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**COLLEGE OF SCIENCE  
FACULTY OF BIOSCIENCES  
DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY**

**CARBOHYDRATE COMPOSITION AND AMYLASE ACTIVITY OF  
SWEETPOTATO (*IPOMOEA BATATAS*) ROOT**

**BY**

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**A THESIS SUBMITTED TO THE DEPARTMENT OF FOOD SCIENCE  
AND TECHNOLOGY, COLLEGE OF SCIENCE IN PARTIAL  
FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF  
DOCTOR OF PHILOSOPHY (Ph.D.)**

**AUGUST 2016**

## DECLARATION

I hereby declare that this submission is my own work towards the PhD. and that, to the best of my knowledge, it contains no material previously published by another person nor material which has been accepted for the award of any other degree of the University, except where due acknowledgement has been made in the text.

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## DEDICATION

This work is dedicated to my wife (Mrs. Matilda Maaboah Owusu-Mensah), Children (Ama Serwaah Owusu-Mensah, Kofi Boakye Owusu-Mensah, Kwaku Dwamena Kenten Owusu-Mensah, and Stephen Kwaku Mensah OwusuMensah), Mad. Margaret Mensah (my mother), Mr. and Mrs. Bandoh (my guardians), and my siblings (Prince Appiah, Eugene Owusu-Mensah, Christiana Owusu-Mensah, and Emmanuel Owusu-Mensah) for their meritorious support in all my endeavours.



## PREFACE

This thesis resulted from my PhD research work at Food Science Department, Kwame Nkrumah University of Science and Technology from August 2010 to September 2015. The work was sponsored by International Potato Center under its Sweetpotato Action for Security and Health in Africa (SASHA) project and Quama Food Processing Co. Ltd, a young and enterprising food firm in Ghana of which I am the Managing Director (MD). With the main objective of evaluating key quality attributes of sweetpotato roots to promote its utilization, the thesis is apportioned into seven chapters. Chapter 1 provides a broad introduction to the work, Chapter 2 a review of pertinent literatures while Chapter 7 summarises the outcomes of the entire research. The main work is detailed in Chapter 3 – 6. Chapter 3 - 5 deals extensively with the key quality attributes of the roots and factors including environment, heating regimes and cooking treatment affecting the levels of the attributes in raw and processed forms while Chapter 6 highlights the development of a fast and accurate technique for the measurement of the attributes in cooked roots. The findings of this study will be beneficial to food scientists, breeders, food and food-related research bodies, food processors and other actors along the root and tuber value chain. It is also anticipated that staff and students working on sweetpotato roots will find this thesis useful in understanding the expression of the quality attributes particularly sugars and amylases.

## ABSTRACT

Selection of sweetpotato clone(s) for production and utilisation depends on its quality attributes especially sugars, dry matter, starch content and amylase activity. The levels of these attributes are however influenced by factors such as the diversified production environments, cooking treatments, and genotypic compositions. The aim of the study is to determine the range of variation and factors of significant influence on concentrations of these attributes during growth and processing of sweetpotatoes. Development of an efficient and accurate technique for rapid assessment of these quality attributes was also relevant to the study. In order to achieve the overall aim, two main approaches were adopted; varying the levels of identified factors that alter the concentration of the attributes, and using near-infrared reflectance spectroscopy technique to develop a prediction model for their assessment. Sugars (sucrose, glucose, fructose and maltose), starch, dry matter content, amylase activity and sweetness were evaluated at harvest, during heating and after cooking. Adaptive Main Effects and Multiplicative Interaction (AMMI) model was employed to ascertain the magnitude of interactions between the factors tested. It was established that concentrations of the quality attributes were significantly affected by all the factors examined. Genotype contributed the highest variability during the growth stage while cooking treatment produced the highest effect during processing. Effect of interaction was more pronounced on amylase activity compared to sugars, dry matter and starch content. Sucrose was the predominant sugar in the raw form while levels of maltose increased substantially during cooking. High precision calibration was also developed for evaluating sugars and starch content in cooked sweetpotato roots. Sweetpotato varieties in Ghana have a wide range of quality attributes, making it a versatile crop for many food applications. Nonetheless recommendation for specific application should consider choice of production environment and processing conditions, which have significant impact on the final quality of the processed product.



## ACKNOWLEDGEMENT

To God be the glory great things He has done. My sincere gratitude goes to my supervisors; Prof. (Mrs.) I. Oduro, Prof. W.O. Ellis and Prof E.E. Carey for their technical directions and cooperation throughout the project. The same thanks go to staff of postharvest/NIRS laboratory (Eric Dery, Thomas Tuffour, Razak Assimah and Kwame Shadrack) and Mr. Ebenezer Obeng, Mr. Yusif Alhassan (SASHA project, CIP-Ghana). Another heartfelt gratitude goes to Mr. Kwadwo Adofo, Dr. Ernest Baafi, Mr. Asamoah, Mr Ewoodzi, all of Sweetpotato National breeding programme – Crop Research Institute, Fumesua, for their technical assistance. May Almighty God bless you for your patience and passion to impact technological ideals into young scientist for the development of our mother country, Ghana.

Finally, I would like to thank my family; wife (Mrs. Matilda Maaboah OwusuMensah), children (Ama Serwaah Owusu-Mensah, Kofi Boakye Owusu-Mensah, Kwaku Dwamena Kenten Owusu-Mensah, and Stephen Kwaku Owusu-Mensah) and Ms. Margaret Mensah (my mother), for their love, advice and financial support. Ayekoo. May the Sovereignty God continue to protect and provide all your needs.

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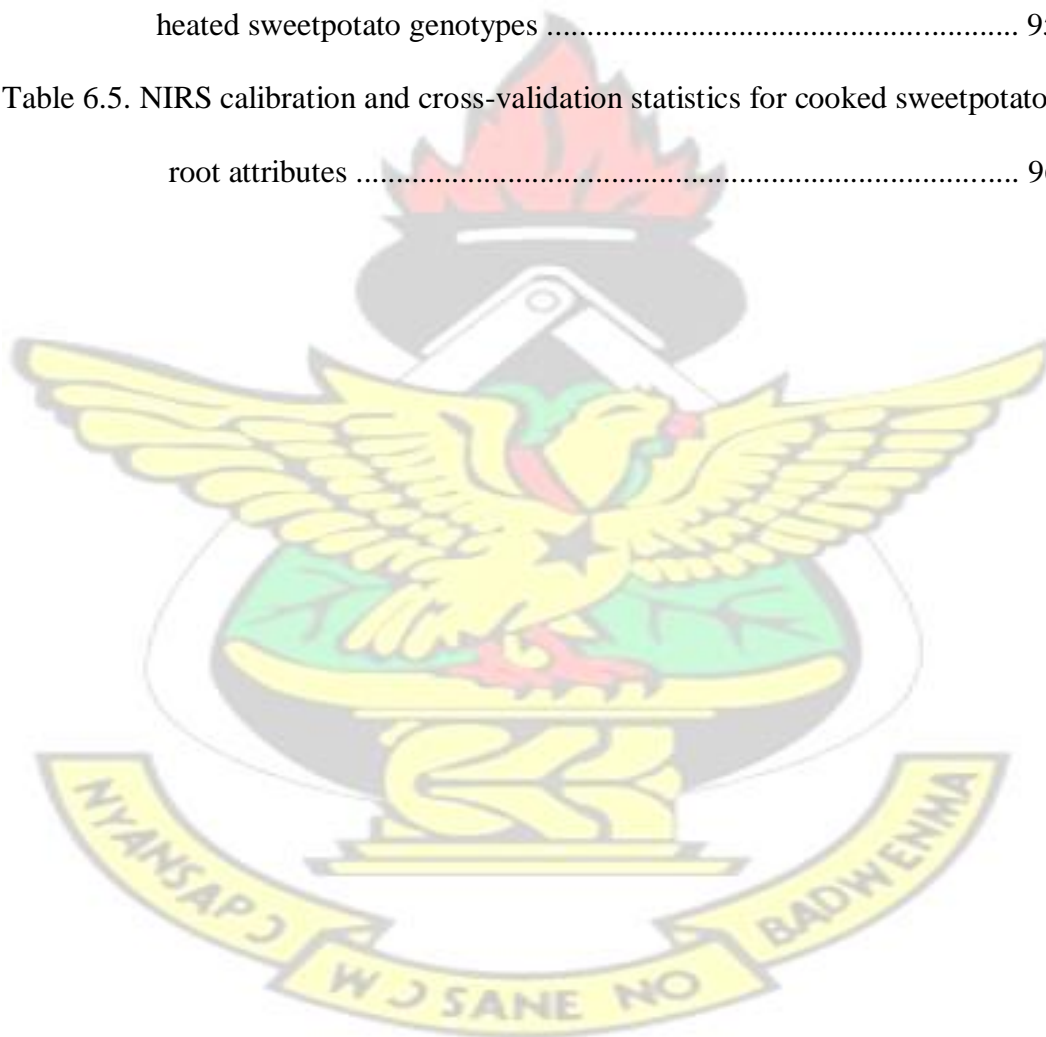
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## CHAPTER ONE

### 1.0 GENERAL INTRODUCTION

#### 1.1 Introduction

Sweetpotato (*Ipomoea batatas*) is among the most important food crops in the world in terms of total production and economic value (Lu *et al.*, 2006; and Fuglie *et al.*, 2002). It is ranked as the seventh crop after wheat, rice, maize, potato, barley, and cassava, and the third most important root and tuber crop in Sub-Saharan Africa (Truong and Avula, 2010; Gibson and Aritua, 2002). In Ghana, sweetpotato is the third most important root and tuber crop with a total production of 90,000 tonnes per year according to Kenyon *et al.* (2006). However current FAOSTAT (2013) production statistics shows an increase in production to 131,999 tonnes. High yielding ability, and short maturity period (3 to 6 months) which also correlates with the savannah rainfall pattern (Truong and Avula, 2010; Akoroda, 2009) are among the key stimulating factors for the current increase in production. Ease of cultivation and ability to thrive even under harsh conditions are other notable contributory factors (Odebode *et al.*, 2008). Moreover, sweetpotato has a wide range of quality attributes, which makes it an exceptionally healthy and versatile crop for the food industry.

Orange Flesh Sweetpotato (OFSP) varieties contain high amounts of  $\beta$ -carotene, a precursor of vitamin A, for combating Vitamin A deficiency in several countries of the developing world (Burri, 2011; Low *et al.*, 2007). The relatively high soluble and insoluble fibre content and polyphenols, also contribute to a number of health benefits including reduced serum low-density lipoprotein cholesterol, risk of cardiovascular disease and incidence of certain cancers and diabetes (Low *et al.* 2007). Despite its high sugar content, sweetpotato has low to moderate

glycemic index depending on the cooking method, for management of diabetes (Allen *et al.*, 2012). Dry matter and starch contents of some varieties are very high and compare favourably with some staple root and tuber crops including yam and cassava. In addition to the high starch content, sweetpotato contains high extractable amylase for starch hydrolysis especially in brewing and syrup production (Hagenimama *et al.*, 1996).

In spite of its high agronomic and varied quality attributes, sweetpotato utilisation is very low compared to similar root and tuber crops in many places (Odebode *et al.*, 2008; Adjeikum, 2003; Wang and Kays, 2000). It is rarely preferred even though it is more nutritious than most staple crops (Kays *et al.*, 2005; Truong and Avula, 2010). Reasons for this low consumption have been largely assigned to the high sugar content (dominant flavour) of the cooked product (Kays *et al.*, 2005; Morrison *et al.*, 1993; Sun *et al.*, 1993). The dominant flavour limits the flavour diversity of sweetpotato processed products (Wang and Kays, 2000; Sun *et al.*, 1993) thereby decreasing its utilisation potential compared to bland starchy staples like some varieties of cassava and yam (Akoroda, 2009; Kays *et al.*, 2005). Sugars present in raw uncooked roots (sucrose, glucose, and fructose) and maltose formed during cooking via starch hydrolysis by amylases (Lewthwaite *et al.*, 1997; Hagenimana *et al.*, 1996; Takahata *et al.*, 1994) are responsible for the distinct flavour of sweetpotatoes (Wang and Kays, 2000; Sun *et al.*, 1993). The sugars also serve as precursors for the synthesis of many aromatic compounds through maillard and caramelization reactions (Yaylayan *et al.*, 1994). Sugar content and amylolytic potential are thus critical in determining the final flavour and acceptability of sweetpotato genotypes for food applications.

Data on sugar profile and amylase activity of officially released sweetpotato varieties in Ghana are however limited despite its high potential toward the development of sweetpotato industry. Moreover, the influence of production environment on accumulation of sweetpotato quality attributes during development and processing has not been largely investigated. Cooking method is also another possible source of diversity in sugar content and amylase function of sweetpotato genotypes. Temperature and time of processing affect the stability and hydrolytic power of native amylases and sugar content of processed products from sweetpotato roots (Wang and Kays, 2000; Takahata *et al.*, 1994; Sun *et al.*, 1993). Information on sugar content and more importantly sweetness of released varieties will aid selection of desirable type(s) for industrial and household utilisation. Availability of preferred varieties will enhance production, marketing, and consumption of sweetpotato thereby creating ready market and generating income for actors (farmers, processors and distributors) along the sweetpotato value chain. To better understand variations and impact of sugars, starch, and amylases in defining what a preferred sweetpotato variety is for specific food applications, the following research questions are posed.

## **1.2 Research questions**

1. What are the ranges of key quality attributes of sweetpotato roots, and are production environments likely to have significant effects on these attributes?
2. Are amylases responsible for maltose formation during processing of sweetpotato roots and what is the inactivation temperature and time regime of  $\beta$ -amylase?
3. Is sugar content of cooked sweetpotato roots dependent on cooking method?



4. Can Near Infrared Reflectance Spectroscopy (NIRS) be calibrated for determination of sugars and starch in cooked sweetpotato roots?

Research objectives were hence formulated to address these research questions.

### 1.3 Objectives

**Main Objective:** To determine the factors affecting the concentration of carbohydrate (sugars and starch) and amylase activity of eleven sweetpotato varieties, and develop a rapid but accurate method for their estimation, and application in the food industry.

#### **Specific Objectives;**

1. To determine sugar profile (sucrose, glucose and fructose), starch content, and amylase activity of eleven sweetpotato varieties in five different production environments.
2. To determine the effect of heat treatment on  $\beta$ -amylase stability and sugars of four sweetpotato varieties at selected temperatures (65°C, 75°C and 85°C) and times (0, 10, 20 and 40 min).
3. To determine the effect of three different cooking methods on sugars and sweetness profile of sweetpotato varieties.
4. To develop a rapid and accurate method for determination of sugar profile, and starch content in cooked sweetpotato roots using Near Infrared Reflectance Spectroscopy (NIR)



## CHAPTER TWO

### 2.0 LITERATURE REVIEW

#### 2.1 Dry matter content of sweetpotato roots

Sweetpotato roots have relatively low dry matter content (30% on average) compared to other major root and tuber crops (Woolfe, 1992). Dry matter (DM) content varies widely in the sweetpotato germplasm and is significantly affected by factors such as variety and environment (Grüneberg *et al.*, 2005). It ranged from 30.2% to 39.2% in ten sweetpotato varieties in Uganda (Nabubuya *et al.*, 2012) and from 18.0% to 30.6% in New Zealand cultivars (Lewthwaite *et al.*, 1997). East African cultivars have relatively higher DM, average of 32.1%, compared to worldwide average figure of 30% (Tumwegamire *et al.*, 2011). Some varieties released in Ghana have comparatively high DM (average of 34%) with values ranging from 21.2% (Apomuden) to 47% (Hi-Starch) (CSIR-CRI, 2005).

