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EFFECTS OF DIFFERENT FERMENTATION MATERIALS ON THE QUALITY OF MIXED AND HYBRID COCOA BEANS

BY

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EFFECTS OF DIFFERENT FERMENTATION MATERIALS ON THE QUALITY OF MIXED AND HYBRID COCOA BEANS

A THESIS SUBMITTED TO THE SCHOOL OF RESEARCH AND GRADUATE STUDIES, KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY, IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF MASTER OF PHILOSOPHY (Mphil. POST HARVEST TECHNOLOGY) DEGREE

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DECLARATION

I hereby declare that this submission is the result of my own work and that it has not been submitted either in part or whole for any other degree elsewhere. Works by other authors have been duly acknowledged.

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DEDICATION

I dedicate this work first of all to God Almighty for his abundant blessing, to my parents Mr and Mrs Bawah Atikpo, my Uncle Osman Egala and Wife Nahid Egala for their immense support and the confidence they had in me right from my primary education till now, I say may the almighty God bless them abundantly.

I also dedicate this work to my wife Aisha Karim and my lovely daughter Jasmine Kokui Atikpo, I say my God almighty bless them abundantly for their love and support, and finally a big thank you to the entire Egala and Atikpo family for their unconditional love.

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ABSTRACT

A field survey on fermentation practices of farmers was conducted in three cocoa districts in Western South of the Western Region of Ghana. The outcome of the survey led to the establishment of an experiment to determine the effects of the use of polythene sheets or fertilizer sacks as alternative materials for fermentation instead of plantain/ banana leaves on the physical quality and chemical composition of the dried cocoa beans. The experiment was a 2 x 3 factorial in a completely randomized design with three replications. The first factor comprised of two cocoa varieties mainly hybrid and mixed varieties whilst the second factor consisted of plantain/ banana leaves, polythene sheets and fertilizer sacks as fermentation materials. The results of the field survey revealed that averagely 58%, 23%, and 17.7% of the farmers used plantain / banana leaves, fertilizer sacks and polythene sheets respectively for fermentation. However, 4% of the farmers in Samreboi used basket for fermentation of their cocoa beans. The purity of the cocoa beans was significantly influenced by the interactive effects of the cocoa varieties and fermentation materials, Hybrid variety or mixed varieties fermented with plantain leaves had the highest purity above 90%, significantly greater than the purity from the other treatment combinations. The least purity was obtained from mixed variety fermented with fertilizer sack (79.63 %). In a similar trend, the pH of the dried cocoa beans was also significantly influenced by the interactions. Mixed varieties fermented with fertilizer sack and polythene sheet had pH of 6.23 and 6.40 compared with 4.90 recorded by mixed varieties fermented with plantain leaves.

All the chemical composition parameters examined in this study were not significantly different between the hybrid and mixed cocoa varieties except pH and

free fatty acids. The study revealed that fermentation of cocoa beans with polythene sheets or fertilizer sacks are not adequately fermented.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

AAS	Atomic Absorption Spectrophotometer
ANOVA	Analysis of Variance
AOAC	Association of Official Analytical Chemists
COCOBOD	Cocoa Board
FFA	Free Fatty Acid
GDP	Gross Domestic Product
HCl	Hydrogen Chloride
KNUST	Kwame Nkrumah University of Science and Technology
NFEs	Nitrogen Free Extracts
OICC	Office International du Cacao et du Chocolat

CHAPTER ONE

1.0 INTRODUCTION

Cocoa (*Theobroma cacao* L) originated from the Amazonia Region of Brazil and grown in tropical countries like Ivory Coast, Ghana, Nigeria, Indonesia, Brazil, Venezuela and Malaysia. Hundreds of thousands of cocoa farmers in Ghana depend on cocoa cultivation for their livelihood. Ghana is the second largest producer of cocoa beans in the world, and in 2005, about 60 % of the country‟s foreign income came from export of cocoa beans.

The cocoa industry alone employs close to about 60 per cent of the national agricultural work force in the country (Assuming-Brempong *et al.*, 2006). It also contributes about 12 per cent of the Gross Domestic Product (GDP).

Ghana is second only to the Ivory Coast in the production of cocoa but produces the best quality cocoa beans (COCOBOD, 2011). Currently, there are six cocoa growing areas in Ghana namely, Ashanti, Brong Ahafo, Eastern, Volta, Central and Western regions (COCOBOD, 2011). There are four (4) methods of cocoa bean fermentation; heap, basket, box and tray methods. The heap method is however the simplest and therefore normally used by small holder farmers. It is done by spreading out fresh plantain leaves in a circle on the ground and heaping fresh cocoa beans on them. The mat of leaves is punctured with a stick to create drainage holes in the mat, this allows easy pulp drainage. The heap of beans is then covered with more leaves and held in place by small logs. This protects the fermenting beans from surface drying, mould

growth and help to maintain the heat generated within the heap. The size of the fermenting heap varies from 300kg – 500kg (Conservation Alliance, 2013).

In recent times however, some farmers in Wassa Amenfi west and Aowin district of the western region sew fertilizer sack into mats, perforated them to allow for the drainage of pulp and used as fermentation material. The use of transparent polyethylene sheet as fermentation material is also gaining popularity. However, unlike the plantain heap method which has received scientific attention in terms of the quality of the dried fermented beans, there is no information on the quality of the beans which have been fermented using any of the two fermentation materials still in the heap methods.

The main objective of this study therefore was to determine the effects of different fermentation materials on the quality of mixed and hybrid cocoa beans.

Specifically, the objectives were to

i. identify fermentation practices of cocoa farmers ii. determine the interactive effects of fermentation materials and cocoa varieties on the physical attributes of the dried cocoa beans iii. determine the interactive effects of fermentation materials and cocoa varieties on the chemical composition of the dried cocoa beans

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 BRIEF HISTORY OF COCOA

Cocoa originated from around the headwaters of the Amazon in South America. Its cultivation and value spread in ancient times throughout central and Eastern

Amazonian and northwards to Central America. It spread to the British, French and Dutch West Indies (Jamaica, Martinique and Surinarn) in the 17th century and to Brazil in the 18th century. From Brazil it was taken to SÃO Tome and Fernando Po (now part of Equatorial Guinea) in 1840; and from there to other parts of West Africa, notably the Gold Coast (now Ghana), Nigeria and the Ivory Coast (World cocoa foundation, 2010). Currently in Ghana, there are six cocoa growing areas namely Ashanti, Brong Ahafo, Eastern, Volta, Central and Western regions.

2.2 FERMENTATION

2.2.1 Fermentation systems

Various cocoa fermentation systems have been developed. These methods of fermentation vary considerably from producing region to region and in some instances even different farmers practice different fermentation techniques depending on the quantity of beans and what system is available to them (Lopez and Dimick, 1995; Afoakwa *et al.*, 2012a). The general methods involve the seeds being placed in some kind of receptacle, confined and weighed down. Most of the world‟s cocoa is fermented on banana leaves covered heaps, in boxes, trays, and baskets and on dry platforms (Lopez and Dimick, 1995; Beckett, 2009; Afoakwa, 2010).

2.2.1.1 Heap fermentation

In this system, beans are piled or heaped on and covered with plantain or banana leaves. Cocoa seeds varying in quantities from 25 to 1000 kg are heaped on a floor covered with plantain leaves and perforated for easy drainage of sweatings. The heap is subsequently covered with the leaves, which are weighed down, by branches or other materials (Lopez and Dimick, 1995). This is done not only to protect the

fermenting mass against insects and conserve heat but also to prevent the entry of rainwater, dust and other foreign matter (Wood and Lass, 1985). The heap ideally must be turned every 72 hours during the fermentation to ensure a uniform fermentation (Baker *et al.*, 1994). According to Baker *et al*. (1994) stated that about 57 % of the Ghanaian farmers do not turn the heaps. This is due to its laborious nature. The duration for this type of fermentation ranges between 4 and 7 days. The heap fermentation system dominates in Ghana and other West African countries (Wood and Lass, 1985; Baker *et al.*, 1994).

2.2.1.2 Tray fermentation

Due to the laborious nature of turning the heap in fermentation, the tray method was developed based on the early observation that when the beans are heaped to ferment, a change in color of the beans occurs up to a depth of about 10 cm when beans are not mixed (Nair, 2010). Based on this, in trays fermentation, reasonable quantities of beans of 10 cm height holding are held in trays stacked one over the other for sufficient development and conservation of heat and there is no mixing

(Thompson *et al.*, 2001).

A typical size of wooden trays is 90 cm x 60 cm x 13 cm. Battens or reapers are fixed at the bottom of the trays with small gaps in between to avert a situation where beans might falling through and also allow for free flow of sweatings (Nair, 2010).

When filled, the trays are stacked one over the other with the last being empty allow for drainage of the sweatings. With the minimum number of trays required for a stack being six the beans of the topmost tray are covered with banana leaves but after 24 hours, the stack of trays is covered with gunnysacks to conserve the heat that

develops. Tray fermentation will normally be completed in day 4 and dried on day 5 (Nair, 2010).

2.2.1.3 Box fermentation

This type fermentation requires relatively large, fixed volume of cocoa and it is usually used on larger farms. A wooden box is subdivided by either fixed or movable internal partitions into compartments measuring approximately 1 m x 1 m x I m. The containers vary in size from country to country and have a holding capacity of 600 to 700 kg of fresh cocoa beans (Lopez and Dimick, 1995). The boxes are arranged in a series of steps raised above ground level to take advantage of gravity to facilitate turning, which is effected by simply removing a movable wall and shoveling the seeds into the box below. The boxes are raised over a drain that holds sweatings. The floor of the box is usually solid and contains holes for drainage and aeration (Lehrian and Patterson, 1983).The beans are covered with banana leaves or jute sacking to maintain the heat and prevent the surface seeds from drying and turning is done every 48 hours.

2.2.1.4 Drying platform fermentation

Fresh cocoa seeds are spread directly on drying platforms to allow for mild anaerobic fermentation while drying and an anaerobic fermentation when the seeds are heaped into piles each night. This type of fermentation is practiced in Ecuador and parts of Central America where the Criollo cocoa is usually grown (Lopez and Dimick,

1995). Though this type of fermentation is adequate for Criollo varieties it is insufficient for Forastero varieties that require longer fermentation times.

2.2.1.5 Basket fermentation

Baskets fermentation is mainly practiced in Nigeria, smallholder farmers in the Philippines and some parts of Ghana (Lopez and Dimick, 1995; Afoakwa, 2010). Small quantities of fresh cocoa beans are placed in baskets lined with plantain leaves with tiny perforations to allow for seepage of sweatings and the surface is covered with plantain leaves and the mass is weighed down. Fermentation usually spans 4 to 6 days and the beans are mixed regularly.

2.2.2 Microbiology of cocoa fermentation

Fermentation of cocoa is a spontaneous microbiological process. The undamaged and healthy beans are sterile but soon as they are removed, they become inoculated with a variety of microorganisms from the pod walls, the labourer‟s hands, the containers used for transporting the beans to the plant, the dried mucilage of the previous fermentation that coats the sweat-boxes, and by insects especially fruit fly (*Drosophila melanogaster*)(Lopez and Dimick, 1995; Thompson *et al.*, 2001; Nair, 2010; Nielsen *et al*., 2007; Nazaruddin, 2006; Afoakwa, 2010).

Fresh cocoa pulp contains sugars (mainly sucrose) and citric acid, which makes it an excellent medium for the growth of microorganisms. During the initial phase of fermentation the low pulp pH of about 3.4 – 4.0 (due to the relatively high content of citric acid), high sugar content(8 – 24%) and the low oxygen tension are most

(Lopez and Dimick, 1995) suitable for the growth of anaerobic yeasts, which initially dominate the fermentation during the first 24 to 36 hours (Thompson *et al.,* 2001) and it has been recorded that yeast population increases rapidly to 107-108 CFU/g and it is followed by a steady decline throughout the rest of the fermentation (Ardhana and Fleet, 2003; Nielsen *et al*., 2007).

