinery they lacked.

According to Sanni, *et al.* (2009), Nigeria has advanced in the provision of equipments for *kokonte* products. High quality peeling blade and handle made of mild steel that peels cassava tubers 35kg/hr, have been manufactured by the National Centre for Agricultural Mechanization (NCAM), Ilorin, Nigeria and gradually replacing the use of knife for peeling of cassava.

A project by Sanni *et al.* (2009), on the „Successes and Challenges of Cassava Enterprises in West Africa“, a case study, was done in Nigeria, Bénin, and Sierra Leone to provide an insight into cassava production, processing and utilization, marketing, equipment development, research, and capacity building in West Africa. As part of the project, the local fabricators were trained on the use of such equipment, especially on finishing and quality control.

In collaboration with local fabricators, a new version of the locally fabricated flash dryer with the product contact surface of stainless steel for cassava flour production has been manufactured. The

new flash dryer, commissioned in August 2008 at Godilogo Farms, produces 250 kg/h of cassava flour (Kuye *et al.* 2008). The design and technology are readily available for transfer to fabricators and Small-Medium Enterprises in African countries. From two flash dryers before 2003, well over 100 flash dryers are now installed in Nigeria. Low-cost in-house dryer using heated drum that also has hole to allow spent air to be vented during drying are also used in Nigeria by cassava pieces processors.

A survey was done by Stumpf (1998), in the Northern Region of Ghana, on methods of drying cassava pieces. The study recorded 38% of respondents that dried on the ground and 55% on a raised wooden platform known as the LEENGA (a drying structure made of a wooden platform held on wooden poles about 1.5 to 2 m in height).

Sanni *et al.* (2009) had a field survey in 2008 and reported the use of two improved sun drying systems by local cassava flour processors in Nigeria. These are the raised poles sun drying system and the concrete with dust protector sun drying system. Nigerian engineers have manufactured low-cost drying facilities, thus sun drying is no longer the only method of drying. Solar dryers as well are also exploited.

Department for International Development's research for Development Project (1996-1999) on the „Development and Orientation of Cassava Chip Production in relation to National and International Markets for Food Consumption and Animal Feed in Ghana, assessed the processing options available for the production of dried cassava. Trials were done in Brong Ahafo and Northern Regions on the use of the mini chipper in producing *kokonte* chips milled into flour and sensory analyses performed. The mini chips showed better storage than traditionally-processed dried cassava pieces. For example, there were significant losses after 5 months for the traditionally-processed dried cassava pieces; but mini chips took more than 8 months to show such losses. Their

project aimed at reducing processing losses through improving the methods of processing cassava to produce quality dried cassava chips.

2.4. STORAGE

The method of storage of the dried cassava pieces is of importance to the quality of the *kokonte* products. The main types of storage structures used by traditional cassava chips processors in the Northern region of Ghana are the improved *Kanbon* (30%) and the *Napogu* (28%). Indoor storage in jute sacks is performed by 22% of the farmers (Stumpf 1998).

The traditional *Kanbon* is cylindrical basket made of straw mats (*zanimats*) and built on a wooden or stone platform or stand which is only slightly raised 0.2 m above the ground. The basket varies in size and can be between 1-2 m high and 1.5-2.5 m in diameter. The traditional *Kanbon* is covered with a grass thatched lid which also serves as an inlet and outlet for cassava chips. In many cases, a small grass door in the basket is used for the same purpose. The inside walls of a traditional *Kanbon* is plastered with a mud cover or cow dung and the cassava chips are loosely arranged in the basket (Stumpf 1998).

The field survey by Stumpf (1998), also found improved version of the *Kanbon* constructed in the same way as described above for the traditional version but the basket is raised well above the ground, approximately 0.5 to 1.1 m, on wooden platform with enough ventilation underneath.

The poles are normally made of rodent-proof with metal sheets and the basket of an improved *Kanbon* varies in diameter from 1.5 to 2.5 m, however, the basket is still plastered with mud or cow dung.

Napogu is the type of cylindrical store that is longer-lasting. It has a mud base covered by a thin layer of cement. The roof is made of grass in a conical shape with an access hole just below it.

Provided the foundation does not subside and crack, its life is considerably longer than that of the traditional woven basket stores. Inside the structure there is commonly a wooden platform covered with zanamats (straw) on which chips are loosely arranged, and underneath there is space where fowl find shade or belongings are stored. *Napogu* is usually between 2 and 2.5 m without roofing and diameter is of between 2.5 and 3.5 m.

Scientists in Ghana have reported the degree of weight loss in stored cassava pieces. For instance, according to Stumpf (1998), *Prostephanus truncatus* infestation of stored dried cassava pieces causes weight loss of between 39% and 50% after 8 weeks of storage. Entsie and Ofosu (2001), have also reported 25% loss of stored *kokonte* (sundried cassava pieces) in the Northern Region of Ghana by *Prostephanus truncatus* infestation. In Togo, Helbig and Schulz (1996) also recorded a 75% loss of dried cassava pieces after 8–12 weeks of storage by *Prostephanus truncatus*. Report by Kumar *et al.* (1996), indicates that unhygienic and unsafe substances such as excreta of the adult and larvae contaminate the end product.

The technology transfer and the adaptation by the traditional *kokonte* processors are not encouraging, thus the problem of contamination of traditional *kokonte* is yet to be resolved. Findings show that most cassava farmers and processors are not motivated to use the mechanized systems due to the fact that power supply for the mechanized systems is expensive (Khan 1964; Malik 1981; Djokoto 1986; Sanni *et al.* 2009). Another problem is that the majority of *kokonte* producers are uneducated thus technology transfer is not well appreciated by the group (Johnson *et al.* 2006).

2.5. TRAINING PROGRAMS FOR *KOKONTE* PROCESSORS IN GHANA

Literature survey indicates that the majority of traditional *kokonte* processors are either uneducated or only had primary education and are mostly subsistent cassava farmers (Johnson *et al.* 2006). This has greater impact on the quality processing of traditional *kokonte* and indicates its inferior nature. The traditional *kokonte* processors use rudimentary processes and do not observe hygienic hand washing rules or any standard of quality processing. The CSIR-Food Research Institute in Ghana together with the European Union realised that the inferiority of traditional *kokonte* has a link to the educational levels of the processors. The major objective of the project titled “Validation of Improved Technology for Processing Cassava *kokonte* with Okper Cassava Producers and Processors Association” was a technology transfer for processing of value-added cassava products (Johnson *et al.* 2006). The trainees were trained on the processing of the improved FRI *kokonte* flour. A motorized slicer Model SAA 100 was used to slice fresh peeled and washed cassava tubers into chips of even sizes to accelerate the drying process as well as for covering in the evening and when it is raining, black polythene was used as a cover. The trainees were trained on the reasons why sun-drying on the bare ground and unhygienic surface should be avoided. The main highlights of the best practices were proper sorting of cassava roots for processing, chipping the cassava to sizes of about 7mm maximum thickness and the use of raised platforms for drying cassava chips. The Okper Cassava Farmers and *Kokonte* Producers' Association in the Eastern Region were trained on this improved technology (Johnson *et al.* 2006). Participants were trained on the lapses in the rudimentary methods used in traditional processed *kokonte*, which result in dark colouration due to the presence of moulds such as *Aspergillus parasiticus* and *Aspergillus flavus* and other pathogenic fungi rendering the final *kokonte* flour unsafe for consumption. These microbes release

secondary metabolites such as aflatoxins into the infested cassava chips, which can cause carcinogenic problems when ingested (Johnson *et al.* 2006).

The International Institute of Tropical Agriculture has also supported the view that appropriate technology transfer through private–public partnership is important. There has been collaboration between local fabricators and Research and Funding Institutions on training the local fabricators on the use of such equipments, especially on finishing and quality control (Sanni *et al.* 2009). A standard to guide the quality production of *kokonte* flour and chips have been considered an immediate necessity, thus quality manuals for *kokonte* production have been published to address cassava product standards in terms of production, processing, storage and distribution (Oti *et al.* 2010).

CSIR-Food Research Institute under the Cassava: Adding value in Africa (C: AVA) project, conducted Recipe development to expand High Quality Cassava Flour usage (CSIR, Food Research Institute (C: AVA) project 2014). Participants included selected senior high schools in Volta Region. Also, 178 domestic bursars from Volta, Eastern, Western, and Central regions in Ghana were trained in High Quality Cassava Flour products. The division also conducted monitoring of High Quality Cassava Flour uptake and shelf - life studies of some composite flour (CSIR, Food Research Institute (C: AVA) project 2014). These have been interventional steps to transfers technology of *kokonte* processing to traditional *kokonte* producers to curb the issues of traditional *kokonte* contamination with microbes as well as unhygienic substances.