Dry matter content is closely related to flesh colour and classification has been made in this regard; white and cream fleshed sweetpotatoes are categorised as high DM ( $\approx 25\%$  to  $35\%$ ) types and orange-fleshed sweetpotatoes (OFSP) with high  $\beta$ -carotene content are classified as low DM ( $\approx 20\%$  to  $25\%$ ) types (Lewthwaite *et al.*, 1997). However, recent breeding efforts have resulted in the development of high dry matter OFSP varieties (Tumwegamire *et al.*, 2011). Sweetpotatoes can also be classified as staple or dessert-type based on the dry matter content. Staple and supplementary staple varieties have high dry matter content with a minimum of 30% while dessert types have lower DM of 24-28% (Lewthwaite *et al.*, 1997). Sweetpotato varieties with high dry matter content and white or yellow flesh colour are generally preferred by Asian and African

consumers (Brabet *et al.*, 1998). Dry matter content has strong correlation with other quality attributes and final acceptability of sweetpotato varieties. Varieties of high dry matter content and low sugar content are suitable for frying. Sogginess and browning which are undesirable characteristics of fried products are reduced when varieties of high dry mass are used for frying. Varieties of high dry matter content are appropriate for production of high quality and less expensive flour since less energy is required for dehydration. Low dry matter varieties are ideal for production of drinks and preparation of soft dishes like “Mpotompoto” (a Ghanaian dish). Approximately 80-90% of sweetpotato dry matter is carbohydrate, which consists mainly of starch and sugars (Woolfe, 1992).

## **2.2 Starch content of sweetpotato root**

Sweetpotato storage roots contain about 70% starch on dry weight basis, (Lu and Sheng, 1990) making it an important agricultural source of starch and starch derivative products. Sweetpotato starch is highly utilized in pharmaceutical and food industries (Woolfe, 1992). The diversity of products derived from sweetpotato root depends on the quality and quantity of starch it contains (Lu *et al.*, 2006). Starch is a polysaccharide of amylose and amylopectin. Quality of sweetpotato starch based products is dependent on the amylose to amylopectin ratio. This ratio also controls the textural and cooking properties of sweetpotato root (Collado, 1999). Starch granule size and distribution determines cooking time and quality of starch noodle (Lu *et al.*, 2006). Sweetpotato starch is useful in biscuits, cakes, juices, ice cream and noodles production (Brabet *et al.*, 1998; Woolfe, 1992). In Japan two novel products, cyclodextrin and oligosaccharide, for food and pharmaceutical industries have been developed from sweetpotato starch

(Woolfe, 1992). Production of glucose syrup from sweetpotato starch and endogenous amylases has been documented (Hagenimana *et al.*, 1996; Hagenimana and Simard, 1994). Varieties of high starch content are used for the production of high quality starch for industrial application. Starch content has a significant positive correlation with DM. Brabet *et al.* (1998) reported a correlation of 0.92,  $p < 0.001$  between starch and DM for 106 sweetpotato clones. Their studies recommended the use of DM content to select clones with high starch content, since the method for determination of DM is simple, fast and relatively cheap. This recommendation is particularly useful when large numbers of clones are being assessed.

### **2.3 Sugar content and sweetness of sweetpotato roots**

Sweetpotato roots contain high concentrations of sugars compared to other root and tuber crops. The sugar content can be as high as 20% dwb in raw tissues and 50% dwb in cooked roots (Wang and Kays, 2003; Morrison *et al.*, 1993). There are different types of sugars in sweetpotato roots. In raw uncooked roots sucrose, glucose, and fructose predominate whilst maltose is the major sugar in cooked sweetpotato products (Kays *et al.*, 2005; Lewthwaite *et al.*, 1997; Morrison *et al.*, 1993) via the hydrolytic activity of native amylase. Sugar content of sweetpotato root is a fundamental component of its eating quality, and preferences vary with ethnicity and geographical location (Kays *et al.*, 2005 and Lewthwaite *et al.*, 1997). Tumwegamire *et al.* (2011) reported that Sub-Saharan Africans prefer less sweet sweetpotato types whilst consumers in developed country like sweet types. The principal reason for consumers to purchase sweetpotato in developed countries is sweetness (Takahata *et al.*, 1994). The majority of clones in United States are either high or very high in sweetness (Kays *et al.*, 2005). Sugars

significantly influence overall flavour of cooked sweetpotato roots and products (Sun *et al.*, 1993; Horvat *et al.*, 1991). Sugars also act as precursors in the synthesis of several key aroma compounds (Wang and Kays 2000).

Individual sugars have different sweetness levels. Sugars at the same concentration have different taste sensation (Lewthwaite *et al.*, 1997; Shallenberger, 1993). It has been established that sucrose has relative sweetness of 1.0; glucose 0.74, fructose 1.73 and maltose 0.33; referred to as sucrose equivalent (SE) (Laurie *et al.*, 2012). Hence comparisons in sweetness among genotypes require both quantification of individual sugars and adjustment for their relative sweetness with reference to sucrose (Laurie *et al.*, 2012; Kays *et al.*, 2005).

### **2.3.1 Classification of sweetpotato roots based on sweetness and sugar content**

Kays *et al.* (2005) categorised the world's sweetpotato germplasm into five groups based on the concentrations and relative sweetness expressed as Sucrose Equivalent (SE) of individual sugars in g/100g dry weight in baked sweetpotatoes: very high  $\geq 38$ , high 29-37, moderate 21-28; low 12-20, nonsweet  $\leq 12$ . Only 9% of the countries surveyed cultivated very high sweet types whilst the majority (62%) cultivated high sweet clones. None of the clones were in the low or non-sweet SE category. Maltose accounted for 42%, whilst sucrose, glucose and fructose contributed 44%, 6%, and 8% of the average percentage of the sucrose equivalent (SE) of 272 clones respectively (Kays *et al.*, 2005).

Sweetpotatoes can also be classified as staple or non-staple based on amount of sugar and sweet perception of the cooked root. Clones of 2% and 5%



(Lewthwaite *et al.*, 1997) sugar concentration or less than 12 SE for instance (Kays *et al.*, 2005) are classified as staple and supplementary staple, whilst those of higher sugar content are termed luxury or dessert types. Morrison *et al.*, (1993) also made a similar classification and grouped sweetpotatoes into four major classes according to initial sugar content and starch hydrolytic potential during cooking. The categories were; low initial sugar concentration/low starch hydrolysis, low initial sugar concentration/high starch hydrolysis, high initial sugar concentration/low starch hydrolysis, high initial sugar concentration/high starch hydrolysis. The first group has low endogenous sugars (sucrose, glucose and fructose) present prior to cooking and produces little or no maltose upon cooking. Such varieties are also classified as non sweet (Kays *et al.*, 2005). The second group produces an appreciable amount of maltose during cooking via the action of amylases (Takahata *et al.*, 1995) resulting in a slightly or moderately sweet product. The third group is quite similar to second group however; there is no or little increase in sugar content during cooking. The fourth group consists of varieties of high initial sugar content coupled with high starch hydrolytic potential. They are typically very sweet dessert types. Jewel variety of 20.6% initial sugar content and 31.0% after baking is a typical example of this group (Morrison *et al.*, 1993)

#### **2.4 Amylolytic potential of sweetpotato root.**

Sweetpotato roots unlike other root and tuber crops contain high extractable amylases (mainly  $\alpha$ -amylase) in addition to high starch content (Dziedzoave *et al.*, 2010; Hagenimana and Simard, 1994). Sweetpotato amylases have important and well-documented influences on fresh sweetpotato root and processing qualities (Dziedzoave *et al.*, 2010; Hagenimana *et al.*, 1996, Hagenimana and



Simard, 1994, Morrison *et al.*, 1993). Amylases are mainly responsible for the breakdown of starch into maltose and short chain oligosaccharide (limit dextrins) resulting in the sweet taste of sweetpotato roots

(Dziedzoave *et al.*, 2010; Lewthwaite *et al.*, 1997; Hagenimana *et al.*, 1996).

Amylase activity is also crucial during starch thinning in brewing and sugar syrup production (Dziedzoave *et al.*, 2010). Sweetpotato flour of high  $\beta$ -amylase activity has been used successfully to increase saccharification in sorghum brewing in place of exogenous enzymes (Etim and EtokAkpan, 1992). Hagenimana and colleagues (1996) used endogenous sweetpotato amylases to hydrolyse 70% and 60% starch of Regal and White Delight sweetpotato varieties at 70°C after 60min of incubation. In the U.S.A.,  $\beta$ -amylase is commercially produced from sweetpotato root for industrial utilisation (Woolfe, 1992).

Dziedzoave *et al.* (2010) identified high  $\beta$ -amylase sweetpotato varieties for glucose syrup production. Hagenimana *et al.* (1996), further showed that sweetpotato endogenous amylases are capable of hydrolyzing starches into maltodextrin, and glucose syrup.

Although there are many efficient methods of starch hydrolysis for energy production, the cost of manufacturing and recent ecological concerns of microbial enzymes for instance pose a challenge (Dziedzoave *et al.*, 2010).

Hence the need to utilise enzymes from plant sources including sweetpotato, which are relatively cheaper and readily available, especially for small-scale indigenous agro-processing firms (Hagenimana *et al.*, 1996). There are three major types of enzymes in sweetpotato root;  $\alpha$ -amylase,  $\beta$ -amylase and starch phosphorylase (Dziedzoave *et al.*, 2010; Hagenimana and Simard, 1994).

#### 2.4.1 Beta-amylase

$\beta$ -amylase is the predominant amylase in sweetpotato root (Chang *et al.*, 1996; Hagenimana and Simard, 1994; Nakamura *et al.*, 1991). It is distributed throughout the root tissue (Hagenimana and Simard, 1994). Despite its higher level, its physiological role in insoluble starch degradation of sweetpotato root is not clear since it is ineffective in attacking raw starch granules (Hagenimana and Simard, 1994).  $\alpha$ -amylase attacks the penultimate glycosidic bond from the nonreducing end of solubilised starch, releasing maltose and low molecular weight limit dextrins (Morrison *et al.*, 1993). The action of  $\beta$ -amylase primarily produces maltose, which contributes to the sweet taste of traditional cooked sweetpotato root.  $\alpha$ -amylase activity in sweetpotato is highly dependent on genotype, maturity period, storage condition and time, and sprouting period (Dziedzoave *et al.*, 2010; Hagenimana and Simard, 1994; Takahata *et al.*, 1995; Morrison *et al.*, 1993). Sumor and Jewel lines displayed high  $\beta$ -amylase concentration of 125  $\mu\text{g/ml}$  and 120  $\mu\text{g/ml}$  respectively compared to 20  $\mu\text{g/ml}$  and 4  $\mu\text{g/ml}$  of '99' and '86' lines following storage (Morrison *et al.*, 1993). Dziedzoave *et al.*, (2010) reported significant differences in  $\alpha$ -amylase activities among four sweetpotato varieties in Ghana at different growing stages. Activity was found to be higher after five months maturity (407.7U/g) than four and half months (302.5 – 366.40U/g) (Dziedzoave *et al.*, 2010). Sweetpotato produced in the forest zone generally showed higher  $\alpha$ -amylase activity than in the savannah zone.  $\alpha$ -amylase activity generally increases during sprouting and storage of sweetpotato roots (Nabubuya *et al.*, 2012; Hagenimana and Simard, 1994; and Morrison *et al.*, 1993).

### 2.4.2 Alpha-amylase

Alpha-amylase is responsible for breakdown of insoluble starch granules to soluble malto-dextrins in plants (Hagenimana and Simard, 1994). It is the only enzyme generally accredited with the ability to bind and attack raw food products. Synthesis of alpha-amylase increases during development, storage and sprouting (Nabubuya *et al.*, 2012; Takahata, *et al.*, 1995; Morrison *et al.*, 1993). In sweetpotatoes,  $\alpha$ -amylase increased in the outer starchy parenchymatous tissues surrounding the cambium layers, suggesting a *de novo* synthesis of the enzyme in cambium or lenticifer layers (Hagenimana and Simard, 1994). Hagenimana and Simard (1994) observed an increase in  $\alpha$ -amylase after 9 days of sprouting, which corresponded with an increase in in-vitro activity measurement. Alpha-amylase activity rose in three sweetpotato varieties during storage and reached to maximum after 90 days (Takahata, *et al.*, 1995). The increase corresponded with a decrease in starch content. Alpha amylase activity is also influenced by size of roots. Large root sizes (6-9cm in diameter) exhibited lower activity compared to small sizes (Carrilho, 1996).

### 2.5 Factors influencing quality attributes of sweetpotato roots

Quality attributes of sweetpotato roots are influenced by factors such as genotype, storage conditions, and cooking treatments (Wang and Kays, 2000; Wang *et al.*, 1998; Lewthwaite *et al.*, 1997; Takahata *et al.*, 1995). Maturity period, environment conditions and their interaction are also known sources of variation (Adu-kwarteng *et al.*, 2014; Dziedzoave *et al.*, 2010; Grüneberg *et al.*, 2005)

### 2.5.1 Genotype and Maturity effects

Genes and hormonal systems responsible for synthesis and accumulation of food reserves differ among crops. In sweetpotato, varying sugar content for example involves altering: genes responsible for the formation of starch, which influences latent pools of mono - and disaccharides; and genes controlling starch hydrolysis and maltose formation (Kays *et al.*, 2005). Sugar content of sweetpotato is genotype dependent (Aina *et al.*, 2009). Assessing the relative sweetness of 272 sweetpotato clones of the world revealed substantial genetic diversity within the genepool (Kays *et al.*, 2005). Majority of the clones (45.58%) were found to be high in sweetness with sucrose equivalent (SE) ranging from 29–37 g/100g dwb. About 33% were in the moderate SE category (21–28g/100g) while 18% were classified, as very high sweet ( $\geq 38$  g/100 g), only seven clones were low, and one non-sweet. Lack of sweetness in sweetpotato can be accounted for by two phenomena: low background levels of initial sugars; and low levels of  $\beta$ -amylase activity for starch conversion during cooking (Morrison *et al.*, 1993).

Quality attributes in sweetpotato roots are also highly affected by maturity period. Delayed harvesting increased total soluble sugar (TSS) content in a study of Ghanaian sweetpotato varieties (Adu-Kwarteng *et al.*, 2014). TSS contents were highest at 5 months maturity for all four cultivars (Hi-starch, Sauti, Otoo and Faara) evaluated. It ranged from 7.36 to 10.33g/100 g on dry weight basis. Sucrose content increased from 3 months to 5 months maturity whilst fructose and glucose concentration varied depending on genotype. Time of harvest also affected TSS in six cultivars grown in Brazil (Woolfe, 1992). Higher concentration of sugars was recorded in roots harvested 6 months after planting than roots harvested at 4 or 8 months (Woolfe, 1992).



### 2.5.2 Curing and Storage effects

Sweetpotato roots are usually placed in a warm humid environment to promote healing of wounds incurred during harvesting and transportation. This process is known as curing. Curing increases the shelf life and preserves the quality attributes of roots after harvest (Woolfe, 1992). It is usually conducted at 30-33°C and very high relative humidity (85-95%) for 5 – 7 days (Woolfe, 1992). Curing can also take place at low relative humidity, but not all genotypes are responsive to this. Wound healing efficiency may be measured by assessing the continuity of lignified layers formed (lignification score), and can be an indicator of storage potential of sweetpotato cultivars (van Oirschott *et al.*, 2006). Curing of the sweetpotato roots prior to storage is a common practice in especially the temperate zones (Kays *et al.*, 2005, Wang and Kays 2000, Sun *et al.*, 1993; Koehler and Kays, 1991). Curing occurs naturally in the tropics following harvesting as the atmospheric conditions present the right temperature required for curing. Amylase activity and sugars increase during curing, resulting in a sweet and moist product when baked (Woolfe, 1992).

Storage, which follows curing, also induces significant changes in quality attributes of sweetpotato roots. Dry matter content, sugars, and amylase contents are highly affected by storage and storage conditions (Woolfe, 1992). Sucrose, dextrins, alpha amylase and sucrose synthase generally increased during storage of Benihayato, Koganesengan and Kyushu 91 cultivars (Takahata *et al.*, 1995). Amylase activities increased dramatically following 90-day storage but decreased constantly up to 150 days (Morrison *et al.*, 1993). However, changes in individual and total sugars were relatively minor. Woolfe (1992) reported an increase in



sucrose, glucose and fructose contents and a decrease in maltose content of three sweetpotato cultivars when baked after curing and storing at 15.6°C and 90% relative humidity for thirty weeks. Reduction in maltose content was attributed to the formation of enzyme inhibitors altering the activity of beta amylase responsible for maltose formation during cooking.