Previous research conducted by Nielsen *et al.,* (2007) shown that Ghanaian cocoa pods have wide range of yeasts and it includes: *Hanseniaspora guilliermondii* (53%), *Pichia guilliermondii* (22%), *Candia intermedia* (7%), *Candida parapsilosis* (6%), *Cryptococcus laurentii* (4%), *Candida silvicola* (2%), *P. membranifaciens* (2%), *Rhodotorula glutinis* (2%) and *Cryptococcus humicola* (2%). Some strains of yeasts produce pectinases which break down the pulp cells so that the draining juices carry away flakes of pulp. However, primarily yeasts are known to produce ethanol from sugars, carbon dioxide and metabolizing citric acid.

This leads to a sharp increase in the ethanol concentration and a decrease in the concentration of fermentable sugars (Lopez and Dimick, 1995; Thompson *et al*, 2001; Ardhana and Fleet, 2003).

Fermentations investigated by Nielsen *et al.* (2007) revealed that approximately 80% of the sugars (glucose, fructose and sucrose) were metabolized within the first 24 hours but the researcher noted an exception where the center of the large heap fermentation were slower and this corroborated similar observations reported by Carr *et al.* (1979). The conversion of glucose and fructose to ethanol causes a moderate rise in temperature at the initial phases of fermentation due to its exothermic nature.

Thompson *et al.* (2001) noted that the decline in yeast population could be attributed to rapidly metabolizing sugars in the pulp to form carbon dioxide and ethanol. Secondly, the production of ethanol creates a toxic environment that represses yeast growth and lastly acetic acid, which is produced from ethanol by the acetic acid bacteria, is also toxic to yeasts (Thompson *et al.*, 2001; Nelson and Cox, 2004). Coupled with the above mentioned processes, the breakdown of pulp parenchyma cells culminates into pockets of gaps which encourages air flow this is corroborated by the research findings of Ardhana and Fleet (2003) that highly pectinolytic filamentous fungi flourished during first 36 hours of fermentation and loss of citric acid by drainage and through microbial metabolism causes a rise in pH these now favours the proliferation of lactic acid bacteria (Lopez and Dimick,
1995; Thompson *et al.*, 2001; Gálvez, 2007; Afoakwa, 2010).

Counts of lactic acid bacteria increases rapidly, but may be present for only a brief period, because they prefer a low oxygen concentration or high concentration of carbon dioxide (Thompson *et al.*, 2001). Their population has been observed to reach 106 to 107 CFU/g in a typical fermentation. The lactic acid bacteria population is usually at its peak for about 3 days then decreases to less than 10% of the total microflora (Thompson *et al.*, 2001). Both homo -fermentative and heterofermentative lactic acid bacteria are present in cocoa fermentations with the former being the majority. Some of the homo-fermentative species included *Lactobacillus plantarum, Lactobacillus casei, Lactobacillus delbrueckii, Lactobacillus acidophilus, Pediococcus cerevisiae, Pediococcus acidilactici,* and The heterofermentative species included *Leuconostoc mesenteroides* and

Lactobacillus brevis (Thompson *et al.*, 2001). In a recent study by Nielson *et al.* (2007), the following lactic acid bacteria were isolated in Ghanaian cocoa beans;*Lactobacillus plantarum, Leuconostoc pseudomesenteroides, Leuconostoc pseudoficulneum* and *Pediococcoccus acidilactici*. With increase in aeration the lactic acid bacteria decrease, which causes a rise in acetic acid bacteria.

Acetic acid bacteria reach around 108 CFU/g during Ghanaian heap fermentations according to Nelson and Cox (2004). Aeration of the fermenting mass by turning clearly influenced acetic acid bacteria growth in the large heap fermentation (Carr *et al.*, 1979). According to Nelson and Cox (2004), following turning of the heap the acetic acid bacteria counts decreased in the outer portions the heap followed by renewed acetic acid bacteria growth while the opposite was observed in the centre of the fermenting mass and this was corroborated by Carr *et al.* (1979) as it was observed that the growth of acetic acid bacteria in the center of the fermenting beans are positively correlated with turning. *Acetobacter syzygii, Acetobacter pasteurianus and Acetobacter tropicalis* were the predominant acetic acid bacteria isolated by Nielsen *et al.* (2007). The aerobic conditions within the fermenting mass make conditions favourable for acetic acid and other aerophilic spore forming bacteria and fungi (Thompson *et al.*, 2001). This results in exothermic oxidation of alcohol culminating in an increase in temperature to about 45 to 50°C.The rise in temperature makes it unfavorable for the survival of acetic acid bacteria (Lopez and Dimick, 1995; Thompson *et al.*, 2001).

These high temperatures coupled with the ethanol and acetic present serves as the final phase of fermentation as this phase is characterised by a high stress factor limiting the growth of many microorganisms (Lehrian and Patterson, 1983; Thompson *et al.,* 2001; Nielsen *et al.,* 2007; Gálvez, 2007; Afoakwa, 2010; Afoakwa *et al.,* 2011a). Also occurring simultaneously in the later phases of fermentation increasing pH and aeration becomes favourable for growth of filamentous fungi which are responsible for off flavours and increased free fatty acids levels and spore forming *Bacillus* spp. often reaching about 108 CFU/g in the later stages of the fermentation (Carr *et al*., 1979; Schwan *et al*., 1995; Ardhana and Fleet, 2003). However, investigations by Nielsen *et al.* (2007) showed that small heap and tray fermentations had no or limited growth of *Bacillus* spp. and these observations were because fermentations were completed after 4 rather than 6 days. *Bacillus* spp. has been shown to have high enzymatic activity (Schwan *et al.,* 1995). They are also responsible for the production of short chain fatty acids (< C14) such as butyric acid and others that produces off flavours, pyrazines and 2,3butanediol (Schieberle, 2000; Thompson *et al.,* 2001; Nielsen *et al.,* 2007; Quao,
2010).

2.2.3 Biochemical and chemical changes during fermentation

The development of chocolate flavour begins with the chemical and biochemical changes occurring within the bean during fermentation and drying. From previous researches, it is imperative to note that pulp preconditioning (pod storage, mechanical and enzymatic depulping and beans spreading), mode of fermentation

and its microbial conditions and drying provide the necessary conditions for complex biochemical reactions to occur (Hodge, 1953; Lopez and Dimick, 1995; Thompson *et al.*, 2001; Nazaruddin, 2006; Nielsen *et al.*, 2007; Afoakwa *et al.*, 2012 ab). However similar cannot be mentioned of immature and unfermented beans as they tend to develop little or no chocolate flavour when roasted while excessive fermentation yields hammy and putrid flavours which are not ideal (Zaibunnisa *et al*., 2000; Reineccius, 2006; Afoakwa, 2010).

Thompson *et al.* (2001) noted that although lactic and acetic acids produced externally by microbial activity affect flavour, chocolate flavour development is predominantly dependent on enzymatic formation of flavour harbingers within the cotyledon and it is highly peculiar to cocoa. These compounds include free amino acids, peptides, reducing sugars, and polyphenols (Kirchhoff *et al.* 1989; Hansen *et al*., 2000; Lee *et al.*, 2003; Hii *et al.*, 2009). The structure of the testa is semi permeable and acts as a natural barrier between microbial fermentation activities outside the bean and chemical reactions within the bean. But with a progression in fermentation there is a migration of ethanol, acetic acid, and water which are metabolites from fermentation from the outside to the inside of the bean (Thompson *et al.*, 2001).

Death of the cocoa bean is a very important process as there is breakdown of cellular integrity which leads to the free interaction of enzymes and substrates also, soluble bean components are usually lost by leaching via the testa and lost by drainage (Hansen *et al*., 1998; Thompson *et al.*, 2001; Afoakwa, 2010). These biochemical reactions responsible for flavour development occur within the cocoa bean on the

onset of bean death. Its long been known that rising temperatures and increasing acetic acid concentrations during fermentation cause seed death but Lehrian (1989) stipulated that the ethanol produced via anaerobic yeast growth phase correlates more with death of the seed. This usually occurs 24 hour after maximum concentrations of ethanol are attained within the cotyledon (Thompson *et al.*, 2001). This averts problems associated with germination, which utilizes a chunk of cocoa butter and other stored nutrients leading to a more stable and desirable product.

Afoakwa (2010) explained that enzymes exhibit different stabilities during fermentation and may be inactivated by heat, acids, polyphenols and proteases. The enzymes associated with cocoa fermentation includes; amino peptidase, cotyledon invertase, pulp invertase and polyphenol oxidase are significantly inactivated, carboxypeptidase is partly inactivated, whereas endoprotease and glycosidases remain active during fermentation (Hansen *et al*., 2000; Thompson *et al.*, 2001;

Afoakwa, 2010).

Research by Kirchhoff *et al.*, (1989) revealed that there is significant difference in composition of the free amino acids released during fermentation as compared with the composition in total seed hydrolysates (reflecting predominantly the proteinbound amino acids) is a striking feature of cocoa fermentation and this is in agreement with findings from other researchers (Forsyth and Quesnel, 1957;

Forsyth and Quesnel, 1963; Biehl *et al.*, 1985; Thompson *et al.*, 2001; Afoakwa, 2010).This is suggestive that there is a fine balance between fermentation times, microbial activity that influences enzyme activity within the cotyledon.

2.2.3.1 Enzymology of cocoa beans

The immediate surroundings of the fermenting mass forms a biocenosis which is affected by pH and temperature, in the fermenting mass that influence cocoa bean enzyme reactions. Enzymes are proteins with powerful catalytic activity, which work over a range of pH and have an optimum pH at which it is most active. Increasing temperatures also accelerates their activation energy (Baker *et al.*, 1994).

Additionally, enzymes have high specificity for both the compound to be converted (substrate specificity) and for the type of reaction to be catalysed (reaction specificity such as hydrolytic and enzymatic reactions in cocoa). Apart from the aforementioned specific factors; moisture is also necessary to allow for enzyme and substrate interaction (Baker *et al.*, 1994), but with the gradual loss of water during drying, enzyme activity is reduced and finally ceased at about 6 to 8% (Thompson *et al.*, 2001).

Temperature of the fermenting mass rises from an ambient of 25°C at the initial stages of fermentation to about 50°C by the 3rd day (Lopez and Dimick, 1995). Increase in temperature of more than 20°C may impact negatively or positively on enzyme activity resulting in fewer flavour precursors and poor chocolate flavour (Thompson *et al.*, 2001). Finally, if appropriate amounts of organic acids are not produced during fermentation, the pH of the cotyledon will not be suitable for

optimal enzyme activity, and the flavour profile of the resulting cocoa will be affected but excessive acid will lead to heightened sourness that can mask the chocolate flavour.

2.2.3.2 Hydrolytic enzyme reactions

During the anaerobic phase of fermentation hydrolytic enzymes namely invertase, glycosidases, and proteases have highest activity (Lopez and Dimick, 1995; Thompson et al., 2001; Afoakwa, 2010). The activity of invertase yields reducing monosaccharides (glucose and fructose) from sucrose that natively cannot partake in non-enzymatic browning reactions that occur during roasting to contribute to chocolate flavour. These reducing sugars represent more than 95% of the total reducing monosaccharides in cocoa beans (Forsyth and Quesnel, 1957; Hansen *et al*., 2000).

In addition to the minuscule amounts of amino acids existing in the unfermented bean, proteases (endo- and exo proteases) account for the hydrolysis of proteins to amino acids and peptides and their activity is dependent on pH and temperature. Proteins in cocoa are vicilin-like globular in nature and are the main target of the proteases. According to Kirchhoff *et al*. (1989) the types and ratio of free amino acids and peptides sequences are unique to cocoa. These amino acids and peptides participate in non-enzymatic browning reactions by forming complexes with reducing sugars during roasting to form important chocolate flavour precursors and also colour formation (Kirchhoff *et al*. 1989; Afoakwa *et al*., 2011a).

Glycosidase is a unique enzyme that hydrolyses anthocyanins to cyanidins and sugars (galactose and arabinose)and has more impact on colour development and some minor flavour components (Lee, 1975; Biehl *et al.*, 1985; Lopez and Dimick, 1995; Thompson *et al.*, 2001; Afoakwa, 2010). Anthocyanins located in specialized vacuoles within the cotyledon and are responsible for the characteristic deep purple colour of the unfermented bean. These compounds are highly affected by the pH of the medium. The colours range from purple in a neutral state, violet in weak alkaline solutions and pink in acidic conditions (Lee, 1975; Konczak and Zhang, 2004).