2.6. MICROBIOLOGICAL ANALYSIS

Ogori and Gana (2003), concerned with the safety and nutrition of meal of retted dried cassava balls and dried cassava chunks performed microbiological examination on the two cassava products in Makurdi Metropolis, Nigeria. The total microbial count on cassava ball and chunks in

the microbiological examination performed did not agree with the U.S.A wheat flour and Germany wheat flour of bacteria load of 10^3 – 10^4 cfu/ml (Rechiter *et al.* 1993; Spincher 1986). The result of microbial analysis showed bacteria count ranged between 3.2×10^3 and 7.3×10^5 cfu/ml. Significantly, high bacteria load was obtained and suggested to have been due to poor processing and drying methods employed in the processing steps.

There was also high mould counts (9.6×10^1 – 3.6×10^3 cfu/ml), an indication of potential spoilage agent and mycotoxins food poisoning (Reiss 1978). During the identification stage, it was observed that there was an increase in Gram positive rods in the cassava chunks flour compared to negative rod of *Proteus* in *Wurukum* and *Fiidi* and *Escherichia* in *Wadata* samples. In a study by Adu-Gyamfi and Appiah (2012), gamma irradiation was used to enhance the quality of some Ghanaian food product which included *kokonte* after investigating the microbial load before and after irradiation. They observed a total viable count of 8.3×10^6 cfu/g for un-irradiated *kokonte* flour.

Also in a case study to develop an appropriate quality assurance system for two cassava-based convenience foods by Johnson *et al.* (2006), they highlighted some difficulties that potential cassava-based factories might face in using the HACCP principles to ensure safe and quality products. They identified the types and sources of hazards, control measures and monitoring procedures for identified critical control points during production of *kokonte* flour. In the washing of raw materials, microbiological kits were used to monitor the hazards in the water source. *E. coli*, *Salmonella*, *Vibrio cholerae*, and fungal spores were detected from the water from storage tanks. At the partial fermentation critical control point, spoilage and pathogenic organisms were detected and during the solar drying, aflatoxins were detected using aflatoxin kits. However, at the village level, there are no microbiological analyses carried out at various critical control points in the processing of traditional *kokonte*.

Kaaya and Eboku (2010), worked on a project to determine mould and aflatoxin contamination of dried cassava chips in Eastern Uganda in association with traditional processing and storage practices. During the survey, it was established that farmers in all the counties in Kumi district harvested and dried cassava for home consumption throughout the exposed year, meaning that the cassava chips were exposed to different weather conditions and resulted in inadequate drying of cassava chips during rainy seasons.

Most of the moulds identified in cassava products are soil borne; implying that farmer practice of drying these products on bare ground predisposes them to fungal infection. Besides, majority of the moulds are known to produce mycotoxins. For instance, *Aspergillus flavus* is a known aflatoxin producing mould species (Klich 2007), *Trichoderma viride* is a known toxigenic mould that produces Trichodermin, *Aspergillus niger* produces oxalic acid and malformin, *Aspergillus fumigatus* produces gliotoxin, fumagilin and veruculogen, *Aspergillus ochraceous* produces ochratoxins, Dextruxin B and Penicillic acid (Adler 2002) while *Penicillium expansum* produces patulin (Moake *et al.* 2005). The results of aflatoxin analysis in cassava show a relatively high (30%) incidence of aflatoxin contaminated cassava samples.

Aryee *et al.* (2006), have reported that at 12% moisture content, cassava products had potential for long shelf life but moisture content greater than 12% allows microbial growth, thus it is important to practice safe moisture content so as to prevent mycotoxins using the Hazard Analysis and Critical Control Point (HACCP) approach (FAO 2001).

Dried cassava pieces develop greenish mould which according to Westby *et al.* (1995), and Wareing *et al.* (2001), can be removed before or after drying the cassava chips in order to improve on the quality of the products but mycotoxins production may have already occurred. Literature, however, shows that the farmers do not remove moulds either before or after drying of

the products. Storage of cassava chips ranges from one week to one year, thus mouldy chips have enough time to produce the highest aflatoxin levels at water activity of 0.996 and temperature of 30 °C (Gqaleni *et al.* 1997) between 5-15 days of storage. Optimum temperatures for aflatoxin production are between 24 and 30 °C with variation between strains and substrates (Klich 2007).

2.7. CHEMICAL ANALYSIS

As reported by FAO (2005), there are no modern processing equipments in the cassava industry in Uganda, thus farmers use indigenous technologies such as hand peeling and slicing and sun drying which are inadequately controlled, labour intensive and produce low quality products. Cassava is sun-dried on virtually any surface in the open and the practices that were positively associated with aflatoxin contamination included the process of drying cassava chips on bare ground; storing by heaping on bare floor and storage in old containers such as *jellycans*.

A study by Kumar *et al.* (1996), focused on the consumption of major nutrients such as starch, sugar and fibre by insects pest on two types of edible cassava chips in India; the plain sun-dried white chips and the parboiled chips, and the their effects on quality. They observed 10% initial damage in plain dried chips and 2% in parboiled chips. Abass *et al.* (1998) and Graffham *et al.* (2000) also reported that the level of moisture content should be $\leq 10\%$ and acid insoluble ash, maximum by mass, should be 0.15 %. Limits for microbial and aflatoxin contents are provided in Table 2.1.

Table 2.1. Limit for biological (microorganisms) and mycotoxin contents in cassava flour

Biological (Microorganisms)	Maximum levels (cfu/g)
-----------------------------	------------------------

Total plate count	10 ⁴
Coliforms	10 ²
<i>Escherichia coli</i>	Nil
<i>Salmonella</i> spp	Nil
Yeast and mould	10 ³
<i>Staphylococcus</i> spp	Nil
<i>Vibrio cholera</i>	Nil
Mycotoxin contents	Maximum levels (µg/kg)
Total Aflatoxins Aflatoxin	10
B ₁	5

Source: Tanzania Bureau of Standards, (2005)

The Ghana Standard Board (GS955:2013), (second edition) standards for unfermented cassava flour; (ISO4833:2003), under which the test method for the thesis was performed provides the microbiological limit per gram for Aerobic Plate Count as 10² minimum and 10³ maximum, *E. coli* Count as zero (0) with Mould and Yeast Count having a minimum of 5 x 10¹ and maximum 10².

CHAPTER THREE

3.0. MATERIALS AND METHOD

During the design of the experiment, market survey indicated that sun-dried cassava pieces for *kokonte* production is not solely produced by *kokonte* producers but has been adopted as secondary occupation for cassava sellers in various markets in order to prevent wastage of cassava tubers. Both open-ended and closed-ended Questionnaires administered to *kokonte* chip

processors in four districts in Volta Region, four markets in Greater Accra Region and *kokonte* chips processed in CSIR – Food Research Institute, Cassava Processing Unit (Pokuase). A total of forty four (44) *kokonte* processors were interviewed; twenty (20) each were from KCPV and KCCA processors while four (4) were from IP processors. All processors interviewed were females.

Three categories of samples analyzed in the study were;

- *Kokonte* Chips processed by *Kokonte* Producers in Volta Region; KCPV
- *Kokonte* Chips processed by Cassava Sellers in Accra Region ; KCCA
- Industry-processed *Kokonte* Chips (CSIR-Food Research Institute) ; IP

3.1. MATERIALS

3.1.1. Equipment and Materials

Mill (Christy and Norris Laboratory Mill (8.000 RPM, Serial Number 45323), Weighing balance (wagtech), Hot-air oven (wagtech), Autoclave (MaCarthy Medical Ltd), Optical microscope (Motic B. Series 30308553), Microscopic slide and slide cover, Incubator (wagtech), Water bath (Grant OLS 200), Desiccators, Tripod stand, Bunsen burner, Measuring cylinder, Petri dish, Hand lens, Paper Towel, Inoculating loop, McCartney Bottles, Zipped plastic bags, Spatula, Aluminum bowl, Kitchen knife, Tray, Pipettes tips, Colony counter (Stuart Scientific, UK), Aluminum cans, Gloves, Cotton, Drying cabinet (wagtech International Ltd.), Safety cabinet (envair, UK), Pipettes (CYAN), Beaker, Solar Dryer, Cassava slicing machine, (< 9 mm).

3.1.2. Chemicals/Reagents

Peptone water [Merk, Darmstadt- Germany], Standard plate count agar [Merk, Darmstadt- Germany], Violet Red Bile lactose agar (VRBLA) [EOS Laboratories], Membrane Lactose Glucuronide agar (MLGA)[Oxoid CM 103M, England], Malt Extract Agar (MEA) [Oxoid CM

0059], Blood agar (Merk, Germany), MacConkey agar (Merk , Germany), Ethanol, absolute Analytical reagent grade (Fisher Chemical, UK), Safranin (Crystal Violet), Iodine.