### **2.5.3 Cooking and cooking treatment**

Although many factors significantly affect quality characteristics of sweetpotato roots, cooking, which causes gelatinization of starch and activation of endogenous amylases for starch degradation, is among the most significant factors affecting quality (Morrison *et al.*, 1993). Cooking changes the texture and flavour profiles of sweetpotato roots. It also increases the digestibility of starch, and reduces the levels of toxic terpenoids, phytoalexins and anti-nutritional trypsin inhibitors (Woolfe, 1992). Sugar content of Sumor sweetpotato variety increased from 8.09% to 26.30% while Jewel reached 36% when baked (Morrison *et al.*, 1993). A total sugar content of 52.78%, 59.50%, and 61.01% were determined in baked Jewel, Centennial, and GA 90-56 respectively (Wang and Kays, 2003). Due to the significant changes of sugar content (mainly maltose) induced by cooking treatment, determination of total available sugars in sweetpotato root should be made after cooking. Consideration should however be given to the conditions (intensity and amount of heat energy) used during cooking, since enzyme activity and starch degradation depend on these parameters. Enzyme activity, for instance, is determined by temperature of reaction medium and time of reaction. Maltose concentration increases rapidly above 50°C, reaching a maximum at 80 – 85°C

(Sun *et al.*, 1993). Changes in reducing sugars were not observed when sweetpotato roots were heated at  $\leq 65$  or  $\geq 85$  °C (Sawai *et al.*, 2009).

Baking, microwaving, boiling, and steaming are the basic methods employed during cooking of sweetpotato roots (Wang and Kays, 2000). Substantial amounts of sweetpotato roots are also fried in some countries, including Ghana. These methods occur at different temperatures and times. Baking is usually performed at 180 - 220 °C for 60 – 90 min, depending on the genotype and tuber size (Chan *et al.*, 2012). Microwave utilizes high temperature short time heating regime to cook food products. Heat is transferred by convection and conduction during baking, whilst electromagnetic waves penetrate food materials causing agitation and friction to produce heat for cooking during microwaving (Wang and Kays, 2000). Conventional baking lasting for an hour usually produces more reducing sugars than microwaving which lasts for about 5 minutes (Sun *et al.*, 1993). The most distinguishing differences between microwave and the other cooking methods are the short heating time, low ambient temperature in the oven, water vapour migration to the surface of the food and rapid heating throughout the sample (Schiffmann, 1994; Woolfe, 1992). Boiling, the preferred cooking method in some countries tends to reduce sugars through leaching (Sun *et al.*, 1993). Boiling is less expensive compared with other methods like frying (Woolfe, 1992). Literature on steaming of sweetpotato roots is obscure. Although sweetpotatoes are cooked by baking, boiling, microwaving, steaming or frying (Wang and Kays, 2000), the most widespread method is baking (Chan *et al.*, 2012). Future research should therefore explore the effect of other cooking methods vis-à-vis common techniques like baking and make recommendations appropriately.

#### **2.5.4 Environmental and Genotype by environment interactions on quality attributes of sweetpotatoes**

Environmental and genotype by environment interactions have been identified as major sources of variation in yield performance and to a lesser extent, quality attributes in crops (Thangavel and Eswaran 2011; Astra *et al.*, 2009). Performance assessment is usually conducted across more than one location in breeding, with the aim of identifying varieties that perform well across environments or in a particular environment (Ceccarelli *et al.*, 2006). Differences in phenotypic expression among genotypes in multi-locational evaluation are mainly due to the interaction between the genotypes and the growing environment (Navabi *et al.*, 2006). Hence genotype and environmental means alone cannot explain all the variations observed in diverse environmental trials. GxE effects have been reported for mainly yield related parameters such as yield and yield components. However, limited information is available on environmental influences on root quality attributes of sweetpotato genotypes, especially sugar profile, starch content and amylase activity. GxE effects on amylase activity have not been reported. Sweetpotato is grown in diverse environments and hence is subject to environmental variation (Manrique and Hermann 2000; Ngeve, 1993). Grüneburg *et al.* (2005) investigated environmental and GxE effect on root quality and nutritional components (root dry matter, starch, and  $\beta$ -carotene; and leaf carotene and chlorophyll) of nine sweetpotato genotypes and concluded that GxE interaction was smaller than genetic variation. Studying the effect of environment on  $\beta$ -carotene concentration showed that the higher the altitude, the higher the concentration

(Manrique and Hermann 2000).

## **2.6 Measurement of quality attributes in sweetpotato roots**

Quality attributes of sweetpotato roots can be measured by both quantitative and qualitative methods. Quantitative measurement employs high performance liquid chromatography (HPLC), Rapid Visco-Analyser (RVA), and spectroscopy, among others (Lu *et al.*, 2006) for measuring specific attributes. These methods are termed traditional methods. Attributes of direct impact on sensory properties and acceptability are usually assessed qualitatively by consumers or trained panel (Laurie *et al.*, 2012; Leksrisonpong *et al.*, 2012). Trained panels develop descriptors for defining individual characteristics influencing sensory perception (Leksrisonpong *et al.*, 2012).

Traditional methods recognised by scientific bodies including Association of Analytical Chemists (AOAC) for quantifying attributes, though very accurate, have some limitations (Bonierbale *et al.*, 2008; Lu *et al.*, 2006, Brenna and Berardo, 2004). High costs of analysis coupled with time required confine the use of spectrophotometer and HPCL to limited number of samples as against large numbers in extensive research or breeding programmes (Bonierbale *et al.*, 2008). Colometric methods for measuring starch properties are prone to interlaboratory variability because of the complexity of the procedure and its reliance on amylose and amylopectin standards for establishing reference curves (Bao *et al.*, 2001). RVA can measure a maximum of five samples per hour (Lu *et al.*, 2006) and HPLC analyses only 5 samples per day (Boniebale *et al.*, 2008). These laboratory methods are too laborious, expensive, complex and time consuming for effectively screening large number of samples. Hence rapid, simple, but accurate method(s)



for evaluating experimental materials in efficient fashion would be welcome. Among such methods is the Near-Infrared Reflectance Spectroscopy (NIRS). NIRS technology is based on the absorption of electromagnetic radiation in the near-infrared region (1100 - 2500 nm) by specific chemical bonds in the sample. A prediction model is then developed between the absorption values (spectral data) and reference values of the same samples. The model is validated and used for estimating the composition of similar samples.

NIRS technology is employed in large-scale routine analysis of some quality traits of many crops because it saves time, and is economical because reagents are not required (Lebot *et al.*, 2011; zum Felde *et al.*, 2007; Lu *et al.*, 2006, Wu *et al.*, 2002; Velasco *et al.*, 1998). It also permits simultaneous determination of multiple traits in one measurement (Lu *et al.*, 2006). In addition, it is an environmentally friendly system because no waste emerges. These qualities make it an ideal choice for rapid estimation of quality attributes (Lu and Sheng, 1990). NIRS technology has been used successfully in screening sweetpotato for dry matter,  $\beta$ -carotene and starch content (Lebot *et al.*, 2011; zum Felde *et al.*, 2009, Ishiguro and Yamakawa, 1998; Katayama *et al.*, 1996; Lu and Sheng, 1990). In 2006, Lu and colleagues, successfully developed NIRS calibration models for determining starch quality parameters and protein content with high coefficients of determination ( $RSQ=0.85-0.92$ ). zum Felde *et al.* (2009) developed NIRS calibration models for screening  $\beta$ -carotene, iron, zinc, starch sugars, protein in sweetpotato germplasm. The calibration showed high prediction accuracy, and it was used to evaluate 1209 sweetpotato accessions in Peru. Results of Lebot *et al.* (2011) further confirmed the potential use of NIRS for estimating starch, sugar, protein and mineral content of sweetpotato roots.

Although NIRS technology has been extensively employed in determining quality attributes in sweetpotato roots, its performance on cooked roots has not been tested. This current study intends to examine this gap by developing a prediction model for evaluating sugars and starch content in cooked sweetpotato using different cooking methods.

## **2.7 Sensory evaluation of quality attributes in sweetpotato roots**

Sweetpotato quality attributes are also evaluated through sensory analysis. Both trained and consumer assessors can be used in sweetpotato screening for desired traits (Leksrisonpong *et al.*, 2012; Chan *et al.*, 2012). The success of any new sweetpotato line depends not only on agronomic traits but also on sensory and utilisation potentials (Tomlins *et al.*, 2007). Taste preference varies with ethnic and geographical locations (Kays *et al.*, 2005), hence selection of sweetpotato variety for a locality should take into account the taste perception of local consumers. However, given the complexity of judging the eating quality of sweetpotatoes using taste panels (Villareal *et al.*, 1979), Kays *et al.* (2005) proposed a mathematical way for assessing and comparing a key quality attribute (sweetness) of sweetpotato roots. This method involves quantifying individual sugars and adjusting their values to relative sweetness. Based on this method, the relative sweetness levels of 272 sweetpotato germplasm accessions from diverse regions of the world have been determined (Kays *et al.*, 2005). This method will also be employed in the study to evaluate the sweetness profile of eleven sweetpotato varieties.

## CHAPTER THREE

### 3.0 CARBOHYDRATE COMPOSITION AND HYDROLYTIC POTENTIAL OF SWEETPOTATOES [*IPOMOEA BATATAS* (L.) LAM.] IN

#### DIVERSE GROWING ENVIRONMENTS

##### 3.1 Introduction

Sweetpotato is an important crop in the world. It is currently attracting attention due to its high nutritional values, ease of cultivation, short maturity period, and its potential for combating food insecurity and generating incomes for poor farmers (Truong and Avula, 2010; Akoroda, 2009). Nonetheless, its per capita consumption is relatively low compared with other root and tuber crops like cassava and yam (Kays *et al.*, 2005; Grabowski *et al.*, 2008). Instability of the root quality attributes such as sugars and amylase activity coupled with the predominant sweet flavour of the processed products are reputed to be among the key factors (Morrison *et al.*, 1993; Sun *et al.*, 1993; Kays *et al.*, 2005). The challenge of sweetpotato as a non-staple crop in many parts of the world is thus not a production constraint but rather lack of expandable utilisation potential (Truong and Avula, 2010).

Sweetpotatoes are usually cultivated in different agro-ecological zones even within a country and hence the occurrence of environmental and genotype by environment interactions (GxE) on the root quality attributes are inevitable (Ceccarelli *et al.*, 2006; Navabi *et al.*, 2006; Manrique and Hermann 2000).

However limited research has been conducted on GxE effects and environmental influences on sweetpotato root quality attributes, particularly sugar profile and amylase activity. Grüneburg *et al.* in 2005 investigated environmental and GxE effects on yield and nutritional components (root dry matter, starch, and

βcarotene) of nine sweetpotato genotypes and concluded that GxE interaction was smaller compared to genetic variation. Dziedzoave *et al.* (2010) investigated effects of two growing environments on β-amylase activities of four sweetpotato varieties, but conclusions were based on genotypic and environmental means alone. The objective of the present study was to determine the effects of genotype, environment and GxE on dry matter content, starch content, sugar profile (sucrose, glucose, and fructose), and amylase activity of eleven officially released sweetpotato varieties in five different production environments.

## **3.2 Methodology**

### **3.2.1 Experimental design**

A factorial design with three replications was employed for the study. The factors were sweetpotato varieties and production environments. The environments were Fumesua (forest zone), Ejura (forest-transition zone), Komenda, Ohawu and Pokuase all in the coastal savannah zone. Eleven varieties were planted in a randomised complete block design in each environment at experimental stations of the Council for Scientific and Industrial Research-Crops Research Institute (CSIR-CRI), Ghana, in 2013. Cultivation was rain-fed, and standard cultural practices were employed. Roots were harvested at four months after planting, and representative undamaged marketable roots selected for evaluation.

### **3.2.2 Materials**

Eleven sweetpotato varieties including Apomuden, Hi-Starch, Okumkom, Santom Pona, Ogyefo, Faara, and Sauti, Ligri, Patron, Bohye and Dadanyuei were selected for the study. The varieties were released by CSIR-CRI, Fumesua, Ghana.



### 3.2.3 Processing of root samples

Six undamaged marketable roots (size: 200 g to 400 g) of each variety from each replicate were selected from the experimental fields after harvest, and transported to the postharvest laboratory of the International Potato Centre (CIP) at Crops Research Institute, Fumesua, for processing. The roots were washed under running clean water, rinsed, air-dried for 6 hours and packed in paper bags. The cleaned roots were then kept for a maximum of seven days at C to 32 °C prior to processing. During processing, four cleaned roots of each sample were peeled with stainless peeler (ceramic knife, Kyocera, USA), quartered longitudinally, and two opposite quarters sliced into 0.3 cm to 0.5 cm thick (Fig 3.1). Approximately 50 g of each sliced sample was weighed into a low-density polyethylene bag and quickly transferred into a freezer of -25 °C. Samples for amylase assay were not peeled. Frozen samples were freeze-dried using a Trueten Biotech freeze dryer (YK-118/50, Taiwan) for 72 hours, milled by a Thomas scientific laboratory miller (3383-L70) and sieved through a 40-mesh screen.

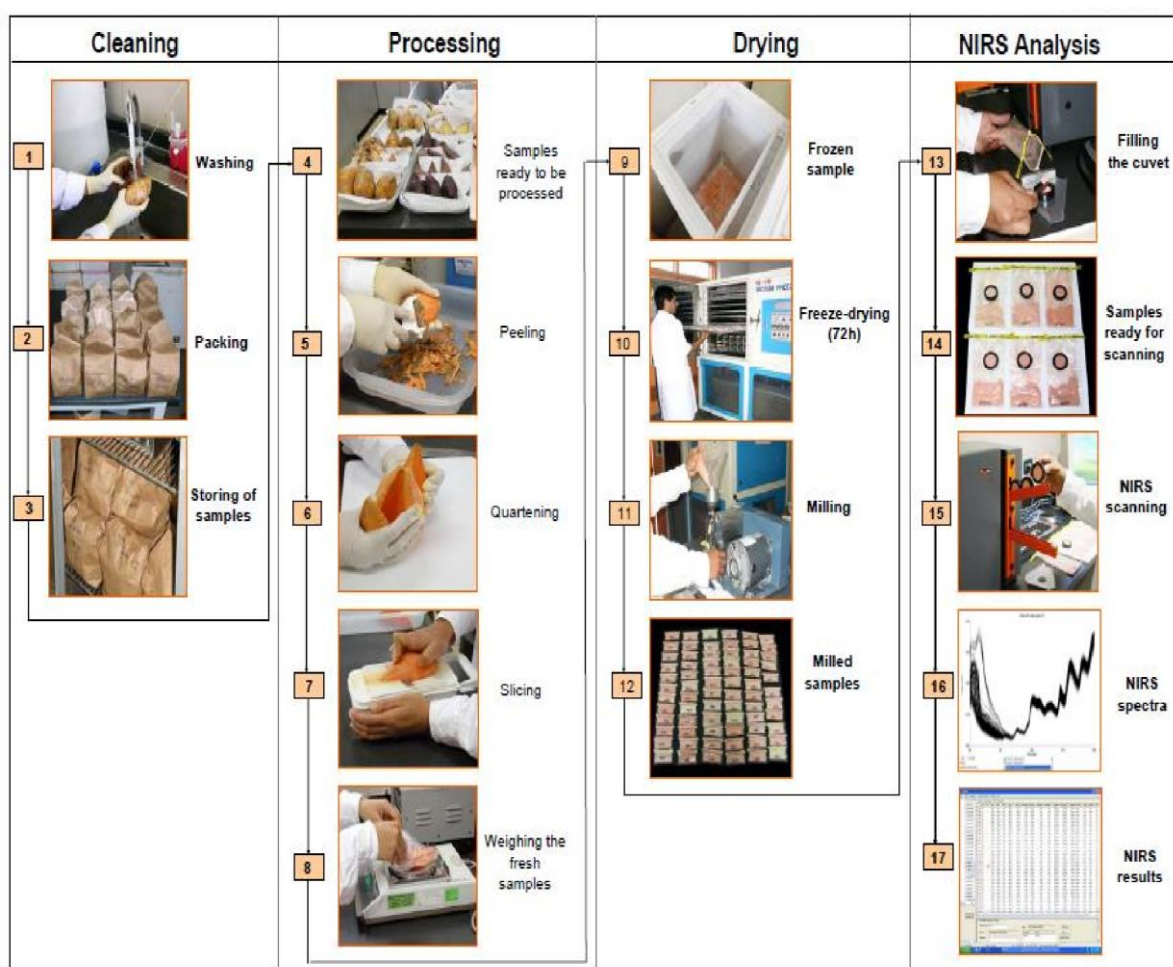


Fig 3.1. Flow diagram of sample preparation and NIRS analysis of sweetpotato roots quality attributes.