2.2.3.3 Oxidative enzyme reactions

Polyphenoloxidase (PPO) is the major oxidase occurring in the aerobic phase of fermentation but continuing well into the drying of cocoa beans and is responsible for some flavour modifications (Thompson *et al.*, 2001). Oxygen facilitates the activity of PPO however; rising temperatures and in sufficient moisture become inhibiting factors for the polyphenol oxidase enzyme during drying. Catechins of which epicatech makes up more than 90% and leucocyanidins are the major classes of polyphenols that is subject to oxidation in cocoa beans. Oxidation of epicatech during the aerobic phase of fermentation and drying is largely responsible for the characteristic brown colour of fermented cocoa beans.

The dihydroxy configuration of polyphenols are oxidized to form quinones which in turn can polymerize with other polyphenols or complex with amino acids and proteins to yield characteristic coloured compounds and high-molecular-weight

insoluble material (Thompson *et al.*, 2001) that result in the reduction in astringency and bitterness.

2.3.4 Factors affecting fermentation

The various methods of fermentation are themselves capable of great differences in detail and this reflects the large number of factors influencing the process.

2.3.4.1 Ripeness of pods

Where the harvesting rounds are done at intervals of three weeks or less, the pods should be at a fairly uniform state of ripeness but where the intervals are longer under ripe and overripe pods may be harvested (Wood and Lass, 1985)

Some trials of the effect of ripeness on fermentation have been reported but the degree of ripeness is not easy to define closely. In a trial conducted in Trinidad, (Wood and Lass, 1985) reported that wholly unripe pods did not ferment normally, the temperature remaining at 35^{O}C, after an initial rise to 40 ^{O}C. The loss of weight during fermentation and drying was far higher than normal so that the yield of dry beans was no more than 21 per cent of the wet weight. The bean size was also smaller at 1.05g compared with 1.34g for overripe beans. This indicates that the unripe pods used in this trial were not fully developed and presumably the pulp was deficient in sugar (Wood and Lass, 1985).

MacLean and Wickens (1951) initiated a similar trial in Ghana, with somewhat similar results. The degree of ripeness was not defined precisely in these experiments

but it seems that pods were very far from ripe, at a stage when the pulp is still firm and the beans have not separated from the pod wall.

In another trial, beans from Amelonado pods which were greenish yellow were fermented and no difference from normal was found either in the fermentation or in the dried beans (Howat *et al.,* 1957).

Conclusion from above trials is that beans from ripe pods and pods in the process of changing colour will ferment properly but beans from less ripe pods will not.

2.3.4.2 Pod diseases

Most pod diseases lead to complete loss of the beans they contain and even when the beans are not destroyed, it is undesirable to use the beans in fermentation. In the case of *Phytophthora* pod rot, the beans may not always be lost as the fungus attacked; the beans can be saved by regular harvesting. I however, the beans are attacked, it leads to a rise in free fatty acid and chocolate made from such beans will not have a normal chocolate flavour (MacLean, 1953).

2.3.4.3 Type of cocoa

There is a basic difference between Criollo and Forastero types in the way they are fermented; Criollo cocoa is fermented for a relatively short period of 2 – 3 days, while Forastero cocoa is fermented for 3 – 7 days, occasionally longer (Wood and Lass, 1985). As a result of this difference, mixed fermentation of the two types should be avoided. This can be arranged in places where Criollo and Forastero trees

are grown separately, but when the hybrid trees contain both white and purple beans they are impossible to segregate. Where this occurs, it is better to ferment for the appropriate period for Forastero cocoa.

2.3.4.4 Variations in pulp / bean ratio

It has recently been realized that fresh beans can vary considerably in the ratio of pulp to bean and in the amount of sugars per bean. These factors vary with the type of tree and also the growing conditions. Data collected in Ghana have the following comparisons (Table below).

Table 2.1: ratio of pulp to bean and in the amount of sugars per bean

	Amazon	Amelonado
Purple / bean ratio (g)	1.53 (12)	0.93 (23)
Sugar / bean (mg)	243 (12)	137 (15)

(The figures in brackets indicate the number of samples analysed)

The differences are significant, but when converted to concentrations the differences is less marked

Analyses carried out in Malaysia showed levels similar to the Amazon figure above but there was less difference between cultivars. Therefore growing conditions also affect the purple / bean ratio and in many countries it is known that the beans are wetter during the wet season and that this in turn affects fermentation.

These differences influence the course of fermentation by affecting aeration and the amount of acetic and lactic acids formed. Beans with more pulp will restrict gas exchange and make the mass of beans more anaerobic and the greater amount of sugars may lead to larger amounts of acids in the cotyledons at the end of fermentation (Carr *et al.,* 1979).

2.3.4.5 Climatic and seasonal differences

As mentioned above, there are seasonal differences in the amount of pulp surrounding the beans. In West Africa, the main crop starts towards the end of the wet season and, as the crop proceeds, the amount of pulp decreases as is revealed by an increasing recovery of dry beans from wet (Wood and Lass, 1985).

Records gathered in the West Indies showed a recovery of about 34 per cent in the wet season and about 38 per cent in the dry season. On an estate in Cameroon where tray fermentation was used, it was found necessary to hold the wet beans harvested in the cooler wet season for six hours in a box to allow more time for sweatings to run off before loading the trays (Wood, 1972).

Apart from seasonal changes, there are countries such as Uganda where ambient temperatures show considerable diurnal variation. This can lead to low temperatures in fermentation so that the fermentary has to be protected from wind and more insulation given to the fermenting beans (Couprie, 1968).

2.3.4.6 Storage of pods

A delay between harvesting and opening pods has been shown to give a more rapid rise of temperature. Several authors have reported this from Trinidad and West Africa (MacLean and Wickens, 1951). More recently the effect of storing pods has been examined in detail (Anon, 1981).

Box fermentation of 500 kg wet beans one with beans from freshly opened pods compared with others. The effect of the delay was to speed up fermentation by about 24hours, as seen by a more rapid rise in temperature. During the delay in opening, there is a loss of moisture which reduced the amount of sweatings by 50 per cent and allowed better aeration at the beginning of fermentation (Wood and Lass, 1985).

In Ghana, farmers harvest for several days before opening in order to gather a sufficient quantity but effect must be to speed up the fermentation and a similar practice in Trinidad has been mentioned.

In Papua New Guinea, farmers have been recommended to delay opening pods for three or four days after harvest for the same reason (Bridgland, 1959).

2.3.4.7 Quantity of cocoa

The heat generated during fermentation is retained by insulation but this becomes more difficult to achieve with small quantities of beans as their surface area is great in relation to their mass. There is therefore a minimum quantity of beans which will ferment satisfactorily (Wood and Lass, 1985).

Various opinions have been given as to this minimum quantity. Rohan (1963) found that heaps containing 70 kg wet beans can be forward. As a rough guide, the weight of wet cocoa should not be less than 90kg when the traditional box or heap methods are used. Small quantities are liable to be more affected by changes in outside temperature and require better insulation.

The maximum quantity that can be fermented will depend on the method employed. In box fermentations, aeration is reduced as the depth increases. In Trinidad, this depth of beans is usually up to 75cm and this figure has been quoted as the minimum desirable depth.

Box fermentation at depths of 42, 68 and 83 cm have been studied in detail These showed greater variation in oxygen profiles with depth during the first two days, but little difference thereafter and there were no clear differences in the flavour of the final product. There were however, differences in the level of acetic acid which was greater in the deeper fermentations (Anon, 1981).

2.3.4.5 Duration

An enquiry conducted into methods of fermentation throughout cocoa – growing countries revealed a wide range of duration of fermentation from 1.5 days up to 10 days (Forsyth and Quesnel, 1957). The major differences lies in the variety of cocoa fermented, which has already been referred to. The enquiry showed that Criollo beans are fermented for 2 – 3 days and Forastero beans for 6 – 8 days, though some

countries which formerly grew Criollo continued to use methods applicable to that type despite a change to Forastero planting material (Wood and

Lass, 1985).

The length of fermentation is also influenced by the method adopted. Box fermentation is normally for six days or more accurately, 144 hours but may be extended to eight days in some countries. Heap fermentation is supposed to last for six days but most farmers in Ghana ferment for a shorter period of 3 – 5 days. Strangely enough there is no reliable information on farmers" methods nor is it clear as to whether the shorter period produces beans of the same quality as six days fermentation. The duration of tray fermentation is only four days.

Under – fermentation will produce beans with more purple pigment, and greater bitterness and astringency will be expected in the final product. Over fermentation will produce beans with a dull dark – coloured nib and little chocolate flavour. Externally, over-fermented beans are much darker than normal and the onset of the putrefactive changes associated with over – fermentation can be spotted by the unpleasant smell accompanying them.

Hammond (1953) reported that some farmers in Ghana held the view that in wet weather, the period of fermentation should be less, and it is also believed that the length of fermentation should be varied according to the time lag between harvesting and breaking the pods. According to Aneani and Asamoah (2004), fermentation duration in Ghana is five and half days on average with a range of four and seven days, which depend on weather and condition of harvested pods. Inadequate sunshine

during the rainy season prompts four days fermentation whilst some farmers extend their fermentation period to even seven days during the dry season. Beans derived from fresh harvested pods are fermented for six to seven days whereas those from exclusively ripe pods go for four days (Aneani and Asamoah, 2004).

2.3.4.9 Turning

The purpose of turning the beans during fermentation is to ensure uniformity. Inevitably, there are differences between one part of the fermenting mass and another so that turning is important to even out these differences. In box fermentation, the wet beans will settle down into a solid mass during the first day, while the sweatings are draining off. Turning this mass of beans is necessary in order to allow air to penetrate (Wood and Lass, 1985).

There are many variations in the frequency of turning, from no turning at all to turning once or twice a day with fermentation in barrels which has been tried in Ivory Coast and Cameroon. The commonest practices are turning everyday or every other day, while some planters turn after one day and then every other day (Wood and Lass, 1985).

Recent research on the problem on acidity has revealed more facts about fermentation which may affect the methods used particularly with regard to the depth of beans in a box, the duration of fermentation and the turning schedule

(Wood and Lass, 1985).

2.4 MATERIALS USED FOR FERMENTATION

Sébastien *et al.,* 2013 indicated that materials used include banana leaves (*Musa paradisiaca* L.), which are the traditional fermentation material commonly used by small-scale cocoa farmers. A heap of cocoa beans was piled on banana leaves spread on the ground and covered by other banana leaves. The second lot of fresh beans was fermented in a wooden box (60 × 60 × 60 cm) with a perforation in the bottom to facilitate pulp drainage.

Others use black tarpaulin (polythene paper), which is spread on sloping ground and covered with a portion of the same black tarpaulin sheet. Black tarpaulin is the most commonly used cocoa fermentation material in Côte d"Ivoire. Black tarpaulin is increasingly used because of its many advantages: low cost, ease of use, and reusability (Sébastien *et al.,* 2013).

Wood and Lass (1985) reported that the bottom and sides of the box or basket should be covered with banana leaves; however, banana leaves on the bottom should be not too thick and should be also perforated by a knife to make sure that the liquid from the cocoa pulp will be well drained. Insufficient drainage of pulp will result in a bad fermentation. The top layer of fermenting cocoa should also be covered with banana leaves or jute bags. This inhibits too much air penetration into the fermenting cocoa also stops too much moisture from being lost. If too much moisture is lost, the cocoa

will not ferment properly. An additional reason for lining the baskets and boxes and covering the fermenting cocoa is the problem of losing heat by dissipation during the fermentation. This would cause the cocoa to not attain the high temperatures required for a good fermentation. Therefore it is very important to cover cocoa beans during fermentation. Jute bags conserve heat better than banana leaves, so jute bags or a combination of banana leaves and jute bags are recommended.