3.1.3. Sampling Area

A total of thirty-six (36) samples were collected from the Greater Accra and the Volta Regions of Ghana. In the Greater Accra Region, samples were purchased from the Dome Market, AccraTema Station Market, Madina Market, Ashiaman Market with the industry-processed samples obtained from the Food Research Institute of the Council for Scientific and Industrial Research (CSIR-FRI), Pokuase. Samples from the Volta Region were purchased from Nkwanta (Nkwanta District), Ho and Deme (Ho Municipality), Akatsi and Torve (Akatsi District), Keta and Dzita (Keta District).

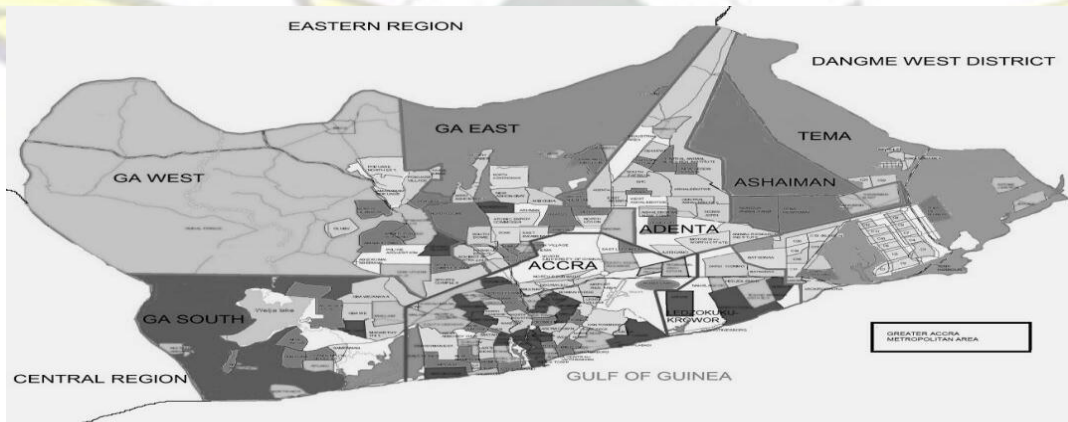


Figure 3.1: Map of Greater Accra, Ghana

Source: <https://sites.google.com/site/ghanaplacenames/visitor/copyright>

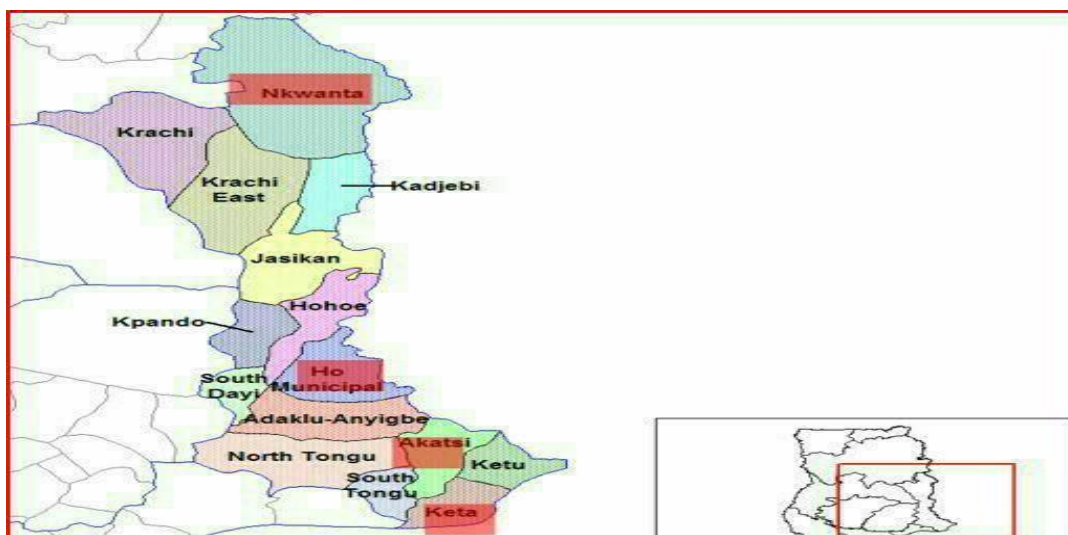


Figure 3.2: Map of Volta Region, Ghana

Source: <https://sites.google.com/site/ghanaplacenames/visitor/copyright>

3.1.4. Sampling Method

Probability sampling method, specifically, the Simple Random Sampling (SRS) was used in the sample collection to ensure variability representative of the population batch and prevented sample bias. Samples were obtained from population batch of dried cassava pieces from *kokonte* processors in Nkwanta (Nkwanta District), Ho and Deme (Ho Municipality), Akatsi and Torve (Akatsi District), Keta and Dzita (Keta District) from the Volta region as well as local cassava sellers in four (4) markets in Greater Accra Region, that is, Dome Market, Accra-Tema Station Market, Madina Market and Ashiaman Market.

Three (3) samples each of *Kokonte* Chips processed by *Kokonte* Producers in Volta Region (KCPV) and *Kokonte* Chips processed by Cassava Sellers in Accra Region (KCCA) were randomly sampled from four (4) traditional *kokonte* producers in Volta Region of Ghana and four (4) markets in Greater Accra, respectively. Three (3) samples each from four (4) Industryprocessed *Kokonte* Chips (IP) batches from CSIR-FRI, Pokuase were also obtained. A total of thirty-six (36)

samples were, thus, obtained for the study. The samples were packed in sterile transparent zipped sealed rubber bags. The bagged samples were placed in a transparent plastic container with cover, transported to the laboratory and kept at room temperature.



Plate 3.1. Kokonte chips processed by kokonte producers in Volta Region, Ghana



Plate 3.2. Kokonte chips produced by cassava sellers in Greater Accra, Region

3.2. INDUSTRY-LEVEL PROCESSING OF *KOKONTE* CHIPS



Figure 3.3. Flow Chart for Industry-Processed *Kokonte* Chips (CSIR-FRI)



Plate 3.3. Industry-level processing of *kokonte* chips at the CSIR-Food Research Institute

3.3. MICROBIOLOGICAL ANALYSIS

The laboratory analysis was carried out in the Microbiology and Quality Control Laboratory of the Animal Research Institute of the Council for Scientific and Industrial Research (CSIR-ARI). Microbiological Analysis performed included Total Viable Count (TVC), Total Coliform Count (TPC), *E. coli* Count (TEC), and the identification of the isolated microbes.

Each of the population batches were milled using the Christy and Norris Laboratory Mill (8.000 RPM, Serial Number 45323) and thoroughly mixed in a sterilized aluminum bowl using a spatula to obtain a homogeneous powder. Two hundred gram (200g) samples were weighed into transparent zipped sealed rubber bags and kept at room temperature.

In the analyses, one (1) gram each of samples was weighed and aseptically transferred into sterile Mac-Cartney Bottles containing 9ml of 0.1 % sterile blank peptone water (Merk, Darmstadt-Germany) and serially diluted using 10-fold serial dilution into seven (7) other MacCartney bottles containing 0.1 % 9ml peptone water. Different sterile pipette tips were used for each dilution.

3.3.1. Total Viable Count (TVC)

One (1) ml of each of the dilutions (10^{-1} - 10^{-7}) was aseptically added to Petri dishes containing molten standard plate count agar (Merk, Darmstadt- Germany), allowed to cool and incubated at 37 °C for 24 hours. Plates showing colonies between 30 and 300 were counted using the colony counter (Stuart Scientific, UK). Counted colonies were multiplied by the dilution factor (Kiiyukia 2003).

3.3.2. Total Coliform Count (TCC)

Using the plate count method, 1ml of each dilution was aseptically placed into 9cm sterile Petri dish containing molten Violet Red Bile lactose agar (VRBLA) (EOS Laboratories), mixed by swirling, allowed to cool and incubated at 37 °C for 24 hours. Using the colony counter, colonies were counted and multiplied by the dilution factor (Kiiyukia 2003).

3.3.3. *E. coli* Count

One (1) ml of each dilution (10^{-1} - 10^{-7}) was aseptically added to molten Membrane Lactose Glucuronide agar (MLGA) (Oxoid CM 103M, England) in sterile Petri dishes, mixed by swirling and allowed to cool before incubation at 37 °C for 24 hours. Plates showing between 30 and 300 colonies were counted using the colony counter (Stuart Scientific, UK) (Kiiyukia 2003).

3.3.4. Mould and Yeast Count

For the yeast and mould count, 100µL of the dilutions (10^{-1} to 10^{-7}) was added onto the surface of Malt Extract Agar (MEA) (Oxoid CM 0059) plates and spread evenly on the entire surface of the plate using a glass spreader. Plates were incubated at room temperature (25 °C to 28 °C) for 5 to 7 days. Using the colony counter, colonies were counted and multiplied by the dilution factor (Kiiyukia 2003).