Adapted from International Potato Center (CIP) - Quality and Nutrition Laboratory, Lima, Peru.

### 3.2.4 Dry matter determination

Fifty grams (50g) of each replicate sweetpotato sliced roots sample was weighed into a low-density polyethylene bag and frozen quickly at  $-25^{\circ}\text{C}$  for 24 hours. The frozen samples were transferred into stainless steel plates and loaded into a pre-cooled freeze dryer (Trueten Biotech; YK-118/50, Taiwan). The samples were dried for 72 hours at which the dried weight was constant. The dried samples were weighed and values recorded. The dry matter content was calculated from the formula (AOAC, 2000);

$$\% \text{ Dry matter content} = \frac{W_1 - W_2}{W_3 - W_2} \times 100$$

Where

$W_1$  = weight of fresh sweetpotato roots and polybag (g)

$W_2$  = weight of poly bag (g)

$W_3$  = weight of dried sweetpotato roots and polybag (g)

### 3.2.5 Sugars and starch content determination

Individual sugars including sucrose, glucose and fructose and starch were determined using Near Infrared Reflectance Spectroscopy (NIRS) technology. NIRS Calibration developed by zum Felde *et al.*, (2009) for raw sweetpotato roots was employed for this determination.

### 3.2.6 Total sugar and sweetness determination

Total sugar determination was based on summation of sucrose, glucose and fructose (Lewthwaite *et al.*, 1997; Morrison *et al.*, 1993). Individual sugars in the raw sweetpotato roots were converted into their relative sweetness with reference to sucrose using the conversion equation: 1.2 fructose + 1 sucrose + 0.64 glucose (Kays *et al.*, 2005).

### 3.2.7 Determination of amylase activity.

The 3,5-dinitrosalicylic acid (DNSA) method for reducing sugars (OwusuMensah *et al.*, 2010; Osman, 2002) was employed to determine the total amylase activity in the freeze-dried sweetpotato roots.

### ***Extraction of sweetpotato amylases***

Exactly 0.75 g of milled sweetpotato root sample was weighed and transferred into a centrifuge tube. 4 mL of sodium-phosphate buffer (pH 8) was added to the sample while mixing using a vortex (MX-S) at ambient temperature. The mixture was then allowed to stand for 30 min with intermittent vortexing after which it was centrifuged for 10 min at 2000 g. The supernatant was decanted, and the volume recorded. One part of the supernatant was diluted with 50 parts of 0.1 M malate buffer (pH 5.5) and used as the sweetpotato amylase extract (SAE). All the extraction processes were performed at  $25 \pm 2$  °C.

### ***Preparation of equilibrated soluble starch***

Approximately 5 g soluble standard starch (Sigma cat. no. S565) was macerated with 100mL of 0.1M malate buffer at pH 5.5. The resultant mixture was heated for 10 minutes at 75 °C – 80 °C to obtain equilibrated starch solution. The solution was then cooled to a temperature of between 55 °C and 60 °C and used for the hydrolysis in the determination of the sweetpotato amylase activity (Osman, 2002).

### ***Determination of total amylase activity***

To 0.25mL pre-equilibrated soluble starch solution, 0.05mL of appropriately diluted SAE was added while mixing and the mixture was incubated for 10 minutes at 60°C. The reaction was terminated by adding 2 mL of 0.1M NaOH solution. 1.0 mL freshly prepared DNSA reagent was added, mixed and heated for 5 min at 80°C. After cooling for appropriately 40 min, the absorbance of the mixture as well as the control and the standard was read at 480nm. The standard and control were treated similarly except that the SAE was added to the control



after the addition of NaOH (Osman, 2002). Amylase activity was calculated according to the following formula:

$$U/g = ABt/ABs*600*EV*DF/10/0.05/0.75/342.3 \text{ (Osman, 2002) in}$$

which,

ABt = the absorbance of test sample at 480nm.

ABs = the absorbance of maltose standard of 600 $\mu$ g.

EV = mL average volume of SAE extracted

DF = dilution factor

10 = min, incubation time in minutes.

0.05 = mL, volume of diluted amylase extract (SAE)

0.75 = g, weight of sweetpotato flour used in amylase extraction.

342.3 = molecular weight of maltose

A unit (U) of amylase activity was defined as the amount of enzyme required to release reducing sugars equivalent to one  $\mu$ mole of maltose/min under the above stated conditions.

### 3.2.8 Statistical Analysis

Experimental means were calculated from triplicate values of each treatment and subjected to analysis of variance using Statistical Analysis System (SAS, 2007). Separation of means was done using LSD at 5% probability level. The additive main effects and multiplicative interaction (AMMI) analysis was used to analyse GxE effects on the attributes measured. The patterns of contribution of varieties and environments to GxE variance were graphed using principal component analysis (PC).

### 3.3 Results and discussion

#### 3.3.1 Variability of genotype, environment and interactions on dry matter content of sweetpotatoes

The percentage variability resulting from genotypic, environmental and interactions (GxE) on root quality attributes of sweetpotato varieties screened is presented in Table 3.1. Generally, the effects of genotype, environment, and genotype by environment were all significant for dry matter content.

Statistically, genotype accounted for the highest proportion of the total variation (92.44%) followed by environment (4.46 %) and GxE (3.09 %). Compared to other quality attributes viz., starch and amylase activity, genotypic influence was more profound on dry matter content (Table 3.1).

**Table 3.1: Percentage variance by genotype, environment and GxE on dry matter, starch, sugars and amylase activity of sweetpotato varieties**

Source of Variation	* Variance (%)					
	Dry matter	Starch	Fructose	Glucose	Sucrose	Amylase activity
<b>Genotype</b>	92.44**	88.26**	83.07 **	84.23 **	88.34 **	65.89 **
<b>Environment</b>	4.46**	5.84**	13.67 **	11.96 **	2.80 **	10.01 **
<b>Genotype x Environment</b>	3.09**	5.90**	3.26 ns	3.81ns	8.45 **	24.06 **

\*\* Significant at  $p < 0.05$ . ns = not significant. \* Calculated from sum Sq.

The results indicate that dry matter content of sweetpotatoes is largely dependent on the genotypic make-up of the variety and less on environment. This finding agrees with report by Moussa *et al*, (2011) which showed that dry matter is greatly influenced by genotype and years of planting compared to environment, which

was not significant. Similarly, genotype by environment interaction (GxE) has been reported to be relatively smaller to genotypic variation for sweetpotatoes (Grüneburg *et al* (2005). It should be noted however that in this current study, though GxE and environmental effects are minimal, their values are statistically significant and hence their contribution to variability in dry matter content of sweetpotatoes should be considered especially in multilocal designs.

The percentage variability resulting from the interactions (GxE) was thus further analysed using the Additive Mean Effect and Multiplicative Interaction (AMMI) model and partitioned into four portions based on Principal Component Analysis (PCA). The percentage variability based on sum of squares were 68.42%, 16.70%, 9.46% and 5.42% for PC1, PC2, PC3 and PC4, respectively (Table A3, Appendix). PC1 had the highest and it was employed to explain the interactions variability observed (Fig 3.2).

#### **3.3.1.1 Dry matter content of the eleven sweetpotato varieties in five production environments**

Dry matter contents of the eleven sweetpotato varieties in five production environments are showed in Table 3.2. Apomuden variety planted at Pokuase environment had the lowest value of 17.93% while Hi-Starch at Ejura environment yielded the highest dry matter content of 46.24%. Significant differences among the varieties were highly remarkable compared to differences between the environments per variety.

**Table 3.2. Mean dry matter content (%) of 11 sweetpotato varieties across**

**five production environments**

Sweetpotato variety	Production sites				
	Ejura (%)	Fumesua (%)	Komenda (%)	Ohawu (%)	Pokuase (%)
<b>Apomuden</b>	20.27 <sup>a</sup> (1.03)	21.45 <sup>b</sup> (0.90)	20.81 <sup>a</sup> (1.54)	17.93 <sup>c</sup> (1.45)	20.62 <sup>a</sup> (0.67)
<b>Bohye</b>	32.90 <sup>a</sup> (0.96)	33.18 <sup>a</sup> (1.45)	28.67 <sup>b</sup> (0.82)	30.06 <sup>c</sup> (0.58)	31.31 <sup>d</sup> (1.01)
<b>Okumkom</b>	35.23 <sup>b</sup> (0.77)	34.80 <sup>b</sup> (1.02)	31.33 <sup>a</sup> (0.65)	28.48 <sup>c</sup> (0.60)	33.23 <sup>d</sup> (1.33)
<b>Patron</b>	35.56 <sup>c</sup> (0.89)	36.13 <sup>c</sup> (1.55)	32.50 <sup>b</sup> (0.88)	32.90 <sup>b</sup> (1.39)	34.04 <sup>a</sup> (1.42)
<b>Santom Pona</b>	36.78 <sup>a</sup> (1.08)	35.23 <sup>b</sup> (1.33)	33.57 <sup>c,d</sup> (0.51)	32.56 <sup>c</sup> (1.44)	34.35 <sup>d</sup> (2.03)
<b>Dadanyuie</b>	35.24 <sup>b</sup> (1.31)	38.97 <sup>a</sup> (2.04)	33.18 <sup>c</sup> (1.56)	33.33 <sup>c</sup> (0.44)	32.63 <sup>c</sup> (2.01)
<b>Ligri</b>	36.51 <sup>a</sup> (1.78)	38.10 <sup>b</sup> (2.23)	33.41 <sup>c</sup> (1.05)	33.19 <sup>c</sup> 1.17	36.24 <sup>a</sup> (0.89)
<b>Faara</b>	36.93 <sup>c</sup> (1.89)	40.63 <sup>b</sup> (1.34)	36.91 <sup>c</sup> (1.01)	38.07 <sup>a</sup> (0.91)	37.83 <sup>a,c</sup> (1.55)
<b>Sauti</b>	35.51 <sup>b</sup> (1.28)	40.41 <sup>c</sup> (3.45)	38.37 <sup>a</sup> (2.76)	40.06 <sup>c</sup> (3.45)	36.98 <sup>d</sup> 1.56
<b>Ogyefo</b>	36.69 <sup>a</sup> (2.34)	42.10 <sup>b</sup> (1.54)	36.58 <sup>a</sup> (1.29)	38.96 <sup>c</sup> 1.78	37.44 <sup>a</sup> 2.91
<b>Hi-Starch</b>	46.24 <sup>b</sup> (1.45)	45.75 <sup>a</sup> (2.09)	44.22 <sup>c</sup> (2.78)	44.24 <sup>c</sup> (3.01)	43.65 <sup>c</sup> (2.01)

Values with the same superscript in a row are not significantly different at  $p < 0.05$ . Standard deviation are presented in brackets

Apomuden and Hi-Starch varieties for instance recorded the lowest and highest dry matter values respectively across all the production environments. In a similar observation, dry matter value (33.10%) for Dadanyuie variety at

Komenda environment was not significantly different from the value obtained at Ohawu (33.32%) and Pokuase (32.63%) environments. Dry matter contents of Bohye, Okumkom, Patron and Ligri varieties were not significantly different in



two environments. Apomuden, Dadanyuie, Faara, Ogyefo and Hi-Starch varieties had statistically the same dry matter values in three environments. This observation confirms early finding (Table 3.1) that dry matter is less dependent on environmental controls.

The overall mean dry matter contents for each variety computed from all the environments, and environmental means calculated from all varieties planted in a particular environment are presented in Fig. 3.2. In terms of variety, Apomuden had the lowest mean dry matter content of 20.22% and Hi-Starch, the highest of 44.82%. Overall mean dry matter content of Ogyefo, Faara, Sauti, and Ligri ranged from 35% to 38% whilst contents of Bohye, Okumkom, Patron, Santom Pona and Dadanyuie varieties were between 31% and 35%. Mean dry matter content of Faara was not significantly different from Sauti and Ogyefo. Similarly mean dry matter content for Patron, Santom Pona and Dadanyuie were statistically the same. Dry matter contents of majority of the varieties were close to the grand mean of 34.77%.

Results obtained for dry matter content revealed that majority of sweetpotato varieties evaluated have high to very high dry matter contents. The grand mean value (34.77%) exceeded average worldwide and East African sweetpotato dry matter contents of 30% and 32% respectively (Tumwegamire *et al.*, 2011; Woolfe, 1992). With regards to production environment, Komenda had the lowest overall mean dry matter content of 33.60%, which was not significantly different from that of Ohawu (33.62%) and Pokuase (34.39%). Fumesua environment had the highest mean dry matter content of 36.98% and was significantly different from all the other environmental means. The relatively high organic matter composition of the soil from the Fumesua environment in the forest zone (Table A10,

appendix) coupled with the high rainfall pattern could have played a role in the high dry matter contents of the varieties since dry matter is basically an organic mass.

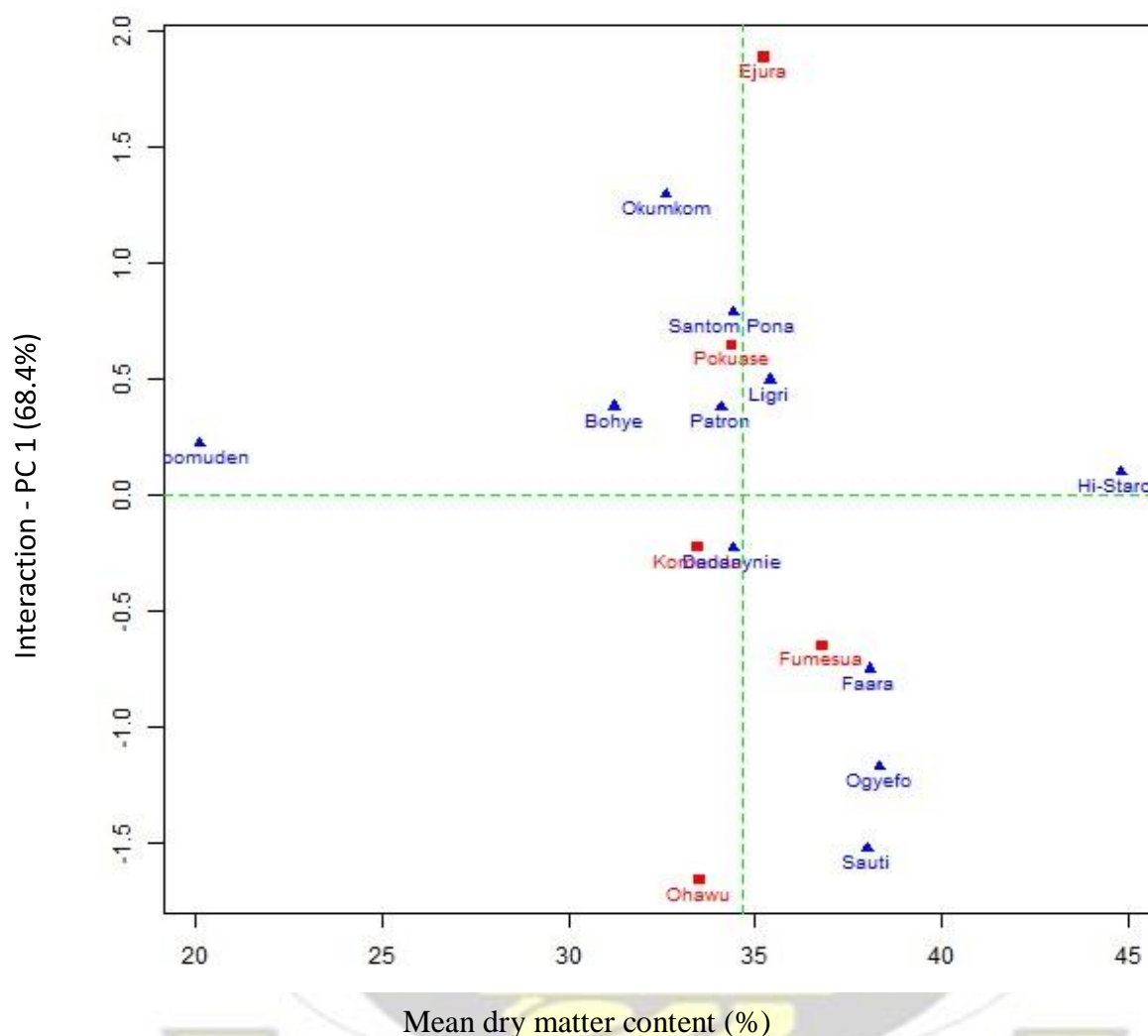


Fig 3.2. Dry matter content of sweetpotato varieties and interactions with environments. ▲ = Variety ■ = Environment.