2.5 QUALITY INDICES OF FERMENTED AND DRIED COCOA BEANS

Generally to develop good quality cocoa products such as chocolates, cocoa butter, cocoa powder and other confectionery cocoa must be free from mould, well fermented, must be low in free fatty acids and less bitterness and astringency and also less acidic. Quality appraisals also rely heavily on the experience of farmers and may lead to a good deal of variation in quality. Since fermentation is a critical step in the development of flavour, it is essential that farmers be trained on the principles of fermentation. The quality of cocoa can be accessed via a number of quantitative and qualitative tests. The most important and easy to assess include cut test to ascertain to some extent the fermentative quality, free fatty acids to assess the level of lipolysis (Thompson *et al.*, 2001).

2.5.1 Cut-test

The cut test is simple, and still the most widely used method is to assess the quality of a random sample of beans from a batch by visual evaluation of the cut surfaces of cocoa beans. Although to some extent the cut test reveals certain defects that are likely to cause off- flavours and degree of fermentation it is extremely subjective and

at best is limited to the measurement of bean defects (Wood and Lass, 1985; Lopez and Dimick, 1995). Results from a cut test assumes that beans showing the least amount of the prescribed defects are in good standing and can produce good chocolate flavour. A standard of 300 beans sampled randomly longitudinally sectioned to expose the surface of the cotyledons, observations are usually made during the day with the aid of the sunlight and sometimes under bright visible light.

The defects most commonly looked for are:

a. Slaty beans – occurrence is due to dried beans without fermentation and it is characterised by a greyish to ash colour. Slaty beans are beans in which more than 50% of the cotyledon is grey or slaty in colour (Fowler, 2009), and have rubbery cotyledon and resistance to cutting. These beans have not undergone fermentation and they have a low level of cocoa flavour with high levels of astringency.
b. Purple beans - a combination of the intensity and occurrence gives an indication of the degree of under fermentation. Purple beans occur when the fermentation has been terminated prematurely (Guehi *et al.,* 2010).
c. Mouldy beans - indicator of poor quality of the product usually;
d. Germinated beans – presence suggest under fermentation and high moisture content. Germinated beans are those where the seed has started to grow before being killed during the fermentation or drying process and the shell has been pierced by the growth of the first root (Lopez and Dimick, 1995).

In the dry germinated bean, the root usually drops out, leaving a hole, which makes the bean more easily attacked by insects and moulds.

e. Flat beans are those which have begun to form, but have not developed or filled out. There is no useful cotyledon in them so they simply add to the shell content, which is waste.

f. Other physical defects such as flat beans, insect infestation and broken beans relate more to the yield rather than flavor quality (Wood and Lass, 1985; Lopez and Dimick, 1995; Thompson *et al.*, 2001). Insect-damaged beans are those which have been penetrated by insects that feed on the cotyledon. These should not be present. Any number will involve loss of material and a risk of contamination with fragments of the insect.

The cut test is not a guarantee of good flavour, and attempts have been made to development a more precise means of flavour or quality determinations. Defective beans are the sum of germinated beans, infested beans and flat beans whiles fully brown beans are well-fermented beans. Results are expressed as a percentage. A batch of cocoa beans with more than 60% fully brown colour beans is considered as good-quality product (Guehi *et al.*, 2010).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 INTRODUCTION

The studies were conducted to evaluate effect of fermentation materials on cocoa bean quality. and comprised two components mainly field survey and laboratory experiment.

3.2 DESCRIPTION OF STUDY AREA

The study was conducted at Samreboi cocoa district which is part of Wassa Amenfi West district of Western Region of Ghana. It is located between Latitude 400‟N and 500 40‟N and Longitudes 10 45‟ W and 20 10‟W.

Average annual rainfall tapers off from 173 mm in the south to 140 mm in the north. The district experiences bimodal rainy season i.e March to July and September to early December. Two dry spells separate the seasons. Temperatures are generally high ranging from 24 degrees Celsius to 29 degrees Celsius.

Maximum temperatures are recorded in March and Minimum in August.

3.3 FIELD SURVEY

3.3.1 Objective

The main objective of the field survey was to identify fermentation practices among farmers particularly fermentation material(s) used during fermentation.

3.3.2 Questionnaire survey

A validated pre-tested questionnaire (Appendix I) was administered to cocoa farmers. The questionnaire for farmers was made up of two sections mainly background information of respondents and fermentation practices.

Simple random sampling technique was used since the exact number of farmers in the districts could not be quantified. Sampling was done in each cocoa district until the 100 farmers were obtained and interviewed. In all, 300 farmers were selected and interviewed in the 3 cocoa districts.

3.4 LABORATORY STUDIES

3.4.1 Experimental Design

A 2 x 3 factorial arrangement in a completely randomized design was used. There were two factors. Factor „A" was varieties with two levels (hybrid and mixed) and factor „B" was fermentation materials with three levels (i.e. plantain leaves, fertilizer sack and polythene). There were six treatments combinations and the experiment was replicated three times.

3.4.2 Experimental Procedure

Cocoa pods (hybrid and mixed) of uniform ripeness were harvested in November 2014 (major season). The harvesting was done by traditional methods (under ambient temperature during the day, 28-30 °C) and the pods transported to the fermentation station where they were stored for 3 days to ensure pulp preconditioning. Pods were

broken using wooden billet which involved one or two sharp blows with the edge of the billet.

About 30 kg of extracted cocoa beans were fermented using the heap fermentation method. It was divided into 3 samples for the three experimental materials to be used (plantain leaves, fertilizer sacks and polyethylene sheets). Fermentation lasted for 6 days with consecutive opening and turning every 48 hours.

Sun drying technique was employed to dry the fermented beans on raised platforms with a wooden mat. Cocoa beans from each material were dried until they reached moisture content below 7% with the help of aqua boy device. The beans were mixed thoroughly to ensure uniformity. Cocoa beans from each fermentation materials were dried separately and sampled for cut test and proximate analysis.

3.5 DATA COLLECTION

3.5.1 Cut Test

The cut test is a visual assessment quality characteristic of cocoa beans. The procedure involved filling three equal sized white calico clothed sampling bags ($5.7dm^3$) with well-mixed beans. The mixed beans were quartered leaving a heap of slightly more than 300 beans, which were used to fill the sampling bags. Each sampling bag thus contained slightly hundred beans and was cut lengthwise through the middle to expose the internal surface of the two cotyledons.

The cut beans were examined in good daylight and the percentage total purple (deep, pale and partly brown/partly purple) beans were determined and recorded.

Percentages of other defective categories (mouldy, slaty, insect infested, flat and germinated beans) were also determined and recorded.

3.5.2 Chemical composition (proximate analysis) of dried cocoa beans Cocoa beans were milled into powder with ceramic mortar and pestle after which the milled beans were defatted. The powders obtained were directly used for proximate analysis by the methods outlined by AOAC (2005) and Pousga *et al.* (2007). The Atomic Absorption Spectrophotometer (AAS) was used to determine the mineral content of the by-product. All the laboratory analysis was carried out at the KNUST Biochemistry Department.

3.5.2.1 Determination of moisture content

The determination was based on the moisture evaporation method used by Pousga *et al.* (2007). In this method, aluminium dishes were washed dried in oven and desiccators for cooling. The weight of each dish was taken. Hundred grammes (100 g) of each sample of cocoa powder were weighed into a sterile aluminium dish; weight of the dish and weight of sun dried sample (in triplicate) were taken. This was transferred into an oven and set at 100ºC and less than 100 mm Hg for approximately 5 hours after which the dish was removed from the oven, covered, cooled in a desiccator and weighed. Then the weight was measured using a measuring scale balance. It was transferred back into the oven for another one hour and then reweighed. The process continued until a constant weight was obtained.

The difference in weight between the initial weight and the constant weight gained was taken as the moisture content. The loss in weight multiplied by 100 over the original weight is the percentage moisture content.

The formula used is presented as follows:

$$\text{Moisture content (g/100g)} = \frac{\text{Lost in weight } (W_2 - W_1)}{\text{Original weight of sample } (W_2 - W_1)} \times 100\%$$

Where W_1= initial weight of empty crucible, W_2 = weight of crucible + SNC before drying, W_3 = final weight of crucible + SNC after drying.

% Total solid (Dry matter) (%) = 100 - moisture (%).

3.5.2.2 Ash content

The ash content represents the minerals component of the sample after all moisture has been removed as well as the organic material. It was determined according to Pousga *et al.* (2007). The method was based on the decomposition of all organic matter such that the mineral elements would not be lost in the process. Approximately 1g of finely ground sample was weighed into porcelain crucible which had been ignited. The crucible was placed in a muffled furnace and heated at 500ºC for four hours, removed and cooled. The ignited residue was moistened with 2 ml distilled water and slowly and carefully 5 ml of 8 N HCl (two parts of conc. HCl was mixed with one part of water). It was transferred again into the cool muffle furnace and the temperature was increased stepwise to 550 ± 5^{O}C. The temperature was maintained for 8 hours until white ash was obtained. It was then brought out and allowed to cool in a desiccator and weighed again. Percentage weight was calculated

as weight of ash multiplied by 100 and divided by the original weight of samples used.

The formula used is presented as:

$$\text{Ash content (\%)} = \frac{\text{Weight of ash}}{\text{Weight of original sample}} \times 100\%$$

3.5.2.3 Crude fibre

Crude fibre content was determined according to methods adopted by Pousga *et al.* (2007). Twenty grams (20 g) of each ground cocoa beans powder samples were defatted separately with diethyl ether for 8 hours and boiled under reflux for exactly 30 min with 200 ml of 1.25% H_2SO_4. It was then filtered through cheese cloth on a fluted funnel. This was later washed with boiling water to completely remove the acid. The residue was then boiled in a round bottomed flask with 200 ml of 1.25% sodium hydroxide (NaOH) for another 30 min and filtered through previously weighed couch crucible. The crucible was then dried with samples in an oven at 100°C, left to cool in a desiccator and later weighed. This was later incinerated in a muffle furnace at 600°C for 2 to 3 hours and later allowed to cool in a desiccator and weighed. The formula used is presented as:

$$\text{Fibre content (\%)} = \frac{\text{Weight of fibre}}{\text{Weight of original sample}} \times 100\%$$

3.5.2.4 Crude Protein determination

Total protein was determined by the Kjedahl method as modified by Pousga *et al,* (2007). The analysis of protein content in a compound by Kjedahl method is based upon the determination of the amount of reduced nitrogen present. Thirty grams

(30g) of each sample was weighed into a filter paper and put into a Kjedahl flask, 10 tablets of Na2SO4 were added with 1 g of $CuSO_4$ respectively. Twenty millilitres (20 ml) of concentrated H_2SO_4 were added and then digested in a fume cupboard until the solution became colourless. It was cooled overnight and transferred into a 500 ml flat bottom flask with 200 ml of distilled water. This was then cooled with the aid of packs of ice block. About 60 to 70 ml of 40% of NaOH were poured into the conical flask which was used as the receiver with 50 ml of 4% boric acid using methyl red as indicator. The ammonia gas was then distilled into the receiver until the whole gas evaporated. Titration was done in the receiver with 0.1 N H_2SO_4 until the solution became colourless. The formula used was presented as:

$$\text{Protein content (\%)} = \frac{Vs - Vb \times 0.01401 \times N \text{ acid } (6.25)}{\text{Weight of original sample used}} \times 100\%$$

Where Vs = Volume (ml) of acid required to titrate sample; Vb = Volume (ml) of acid required to titrate blank; N acid = normality of acid.

3.5.2.5 Determination of pH

The pH determination was in accordance with the procedures of the Office International du Cacao et du Chocolat (OICC) (1972). 10 g of ground cocoa beans was extracted with 90 ml boiling deionized water. The sample was extracted for 10 min, cooled to 25°C, and the pH was determined using a Mettle-Toledo pH meter.