3.3.5. Isolation and Identification of microbes

From each sample, a sterile loopful of the 10^{-1} was aseptically streaked onto Blood agar (Merk Germany) and MacConkey agar (Merk, Germany) using plate-out technique (Heritage *et al.* 1996). Cultures were incubated aerobically and non-aerobically at 37 °C for 24 hours in a bacteriological

incubator. Impure cultures on primary media were purified by sub-culturing onto selected secondary media. Colonial morphology of organisms based on the physiological characteristics was studied for size, shape, outline, colour-change in medium. Standard bacteriological techniques including staining, cell morphology and biochemical tests such as Catalase and Indole were used to isolate and identify the foodborne microbes (Kiiyukia 2003).

3.4 MOISTURE DETERMINATION ANALYSIS

About two grams of each sample was weighed into empty aluminum cans in triplicate. The empty aluminum cans were weighed and the weights recorded. The aluminum cans containing the weighed samples were arranged on a metallic tray and dried in an electric hot-air oven for six hours at a temperature of 105 °C. The dried samples were then placed in desiccators to cool and weighed. The weights of the cans with the dry sample were recorded and the weight of dry matter calculated with the formula below.

The percentage dry matter was calculated as;

$$\% \text{ Dry Matter} = \frac{\text{Weight of Dry Matter}}{\text{Weight of Sample}} \times 100$$

The moisture content was calculated as;

$$\% \text{ Moisture} = 100 - \% \text{ Dry Matter}$$

3.5. STATISTICAL TOOL

The one - way analysis of variance (ANOVA) was used to determine the significant differences between the means of three *kokonte* samples processed from three processing methods in Ghana.

This analysis tests the null hypothesis:

$$H_0 : \mu_1 = \mu_2 = \mu_3 = \dots = \mu_k$$

Where μ =group mean and k = number of groups.

The alternative hypothesis is that “the population means are not all equal” (Laerd Statistics, Lund Research Ltd. 2013).

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CHAPTER FOUR

4.0. RESULTS

4.1. MICROBIAL ANALYSIS

4.1.1. Microbial Load on Samples from Accra

Table 4.0.1 shows the Log (cfu/g) of microbial loads on the dried cassava chips sampled from the four markets from the Greater Accra Region. The TVC of the samples ranged from log 6.602 to 7.903 cfu/g, with a mean of $\log 7.440 \pm 0.573$ cfu/g. The TCC ranged from log 0.000 to 1.845 cfu/g, with an average of $\log 1.163 \pm 0.733$ cfu/g. The load of *E. coli* ranged from log 0.000 to 1.301 cfu/g. Moulds and yeasts recorded loads greater than log 6.00 cfu/g (Table 4.0.1).

Table 4.0.1. Microbial Counts of Samples from Accra

ID	TVC	[Log (cfu/g)]			Yeast and moulds
		TCC	<i>E. coli</i>		

ASM	7.903	1.699	1.000	>6
	7.845	1.602	1.000	>6
	7.778	1.000	0.000	>6
ATSM	6.699	0.000	0.000	>6
	6.778	0.000	0.000	>6
	6.602	0.000	0.000	>6
MDM	7.699	1.699	1.301	>6
	6.602	1.301	1.000	>6
	7.845	1.602	0.000	>6
DOM	7.845	1.602	0.000	>6
	7.903	1.845	1.301	>6
	7.778	1.602	1.000	>6 Mean
<hr/>				
7.440 ± 0.573		1.163 ± 0.733	0.550 ± 0.584	>6

TVC = Total Viable Count, TCC = Total Coliform Count, *E.coli* = *Escherichia coli*

KCCA = Kokonte Chips processed by Cassava Sellers in Accra Region

ASM = Ashiaman Market

ATSM = Accra -Tema Station Market

MDM = Madina Market

DOM = Dome Market

4.1.2. Microbial Isolates from Accra

Table 4.0.2 shows the types of microbes isolated from the samples from Accra. Main bacteria genera were *Bacillus*, *Corynebacterium*, *Escherichia*, and *Enterobacter* while the main fungal genera were *Rhizopus*, *Aspergillus*, *Mucor*, *Fusarium*, and *Cryptococcus*.

TABLE 4.0.2. Microbial Isolates on samples from Accra

ID	BACTERIAL ISOLATES	FUNGAL ISOLATES
ASM	<i>Bacillus cereus</i> , <i>Bacillus subtilis</i> , <i>Corynebacterium glutamicum</i> , <i>Enterobacter</i> spp	<i>Rhizopus</i> spp., <i>Aspergillus niger</i> , <i>Mucor</i> spp., <i>Fusarium</i> spp.
ATSM	<i>Bacillus cereus</i> , <i>Bacillus licheniformis</i>	<i>Rhizopus</i> spp., <i>Aspergillus niger</i> , <i>Mucor</i> spp., <i>Cryptococcus</i> spp.

MDM	<i>Bacillus cereus</i> , <i>E. coli</i> , <i>Enterobacter</i> spp.	<i>Rhizopus</i> spp., <i>Aspergillus niger</i> , <i>Mucor</i> spp., <i>Cryptococcus</i> spp.
DOM	<i>Bacillus cereus</i> , <i>E. coli</i> , <i>Enterobacter</i> spp.	<i>Rhizopus</i> spp., <i>Mucor</i> spp., <i>Aspergillus niger</i> , <i>Cryptococcus</i> spp.

KCCA = Kokonte Chips processed by Cassava Sellers in Accra Region

ASM = Ashiaman Market

ATSM = Accra -Tema Station Market

MDM = Madina Market

DOM = Dome Market

4.1.3. Microbial Load on Samples from the Volta Region

Table 4.0.3 shows the Log (cfu/g) microbial profile of dried cassava chips sampled from *kokonte* producers in four districts in Volta Region, Ghana. The TVC of the samples ranged from log 7.301 to log 7.778 cfu/g with the TCC ranging from log 1.000 to 1.845 cfu/g. In the case of the *E. coli*, the counts ranged from log 1.000 to 1.477 cfu/g. Counts for the Moulds and yeasts were greater than log 6.00 cfu/g.

Table 4.0.3. Microbial Counts of Samples from the Volta Region

	[Log (cfu/g)] ID	TVC		
		TCC	<i>E. coli</i>	Mould and Yeast
NKD	7.699	1.699	1.301	>6
	7.602	1.301	1.301	>6
	7.778	1.602	1.000	>6
KTD	7.602	1.477	1.000	>6
	7.477	1.699	1.477	>6
	7.301	1.845	1.301	>6
AKD	7.778	1.301	1.000	>6
	7.301	1.000	1.000	>6
	7.699	1.602	1.000	>6
HOD	7.699	1.602	1.301	>6
	7.301	1.000	1.000	>6

	7.477	1.301	1.000	>6
Mean	7.560 ± 0.184	1.452 ± 0.273	1.140 ± 0.180	>6

TCC = Total Coliform Count, TVC = Total Viable Count, *E.coli* = *Escherichia coli*

KCPV = Kokonte Chips processed by Kokonte Producers in Volta Region

NKD = Nkwanta District, KTD = Keta District, AKD = Akatsi District, HOD = Ho District

4.1.4. Microbial Isolates from the Volta Region

Table 4.0.4 shows the microbial isolates in dried cassava chips from four districts in the Volta Region, Ghana. The main bacteria genera were *Salmonella*, *Bacillus*, and *E. coli* while that of the fungi were *Aspergillus* and *Mucor*.

TABLE 4.0.4. Microbial Isolates on samples from the Volta Region

ID	BACTERIAL ISOLATES	FUNGAL ISOLATES
NKD	<i>Staphylococcus aureus</i> , <i>E. coli</i> , <i>Salmonella</i> spp., <i>Corynebacterium</i> spp.	<i>Aspergillus niger</i> , <i>Rhizopus</i> spp., <i>Mucor</i> spp.
KTD	<i>Bacillus subtilis</i> , <i>Salmonella</i> spp.	<i>Rhizopus</i> spp., <i>Aspergillus niger</i> ,
AKD	<i>Proteus</i> spp., <i>E. coli</i> , <i>Bacillus cereus</i> <i>Rhizopus</i> spp., <i>Aspergillus niger</i> , <i>Mucor</i> spp., <i>Fusarium</i> spp.	<i>Salmonella</i> spp., <i>Streptococcus</i> spp.
HOD	<i>Corynebacterium</i> spp, <i>E. coli</i> , <i>Bacillus cereus</i> , <i>Salmonella</i> spp., <i>Proteus</i> spp, <i>Enterobacter</i> spp.	<i>Rhizopus</i> spp., <i>Aspergillus niger</i> , <i>Mucor</i> spp., <i>Fusarium</i> spp.

KCPV = Kokonte Chips processed by Kokonte Producers in Volta Region

NKD = Nkwanta District, KTD = Keta District, AKD = Akatsi District, HOD = Ho District

4.1.5. Microbial Load on Industry - Processed Samples

Table 4.0.5 indicates the microbial loads in four IP samples from the CSIR-FRI cassava chips processing unit. The mean TVC was $\log 2.038 \pm 0.409$ cfu/g. Counts of moulds and yeasts ranged from $\log 1.000$ to 1.845 cfu/g.