### 3.3.1.2 Genotype by environment (GxE) Interaction effect on dry matter contents of eleven sweetpotato varieties

Interactions between varieties and environments, and within the varieties and environments are presented in Fig 3.2. The proximity of Apomuden and HiStarch to the origin (0,0) indicated that dry matter contents of these varieties were very stable across the environments. They exhibited less variation and hence produced

dry matter values, which were not significantly different between the production environments. Okumkom and Sauti showed the highest interaction effects of +1.3 and -1.5, respectively. The remaining six varieties recorded minimal interaction scores ranging from -1 to +1. Okumkom, Santom Pona, Ligri, Patron, Apomuden and Hi-starch were located on the same side of the PC1 axis, indicating that they had a similar interaction pattern to the environments. Okumkom and Sauti, on the other hand, were located on opposite side of the graph showing different response patterns to the environments with regards to accumulation of dry mass. Santom Pona, Ligri, Patron and Bohye were located close to Pokuase environment and hence exhibited positive interactions whilst Dadanyuie exhibited similar pattern to that of the Komenda environment. Similarly, the interactive effect of Faara was parallel to that of Fumesua environment. Komenda, Fumesua and Ohawu environments were located on the same side of the interaction axis, which showed that these environments displayed similar interactive outputs on dry matter content of sweetpotato roots. The result from GxE analysis establishes that dry matter content of Apomuden and Hi-Starch varieties are less dependent on the major production environments evaluated, and thus the effect of environment can be ignored when dry matter content is the main attribute of interest. Okumkom and Sauti varieties showed high deviation across the environments. The effect of GxE should therefore be considered regarding the dry matter content of these sweetpotato varieties.

### **3.3.2. Effects of genotype, environment and GxE on starch content of sweetpotatoes**

The results from Table 3.1 depict that genotype, environment and GxE significantly influenced starch content of the sweetpotatoes. Genotype contributed

the highest of 88.26% whilst environment and GxE together accounted for 11.74% of the total variation. According to Grüneburg *et al.* (2005), effect of GxE interaction on starch content of sweetpotatoes is usually smaller compared to genetic variability. From this result, it could be deduced that starch content of sweetpotato roots is highly dependent on the genotypic composition of the variety and that differences in production environments do not substantially, though significant, determine its content. A similar observation was also noticed in the dry matter content evaluation, which indicates that these two quality attributes respond similarly when sweetpotatoes are subjected to different production environments.

#### **3.3.2.1 Starch content of eleven sweetpotato varieties in five production environment**

Starch content of sweetpotato varieties across the five production environments are presented in Table 3.3. Starch content ranged from 34.79% to 75.15%; with Apomuden and Hi-starch recording the lowest and highest mean values across the environments. Starch content values were not substantially different, though significantly ( $P < 0.05$ ), from each other across the environment for most of the varieties. The lowest starch content values were observed in Ohawu production environment for majority of the varieties, which resulted in its lowest overall mean (Fig 3.3). On the other hand, Fumesua and Pokuase environments produced sweetpotato varieties with significantly higher starch contents (Fig 3.3). This implies that environmental conditions at these production environments are favourable for synthesis of starch in majority of the sweetpotato varieties.



**Table 3.3: Mean starch content (% dry weight basis) of 11 sweetpotato varieties across five production environments**

Sweetpotato varieties	Production environment				
	Komenda	Ohawu	Pokuase	Ejura	Fumesua
<b>Apomuden</b>	46.11 <sup>a</sup> (2.45)	46.75 <sup>a</sup> (1.98)	45.88 <sup>a</sup> (2.31)	34.79 <sup>b</sup> (1.67)	47.01 <sup>a</sup> (1.52)
<b>Bohye</b>	61.23 <sup>d</sup>	68.09 <sup>c</sup>	61.49 <sup>b</sup>	67.93 <sup>c</sup>	65.75 <sup>d</sup>
<b>Dadanyuie</b>	65.31 <sup>e</sup>	67.81 <sup>c</sup>	68.42 <sup>c</sup>	71.62 <sup>d</sup>	69.36 <sup>c</sup>
<b>Faara</b>	62.55 <sup>a</sup>	70.21 <sup>e</sup>	64.54 <sup>d</sup>	69.46 <sup>e</sup>	67.79 <sup>f</sup>
<b>Ligri</b>	67.43 <sup>a</sup>	69.53 <sup>c</sup>	67.66 <sup>a</sup>	68.74 <sup>a,c</sup>	67.72 <sup>a</sup>
<b>Okumkom</b>	63.58 <sup>c</sup>	56.82 <sup>b</sup>	65.86 <sup>a</sup>	64.08 <sup>c</sup>	64.99 <sup>c,a</sup>
<b>Patron</b>	64.81 <sup>b</sup>	69.42 <sup>e</sup>	64.80 <sup>b</sup>	69.14 <sup>c</sup>	60.92 <sup>a</sup>
<b>Santom Pona</b>	67.15 <sup>a</sup>	64.28 <sup>b</sup>	70.11 <sup>c</sup>	67.30 <sup>a</sup>	66.93 <sup>a</sup>
<b>Sauti</b>	70.95 <sup>e</sup>	68.63 <sup>f</sup>	69.26 <sup>f,g</sup>	70.20 <sup>e,g</sup>	70.07 <sup>b</sup>
<b>Ogyefo</b>	73.10 <sup>c</sup>	73.32 <sup>c</sup>	71.03 <sup>b</sup>	74.13 <sup>c</sup>	74.93 <sup>e,a</sup>
<b>Hi-Starch</b>	67.79 <sup>c</sup>	73.08 <sup>b</sup>	75.15 <sup>a</sup>	74.05 <sup>e,b</sup>	74.93 <sup>e,a</sup>
	(2.05)	(1.71)	(1.63)	(2.03)	(1.81)

Values with the same superscript in a row are not significantly different at  $p < 0.05$ . Standard deviation are presented in brackets.

### 3.3.2.2 Interaction effect on starch content of sweetpotato varieties

Results from the analysis of variance (ANOVA) on genotype by environment interactions of sweetpotato starch content are presented in Table A5 (appendix). The outcome showed that the first and second principal components (PC1 and 2) were significant at  $p < 0.05$ . PC1 explained 53.34% and PC2 28.32% of the total variation respectively. The biplots of the two principal components are presented in Fig 3.3 and

Fig 3.4.



was farthest from the origin, in the opposite direction from Ohawu, which was also far from the origin, due to its high contribution to GxE.

Hi-Starch, given its distance from the origin, also contributed fairly strongly to GxE, its placement opposite the origin from Komenda, due to its lower starch content at Komenda. Contrast to negligible effect of GxE on dry matter content of Hi-Starch and Apomuden (Fig 3.2), starch content of these varieties was strongly influenced by GxE, hence the difference in percentage variability of GxE on dry matter and starch content (Table 3.1) In general, the outcome of the study shows that starch content of Ghanaian sweetpotato varieties is relatively high, a trend which reflects the eating pattern of most of the populace in Ghana who obtain a higher percentage of their energy requirement from carbohydrates in roots and tubers. Moreover, starch synthesis was significantly dependent on GxE during growth stages of sweetpotato varieties.

The stability and performance of the starch content of each variety in an environment were also investigated (Fig 3.4). The outcome reveals that starch content of Santom Pona, Ligri, Faara, Dadanyuie and Bohye were the most stable across the five production environments since their PC scores were close to the origin. Hi-Starch and Apomuden varieties were the least stable. For dry matter content, Hi-Starch and Apomuden were the most stable varieties. This sharp contrast in stability performance between starch content and dry matter indicates that, differences resulting from GxE effect varied substantially among quality attributes. Santom Pona, Okumkom and Apomuden were in the same quarter with Fumseua, Pokuase, and Ejura environments indicating their favourable responses to these environments. Patron and Hi-starch clustered in the quadrant opposite

Komenda environment. Moreover, Fumesua, Pokuase and Ejura are located in the same quarter of the biplot.

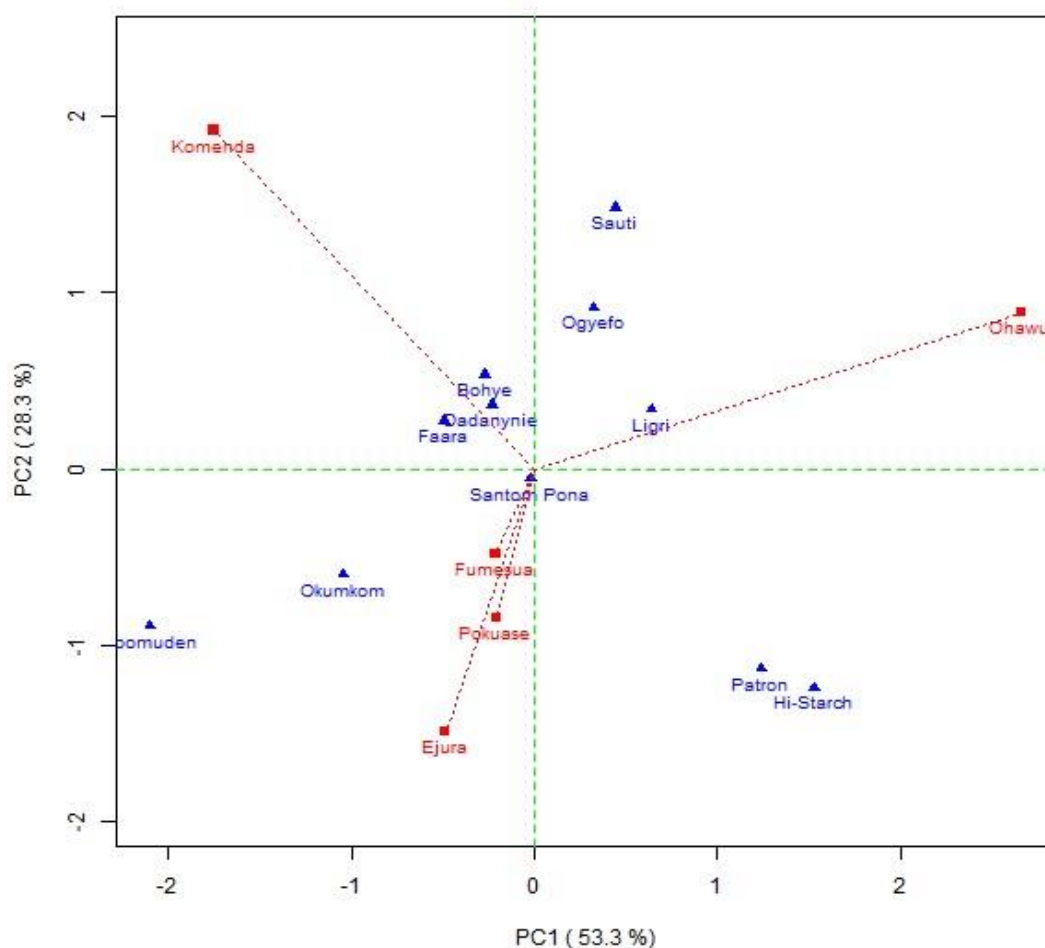


Fig 3.4. Stability and performance of sweetpotato varieties over environments on starch content. ▲ = Varieties ■ = Environments

This observation shows that these environments are not significantly different from each other and therefore produce similar effect on starch accumulation of sweetpotatoes during growth. However, the consistency of the effect of these environments on root starch content and their relevance to sweetpotato production and utilization over several seasons will require further study.

Starch content is considered as an important attribute influencing cooking and textural qualities of sweetpotato products (Lu *et al.*, 2006; Brabet *et al.*, 1998). It



determines the quality of sweetpotato starch-based food products (Collado, 1999). Varieties with high starch content such as Hi-Starch, Ogyefo, and Sauti would be useful for starch production provided their yields were high enough to compete with other starch sources, and if their starch quality attributes are suitable for intended utilization. Short maturity period and wide adaptation will make sweetpotato an economically viable raw material source for starch production compared to other root and tuber crops. High starch varieties can also be used for production of flour for bread and other pastries since less energy will be required for dehydration compared to varieties with lower dry matter and starch content. Glucose syrup and confectioneries including candies can be manufactured from the high starch and high dry matter varieties. Varieties with high starch content are usually used as staples, and typically have good eating qualities.

### **3.3.2.3 Correlation between starch content and dry matter content**

Starch content showed strong positive correlation ( $r=0.864$ ,  $p<0.05$ ) with dry matter (Fig 3.5). Hi-Starch, which had the highest dry matter content for instance, also had the highest starch content while Apomuden of the lowest dry matter content also recorded the lowest starch content (Table 3.2 and 3.3). The trend was similar for majority of the varieties. This finding support early reports that dry matter content is mainly starch constituent for many root and tubers crops (Woolfe, 1992). Hence for a rapid screening of quality attributes, starch content could be estimated from dry matter content values using the equation:  $y=0.7279x - 13.161$ . However, factors other than starch may also contribute differentially to the dry matter content of sweetpotato.

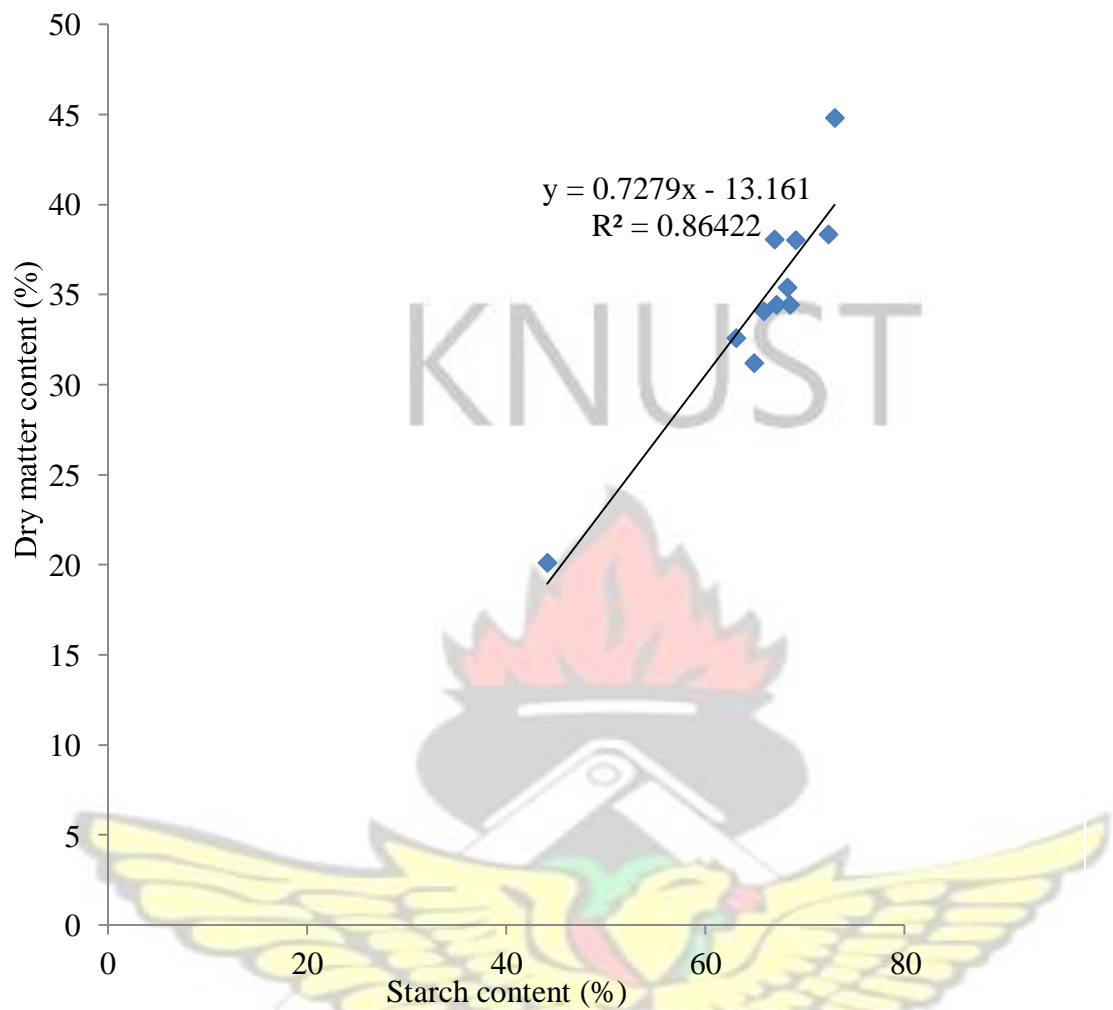


Fig 3.5. Correlation between dry matter content and starch content of sweetpotato roots

### 3.3.3 Effect of genotype, environment and GxE on sugars of sweetpotatoes

Predominant sugars identified and quantified in the uncooked sweetpotato roots were sucrose, glucose and fructose (Table 3.4). These sugars have already been identified by a number of food scientists including Kays *et al.*, (2005), Lewthwaite *et al.*, (1997) and Morrison *et al.*, (1993) as the main sugars in the uncooked roots.