3.5.2.6 Determination of Free fatty acid

Free fatty acid content was determined using titration method (ISC 1998). Fat obtained from extraction is dissolved in warm ethanol and then titrated using alkali

solution (NaOH 0.1N). Free fatty acid was calculated and expressed as the percentage of mass per mass using the following formula:

$$\% \text{ free fatty acid content} = \frac{V \times N}{M} \times \frac{100}{100 - mc}$$

V = the volume, in ml, of NaOH

N = the normality of NaOH solution

M = the mass, in grams, of cocoa bean lipid and Mc = moisture content

3.5.2.7 Determination of fat content

The method employed was the Soxhlet extraction technique adopted by Pousga *et al.* (2007). Twenty grams (20g) of the samples were weighed and carefully placed inside a fat free thimble. This was covered with cotton wool to avoid loss of the sample. The loaded thimble was put in the Soxhlet extractor and about 200 ml of petroleum ether poured into a weighted fat free soxhlet flask with the flask attached to the extractor. The flask was placed on a heating mantle such that the petroleum ether in the flask refluxed. Cooling was achieved by a running tap connected to the extractor for at least 6 hours after which the solvent was completely siphoned into the flask. Rotary vacuum evaporator was used to evaporate the solvent leaving behind the extracted lipids in the soxhlet. The flask was removed from the evaporator and dried to a constant weight in the oven at 60°C. The flask was then cooled in a desiccator and weighed. Each determination was done in triplicate.

The amount of fat extracted was calculated by the formula presented below:

$$\text{Ether Extract (EE)}\% = \frac{\text{Weight of extracted lipids(g)}}{} \times 100\%$$

Weight of dry sample (g)

3.5.2.8 Nitrogen- free Extracts (NFEs)

Nitrogen-free extracts (NFEs) represents the soluble carbohydrate of a feed, such as starch and sugars. This is determined simply by subtracting the average of each of the other components (per cent crude protein, crude fat, crude fibre, moisture and ash) from 100 (Crampton *et al*., 1969).

3.6 DATA ANALYSIS

The data gathered from the field survey were coded and analysed using descriptive statistics of Statistical Product and Service Solutions (SPSS) software (version 19.0). The descriptive statistics used were frequencies, percentages and means.

The data recorded from cut test and proximate analyses were subjected to Analysis of Variance (ANOVA) using GenStat statistical package 9th edition. Treatment means were compared using the least significant difference (Lsd) at 5 % level of probability. Square root transformation was used to transform count data where appropriate before the analysis.

CHAPTER FOUR

4.0 RESULTS

4.1 FIELD SURVEY

4.1.1 Demographic characteristics of the farmers

Majority of the respondents (cocoa farmers) in the three cocoa growing districts were males whilst the minorities were females (Table 4.1). With regards to age distribution among the farmers, Samreboi cocoa district had 36% of the farmers aged 40 – 49 years followed by 50 or above years with 30%, 30 – 39 years with 23% and 18 – 29 years with 11% of the farmers. In Asankrangwa cocoa district, 42% of the farmers were in the age group of 30 – 39 years whereas 27%, 23% and 8% were aged 40 – 49, 50 or more and 18 – 29 years, respectively, Enchi cocoa district had age distribution in order of 50 or more years (32.3%) > 40 – 49 years (32%) > 30 – 39 years (21%) > 18 – 29 years (2%) (Table 4.1).

Averagely, 38.3% of the farmers had no formal education, 25.3% of them had the Middle School Leavers Certificate, 20.3% reported to have schooled up to Junior Secondary School, 7.8% had primary education, 7% had Senior Secondary School education and the remaining 1.8% of the farmers had Tertiary education. Averagely, 79% of the farmers in the three cocoa growing districts were married, 8.7% were divorced, 6.7% were singles and 5.6% were widows/widowers. Majority (85%) of the farmers who participated in this owned a farm whilst the minority were not the owners (Table 4.1).

Table 4.1: Background characteristics of farmers

	Cocoa District			
Variables	Samreboi	Asankrangwa	Enchi	Total

	Freq.	%	Freq.	%	Freq.	%	Freq.	%
Sex of farmers								
Male	66	66.0	78	78.0	72	72.0	216	72.0
Female	34	34.0	22	22.0	28	28.0	84	28.0
Total	100	100.0	100	100.0	100	100.0	300	100.0
Age of farmers(years)								
18 -29	11	11.0	8	8.0	2	2.0	21	7.0
30 -39	23	23.0	42	42.0	21	21.0	86	28.7
40 – 49	36	36.0	27	27.0	33	33.0	96	32.0
50 or more	30	30.0	23	23.0	44	44.0	97	32.3
Total	100	100.0	100	100.0	100	100.0	300	100.0
Educational level								
No formal education	39	39.0	26	26.0	50	50.0	115	38.3
Primary	13	13.0	9	9.0	1	1.0	23	7.8
MSLC	21	21.0	28	28.0	27	27.0	76	25.3
JSS	19	19.0	22	22.0	20	20.0	61	20.3
SSS	8	8.0	11	11.0	2	2.0	21	7.0
Tertiary	-	-	4	4.0	-	-	4	1.3
Total	100	100.0	100	100.0	100	100.0	300	100.0
Marital status								
Married	80	80.0	83	83.0	74	74.0	237	79.0
Single	5	5.0	11	11.0	4	4.0	20	6.7
Divorced	11	11.0	2	2.0	13	13.0	26	8.7
Widow / Widower	4	4.0	4	4.0	9	9.0	17	5.6
Total	100	100.0	100	100.0	100	100.0	300	100.0
Ownership of farm(s) Yes	91	91.0	86	86.0	96	96.0	273	91.0
No	9	9.0	14	14.0	4	4.0	27	9.0
Total	100	100.0	100	100.0	100	100.0	300	100.0

4.1.2 Postharvest practices of farmers

Eighty three per cent of the farmers in Samreboi cocoa district delayed breaking cocoa pods after harvest whilst 17% of the farmers did not. With regards to days between harvest and pod breaking, 32.5 %, 22.9%, 15.7% and 13.3% of farmers waited for 5days, 3 days and 6 days, respectively. The remaining 9.6%, 3.6% and 2.4% of the farmers waited for 7 days, 2 days and 1 day (Table 4.2).

In Asankrangwa cocoa district, 68% of the farmers delayed pod breaking after harvest whereas 32% did not delay. Among those who delayed pod breaking, 29.5%, 23.5%, 20.5%, 10.3%, 7.4%, 5.9% and 2.9% waited for 4, 3, 5, 2, 7, more than 7 and 6 days, respectively (Table 4.2).

In Enchi cocoa district, 96% of the farmers practiced delay in pod breaking and only 4% of the farmers do not practice delay in pod breaking. 48.4% of the farmers used 5 days, 22% of the farmers used 6 days and 20% of the farmers used 7 days. 7.4%, 1.1% and 1.1% of the farmers used 4, 3 and 2 days respectively (Table 4.2).

Table 4.2: Practice of delay in pod breaking after harvesting of pod by the farmers

(Those values in parenthesis are their respective percentages)

	Interval between harvesting and pod breaking								
Cocoa district	day	days	days	days	days	days	days	> 7 days	Total
Samreboi	(2.4)	(3.6)	(15.7)	(22.9)	(32.5)	(13.3)	8(9.6)	-	83
Asankrangwa	-	(10.3)	(23.5)	(29.5)	(20.5)	(2.9)	(7.4)	(5.9)	68
Enchi	-	(1.1)	(1.1)	(7.4)	(48.4)	(22.0)	(20)	-	95
Total	2	11	30	46	87	34	32	4	246
Percentage	0.8	4.5	12.2	18.7	35.4	13.8	13.0	1.6	100

Table 4.3: Reasons farmers practiced delay in harvesting and pod breaking

Reasons	Cocoa District							
	Samreboi		Asankrangwa		Enchi		Total	
	Freq.	%	Freq.	%	Freq.	%	Freq.	%
Inadequate hands for pod gathering and breaking	46	55.4	32	47.1	50	52.6	128	52.0
Improves fermentation	27	32.5	21	30.9	33	34.7	81	32.9
Improves fermentation and drying of beans	10	12.1	15	22.0	12	12.7	37	15.1
Total	83	100.0	68	100.0	95	100.0	246	100.0

Among the reasons why farmers practice delayed in pod breaking after harvest were inadequate hands for pod gathering and breaking (52%), improves fermentation (32.9%) and improves fermentation and drying of beans (15.1%) (Table 4.3).

Seventy two percent (72%) of the farmers in the Samreboi cocoa district used plantain/banana leaves in fermenting their cocoa beans whilst 16% of them used fertilizer sacks, 8% used polythene sheets and 4% used basket (Figure 4.1). In the Asankrangwa cocoa district, 60%, 28% and 12% of the farmers used plantain/banana leaves, fertilizer sacks and polythene sheets respectively, in fermenting their cocoa beans (Figure 4.1). Similarly, in the Enchi cocoa district, 42% of the farmers used plantain/banana leaves in fermenting their cocoa beans, 33% used polythene sheets and 25% used fertilizer sacks (Figure 4.1).

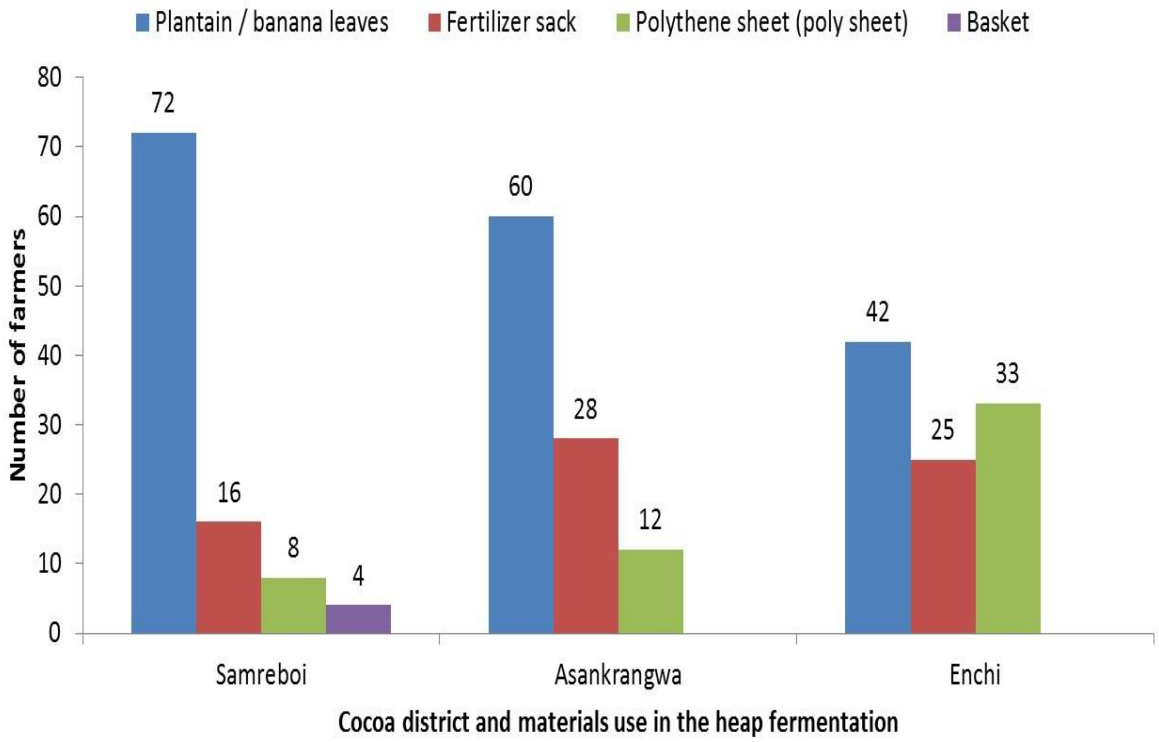

Figure 4.1: Cocoa district and materials use in the heap fermentation by farmers

Majority of the farmers used plantain /banana leaves because they were readily available and easy to handle. Those who used basket said it was more durable. Fertilizer sack was used because they were more durable and easy to handle. Polythene sheet was used because it was easy to obtain, more durable and easy to handle.