TABLE 4.0.5. Microbial Counts of the Industry-Processed Samples

ID	[Log (cfu/g)]			
	TCC	TVC	<i>E. coli</i>	Moulds and Yeasts
IP 1	0.000	2.000	0.000	1.000
	0.000	2.000	0.000	1.477
IP 2	0.000	1.845	0.000	1.000
	0.000	2.602	0.000	1.301
IP 3	0.000	2.477	0.000	1.845
	0.000	2.000	0.000	1.000
IP 4	0.000	2.000	0.000	1.000
	0.000	2.301	0.000	1.000
IP 4	0.000	2.602	0.000	1.301
	0.000	1.845	0.000	1.477
IP 4	0.000	0.000	1.000	1.000
	0.000	1.301	0.000	1.000
Mean	0.000 ± 0.000	2.038 ± 0.409	0.000 ± 0.000	1.200 ± 0.281

Mean \pm standard deviation, TCC = Total Coliform Count, TVC = Total Viable Count

IP 1 = Industry Processed *kokonte* chips Sample 1

IP 2 = Industry Processed *kokonte* chips Sample 2

IP 3 = Industry Processed *kokonte* chips Sample 3

IP 4 = Industry Processed *kokonte* chips Sample 4

4.1.6. Microbial Isolates from Industry - Processed Samples

Table 4.0.6 shows the microbial isolates from the Industry processed samples from the CSIRFRI, cassava chips processing unit. The only bacteria genus isolated was *Bacillus* while only *Aspergillus niger* was the type of fungus isolated.

TABLE 4.0.6. Microbial Isolates on the Industry Processed Samples

ID	BACTERIAL ISOLATES	FUNGAL ISOLATES
IP1	<i>Bacillus subtilis</i>	<i>Aspergillus niger</i>
IP2	<i>Bacillus subtilis</i>	<i>Aspergillus niger</i>
IP3	<i>Bacillus cereus</i>	<i>Aspergillus niger</i>
IP4	<i>Bacillus subtilis</i>	<i>Aspergillus niger</i>

IP 1 = Industry Processed *kokonte* chips Sample 1

IP 2 = Industry Processed *kokonte* chips Sample 2

IP 3 = Industry Processed *kokonte* chips Sample 3

IP 4 = Industry Processed *kokonte* chips Sample 4

4.2. SUMMARY OF MICROBIAL LOADS ON THE DIFFERENT SAMPLES

Figure 4.1 summarizes the microbial load in the samples from the three treatment groups. The IP samples had lower TVC and absence of coliforms. Hypothesis testing using the One-way ANOVA indicated that the differences among the means of TVC are significant, p-value of 0.001. The microbial counts on the IP samples differed significantly from the KCPV and the KCCA; however, the KCCP did not significantly differ from KCPV. The differences among the

means of TCC were significant with p-value of 0.000. All samples differed significantly from each other. The differences among the means of *E. coli* were significant with p-value of 0.000.

This analysis tests the null hypothesis:

$$H_0: \mu_1 = \mu_2 = \mu_3 = \dots = \mu_k$$

Where μ =group mean and k = number of groups.

From the one – way ANOVA results the alternative hypothesis that states that “the population means are not all equal” (Laerd Statistics, Lund Research Ltd, 2013), is accepted. There are significant differences amongst the three treatment groups of *kokonte* samples analyzed.

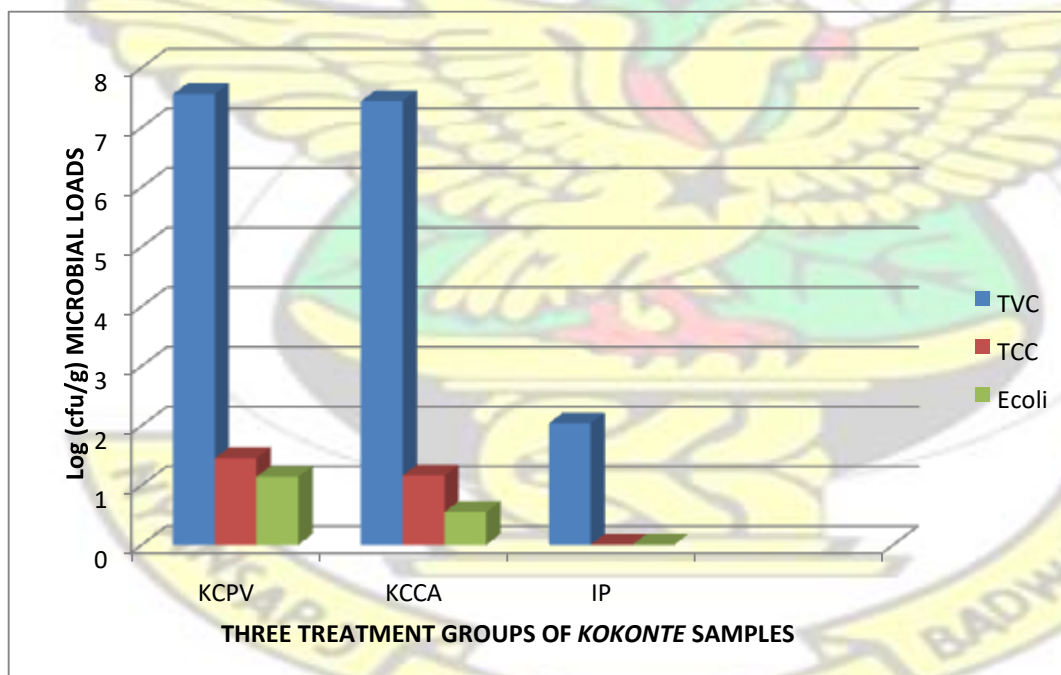


Figure 4.1. Levels of microbial contaminations Log (cfu/g) in the three treatment groups

Kokonte Chips processed by Kokonte Producers in Volta Region = KCPV

Kokonte Chips processed by Cassava Sellers in Accra Region = KCCA

Kokonte Chips processed by the CSIR-FRI (Industry Processed) = IP

4.3. MOISTURE CONTENT ANALYSIS

4.3.1. Moisture Content of Samples from the Greater Accra Region (KCCA)

Table 4.0.7 shows the percentage moisture contents of the dried cassava chips from four districts in the Greater Accra Region. The values of percentage moisture of KCCA ranged from 8.000 to 9.667%.

TABLE 4.0.7. Average Moisture Contents of KCCA

Sample Code	% Dry Matter	% Moisture
ASM	92.144	8.000
ATSM	90.311	9.667
MDM	90.410	9.667
DOM	91.159	9.000
MEAN	91.006 ± 0.848	9.084 ± 0.788

KCCA = Kokonte Chips processed by Cassava Sellers in Greater Accra Region

ASM = Ashiaman Market; ATSM = Accra -Tema Station Market; MDM = Madina Market; DOM = Dome Market

4.3.2. Moisture Content of Samples from the Volta Region (KCPV)

Table 4.0.8 shows the percentage moisture contents of the dried cassava chips from four markets in the Greater Accra Region. The values of percentage moisture of KCPV ranged from 3.663 to

4.644%.

TABLE 4.0.8. Average Moisture Contents of KCPV

Sample Code	% Dry Matter	% Moisture
NKD	96.300	3.663
KTD	95.601	
4.399 AKD	95.356	
4.644 HOD	95.724	
4.276		
MEAN	91.006 ± 0.848	9.084 ± 0.788

KCPV = *Kokonte* Chips processed by Kokonte Producers in Volta Region

NKD = Nkwanta District, KTD = Keta District, AKD = Akatsi District, HOD = Ho District

4.3.4 Average Moisture Contents of IP

Table 4.0.9 shows the percentage moisture contents of the dried cassava chips from four batches of IP samples. The values of percentage moisture of IP ranged from 5.940 to 5.985%.

TABLE 4.0.9. Average Moisture Contents of IP

Sample Code	% Dry Matter	% Moisture
IP1	94.018	5.982
IP2	94.015	5.985
IP3	94.060	5.940
IP4	94.031	5.969
MEAN	94.031 ± 0.0168	5.9690 ± 0.0252

IP 1 = Industry Processed *kokonte* chips Sample 1

IP 2 = Industry Processed *kokonte* chips Sample 2
IP 3 = Industry Processed *kokonte* chips Sample 3
IP 4 = Industry Processed *kokonte* chips Sample 4

4.4. BIO-DATA OF RESPONDENTS

This section summarizes the outcome of questionnaires administered to *kokonte* chip processors in four districts in Volta Region, four markets in Greater Accra Region and *kokonte* chips processed in CSIR – Food Research Institute, Cassava Processing Unit (Pokuase). A total of forty-four (44) *kokonte* processors were interviewed; twenty (20) each were from KCPV and KCCA processors while four (4) were from IP processors. All processors interviewed were females.