The synergistic effect of these sugars control

**Table 3.4 Overall mean individual and total sugar content (%dwb) of 11 sweetpotato varieties.**

Sweetpotato variety	Sugars (%dwb)			
	Sucrose	Glucose	Fructose	*Total Sugar
<b>Apomuden</b>	23.20 (3.67) <sup>a</sup>	8.87 (2.57) <sup>a</sup>	5.32 (2.23) <sup>b</sup>	37.38 (3.82) <sup>c</sup>
<b>Okumkom</b>	11.39 (2.14) <sup>b,d</sup>	5.17 (2.89) <sup>b</sup> (2.78) <sup>c</sup>	3.23 (3.40) <sup>d</sup>	19.79
<b>Patron</b>	14.37 (2.78) <sup>c</sup>	2.64 (2.71) <sup>c</sup>	1.25 (1.99) <sup>d</sup>	18.27 (4.22) <sup>a</sup>
<b>Bohye</b>	11.92 (2.11) <sup>b</sup>	3.56 (2.67) <sup>d</sup>	1.94 (2.09) <sup>e</sup>	17.41 (3.07) <sup>a,b</sup>
<b>Ligri</b>	11.93 (2.64) <sup>b</sup>	3.03 (2.39) <sup>e</sup>	1.47 (2.15) <sup>f</sup>	16.43 (2.51) <sup>b,e</sup>
<b>Dadanyuie</b>	10.17 (1.89) <sup>d,e</sup>	3.96 (3.16) <sup>d</sup>	2.21 (3.20) <sup>g</sup>	16.35 (2.43) <sup>b,e</sup>
<b>Faara</b>	9.67 (2.03) <sup>e</sup>	3.76 (2.78) <sup>d</sup>	2.04 (1.78) <sup>g</sup>	15.47 (2.37) <sup>e</sup>
<b>Santom Pona</b>	10.66 (1.98) <sup>d,e</sup>	3.03 (1.72) <sup>e</sup>	1.13 (2.98) <sup>f,d,i</sup>	15.38 (2.07) <sup>e</sup>
<b>Sauti</b>	8.50 (1.25) <sup>e,f</sup>	2.57 (1.60) <sup>c,f</sup>	1.13 (0.65) <sup>f,d,i</sup>	12.20 (2.51) <sup>f</sup>
<b>Hi-Starch</b>	8.70 (0.74) <sup>e,f</sup>	2.21 (2.18) <sup>f</sup> (0.53) <sup>i</sup>	0.86 (1.43) <sup>f</sup>	11.77
<b>Ogyefo</b>	9.34 (1.03) <sup>e,f</sup>	1.55 (1.78) <sup>g</sup> (0.42) <sup>j</sup>	0.34 (3.96) <sup>f</sup>	11.23

Standard deviations are presented in brackets. \* Total sugar content was calculated from the three main sugars in raw sweetpotato roots, namely; sucrose, glucose and fructose. Values with the same superscript in column are not significantly different at  $p < 0.05$ .

the overall sweet sensation of the root (Kays *et al.*, 2005). The concentration of the three main sugars in the sweetpotato roots during growth was evaluated in the production environments under study. Results from the analysis of variances (Table 3.1) showed that except for sucrose, genotype by environment (GxE) interaction was not significant for sugars in uncooked sweetpotato roots. Genotypic effect was highly significant ( $P < 0.05$ ) and contributed the highest (83% to 88%) of the overall variation in the sugars. Environmental effect accounted for 13.67% and 11.96% in fructose and glucose contents, respectively. Only 2.80% of the variance

observation in sucrose content resulted from the differences in the production environments. It could therefore be deduced from this study that genotypic composition rather than interaction with environmental conditions significantly controls synthesis of sugars during development of sweetpotato roots. Hence research programmes aiming at altering the sugar levels of sweetpotato roots during growth should target the genomics rather than other possible influential factors such as soil fertility.

#### **3.3.3.1 Genotypic effect on sugars and sweetness of sweetpotato varieties**

Percentage mean sugar viz., fructose, glucose and sucrose as well as total sugar content in uncooked roots of sweetpotato varieties screened in the current study are shown in Table 3.4. Sucrose was the predominant sugar in all the varieties and ranged from 8.50% (Sauti) to 23.20% (Apomuden). Glucose content was lowest (1.55%) in Ogyefo variety and highest in Apomuden variety (8.87%). Fructose was the lowest sugar and the minimum value, 0.34%, was recorded in Ogyefo and maximum content, 5.32%, in Apomuden. The summation of the three major sugars (total sugar) yielded between 11.23% to 37.38% for Ogyefo and Apomuden respectively. Apart from Apomuden, all the varieties had less than 20% total sugar content. A wide variation was observed between the sugar profile of Apomuden variety and the rest of the varieties. Similar findings confirming the dominance of sucrose in raw sweetpotato roots has been reported (Lewthwaite *et al.*, 1997; Morrison *et al.*, 1993).

In order to estimate the overall sweetness of the sweetpotato varieties, individual sugars were converted into sucrose equivalent (SE) based on relative sweetness factors (Table 3.5). The SE of individual variety ranged from 10.72% (Ogyefo) to



35.25% (Apomuden). The overall mean for the varieties was 16.50%. In general about 71% of the SE was realised from sucrose whilst fructose and glucose contributed 14.20% and 14.23%, respectively. Fructose increased the total SE by 3.00% whilst glucose decreased the level by 6.83%, generally, based upon their sweetness factors (Shallenberger, 1993). Hence sweetpotato variety with high fructose concentration is likely to be sweeter than those with less fructose concentration.

Results from the sugars and sweetness evaluation established that majority of Ghanaian sweetpotato varieties are low in sweetness (13 – 20% SE). Apomuden was the only high sweet variety (35%) whilst Sauti, Hi-starch and Ogyefo can be classified as non-sweet ( $\leq 12$ ) based on the classification of Kays *et al.* (2005).

However, due to the formation of maltose via the action of  $\alpha$ -amylase (E.C. 3.2.1.1) and  $\beta$ -amylase (E.C.3.2.1.2) during cooking (Kay *et al.*, 2005; Takahata *et al.*, 1994), the sugar and sweetness levels may vary after cooking. Sugar content in Jewel and Sumor varieties for instance increased from 20.6% to 31.0% and from 8.1% to 26.3% during baking as reported by Morrison *et al.*, (1993) in their studies on sweetpotato varieties. On the other hand, baking treatment decreased the sugar concentration from 7.99% to 6.85% in the 99 sweetpotato line (Morrison *et al.*, 1993). Effect of cooking treatment on sugar profile and sweetness of the sweetpotato varieties was therefore evaluated extensively in Chapter five.

**Table 3.5. Mean sucrose equivalent (SE) of individual and total sugar (%) and levels of sweetness of 11 sweetpotato varieties**

Sweetpotato variety	Fructose	SE (%)		*Total	#Category of sweetness
		Glucose	Sucrose		
Apomuden	6.38	5.67	23.20	35.25	High

	(1.31)	(1.67)	(2.23)	(3.45)	
Okumkom	3.88	3.31	11.39	18.57	Low
	(1.27)	(0.43)	(2.31)	(3.71)	
Patron	1.51	1.69	14.37	17.57	Low
	(0.33)	(0.26)	(2.07)	(4.02)	
Bohye	2.33	2.28	11.92	16.53	Low
	(0.51)	(0.78)	(1.32)	(3.29)	
Ligri	1.77	1.94	11.93	15.63	Low
	(0.41)	(0.61)	(2.70)	(3.81)	
Dadanyuie	2.65	2.54	10.17	15.36	Low
	(0.34)	(0.12)	(2.34)	(4.10)	
Faara	2.45	2.41	9.67	14.52	Low
	(0.79)	(0.57)	(1.87)	(3.01)	
Santom	2.02	1.94	10.66	14.62	Low
Pona	(0.55)	(0.09)	(2.06)	(2.89)	
Sauti	1.36	1.64	8.50	11.50	Non Sweet
	(0.07)	(0.34)	(1.91)	(2.39)	
Hi-Starch	1.03	1.42	8.70	11.15	Non Sweet
	(0.31)	(0.12)	(1.88)	(2.78)	
Ogyefo	0.40	0.99	9.34	10.7	Non Sweet
	(0.02)	(0.22)	(1.73)	(2.32)	

\*Total SE was calculated from the three main sugars namely; sucrose, glucose and fructose. #

Categorisation was based on method adopted by Kays *et al.*, 2005. Standard deviation are presented in brackets.

### 3.3.4 Effects of genotype, environment and GxE on total amylase activity of sweetpotato roots.

Response of total amylase activity to genotype, environment and GxE interactions was significant at  $p < 0.05$  (Table 3.1). The genotypic effect was more profound and contributed 65.89% of the total variation. While environmental impact accounted for 10.01%, GxE resulted in 24.06% of the differences in the amylase activities of the varieties. Generally, wide variations were observed among the activities across the five production environments in all varieties (Table 3.6) compared to the other quality attributes including dry matter. This trend resulted in the highest GxE contribution as recorded in Table 3.1. Dadanyuie for instance obtained 949.67U/g of amylase activity in Ejura and 631.71U/g in Pokuase. A similar trend was noted in amylase activity of Apomuden, Santom Pona and Sauti.

It could also be inferred from the results (Table 3.6) that majority of the varieties evaluated contained substantial amounts of amylase and would therefore be a good source of amylase for industrial application. Varieties including Dadanyuie, Santom Pona and Ligri of high amylolytic power, generally, could be an excellent source of enzymes for glucose syrup, brewing, and like industries. Hagenimana *et al.*, (1996) stated that sweetpotatoes are potential source of maltodextrins, syrup, and glucose conversion. Low manufacturing cost and recent ecological barriers of microbial enzymes could make sweetpotato an attractive alternative source of amylases for most especially developing countries, which require less expensive and readily available source of amylase for their cottage food industries (Dziedzoave *et al.*, 2010). However, to maximise the activity of amylase in a preferred variety, cultivation should be done in an environment that facilitates high synthesis of amylases in that variety.

**Table 3.6. Mean amylase activity (U/g) of 11 sweetpotato varieties over five**

**production environments**

Sweetpotato varieties	Production environment				
	Ejura	Fumesua	Komenda	Ohawu	Pokuase
Apomuden	336.74 <sup>a</sup> (7.56)	454.10 <sup>b</sup> (6.34)	467.94 <sup>b</sup> (7.89)	602.93 <sup>c</sup> (5.67)	457.40 <sup>b</sup> (8.34)
Bohye	221.93 <sup>c</sup> (8.90)	387.06 <sup>b</sup> (10.56)	257.61 <sup>a</sup> (9.98)	487.98 <sup>d</sup> (6.83)	391.54 <sup>b</sup> (10.61)
Dadanyuie	949.67 <sup>b</sup> (11.45)	882.05 <sup>a</sup> (12.78)	764.99 <sup>c</sup> (12.71)	862.92 <sup>d</sup> (10.86)	631.71 <sup>c</sup> (9.08)
Faara	717.69 <sup>d</sup> (15.81)	678.32 <sup>c</sup> (17.02)	641.23 <sup>b</sup> (10.41)	794.95 <sup>a</sup> (9.73)	799.82 <sup>a</sup> (13.45)
Hi-Starch	229.05 <sup>a</sup> (4.98)	414.26 <sup>b</sup> (5.89)	226.73 <sup>a</sup> (5.76)	361.41 <sup>c</sup> (97.21)	391.91 <sup>d</sup> (6.67)
Ligri	722.92 <sup>b</sup> (8.55)	927.14 <sup>a</sup> (7.39)	826.50 <sup>c</sup> (14.21)	859.24 <sup>d</sup> (7.71)	643.20 <sup>c</sup> (5.61)
Ogyefo	841.58 <sup>c</sup> (11.89)	804.10 <sup>d</sup> (13.56)	723.32 <sup>e</sup> (8.89)	665.39 <sup>a</sup> (7.76)	613.46 <sup>b</sup> (6.19)
Okumkom	711.40 <sup>a</sup> (6.31)	779.25 <sup>b</sup> (7.71)	659.76 <sup>c</sup> (9.01)	827.40 <sup>d</sup> (14.61)	642.50 <sup>e</sup> (12.89)
Patron	444.04 <sup>c</sup> (4.32)	489.81 <sup>b</sup> (7.11)	422.98 <sup>c</sup> (5.57)	466.23 <sup>d</sup> (4.21)	247.15 <sup>a</sup> (3.78)
Santom Pona	935.89 <sup>b</sup> (7.89)	949.01 <sup>b</sup> (15.78)	649.63 <sup>a</sup> (12.67)	823.32 <sup>c</sup> (14.29)	644.37 <sup>a</sup> (13.32)
Sauti	678.36 <sup>c</sup> (8.81)	809.24 <sup>c</sup> (8.81)	632.67 <sup>b</sup> (8.91)	746.95 <sup>d</sup> (5.89)	557.13 <sup>a</sup> (7.89)

Standard deviations are presented in brackets. Values with the same superscript in a row are not significantly different at  $p < 0.05$ .



### **3.3.4.1 Genotype by environment interactions on amylase activity of sweetpotato varieties**

Analysis of the genotype by environment interactions revealed that the first four principal components (PC) were all significant, however the first (PC1) and the second (PC2) principal components accounted for the highest (86%) variability (Table 8A, Appendix). The biplot of PC1 versus mean amylase activity of the sweetpotato varieties and environments are presented in Fig 3.6. Dadanyuie and Santom Pona had the highest amylase activity but their distance from the origin indicated that they were not consistently high over all the environments unlike Ligri, which recorded a relatively high and stable amylase activity across the production environments (Fig 3.6). Mean activity values of Patron, Apomuden, Hi-starch, and Bohye were all lower than the grand mean. PC1 scores of Okumkom, Ligri, and Sauti genotypes were closer to the zero origin and hence showed little GxE. Fumesua environment recorded the highest mean value of 689U/g followed by Ohawu (682U/g), Ejura (616U/g), Komenda (570U/g) with Pokuase recording the lowest of 547U/g. The study revealed that, Komenda environment, with the lowest PC1 value, made little contribution to GxE. Therefore it could be selected as an environment for production of sweetpotato with consistent amylase content compared to the overall mean.