Table 4.4: Fermentation materials and reasons for their use by farmers

Fermentation materials	Reasons for use Easy to come by	More durable	Easy to handle	Total
Plantain /banana leaves	(65)(47)(39)	(0)(0)(0)	(7)(13)(3)	(72)(60)(42)
Baskets	(0)(0)(0)	(4)(0)(0)	(0)(0)(0)	(4)(0)(0)

Fertilizer sack	(0)(0)(0)	(16)(26)(20)	(0)(2)(5)	(16)(28)(25)
Polythene sheet	(1)(2)(9)	(6)(8)(19)	(1)(2)(5)	(8)(12)(33)
Total	(66)(49)(48)	(26)(34)(39)	(8)(17)(13)	(100)(100)(100)

(Figures in a cell followed in order of Samreboi, Asankrangwa and Enchi cocoa districts, respectively)

With regards to number of days for fermentation, 64%, 22%, 6%, 2% and 1% fermented their cocoa for 6 days, 7days and the remaining 5 days, 4 days and 3 days, respectively (Table 4.5).

In Asankrangwa cocoa district, 60%, 31%, 7% and 2% fermented their cocoa beans for 4 days, respectively (Table 4.5).

Forty nine percent (49%), 45% and 5% fermented their cocoa for 7 days,6 days,5days and 3 days, respectively (Table 4.5).

Table 4.5: Number of days farmers use for fermentation of cocoa beans

	Days for fermentation					
Cocoa district	3 days	4 days	5 days	6 days	7 days	Total
Samreboi	1	2	6	64	27	100
Asankrangwa	-	2	7	60	31	100
Enchi	1	-	5	45	49	100

Total	2	4	18	169	107	300
Percentage	0.7	1.3	6.0	56.3	35.7	100

Source: Field data, 2015

Table 4.6 presents the results of the Pearson Chi-Square test of association between the delay in pod breaking after harvest and number of days for fermentation in Samreboi cocoa district. The Chi-Square value of 44.5 with p-value of 0.01 indicates that there exists a significant association between the delay in pod breaking after harvest and number of days for fermentation in Samreboi cocoa district.

Table 4.6: Influence of interval between harvest and pod breaking on number of days for fermentation at Samreboi District

Interval between harvest and pod breaking	Number of days for fermentation						Chi- square test
	3 days	4 days	5 days	6 days	7 days	Total	
1 day	0	0	0	1	1	2	$\chi^2 = 44.5$
2 days	0	0	2	1	0	3	
3 days	0	1	0	10	2	13	p - value = 0.01
4 days	0	0	0	13	5	18	

5 days	1	1	1	19	5	27
6 days	0	0	0	8	3	11
7 days	0	0	0	5	4	9
Total	1	2	3	57	20	83

Table 4.7 gives the results of the Pearson Chi-Square test of association between the delay in pod breaking after harvest and number of days for fermentation among farmers in Asankrangwa cocoa district. The Chi-Square value of 10.8 with p-value of 0.54 indicates that there is no association between the delay in pod breaking after harvest and number of days for fermentation in Asankrangwa cocoa district.

Table 4.7: Influence of interval between harvest and pod breaking on number of days for fermentation at Asankrangwa District

Interval between harvest and pod breaking	Number of days for fermentation			Total	Chi- square test
	5 days	6 days	7 days		
2 days	1	4	2	7	$\chi^2 = 10.8$
3 days	0	12	4	16	

4 days	3	10	6	19	p - value = 0.54
5 days	1	9	5	15	
6 days	0	1	1	2	
7 days	2	3	0	5	
More than 7 days	0	2	2	4	
Total	7	41	20	68	

Table 4.8 shows the results of the Pearson Chi-Square test of association between the delay in pod breaking after harvest and number of days for fermentation in Enchi cocoa district. The Chi-Square value of 37.3 with p-value of <0.01 indicates that there exists an association between the delay in pod breaking after harvest and number of days for fermentation in Enchi cocoa district.

Table 4.8: Influence of interval between harvest and pod breaking on number of days for fermentation at Enchi District

Interval between harvest and pod breaking	nber of day fermentatic			Total	Chi- square test
	3 days	5 days	6 days	7 days	

2 days	0	0	0	1	1	$\chi 2 = 37.3$
3 days	0	1	0	0	1	
4 days	0	0	6	1	7	
5 days	0	1	22	23	46	p - value = < 0.01
6 days	1	0	10	10	21	
7 days	0	2	5	12	19	
Total	1	4	43	47	95	

4.2 EFFECT OF COCOA VARIETY AND FERMENTATION MATERIALS ON THE PHYSICAL QUALITY OF COCOA BEANS (CUT TEST)

4.2.1 Per cent mouldiness of beans

There was significant effect of cocoa variety and fermentation material interaction on percent mouldiness in the cocoa beans (Table 4.9). Hybrid cocoa variety with the various fermentation materials produced no mould. In contrast, the mixed cocoa varieties with plantain leaves, fertilizer sack and polythene sheet produced 1.83 %, 2.23 % and 1.80 % mouldiness which were similar but significantly different from those of the hybrid combinations. Cocoa bean mouldiness was also significantly affected by the cocoa variety. The mixed variety had the highest per cent mouldiness (1.96 %), significantly greater than that in the hybrid which recorded no mould in the beans (Table 4.9). The fermentation materials did not influence per cent mould in the beans.

Table 4.9: Effect of cocoa variety and fermentation materials on the per cent mouldiness of dried cocoa beans

	Per cent mouldiness
	Fermentation Material (FM)

Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	0.00	0.00	0.00	0.00
Mixed	1.83	2.23	1.80	1.96
Mean	0.92	1.12	0.90	
Lsd (0.05)	V = 0.62	FM = 0.76	V × FM = 1.08	
CV (%) = 6.81				

4.2.2 Per cent purple beans

The combined effect of cocoa variety and fermentation materials significantly ($p<0.05$) affected the percent purple beans (Table 4.10). Significantly higher percentage of purple beans were recorded by both hybrid and mixed varieties fermented with either fertilizer sack or polythene sheet as compared to both hybrid and mixed varieties fermented with plantain leaves which recorded the least per cent purple beans (Table 4.10). The variety of cocoa did not affect the percentage of purple beans produced. Fermentation materials however significantly affected the per cent of purple beans produced.Using Polythene sheet as the fermentation material resulted in the highest per cent purple beans (43 %) which was significantly greater than that from the plantain leaves but similar to that from the fertilizer sack (Table 4.10).

Table 4.10: Effect of cocoa variety and fermentation materials on purple dried cocoa beans

	Per cent purple beans			
		Fermentation Material (FM)		
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	13.67	35.00	42.33	30.33

Mixed	12.67	42.00	43.67	32.78
Mean	13.17	38.50	43.00	
Lsd (0.05)	V = 9.38	FM = 11.49	V × FM = 16.25	
CV (%) = 2.83				

4.2.3 Per cent slaty beans

The interactive effects of cocoa variety and fermentation material had significant influence on the per cent slaty beans. Significantly higher percentage of slaty beans was recorded by the mixed variety fermented with polythene sheet as compared to the other treatment combinations (Table 4.11).

Percent slaty beans was significantly influenced by the fermentation material such that polythene sheet resulted in the highest (1.17%) whiles plantain leaves led to the least (0.17%).

Table 4.11: Effect of cocoa variety and fermentation materials on the per cent slaty of cocoa beans

Per cent slaty beans				
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	0.00	0.00	0.00	0.00

Mixed	0.33	0.67	2.33	1.11
Mean	0.17	0.34	1.17	
Lsd (0.05)	V = 0.66	FM = 0.80	V × FM = 1.13	
CV (%) = 1.12				

4.2.4 Percent purity of beans

Cocoa variety x fermentation material interaction had significant effect on the purity of the dried cocoa beans. Either hybrid variety or mixed varieties fermented with plantain leaves had the highest purity above 90%, significantly greater than the purity from the other treatment combinations. The least purity was obtained from mixed variety fermented with fertilizer sack (79.63 %).

Fermentation material also had significant ($p<0.05$) effect on the purity of the dried cocoa beans. Plantain leaves had 99.38% purity, significantly higher than those from Fertilizer sack (82.85%) and polythene sheet (83.92%) (Table 4.12).

Table 4.12: Effect of cocoa variety and fermentation materials on the purity test of dried cocoa beans

	Per cent purity			
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	99.40	86.07	85.73	90.40

Mixed	99.37	79.63	82.10	87.03
Mean	99.38	82.85	83.92	
Lsd (0.05)	V = 4.69	FM = 5.75	V × FM = 8.13	
CV (%) = 5.04				

4.3 EFFECT OF COCOA VARIETY AND FERMENTATION MATERIALS ON THE CHEMICAL COMPOSITION OF COCOA BEANS (PROXIMATE ANALYSIS)

4.3.1 Moisture content of beans

Moisture content in the dried cocoa beans was not significantly affected by cocoa variety, fermentation or their interactions. Moisture content ranged from 6.23 to 6.63 (Table 4.13).

Table 4.13: Effect of cocoa variety and fermentation materials on moisture content of cocoa beans

	Moisture content (%)			
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	4	3.67	4	3.89
Mixed	3.83	4.02	3.67	3.84
Mean	3.92	3.85	3.84	
Lsd (0.05)	V = 0.49	FM = 0.59	V × FM = 0.84	
CV (%) = 11.95				

4.3.2 pH of beans

Combination of cocoa variety and fermentation material had significant ($p<0.05$) effect on the pH in the dried cocoa beans (Table 4.14). Mixed varieties fermented with fertilizer sack and polythene sheet had pH of 6.23 and 6.40 which were similar

but significantly different from the other combinations. Hybrid variety had pH of 5.38 which was lower and different from the mixed varieties which recorded 5.84. pH in the dried cocoa beans was also significantly ($p<0.05$) influenced by the fermentation material. Fertilizer sack and polythene sheet had pH of 5.95 and 5.94 respectively which were significantly different from plantain leaves which had 4.95 (Table 4.14).

Table 4.14: Effect of cocoa variety and fermentation materials on pH of dried cocoa beans

pH in the dried cocoa beans				
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	5.00	5.67	5.47	5.38
Mixed	4.90	6.23	6.40	5.84
Mean	4.95	5.95	5.94	
Lsd (0.05)	V = 0.28	FM = 0.13	V × FM = 0.48	
CV (%) = 4.58				

4.3.3 Ash content of beans

The ash content of the dried beans was neither influenced ($p>0.05$) by cocoa variety, fermentation material nor their combined effects. The percent ash content ranged from 2.53 % to 5.24 % (Table 4.15).

Table 4.15: Effect of cocoa variety and fermentation materials on ash content of cocoa beans

Ash content (%)		
	Fermentation Material (FM)	
	Polythene sheet	Mean

Cocoa Variety (V)	Plantain leaves	Fertilizer sack	2.90	2.76
Hybrid	2.57	2.82		
Mixed	2.53	5.24	2.65	3.47
Mean	2.55	4.03	2.78	
Lsd (0.05)	V = 2.23	FM = 2.73	V × FM = 3.87	
CV (%) = 6.81				

4.3.4 Fibre and protein contents of beans

The fibre content of the dried beans was neither influenced (*p>0.05*) by cocoa variety, fermentation material nor their combined effects. The per cent fibre content ranged from 17.63 % to 23.61 % (Table 4.16). Similarly, the protein content of the dried beans was neither influenced (*p>0.05*) by cocoa variety, fermentation material nor their combined effects. The percent protein content ranged from 10.43 % to 14.71 % (Table 4.17).

Table 4.16: Effect of cocoa variety and fermentation materials on fiber content of cocoa beans

Fibre content (%)				
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	22.71	17.63	23.61	21.32
Mixed	20.48	18.54	21.60	20.21
Mean	21.60	18.09	22.61	
Lsd (0.05)	V = 6.91	FM = 0.59	V × FM = 0.84	
CV (%) = 11.95				

Table 4.17: Effect of cocoa variety and fermentation materials on protein content of dried cocoa beans

Protein content (%)
Fermentation Material (FM)

Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	14.28	14.29	14.05	14.21
Mixed	14.71	10.43	14.20	13.11
Mean	14.50	12.36	14.13	
Lsd (0.05)	V = 2.53 (ns)	FM = 3.09 (ns)	V × FM = 4.38 (ns)	
CV (%) = 1.76				

4.3.5 Fat content of beans

The interaction of cocoa variety and fermentation material significantly ($p<0.05$) affected the fat content in the dried cocoa beans. All the interaction effects had similar fat content in the range of 41.33 % to 46.40% except hybrid variety fermented with plantain leaves which had 34.22% as the least (Table 4.18). Fermentation material had significant (*$p<0.05$*) effect on the fat content in the dried cocoa beans. The highest bean fat content was recorded from fertilizer sack (45.52%) fermentation whiles the least was recorded from plantain leaves (37.78%) fermentation (Table 4.18). The cocoa varieties however did not affect the fat content of the cocoa bean.