4.4.1. State of Cassava Tubers Used In Processing *Kokonte*

Table 4.10 summarizes the state of cassava tubers used in processing *kokonte*. Out of twenty (20) participating KCPV processors (*Kokonte* chip processor in Volta Region), twelve (12) processors representing 60% processed fresh cassava tubers mostly harvested from their farms or other times purchased from the market. Eight processors representing 40% processed both fresh and partially-rotten cassava tubers. All twenty (20) participating KCCA processors (*Kokonte* chip processed by cassava sellers in Greater Accra Region), representing 100%, processed partially-rotten cassava tubers from the cassava stock they market. Four (4) IP processors (CSIR-FRI *kokonte* processors) representing 100% processed fresh cassava tubers for *kokonte* chip production.

Table 4.10. State of Cassava Tubers Used In Processing *Kokonte*

Cassava Tubers	State of Processors	Number of Processors	Percentage of
KCPV Processors	Fresh cassava tubers	12	60%
	Mixture of fresh and partially - rotten cassava tubers	8	40%
KCCA Processors 20	Partially - rotten cassava tubers		100%
IP Processors	Fresh cassava tubers	4	100%

4.4.2. Washing of Cassava during Processing of *Kokonte* Chips

Table 4.11 summarizes the washing of cassava during *kokonte* chips processing. Out of twenty (20) participating KCPV processors (*Kokonte* chip processor in Volta Region), 90% washed the cassava tubers using pipe-borne water after peeling and after chipping while 10% washed cassava chips using pipe-borne water only after peeling cassava. All twenty (20) participating KCCA processors (*Kokonte* chip processed by cassava sellers in Greater Accra Region), representing 100% washed the tubers only after chipping of the cassava tubers. Four (4) IP processors (CSIR- FRI *kokonte* processors) representing 100% washed the cassava tubers severally (3 to 4 times) using pipe-borne water after peeling.

Table 4.11. Washing of Cassava during Processing of *Kokonte* Chips

	Washing of Cassava Tubers	Number of Processors	Percentage of Processors
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KCPV Processors	After peeling and after chipping	18	90%
cassava tubers			
	After peeling	2	10%
cassava tubers			
KCCA Processors	After chipping	20	100%
cassava tubers			
IP Processors	Severally (3 to 4 times) after peeling cassava tubers	4	100%

4.4.3. The Chipping Methods used in Processing of *Kokonte* Chips

Table 4.12 summarizes the chipping method used in *kokonte* chips production. All twenty (20) participating KCPV processors (*Kokonte* chip processor in Volta Region), cut cassava into pieces using a heavy kitchen knife. All the twenty (20) participating KCCA processors (*Kokonte* chip processed by cassava sellers in Greater Accra Region) also cut cassava into pieces using heavy kitchen knife. Four (4) IP processors (CSIR- FRI *kokonte* processors) representing 100%, used slicing machine to slice cassava into chips.

Table 4.12. Chipping Methods used in *Kokonte* Chips Production

Processors	Processors	Chipping Method	Number of	Percentage of
KCPV Processors		Use of heavy kitchen knife to chip	20	100%
KCCA Processors		Use of heavy kitchen knife to chip	20	100%
IP Processors		Use slicing machine to slice cassava into chips	4	100%

4.4.4. The Drying Methods used in Processing of *Kokonte* Chips

Table 4.13 summarizes the drying methods used in *kokonte* chips processing. Out of twenty (20) participating KCPV processors (*Kokonte* chip processor in Volta Region), 40% sun-dried cassava pieces on sheets laid on the ground whereas 60% sun-dried cassava pieces on raised pole structures. Out of twenty (20) participating KCCA processors (*Kokonte* chip processed by cassava sellers in Greater Accra Region), 90% sun-dried the cassava pieces on table-tops while only 10% sun-dried the cassava pieces on sheets laid on ground. Four (4) IP processors (CSIR-FRI *kokonte* processors) representing 100% dried the cassava chips in solar dryer.

Table 4.13. Drying Methods of *Kokonte* Chips / Pieces

Cassava chips/pieces	Drying Method Processors	Number of Processors	Percentage of
KCPV Processors	Sun - drying on sheets laid on ground	8	40%
	Sun - drying on raised poles structures	12	60%
KCCA Processors	Sun – drying on table – tops	18	90%
	Sun - drying on sheets laid on ground	2	10%
IP Processors	Solar – drying in solar dryer	4	100%

4.4.5. Availability of Extension Messages/Advices on Hygienic *Kokonte* Chip Processing

Table 4.14 summarizes the availability of extension messages on hygienic *kokonte* chip processing. Out of twenty (20) participating KCPV processors (*Kokonte* chip processor in Volta Region), 80% had extension messages/advices on improved drying methods and improved chipping methods while 20% had no external messages/advices on *kokonte* chip processing. Out of twenty (20) participating KCCA processors (*Kokonte* chip processed by cassava sellers in

Greater Accra Region), 100% of processor, had no external messages/advices on *kokonte* chip processing. Four (4) IP processors (CSIR- FRI *kokonte* processors) representing 100%, had access to *kokonte* processing manual.

Table 4.14. Extension Advices/Messages on Hygienic *Kokonte* Chip Processing

Processors	Availability of Extension Messages Processors	Number of	Percentage of
KCPV Processors	Extension messages on improved drying methods	16	80%
	No extension message	4	20%
KCCA Processors	No extension messages	20	100%
IP Processors	Availability of kokonte processing manual	4	100%

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CHAPTER FIVE

5.0. DISCUSSION

Food safety is critical in preventing outbreak of food-borne diseases. The quality of food not only relates to correct portions of nutrient formulations, but includes the hygienic conditions surrounding food preparation and food handling. Thus intrinsic and extrinsic factors must be controlled in the safety processing of food. Hygienic practices have been reported to be poor amongst food handlers in Ghana, Addo *et. al.* 2007. The health dangers in the processing, preservation and storage of dried cassava pieces are microbial contaminations and subsequent toxin productions. Many argue that prepared *kokonte* from sun-dried cassava flour are cooked and thus microbes are destroyed eventually. According to Kamimura *et. al.* (2010), at a temperature of 100-120 °C, mycotoxins remained almost 100% after 45 minutes heating. Most bacteria toxins are also heat resistant Kamimura *et. al.* (2010). Although the body has detoxification mechanisms to

eliminate relatively normal toxin levels, unacceptable levels pose dangerous health effects. Local *kokonte* processors must be supported by governmental organizations to ensure the mechanization of labour and technology transfer through training program in order to prevent the introduction of food pathogens in the product and ensure food safety for traditional *kokonte* production as the poor quality of locally processed *kokonte* chips not only hinder the market-share of such economically important food commodity but most importantly endanger the health of consumers with heat-resistant microbial toxins.

Cassava (*Manihot esculenta* Crantz) is highly perishable due to its high moisture content. Dry matter of fresh cassava as reported by Barima *et al.* (2000), ranges between 17% and 47%. The results of this study indicated a mean dry matter content of 95.75% and 91.01% for KCPV and KCCA, respectively while that of the Industry - processed samples (IP) was observed to be 94.031%. Low moisture contents were obtained for the three categories of samples; KCCA, KCPV and IP were 9.084%, 4.246, and 5.969, respectively. The result from this study agrees with Kenoyer (1975), who stated that „Sun drying though one of the oldest food preservation methods known to man is still highly efficient in areas of strong sunlight and low humidity“, probably, the reason why sun-drying is common in Ghana. Quintson (2015) stated that 95% of *kokonte* producers in the Hohoe Municipality of the Volta Region of Ghana sun-dry their *kokonte* pieces with none using the solar drying method. The bio-data of this study confirmed the data survey of Stumpf (1998), in the Northern Region of Ghana, on the method of drying cassava pieces. In this study 40% of KCPV processors sun-dried their cassava pieces on sheet laid on the ground whereas 60% of KCPV processors interviewed sun-dried their cassava pieces on raised-pole structures. A study by Stumpf (1998), recorded 38% of *Kokonte* processors who dried cassava on the ground while 55% dried their *kokonte* chips on raised wooden platform known as Leenga. Thus the improved sun-drying system is gradually being adopted by local *kokonte* processors. The solar drying is still not exploited by the local *kokonte* processor.

Water activity (a_w) is the term used to describe the water requirements of microorganisms in food or environment and is defined as the ratio of water vapour pressure of the food substrate to the vapour pressure of pure water at the same temperature (Jay 2000). Growth of microbes is facilitated when water is available in the food to participate in chemical as well as biochemical reactions. Moisture reduction techniques are means of extending the shelf-life of cassava.