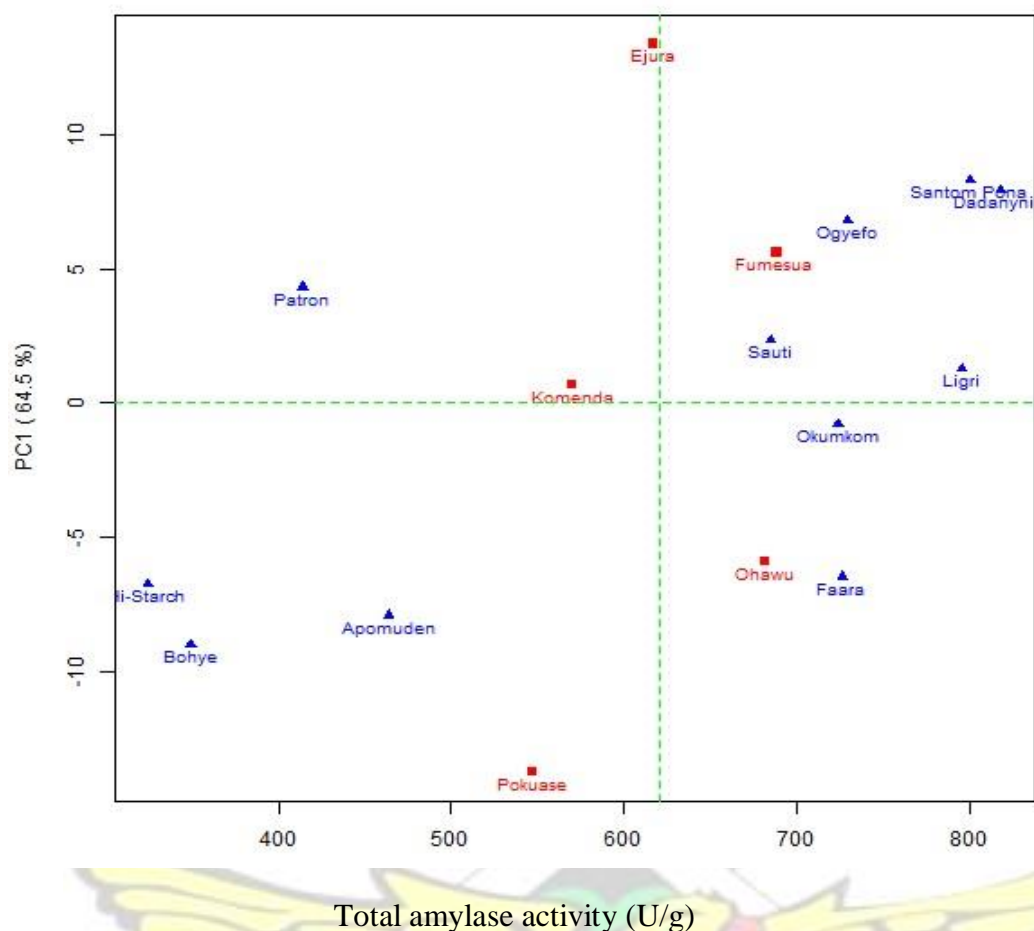


Fig 3.6. Sweetpotato amylase activity and their interaction levels in five production environments. ▲ = variety ■ = environment.

Fig 3.7 depicts the stability and the performance of the varieties and the production environments. The central location of Okumkom on the biplot makes it the stable variety. Ligri had a strong affinity for Komenda environment where its activity was not significantly different from the overall mean. Apomuden and Patron performed better in terms of amylase activity at Ohawu and Fumesua environments, respectively. Similarly, Dadanyuie, Ogyefo and Santom Pona performed better at Ejura environment. The amylase activity of Bohye, HiStarch and Faara were highest at Pokuase environment. Komenda and Fumesua were located in the same quadrant, showing similar contributions to GxE

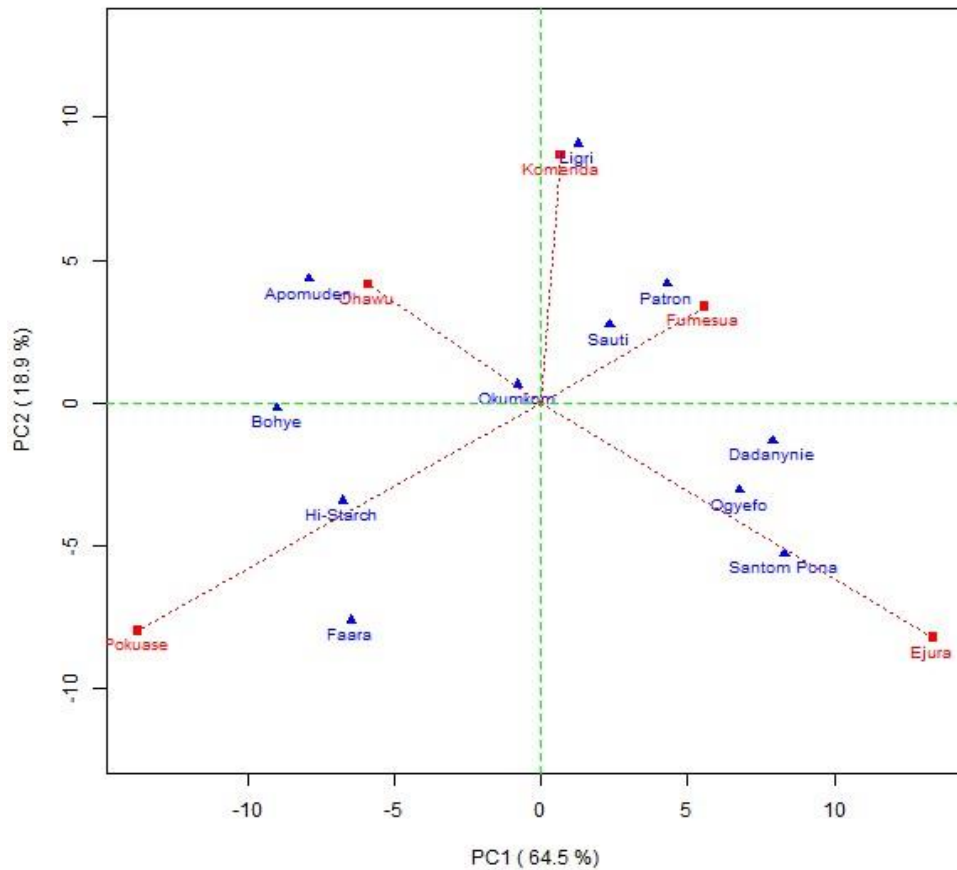
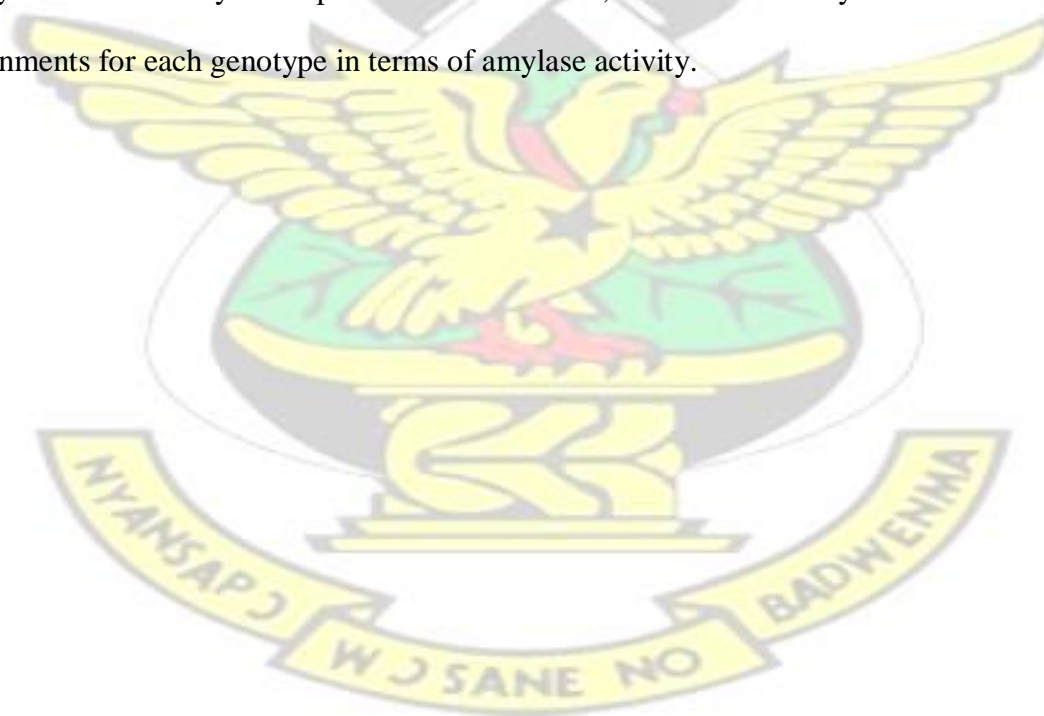


Fig 3.7. Stability and performance of sweetpotato varieties over environments on amylase activity. ▲ = Varieties ■ = Environments

The results from the study establish that amylase activity of sweetpotatoes is greatly controlled by the genotypic composition of the variety. Nevertheless contributions from environment and more especially GxE were highly significant and relatively high, in terms of magnitude of variability, among the key quality attributes (dry matter, starch and sugar contents) evaluated. Some varieties had higher affinity for specific environment(s) where they produced consistent amylase activity. Ligri and Apomuden for instance had higher amylase activities at Fumesua and Ohawu respectively. Such environments may have good soil and weather conditions conducive for higher synthesis of amylases in these varieties (Table 10A, appendix). Environmental conditions at Komenda hardly influenced the amylase activity of the roots. Komenda was hence selected as the most stable location (Fig 3.6).

Comparative analysis of overall means of amylase activity for individual environments may support earlier report by Dziedzoave *et al*, (2010) that amylase activity is highly dependent on production environment of which Fumesua (forest zone) produced the highest compared to coastal savannah (Okyereko). Overall mean amylase activity of sweetpotatoes planted at Fumesua in this study was higher than those planted at Pokuase (coastal savannah zone), and Komenda (coastal savannah) but was not significantly different from overall mean recorded at Ohawu, which is also in the coastal savannah zone. Means of genotypes and environments alone cannot completely explain GxE effects in multi-environment trials, (Thangavel *et al.*, 2011; Asrat *et al.*, 2009). Occurrence of GxE is inevitable in multi-locational trials (Ceccarelli *et al.*, 2006). AMMI biplots were therefore employed in this study to explore the interactions, and also identify suitable environments for each genotype in terms of amylase activity.





## CHAPTER FOUR

### 4.0 THERMAL STABILITY OF $\alpha$ -AMYLASE ACTIVITY AND SUGAR

#### PROFILE OF SWEETPOTATO VARIETIES DURING PROCESSING.

##### 4.1 Introduction

Amylases are of fundamental importance to processing and eating qualities of sweetpotato roots (Dziedzoave *et al.*, 2010; Kays *et al.*, 2005; Hagenimana *et al.*, 1996; Morrison *et al.*, 1993; Woolfe, 1992). Literature is replete with information on the presence and role of amylases in sweetpotato roots

(Nabubuya *et al.*, 2012; Dziedzoave *et al.*, 2010; Hagenimana *et al.*, 1994; Takahata *et al.*, 1994 and Morrison *et al.*, 1993). There are three major types of amylases in sweetpotato root;  $\alpha$ -amylase,  $\beta$ -amylase and starch phosphorylase. However  $\beta$ -amylase is the most abundant (Nabubuya *et al.*, 2012; Hagenimana and Simard, 1994; Takahata *et al.*, 1994) and important during processing (Morrison *et al.*, 1993). Sweetpotato flour with high  $\beta$ -amylolytic activity has been successfully used to increase wort extract of sorghum beer (Etim and EtokAkpan, 1992).

Utilisation of native amylases in intact roots involves heating the roots to gelatinise the starch fraction, and activate the amylases for starch degradation to maltose and limit dextrins (Sawai *et al.*, 2009; Sawai *et al.*, 2004; Hagenimana and Simard, 1994). Rapid heating of sweetpotato mashes to 70°C may be optimal for starch conversion and subsequent ethanol production (Hagenimana *et al.*,

1994). Takahata *et al.* (1994) investigated the effects of temperature on  $\beta$ amylase activity of six sweetpotato lines of varying maltose content and reported different response patterns between high, moderate and low maltose line.

However, the percentage variability of the temperature, time and more especially interactions with  $\beta$ -amylase activity were not fully investigated. This current study therefore sought to examine the effect of duration of heating and temperature on  $\beta$ -amylase activity and concentrations of other key sugars including sucrose during processing of four sweetpotato varieties. The presence and synergistic effect of  $\alpha$  –amylase, at relatively higher temperature, in sugar formation was also investigated. Such information would be useful in controlling free sugar composition during processing of sweetpotato roots.

## 4.2 Methodology

### 4. 2.1 Experimental Design:

A factorial design with three replications was used for all the experimental runs. The main factors were genotype, temperature, and heating time. Variables measured included  $\beta$ -amylase activity,  $\alpha$ -amylase activity, sucrose, maltose, glucose and fructose.

**Table 4.1. Amylase activity, dry matter content, and yield of the four sweetpotato varieties**

<b>Sweetpotato variety</b>	<b>*Amylase activity (U/g)</b>	<b>*Dry matter content (%)</b>	<b>+Yield (t/ha)</b>
<b>Apomuden</b>	484.13	21.90	30.00
<b>Santom Pona</b>	800.44	34.40	17.00
<b>Ligri</b>	791.57	34.00	18.40
<b>Hi-Starch</b>	324.74	40.00	18.00

\* Figures from Chapter three; + sourced from CRI, 2012.

#### 4. 2.2 Materials

Four sweetpotato varieties viz., Ligri, Santom Pona, Hi-Starch, and Apomuden with varying dry matter content, and yield (Table 4.1) were supplied by the Council for Scientific and Industry Research (CSIR) – Crops Research Institute (CRI), Fumesua, Ghana. They were planted in 2014 at the experimental field of CRI in Ohawu, the site that yielded relatively higher amylases activities generally (Fig 3.6) and moreover it was the site that was readily available for this experiment. The site falls within the coastal savannah agro-ecological zone. The roots were harvested at four months and stored under room conditions (25°C – 27°C) for two weeks prior to analysis.

#### 4.2.3 Methods

Four medium-sized intact roots of each variety were washed, rinsed and airdried. The roots were then quartered, rinsed in de-ionised water and dried with paper towel. Each quarter was sliced along its longitudinal axis into 1.0 cm thickness and composite samples within each variety divided into 13 subsamples of 50g each. One sub-sample was denoted as raw. The rest were tightly wrapped with aluminium foil and separated into four groups. Each group from the varieties were placed in a force-air temperature controlled oven (Genlab MINI/50/DKG) and heated at varying temperatures; 65°C, 75°C and 85°C for 0, 10, 20 and 40min. Termination of heating process was aided by submerging the sealed heated sweetpotato root samples into ice bath for 20min (Dutta *et al.*, 2006). The samples were peeled and frozen at -25°C to -28°C and freeze dried using Truten Biotech freeze dryer (YK-118/50, Taiwan) for 72 hours. The dried samples were milled using Thomas Scientific laboratory Miller (model 3383

L70) and sieve through 40 mesh receiver (Thomas scientific, cat no. 3383N20)

#### **4.2.4 Amylase extraction and determination**

##### ***Extraction of amylases***

To 0.5g of freeze-dried and milled sweetpotato root sample was added 5mL of 1M Tris/HCL buffer (pH 8.0) containing disodium EDTA (20mM) and cysteine HCL (100mM). Amylase was extracted for a maximum of 1hr with intermittent vortexing at room temperature. The extracts were obtained by centrifuging the mixture in a Shikang laboratory bench centrifuge (800D, China) at 4,000r/min for 10min. The supernatant was decanted into another labelled centrifuge tube. Approximately 0.1mL of the supernatant was added to 2.0mL dilution buffer (MES buffer, pH 6.2) and was used for total  $\beta$ -amylase assay. For  $\alpha$ -amylase, 1.5mL of sodium malate (1M, pH 5.4) was added. (Beta-amylase assay procedure 2010 and Alpha-amylase procedure, 2012, Megazyme International, Ireland)

##### ***Amylase assay***

Amylase extracts (0.1mL) of each sample was dispensed to 15mL polypropylene tubes and pre-incubated for 5min at 50°C and 60°C for  $\beta$ - and  $\alpha$ -amylase respectively together with the substrates; *p*-Nitrophenyl  $\beta$ -Maltotrioside (PNP  $\beta$ G3) – for  $\beta$ -amylase and Block *p*-nitrophenyl Maltoheptaoside (BPNPG7) – for  $\alpha$ -amylase. To each tube containing the amylase extracts, 0.1mL of the substrate solution was added, stirred and incubated at 50°C and 60°C for exactly 10min from time of addition. 1.5 mL of stopping reagents (1% Trizma base.) of pH 11.0 was added and stirred. The absorbance of the reaction mixtures and the reagent blanks were read at 400nm. One unit of activity was defined as the amount of



enzyme required, in the presence of excess  $\beta$ -glucosidase (for  $\beta$ -amylase) and  $\alpha$ -glucosidase (for  $\alpha$ -amylase), to release one micromole of *p*-nitrophenol from PNP $\beta$ -G3 or BPNPG7 in one minute under the defined assay conditions and termed a Betamyl-3 and Ceralpha Unit correspondingly. Betamyl-3 unit was converted to International unit (U/g) using a factor of 193.9. Ceralpha unit was also expressed as International unit (U/g) on starch substrate with a factor of 4.1 (Beta-assay procedure 2010 and Alpha-amylase procedure, 2012; Megazyme International, Ireland).