Table 4.18: Effect of cocoa variety and fermentation materials on fat content of dried cocoa beans

Fat content (%)				
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	34.22	46.40	43.50	41.37

Mixed	41.33	44.63	45.00	43.65
Mean	37.78	45.52	44.25	
Lsd (0.05)	V = 3.41	FM = 4.18	V × FM = 5.91	
CV (%) = 7.64				

4.3.6 Free fatty acid content in dried beans

The combined cocoa variety and fermentation material interaction had significant effect on the free fatty acid in the dried cocoa beans. All the combinations had similar free fatty acid content except mixed varieties fermented with polythene sheet which had 8.65%, the least (Table 4.19).

Cocoa variety had significant ($p<0.05$) effect on the free fatty acid in the dried cocoa beans. Hybrid variety had free fatty acid of 3.93% which was lower and different from mixed variety which recorded 5.84% (Table 4.19).

Free fatty acid in the dried cocoa beans was not significantly influenced by the fermentation material (Table 4.19).

Table 4.19: Effect of cocoa variety and fermentation materials on free fatty acid content of dried cocoa beans

Free fatty acid content (%)				
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	3.82	4.44	3.55	3.93
Mixed	4.79	4.08	8.65	5.84

Mean	4.31	4.26	6.10
Lsd (0.05)	V = 1.58 (s)	FM = 1.95 (ns)	V × FM = 2.75 (s)
CV (%) = 3.09			

4.3.7 Nitrogen free extracts content

The interaction of cocoa variety and fermentation had significant ($p<0.05$) effect on the nitrogen free extracts content in the dried cocoa beans. The two levels of cocoa variety fermented with either plantain leaves or fertilizer sack had similar nitrogen free extracts in the range of 10.75 - 18.41% but only hybrid variety fermented with plantain leaves was different from hybrid fermented with polythene sheet and mixed varieties fermented with polythene sheet (Table 4.20).

Fermentation material had significant effect on the nitrogen free extract content in the dried cocoa beans. The highest bean nitrogen free extract content was recorded from the plantain leaves (15.37%) fermentation whiles the least was recorded for those fermented with polythene sheet (6.31%) (Table 4.20). The cocoa varieties however did not affect the nitrogen free extract content of the cocoa bean.

Table 4.20: Effect of cocoa variety and fermentation materials on nitrogen free extracts content of cocoa beans

Nitrogen free extracts content (%)				
	Fermentation Material (FM)			
Cocoa Variety (V)	Plantain leaves	Fertilizer sack	Polythene sheet	Mean
Hybrid	18.41	10.75	8.39	12.52
Mixed	12.33	13.05	4.23	9.87

Mean	15.37	11.90	6.31
Lsd (0.05)	V = 5.61	FM = 6.87	V × FM = 9.72
CV (%) = 4.78			

CHAPTER FIVE 5.0 DISCUSSION 5.1 FERMENTATION PRACTICES OF COCOA FARMERS

Majority of the farmers practiced pods storage before pod breaking. The farmers had several reasons for practicing pods storage notable among them were inadequate hands for pod gathering and breaking, improves fermentation and drying of beans. According to Are and Gwynne-Jones (1974) and Mossu (1992) pods should not be stored for more than six days after harvesting. This is because pod storage, as a method of pulp preconditioning, results in changes in the properties of the pulp in the beans prior to fermentation. This therefore allows substrate metabolization during fermentation causing the production of acids by lactic acid bacteria, yeasts and acetic

acid bacteria (Biehl *et al.,* 1989; Meyer *et al.,* 1989). The technique of pod storage reduces the formation of acids throughout the fermentation without enhancing the degradation of acids at the end of fermentation. During pod storage, the beans within the pod lose moisture which allows more air to penetrate the pods into beans before the pods are broken to start the fermentation. This facilitates the fermentation process with accompanied faster temperature rises and thus resulting in improved quality of the cocoa beans (Biehl *et al.,* 1989; Meyer *et al.,* 1989; Nazaruddin *et al.,* 2006). Previous research revealed that fermentation of beans from stored pods are more rapid and result in higher brown bean counts (Afoakwa *et al.,* 2012). Pod storage has also been reported to reduce pulp volume and acidity, affecting subsequent reductions in the polyphenolic and anthocyanin components, resulting in enhanced bean flavour quality (Biehl *et al.,* 1990; Afoakwa *et al.,* 2013).

More than half of the farmers used plantain/banana leaves in fermenting their cocoa beans. Between 16 – 28% of the farmers use fertilizer sacks whiles 8 – 33% used polythene sheets. This could be related to the drainage of pulp surrounding the beans which if not properly done could result in bad fermentation, (Wood and lass 1985).

Concerning the number of days for fermentation of the cocoa beans, 56.3% of the farmers use 6 days, Quality corresponds with fermentation period, where an optimal quality is achieved by 5 to 7 days of fermentation. Proper fermentation results in a better flavour, removes the bitterness in the beans and makes it easier for the grinders to remove the bean coat (Ruf and Yoddang, 2009; Taher, 1996). There were significant associations between pod storage and number of days used for fermentation among the farmers in Samreboi and Enchi cocoa districts. This could

be explained by the fact that pod storage as a means of pulp preconditioning cocoa beans prior to fermentation influences the activity of polyphenol oxidase during fermentation and drying, resulting in modifications in the polyphenolic and anthocyanin concentrations the effects of which are expected to influence the fermentative quality of the beans (Afoakwa *et al.*, 2012).

5.2 EFFECTS OF FERMENTATION MATERIALS ON THE PHYSICAL ATTRIBUTES AND CHEMICAL COMPOSITION OF THE DRIED COCOA BEANS

Fermentation materials used in this study did not influence moisture content in the dried cocoa beans which ranged from 6.30 – 6.60%. The moisture levels of the cocoa beans were considered normal compared with the acceptable limits (6 – 7%) for long term storage of cocoa (Wood and Lass, 1985; Dand, 1997; Fowler, 2009). These relatively lower moisture content attained was to ensure that virtually all microbial and enzymatic reactions had ceased. Although fermentation reduced the water content of the beans there was still considerable amount of moisture lost during drying, thus confirming previous findings (Páramo *et al*., 2010).

Mouldiness of cocoa beans obtained from plantain, fertilizer sack and polythene sheet were 0.92, 1.12 and 0.90% respectively. Purple beans in the dried cocoa beans were significantly influenced by the fermentation materials. Plantain/banana leaves produced purple beans of 13.17% compared to fertilizer sack and polythene sheet (38.50 and 43.00% respectively). Guehi *et al., (*2010) reported that purple beans occur when the fermentation has been terminated.

The number of slaty beans counted differed significantly among the fermentation materials. Plantain/banana leaves had the lowestt percentage of slaty beans. Slaty beans are beans in which more than 50% of the cotyledon is grey or slaty in colour (Fowler, 2009) and have rubbery cotyledon and resistance to cutting (Guehi *et al.*, 2010). These beans have not undergone fermentation and they have a low level of cocoa flavour with high levels of astringency. This means that cocoa beans produced from fertilizer sack and polythene sheet were not well fermented. This could partly be due to the insufficient drainage of pulp in the fermentation with fertilizer sack and polythene sheet result in a bad fermentation (Wood and Lass, 1985).

Fermentation materials had significant influence on the all other defects in the dried cocoa beans. Plantain/banana leaves, fertilizer sack and polythene sheet had 2.42, 1.17 and 1.98%, respectively which may be due easy damage of plantain leaves by insects and rodents and accessing the drained pulp from the fermenting beans. The purity of the dried cocoa beans differed significantly among the fermentation materials. Cocoa beans produced from plantain / banana leaves had 99.38% purity, fertilizer sack had 82.85% and polythene sheet had 83.92%. This supports the assertion that insufficient drainage of pulp result in a bad fermentation (Wood and Lass, 1985) which ultimately affects the purity of the dried cocoa beans.

Moisture composition in the dried cocoa beans did not vary significantly among the fermentation materials. The moisture content in the dried cocoa beans ranges from 3.84% to 3.92% and these are slightly higher than the 3.2%t reported by Dand (1997) . This gives an indication of the moisture composition in the bean; however this

varies depending on the type of bean, the quality of the fermentation and drying and the subsequent processing of the bean.

In this study, fermentation with plantain/banana leaves gave the least pH of 4.95 which indicates that the beans were well fermented compared with fertilizer sack (5.95) and polythene sheet (5.94). During fermentation, the rate of diffusion of organic acids into the cotyledons, timing of initial entry, duration of the period of optimum pH and final pH are crucial for optimum flavour formation (Biehl *et al*., 1985). Beans of higher pH (5.5-5.8) are considered unfermented - with low fermentation index and those of lower pH (4.75- 5.19), well fermented.

The ash, fiber, protein and free fatty acids contents did not vary significantly among the fermentation materials. Crude protein content from fermentation materials ranged 12.36 to 14.50% and this was lower than reported values of 15.2– 19.8% (Aremu *et al.,* 1995; Afoakwa *et al.,* 2008). The ash content of the cocoa beans from the fermentation materials were from 2.55% to 4.03% compared to the reported value of 2.63% (Aremu *et al.,* 1995).

5.3 THE IMPACT OF COCOA VARIETIES ON THE PHYSICAL ATTRIBUTES AND CHEMICAL COMPOSITION OF THE DRIED COCOA BEANS

Hybrid cocoa beans had 6.39% moisture compared to the mixed varieties which had 6.49%. The difference between hybrid and mixed cocoa varieties could contribute to the variation in mouldiness of the cocoa beans as hybrid variety had no moulded beans whilst the mixed varieties had 1.96% (Asare, 2010). The purple beans did not

significantly vary between the hybrid and mixed cocoa varieties. Hybrid cocoa beans had 30.33% whilst mixed varieties had 32.78%. This confirms the report of Guehi *et al. (*2010) purple beans occur when the fermentation has been terminated prematurely and not variety dependent. In this study all the beans were fermented on the same days interval however, the higher figure recorded by the mixed varieties could be attributed to different composition of cocoa beans fermented from the mixed varieties. According to Wood and Lass (1985) types of cocoa for fermentation vary for instead Criollo cocoa is fermented for a relatively short period of 2 – 3 days while Forastero cocoa is fermented for 3 – 7 days, occasionally longer. There was a significant variation in slaty beans between the varieties used in this study, the hybrid cocoa beans had no slaty beans but mixed varieties had 1.11% and this may be due to inadequate fermentation of the beans from the mixed varieties (Wood and Lass, 1985). All other defects summation weevil infested, flat, holed and germinated beans did not vary significantly between the hybrid and mixed cocoa varieties. The purity of cocoa beans from the hybrid and mixed varieties were similar.

All the chemical composition parameters examined in this study were not significantly different between the hybrid and mixed cocoa varieties except pH and free fatty acids. The similarity in moisture, ash, fiber, protein, fat and nitrogen free extracts contents may be attributed to the genetic composition of the varieties used. However, the variation in pH and free fatty acids may be linked to the per cent mouldiness beans recorded by the varieties as observed by the Woo and Lass (1985) that moulds inside the beans can also increase the free fatty acid content.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATION

6.1 CONCLUSION

Majority of the farmers practiced delayed pod breaking after harvest in Samreboi, Asankrangwa and Enchi cocoa districts.

Most farmers used plantain banana leaves although some also used fertilizer sacks and polythene sheets for fermentation.

Majority of the farmers used 6 days for fermentation.