In this study, the mean moisture content of 4.25% (± 0.42) was obtained for KCPV, 9.08% (± 0.79) moisture content for KCCA and 5.9690 ± 0.0252 for IP. The mean percentage moisture for KCPV was significantly different from KCCA with a p-value of 0.000 ($p < 0.05$). Although the moisture content in the KCPV and KCCA were low, ($<12\%$), KCCA had higher moisture content. This may be due to lack of spacious drying areas for sun-drying cassava pieces in the market. KCCA samples are dried several days to weeks to acceptable moisture content to reduce the moisture content. The long period of sun-drying on unhygienic drying surfaces in the market premises, offers opportunity for microbial contamination, which explains the high count of Total

Viable Counts and Total Coliform Count in the local “kokonte” chips . Cassava pieces are sundried from a week to over 4 weeks depending on the weather conditions. The sizes of *kokonte* chips also affect the quality of the end product. Although there has been the introduction of motorized cassava slicing machine to the local *kokonte* processors (80% of KCPV have had messages/advice from external officers on improved methods of drying and improved chipping methods), the use of heavy kitchen knife to cut cassava into unequal sizes is the method of chipping used by the local processors. All (100%) the KCPV processors and KCCA processors used heavy kitchen knife in cutting cassava into pieces before drying. Unlike these local processors, the IP *kokonte* chip processors used the slicing machine to slice cassava into light-weight (< 9 mm) cassava chips with a drying period of three (3) to five (5) days. The sizes of the

samples from KCPV and KCCA were noted to be large and unequal and might have prolonged the drying process. Department for International Development (DFID), Research and Development Project Record (<http://www.dfid.gov.uk/R4D/Project/1368/Default.aspx>) on “Development and orientation of cassava chip production in relation to national and international markets for food consumption and animal feed in Ghana” developed a system of drying chip based on tray and polythene sheet drying of mini-chips that produces a high quality product after, typically, 2 days. A mini-chipping machine, designed by the International Institute of Tropical Agriculture was also used to obtain cassava chips. In participatory trials in Brong Ahafo and Northern Regions, sun-dried *kokonte* showed significant losses due to contaminations after 5 months; but mini chips took more than 8 months to show such losses. The bulky unequal sizes of samples from KCPV and KCCA thus reduced the rate of drying and prolonged inefficient sundrying of cassava pieces. This offered opportunity to microbes introduced during unhygienic processing of cassava pieces to use the available water during the period of sun-drying of cassava pieces for chemical and biochemical reaction for their proliferation. Therefore, in spite of the low moisture content in the KCPV and KCCA, high bacteria and mould counts were observed in this study.

Moisture content of 5.969% was obtained for samples from the IP samples compared to the moisture contents of 4.245% and 9.084 % for KCPV and KCCA, respectively. This indicates that significant amount of moisture was evaporated from the samples at a temperature of 50 °C for a minimum of 3 days and maximum of 5 days. The use of solar dryer is quicker, less exposure of the cassava pieces to microbial contamination and less time consuming compared to the traditional sun-drying method. However, all three categories of samples had moisture contents less than 12%

and thus the locally processed cassava pieces would have had good shelf-life if there were no significant microbial contaminations.

All the samples of KCPV and KCCA, after plating on Malt Extract Agar (MEA), had uncountable levels of moulds (Tables 4.0.1 and 4.0.3) with isolates of *Aspergillus niger*, *Rhizopus* spp., *Mucor* spp., *Fusarium* spp. These mould counts were higher compared to the mean mould count of 5.0×10^4 cfu g⁻¹ obtained by Kaaya (2010).

The mould and yeast isolates in the KCPV as well as the KCCP are in agreement to the mould and yeast isolates according to [Wareing et al. \(2001\)](#) and [Essono et al. \(2007\)](#), who reported presence of *Rhizopus*, *Mucor*, *Penicillium*, *Aspergillus* and *Fusarium* species observed in cassava products. However, high levels of moulds ($> 6 \log \text{ cfu g}^{-1}$) recorded exceeds the minimum of 5×10^1 and maximum of 10^2 cfu/g mould count set by Ghana Standard Board (GS955:2013) for unfermented cassava flour indicating that the sample from KCPV and KCCA, which are milled into *kokonte* flour (unfermented cassava flour) may be unsafe for human consumption.

The Florida Solar Energy Center (FSEC) (2007-2014), publication on mould growth, explained the importance of the four critical requirements for mould growth; available mould spores, available mould food, appropriate temperatures and considerable moisture, and indicated prohibition of mould growth with the removal of any one of the mentioned requirements. Food borne yeasts and moulds will out compete common bacteria under harsh growth condition of relatively low moisture (water activity of ≤ 0.85) and nutrient (Wilkins 2014).

In the processing of sun-dried cassava pieces, the cassava provides sufficient nutrients to support mould growth and the elimination of mould spores is not possible as mould spores are ubiquitous. Thus, the reduction of moisture to acceptable level at appropriate temperature is a means of

controlling mould growth, in this case by sun-drying. However, because the drying periods were prolonged from a week to over 4 weeks, mould growth was enhanced.

The IP samples recorded a mean of 1.200 ± 0.281 (Log cfu/g) with *Aspergillus niger* isolates compared to the high fungal load (6 log cfu/g) of the locally processed cassava pieces (KCPV and KCCA). *Aspergillus niger* are ubiquitous in nature and usually grow on starchy foods. This could be a contributing factor for their presence in the IP samples. Mean Total Viable Count of 7.560 ± 0.184 log cfu/g, 7.440 ± 0.573 log cfu/g and 2.038 ± 0.409 log cfu/g were recorded for KCPV, KCCA and IP samples, respectively. The microbial counts of the locally processed samples (KCPV and KCCA) agree with the results for dried cassava chips by Gacheru *et al.* (2016), who obtained TVC of 5.16 - 8.04 log cfu g⁻¹ and 4.81- 7.21 log cfu g⁻¹ respectively with p-value of 0.029. Hypothesis testing using the One-way ANOVA showed significant differences among the TVC means of three categories of samples (KCPV, KCCA, and IP samples), p-value of 0.001. The IP samples differed from KCPV and KCCA; however, the KCCA did not significantly differ from the KCPV. This may be due to the unhygienic processing methods used.

For example the drying surfaces used for KCCA were observed to be top of roofing sheets of market kiosks, table tops and sacks while the KCPV were dried on the bare ground, road sides and roof tops exposing the cassava pieces to bacterial isolates such as *Corynebacterium* spp., *E. coli*, *Bacillus cereus*, *Salmonella* spp., *Proteus* spp., *Streptococcus* spp., *Bacillus subtilis*, *Staphylococcus aureus*, *Enterobacter* spp., and *Bacillus licheniformis*.

The mean Total Viable Count was high for KCPV (7.560 ± 0.184 log cfu/g) and KCCA (7.440 ± 0.573 log cfu/g). The result exceeds the microbiological limit per gram for Aerobic Plate Count provided by Ghana Standard Board (GS955:2013) for unfermented cassava flour; ISO4833:2003

(minimum of 10^2 and maximum of 10^3 cfu/g). The bacterial isolates in the IP samples were *Bacillus subtilis*, *Bacillus cereus*. *Bacillus cereus* is found in dust, soil and spices. The life cycle of *Bacillus* spp. involves a dormant stage where they exist as spores and could have easily contaminated the IP samples. These spores are very resistant to chemicals, heat and other treatments (Public Health of Canada 2004). There was significant difference, p-value of 0.000, among the mean Total Coliform Count for KCPV, KCCA and IP samples (1.452 ± 0.273 log cfu/g, 1.163 ± 0.733 log cfu/g and 0.000 ± 0.000 log cfu/g respectively (Table 4.1 and 4.3). All the samples differed significantly from each other. Among the three genera of coliforms; *Escherichia*, *Klebsiella* and *Enterobacter* were isolated in both KCPV and KCCA samples. Coliform bacteria are aerobic or facultative anaerobic, Gram-negative, non-spore-forming, rodshaped bacteria commonly found in the intestinal tract, and known as “indicator” microorganisms (Yousef and Carlstrom 2003). As indicator microorganisms, presence of coliforms in the dried cassava pieces indicates poor sanitary conditions as coliforms may originate from either faecal or non-faecal contamination according to Yousef and Carlstrom (2003). In this case coliforms isolated might largely have been introduced to the cassava pieces from the dropping of birds as well as the faecal matter of household animals due to poor drying conditions of the cassava pieces. According to the Ghana Standard Board (GS955:2013) for unfermented cassava flour; ISO4833:2003, the minimum *E. coli* count is 0 cfu/g and maximum is 0 cfu/g. Thus *E. coli* must not be present in food. The means of *E. coli* count for KCPV and KCCA were 1.140 ± 0.180 log cfu/g and 0.550 ± 0.584 log cfu/g, respectively making them unsafe for human consumption. The differences among the means of *E. coli* count in the three categories of samples (KCPV, KCCA and IP samples) were significant with p-value of 0.000.

From the one – way ANOVA results the alternative hypothesis that states that “the population means are not all equal” (Laerd Statistics, Lund Research Ltd. 2013), is accepted. There are significant differences amongst the three treatment groups of *kokonte* samples analyzed.

The interventions by food processing organizations and funding agencies in curbing the unhygienic processing of dried cassava pieces, through workshops, encouraging co-operative formations, provision of basic processing equipments such as solar dryers, will not only increase productivity but also ensure hygienic processing of the traditional *kokonte* products.



CHAPTER SIX

6.0. CONCLUSIONS AND RECOMMENDATIONS

6.1. CONCLUSIONS

The study revealed that the microbial profile of locally processed dried cassava pieces (KCPV and KCCA) exceeded the accepted limit for human consumption as the counts were above the set codex standards for contaminant and toxins (CODEX STAN 193 - 1995) for edible cassava flour. However, the industry-level(IP) processed cassava chips, showed no coliform count and recorded low bacterial, mould and yeast counts that were below the set codex standards for contaminant and toxins (CODEX STAN 193 - 1995) for edible cassava flour.

6.2. RECOMMENDATIONS

Traceability is important for improving the safety and quality of food products. Further analyses on sources of microbial contamination along the processing stages of traditional *kokonte* chips should be carried out to ensure production of high quality traditional *Kokonte* chips.

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APPENDICES

Appendix 1

Questionnaires for Kokonte producers in Volta Region, Ghana.

1. Name of *kokonte* producer _____ Date _____
District _____ Village _____

2. Who are involved in commercial *kokonte* production in your area?

Kokonte Producers _____

Cassava Farmers _____

Cassava Sellers _____

Both Kokonte Producers and Cassava Farmers _____

3. What is the state of cassava processed for *kokonte*?

Fresh cassava tubers _____

Partially- rotten cassava tubers _____

Rotten cassava tubers _____

4. What processes do you use in the production of Kokonte?

5. Do you wash the cassava? If yes, do you wash; Before peeling?

After peeling?

After cutting into Pieces?

Before / after peeling / after cutting into Pieces?

6. Source of water for washing cassava

7. Where do you dry your chips?

On Sheets on the ground _____

Other means of drying _____

8. How long do you dry the cassava chips (days/weeks)? _____

9. Where do you store the cassava chips?

In sacks _____

On the ground _____

Raised platform _____

Floor/Room _____

In _____ storage structures such as _____

Kanbon _____

Napogu _____

10. Are any extension messages on *kokonte* chip processing available?

No _____ Yes _____

11. If yes, what kind of advises?

Appendix 2

Questionnaires for Market – processed sun dried cassava pieces sellers in Greater Accra, Ghana.

1. Name of dried cassava pieces seller _____ Date _____

District _____

2. Is dried cassava pieces production your main occupation?

3. What is the state of cassava processed for *kokonte*?

Fresh cassava tubers _____

Partially- rotten cassava tubers _____

Rotten cassava tubers _____

4. What processes do you use in the production of Kokonte?

5. Do you wash the cassava? If yes, do you wash; Before peeling?

After peeling?

After cutting into Pieces?

Before / after peeling / after cutting into Pieces?

6. Source of water for washing cassava

7. Where do you dry your chips?

Leenga _____

On the ground _____

Both ways _____

Other means of drying _____

8. Are any extension messages on *kokonte* chip processing available?

No _____ Yes _____

9. If yes, what kind of advises?

Appendix 3.

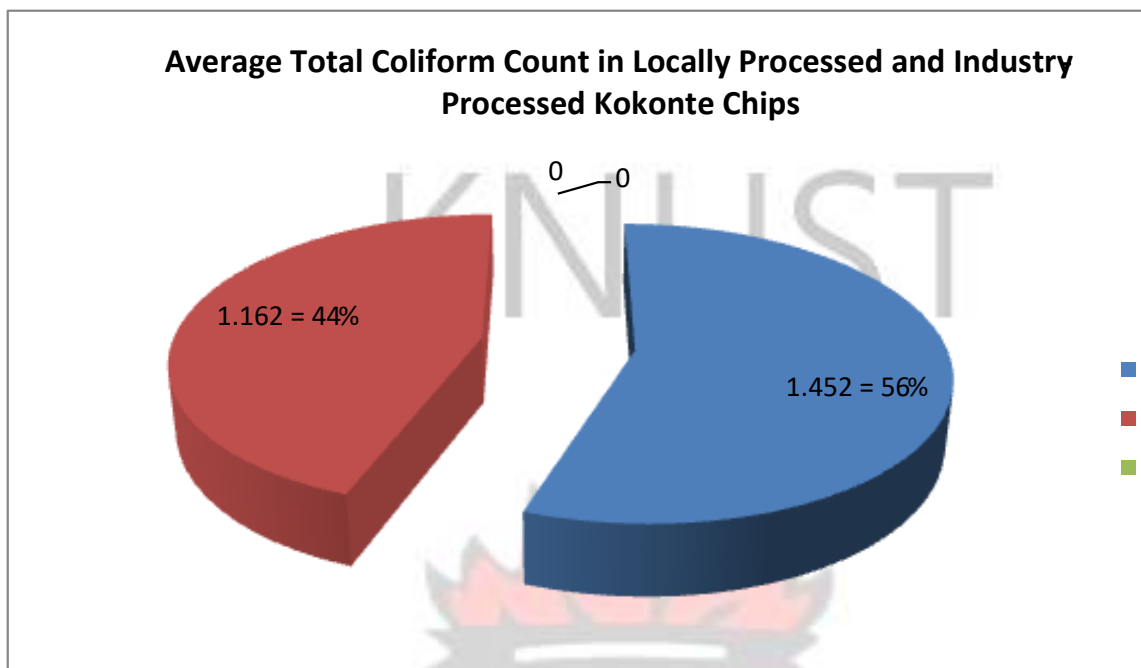


Figure 4.2. Levels of microbial contaminations (TCC Log (cfu/g)) in locally processed samples and Industry- level processed *kokonte* chips

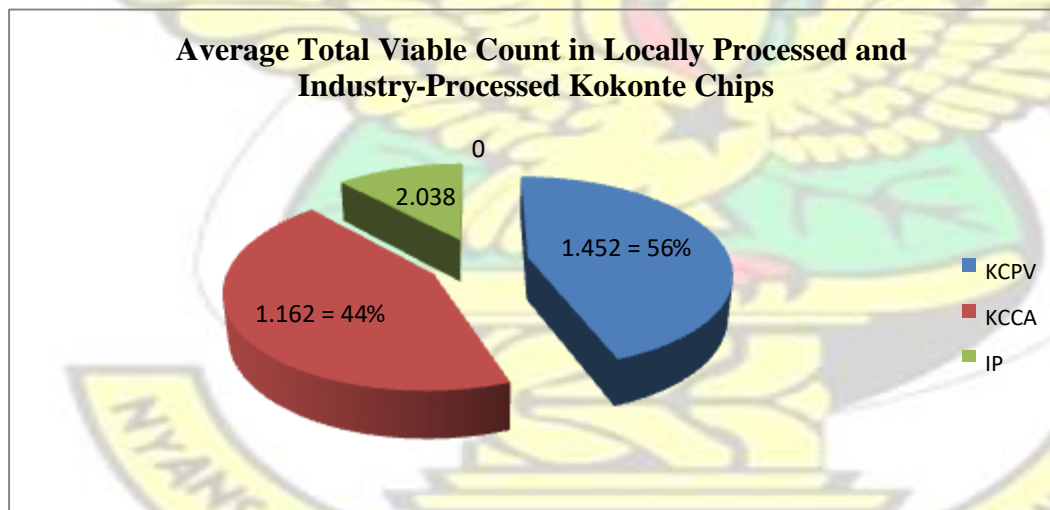


Figure 4.3. Levels of microbial contaminations (TVC Log (cfu/g)) in locally processed samples and Industry- level processed *kokonte* chips

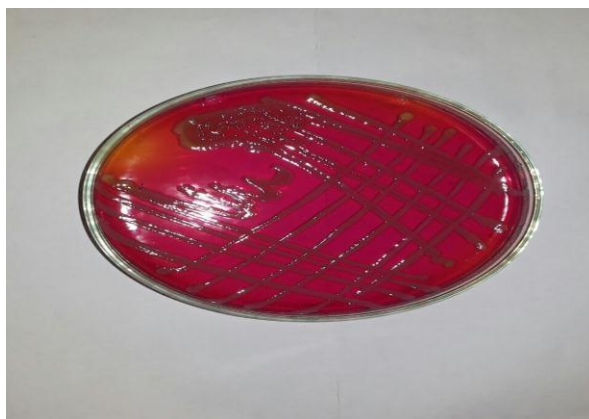


Plate 4.1. A representative plate showing *Salmonella* spp. on XLD Agar



Plate 4.2. A representative plate showing *E. coli* on EMB Agar

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