Fermentation materials used in this study did not influence moisture content in dried cocoa beans. Mouldiness of cocoa beans, purple beans production and number of slaty beans were less with the use of plantain leaves than the other fermentation materials.

Fermenting with plantain / banana leaves gave the least pH of 4.95 although for ash, fiber, protein and free fatty acids contents, all the fermentation materials were produced similar effects.

The hybrid and mixed cocoa varieties differed in mould content such that the hybrid did not have any mouldy beans compared with the mixed variety

The chemical composition such as moisture, ash, fiber, protein, fat and nitrogen free extracts content were not significantly different between the hybrid and mixed cocoa varieties, but, contrary observation was made for pH and free fatty acids. The use of polyethylene sheets or fertilizer sack should be discouraged among farmers.

6.2 RECOMMENDATIONS

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

Further studies should be conducted to ascertain the effects the number of turning has on cocoa beans quality fermented with polyethylene sheets and fertilizer sack

Additional studies should be conducted to evaluate the effects of different days of fermenting of cocoa beans on the cocoa bean quality using fertilizer sack and polyethylene sheets as fermenting materials.

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APPENDICES

SAMPLE OF QUESTIONNAIRE

KWAME NKRUMAH UNIVERSITY OF SCIENCE & TECHNOLOGY

COLEGE OF AGRICULTURE AND NATURAL RESOURCES

DEPARTMENT OF HORTICULTURE

Questionnaire №:……

Date:……

Time started:..…… Time ended:…….

Name of Community:…… Name of cocoa District……

Name of Political/AdministrativeDistrict: …….

INFORMED CONSENT FOR FARMER

The researcher is a student at the Department of Horticulture, Kwame Nkrumah University of Science and Technology, Kumasi. I am conducting a study to "the use of „fertilizer sacks" and polyethylene sheet as an alternative material for the fermentation of cocoa beans and its impact on cocoa bean quality". I wish that you take full participation in the survey on fermentation practices of farmers. However, participation in this study is completely voluntary and you reserve the right to decide not to respond to certain questions or withdraw at any time in the course of the survey without any penalty. This interview is anonymous and your responses are completely confidential. Notice that whatever information you disclose will only be used for academic purposes and will be treated as strictly confidential as possible and that will be reported in a way that no one will know your specific responses.

Tick **(√)**, mark **(×)** or write where appropriate. If you have any question and queries concerning this research, please do not hesitate to contact the researcher at

+322 (0) 266166124.

SECTION A: BACKGROUND CHARACTERISTICS OF FARMER

1. Sex: i. Male ( ) ii. Female ( )
2. Age: i. 18 – 27 ( ) ii. 30 – 39 ( ) iii. 40 – 49 ( ) iv. 50 or more years ( )
3. Educational Qualification: i. No formal education ( ) ii. Primary ( ) iii. MSLC ( ) iv. JSS ( ) v. SSS ( ) vi. Tertiary ( )
4. Marital status: i. Married ( ) ii. Single ( ) iii. Divorced ( ) iv. Widow ( ) v. Widower ( )
5. Do you own the cocoa farm? i. Yes ( ) ii. No ( )
6. If No, what is your relationship with the owner?
   i. Family relation ( ) ii. Care taker ( ) iii. Other (specify)
   ……..
7. If yes, how did you come by it?
   i. Purchased ( ) ii. Share cropping ( ) iii. Rented land ( ) iv. Leasehold ( ) v. Gift ( ) vi. Family inheritance ( ) vii. Family land ( )
   v. Other (specify) ……

SECTION B: FERMENTATION PRACTICES

8. Do you delay between harvesting and pod breaking? i. Yes ( ) ii. No ( )

9. a) If yes, how many days do you delay between harvesting and pod breaking?

i. 1 day ( ) ii. 2 days ( ) iii. 3 days ( ) iv. 4 days ( ) v. 5 days ( ) vi. 6 days ( ) v. 7 days ( )

vi. More than 7 days ( )

b). Give reasons for your answer

……

……

10. What methods of fermentation do you adopt on your farm?

i. Heap ( ) ii. Tray ( ) iii. Basket ( ) iv. Box ( ) v. Other (specify) .....

11. What material(s) do you use for fermentation of the beans? i. Plantain/banana leaves ( ) ii. Baskets ( ) iii. Box ( ) iv. Polythene sheet (polysack) ( ) v. Fertilizer sacks ( ) vi. Jute sack ( )

v. Other (Specify): ……

12. Why do you choose this material? i. Easy to come by ( ) ii. More durable ( ) iii. Easy to handle ( )

iv. Other (Specify): ……

13. How many days do you ferment your cocoa?

i. 3 days ( ) ii. 4 days ( ) iii. 5 days ( ) iv. 6 days ( ) v. 7days ( ) vi. Specify others…….....

14. Why this number of days?

i. Better fermentation ( ) ii. Better bean colour ( ) iii. Weight gain ( ) iv. Convenience ( )

v. Other (Specify): …….

15. How often do you turn your cocoa?

i. No turning ( ) ii. 1turning ( ) iii. Every 2 days ( ) Specify others……

16. Why this number of turnings?

i. Improve fermentation ( ) ii. Lack of time ( ) iii. Specify others……..

17. What is colour of your beans after fermentation?

i. Purple coloured ( ) ii. Dark coloured ( ) iii. Dull dark coloured ( ) iv. Much darker than normal ( )

SECTION C: QUALITY CHARACTERISTICS OF COCOA BEANS

18. How do your beans look like after drying?

a). Slaty i. Yes ( ) ii. No ( )

b). Purple i. Yes ( ) ii. No ( )

c). Germinated i. Yes ( ) ii. No ( )

d). Mouldiness i. Yes ( ) ii. No ( )

21. Do you think fermentation method and materials you used influence the qualities below?

a). Slaty i. Yes ( ) ii. No ( )

b). Purple i. Yes ( ) ii. No ( )

c). Germinated i. Yes ( ) ii. No ( )

d). Mouldiness i. Yes ( ) ii. No ( )

22. Kindly explain how you think fermentation method and materials you used influence the below quality?

a). Slaty: ……

……..…… b).

Purple:……

……..

c). Germinated: ……..……..

……

d). Mouldiness: ……

……

Thank you for your participation

APPENDIX II

ANALYSIS OF VARIANCE

Fact A = Cocoa Variety (CV)

Fact B = Fermentation material (FM)

Interaction = CV*FM

Analysis of Variance Table for Moisture content determined by aqua boy

Source	DF	SS	MS	F	P
Rep	2	0.00444	0.00222		
CV	1	0.04500	0.04500	0.56	0.4729
FM	2	0.27444	0.13722	1.70	0.2321
CV*FM	2	0.07000	0.03500	0.43	0.6604
Error	10	0.80889	0.08089		
Total	17	1.20278			

Grand Mean 6.4389 CV 4.42

CUT TEST (PHYSICAL ATTRIBUTES)

Analysis of Variance Table for Mouldiness

Source	DF	SS	MS	F	P
Rep	2	3.1078	1.5539		
CV	1	17.2089	17.2089	48.81	0.0000
FM	2	0.1744	0.0872	0.25	0.7855

CV*FM 2 0.1744 0.0872 0.25 0.7855

Error 10 3.5256 0.3526

Total 17 24.1911

Grand Mean 0.9778 CV 6.07

Analysis of Variance Table for Purple

Source DF SS MS F P

Rep 2 456.44 228.22

CV 1 26.89 26.89 0.34 0.5745

FM 2 3104.11 1552.06 19.44 0.0004

CV*FM 2 50.78 25.39 0.32 0.7347

Error 10 798.22 79.82

Total 17 4436.44

Grand Mean 31.556 CV 2.83

Analysis of Variance Table for Slaty

Source DF SS MS F P

Rep 2 0.1111 0.05556

CV 1 5.5556 5.55556 14.29 0.0036

FM 2 3.4444 1.72222 4.43 0.0419

CV*FM 2 3.4444 1.72222 4.43 0.0419

Error 10 3.8889 0.38889

Total 17 16.4444

Grand Mean 0.5556 CV 1.12

Analysis of Variance Table for All other defects

Source	DF	SS	MS	F	P
Rep	2	1.5478	0.7739		
CV	1	2.1356	2.1356	1.15	0.3083
FM	2	4.8344	2.4172	1.30	0.3138
CV*FM	2	42.8344	21.4172	11.56	0.0025
Error	10	18.5322	1.8532		
Total	17	69.8844			

Grand Mean 1.8556 CV 7.34

Analysis of Variance Table for Percentage purity

Source	DF	SS	MS	F	P
Rep	2	26.41	13.205		
CV	1	51.00	51.005	2.56	0.1409
FM	2	1027.41	513.707	25.74	0.0001
CV*FM	2	30.88	15.440	0.77	0.4870
Error	10	199.54	19.954		
Total	17	1335.25			

Grand Mean 88.717 CV 5.04

CHEMICAL COMPOSITION (PROXIMATE ANALYSIS)

Analysis of Variance Table for Moisture content

Source	DF	SS	MS	F	P

Rep 2 0.36943 0.18472

CV 1 0.01027 0.01027 0.05 0.8307

FM 2 0.02443 0.01222 0.06 0.9447

CV*FM 2 0.38888 0.19444 0.91 0.4330

Error 10 2.13383 0.21338

Total 17 2.92685

Grand Mean 3.8650 CV 11.95

Analysis of Variance Table for pH

Source DF SS MS F P

Rep 2 0.86111 0.43056

CV 1 1.68056 1.68056 23.82 0.0006

FM 2 0.66778 0.33389 4.73 0.0358

CV*FM 2 0.27444 0.13722 1.94 0.1934

Error 10 0.70556 0.07056

Total 17 4.18944

Grand Mean 5.8056 CV 4.58

Analysis of Variance Table for ash content

Source DF SS MS F P

Rep 2 4.7717 2.38587

CV 1 2.2543 2.25427 0.50 0.4960

FM 2 7.6252 3.81262 0.84 0.4584

CV*FM 2 6.6027 3.30136 0.73 0.5055

Error 10 45.1709 4.51709

Total 17 66.4249

Grand Mean 3.1206 CV 6.81

Analysis of Variance Table for fibre content

Source	DF	SS	MS	F	P
Rep	2	76.993	38.4965		
CV	1	5.544	5.5445	0.13	0.7280
FM	2	67.508	33.7540	0.78	0.4847
CV*FM	2	9.159	4.5795	0.11	0.9007
Error	10	433.181	43.3181		
Total	17	592.386			

Grand Mean 20.761 CV 3.17

Analysis of Variance Table for protein content

Source	DF	SS	MS	F	P
Rep	2	5.435	2.71747		
CV	1	5.390	5.39014	0.93	0.3571
FM	2	15.583	7.79133	1.35	0.3033
CV*FM	2	17.292	8.64588	1.50	0.2704
Error	10	57.830	5.78304		
Total	17	101.530			

Grand Mean 13.659 CV 1.76

Analysis of Variance Table for fat content

Source	DF	SS	MS	F	P
Rep	2	183.554	91.777		
CV	1	23.461	23.461	2.22	0.1669
FM	2	206.927	103.463	9.80	0.0044
CV*FM	2	60.566	30.283	2.87	0.1036
Error	10	105.561	10.556		
Total	17	580.069			

Grand Mean 42.514 CV 7.64

Analysis of Variance Table for free fatty acid content

Source	DF	SS	MS	F	P
Rep	2	11.4635	5.7318		
CV	1	16.3401	16.3401	7.14	0.0234
FM	2	13.2067	6.6034	2.89	0.1024
CV*FM	2	24.3148	12.1574	5.32	0.0268
Error	10	22.8730	2.2873		
Total	17	88.1982			

Grand Mean 4.8872 CV 3.09

Analysis of Variance Table for Nitrogen Free Extracts content

Source	DF	SS	MS	F	P
Rep	2	27.796	13.898		
CV	1	31.469	31.469	1.10	0.3186
FM	2	250.581	125.290	4.39	0.0429
CV*FM	2	57.869	28.934	1.01	0.3976
Error	10	285.650	28.565		
Total	17	653.364			

Grand Mean 11.194 CV 4.78