#### 4.2.5 Sugars determination

Freeze-dried and milled sweetpotato samples were sent to the Quality Plant Product Laboratory (Department of Crop Science, University of Gottingen, Germany) for analysis of sugars using High Performance Liquid Chromatography (HPLC). Water extract of the freeze-dried samples (0.1g in 100mL) was incubated in a water bath at 60°C for 1h and treated with 0.2mL Carrez I and Carrez II solution to remove proteins. The mixture was purified by centrifugation (Sorvall RC-5B Refrigerated Superspeed, GMI, Ramsay, USA) for 10min at 10000 rpm. Sugars were determined from the membrane-filtered supernatant (pores size 0.45  $\mu$ m). Glucose, fructose, sucrose, and maltose were separated using a LiChrospher 100 NH<sub>2</sub> (5  $\mu$ m) 4 x 4 mm pre-column in combination with a LiChrospher 100 NH<sub>2</sub> (5  $\mu$ m) 4 x 250 mm separation column (Merck KGaA, Darmstadt, Germany) and an acetonitrile: pure water solution (80:20 v/v) as mobile phase at a flow rate of 1.0 mL min<sup>-1</sup> at 20 °C and an injection volume of 20  $\mu$ L. Sugars were detected with a Knauer differential refractometer 198.00 (Knauer, Berlin, Germany).

#### 4.2.6 Statistical Analysis

Statistical Analysis System (SAS, 2007) was used to analyse the experimental data obtained from the triplicates samples. Means were separated using LSD at 5% probability level and results presented by graphs and tables.

#### 4.3 Results and discussion

##### 4.3.1 Temperature and time variability, and their interactions on $\beta$ -amylase activity of sweetpotatoes

The results of percentage variability due to temperature, time, genotype and their interactions on  $\beta$ -amylase activity during processing of sweetpotatoes are presented in Table 4.2. Temperature and genotype accounted for the largest proportion of the experimental variance of 50.37% and 25.94% respectively. Heating time contributed for 11.25% while genotype by temperature interaction accounted for 7.95%. Temperature by time interaction and genotype by time interaction account for 2.49% and 1.44% respectively. The combined interaction effect of all the factors was also significant and explained 0.56% of the variance observed.

**Table 4.2. Percentage variance of temperature, genotype, time and interactions on  $\beta$ -amylase activity of sweetpotato root**

**+Variance      F Value**

	(%)	
<b>Temperature (Temp)</b>	50.37**	40775.60
<b>Genotype (GT)</b>	25.94**	20996.90
<b>Time</b>	11.25**	9103.84
<b>GT*Temp</b>	7.95**	6433.85
<b>Temp*Time</b>	2.49**	2021.17
<b>GT*Time</b>	1.44**	1167.02
<b>GT*Temp*Time</b>	0.56**	452.29

\*\* Significant at p< 0.05. + Calculated from sum of squares.

These current findings correspond well with earlier reports that temperature has profound effect of  $\beta$ -amylase activity during processing (Takahata *et al.*, 1994). It decreased the activity of beta amylase when six sweetpotato lines of varying maltose content were heated. Hagenimama *et al.* (1994) reported similar findings that  $\beta$ -amylase activity decreased greatly when heated outside its optimum temperature. However, the percentage contributions from each of the factors (temperature, genotype and time) and their interaction have not been evaluated prior to this current work. The results of this study indicate that thermal stability of  $\beta$ -amylase in sweetpotato roots is also significantly determined by the interaction between variety and the processing conditions including temperature and time.

#### 4.3.2 Thermal stability of $\beta$ -amylase activity in four sweetpotato varieties.

Rates of thermal stability of  $\beta$ -amylase activity in four sweetpotato varieties profiled in the current study are presented in Fig 4.1 (A, B and C). Temperaturetime combined treatment generally had a negative effect on  $\beta$ -amylase activities in all the varieties. Activity generally decreased when temperature and heating time were increased. The effect was more profound at 75°C and 85°C for longer

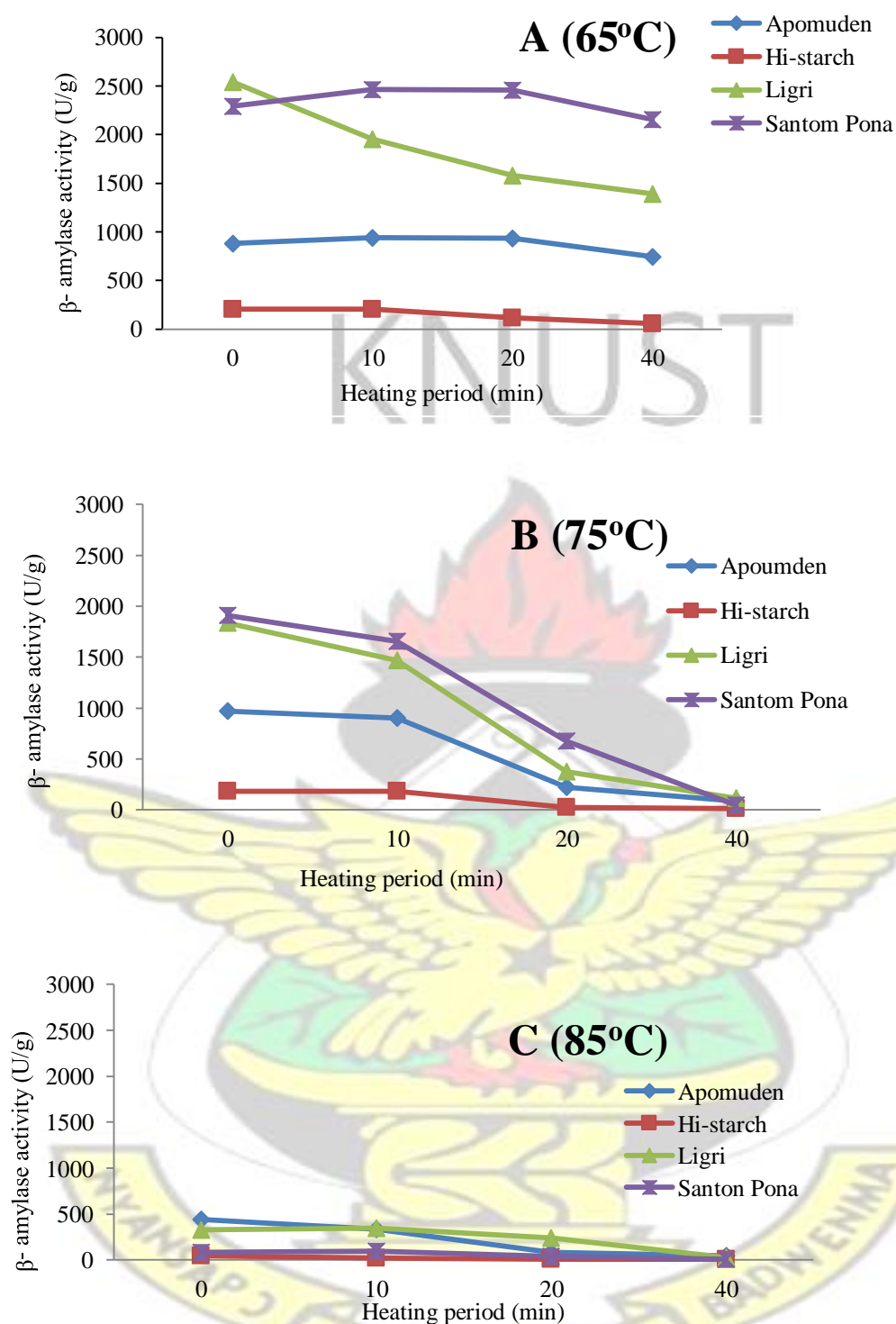


Fig 4.1. Changes in  $\beta$ -amylase activity during temperature-time heating regimes of four sweetpotato varieties: Santom Pona; Ligri; Apomuden; Hi-Starch. A – 65 °C; B - 75°C; and C - 85°C.

LSD=22.07

holding times (40 min), where all the varieties lost nearly all of their  $\beta$ -amylase activity.  $\beta$ -amylase activities were relatively stable at 65°C at the initial stages of



the heating. Variations were however observed among the varieties after 20min of heating. Santom Pona and Ligri varieties had the highest amylase activity, Apomuden moderate and Hi-Starch the lowest at the initial stages of the processing process. The differences were maintained throughout the heating times except Ligri, which dropped from first to second rank after 40min of heating. The high amylase potential and retention rate of Santom Pona could make it a good alternative source of amylase for food applications, which employ relatively mild processing temperature. At 75°C, beta amylase activity decreased steadily with time in all the varieties. Hi-Starch completely lost its activity whilst Apomuden, Ligri and Santom Pona retained 30% to 50% of its activity after 20 min of heating. All the varieties lost most of their activity after 40 min of heating. Beta-amylase activities were greatly reduced (less than 500 U/g) at the beginning of heating at 85°C. Only Apomuden and Ligri retained some activity and maintained the activity for 10 min and 20 min respectively. Activities in Santom Pona and Hi-Starch varieties were not detected even at the beginning of the 85°C treatment. From the study, Ligri and Apomuden retained some activities even at 85°C and can hence withstand high temperatures during mashing and liquefaction processes of starches. It should be noted that gelatinization, a rate determining step in starch conversion, occurs at temperatures  $\geq 60^{\circ}\text{C}$  (Sawai *et al.*, 2009; Takahata *et al.*, 1994). Hence raw materials selected for brewing and syrup production should have the ability to retain high amylase activity during heating.

#### **4.3.3 Variability of temperature, genotype and interactions on sugar profile of sweetpotato roots**

Data on the influence of temperature, genotype and their interaction on sugar profile (sucrose, maltose, glucose and fructose) of sweetpotato roots during heating are shown in Table 4.3. Genotypic influence was the most intense and accounted for 99.71%, 99.01 and 97.84% of the variance observed in glucose, fructose and sucrose content respectively. The lowest effect was recorded in maltose (54.89%). Conversely, effect of temperature was highest on maltose (31.56%) formation. The rest of the sugars had less than 1.50% of the variance resulting from temperature. On the other hand, the interaction effect was highest for maltose content (13.56%) and least for fructose (0.27%). Sucrose and glucose recorded 1.08% and 0.83% respectively.

The results indicate that except for maltose content, sugars in sweetpotatoes are greatly determined by the genetic make up of the variety and do not substantially respond to temperature during heating. Hence, the final concentration of these sugars (sucrose, glucose and fructose) is at least 97% proportional to the initial concentrations during processing of sweetpotato roots (Table 4.3). Variation in endogenous sugars (sucrose, glucose and fructose) have been reported to be minimal after cooking (Lewthwaite *et al.*, 1997; Babu 1994; Morrison *et al.* 1993; Takahata *et al.* 1992; Picha 1985), though their concentrations during heating have not been deeply investigated prior to this studies. Sun *et al.* (1993) reported that fructose, glucose and sucrose levels in Jewel sweetpotato cultivar were not affected by baking temperature. Variation in maltose content, however, has been extensively documented (Sawai, *et al.*, 2009, Lewthwaite *et al.*, 1997; Babu 1994; Morrison *et al.*, 1993, Takahata *et al.*, 1994, Picha 1985). Reasons for the increase

in maltose content have been largely assigned to the increase in temperature, which activates the hydrolytic ability of native amylases found in the root resulting in increased maltose content in the final product (Takahata *et al.*, 1994; Morrison *et al.*, 1993)

**Table 4.3. Source of variation and percentage variance of sugars during heating of sweetpotato roots**

Source of variation	<sup>+</sup> Variance (%)			
	Sucrose	Maltose	Glucose	Fructose
<b>Genotype</b>	97.84**	54.89**	99.71**	99.01**
<b>Temperature</b>	1.09**	31.56**	0.22**	0.91**
<b>Genotype x Temperature</b>	1.08**	13.56**	0.83**	0.27**

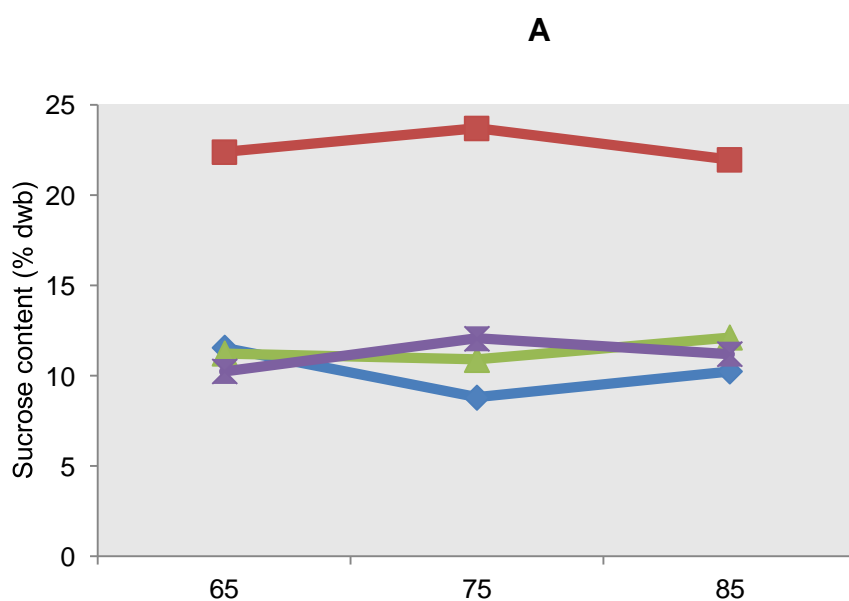
<sup>+</sup> Percentage variance was calculated from sum of squares. <sup>\*\*</sup> Significant at p=0.0001

#### 4.3.4 Effect of temperature on sugar profile of four sweetpotato varieties

Variation in sugar profile of the four sweetpotato varieties during heating is presented in Figure 4.2 and 4.3. Sucrose, glucose and fructose concentrations in all the varieties were relatively stable throughout the heating period. Interaction effect was also minimal except at 75 °C for sucrose contents. In all, Apomuden variety recorded the highest content of sucrose, fructose and glucose contents at 65 °C and maintained the levels during the heating process. Effect of temperature was marked on maltose content, an observation confirming early conclusion that maltose content is greatly affected by temperature (Table 4.3). Maltose, which was barely present in all the varieties prior to heating, increased to 6.76% in Apomuden for instance at 75°C. Maltose content in Santom Pona began to

increase substantially after 75°C. The presence and stability of beta amylase activity detected initially accounted for the variation in maltose content. Nevertheless, it should be noted that the optimum temperature of  $\beta$ -amylase falls below 75°C; hence the difference in maltose content at higher temperatures may be due to the synergistic activity of  $\alpha$ -amylase (Takahata *et al.*, 1994; Ikemiya and Deobald 1966). Alpha amylase, which is relatively heat stable with optimum temperature of 71°C, rapidly degrades starch to lower molecular weight dextrins which are simultaneously hydrolysed into maltose by  $\beta$ -amylase (Truong and Avula, 2010; Hagenimana and Simard, 1994).

Thermal stability of  $\alpha$ -amylase activity in the eleven sweetpotato varieties under study was therefore assessed in order to ascertain its presence and synergistic contribution to  $\beta$ -amylase in sugar formation. The results revealed that the sweetpotato varieties contained considerable activity of  $\alpha$ -amylase (Fig 4.4). Alpha amylase activity has been detected in several sweetpotato varieties including Kakamega, Naspot 1, Naspot 2, Naspot 9 and 10 (Nabubuya *et al.*, 2012).





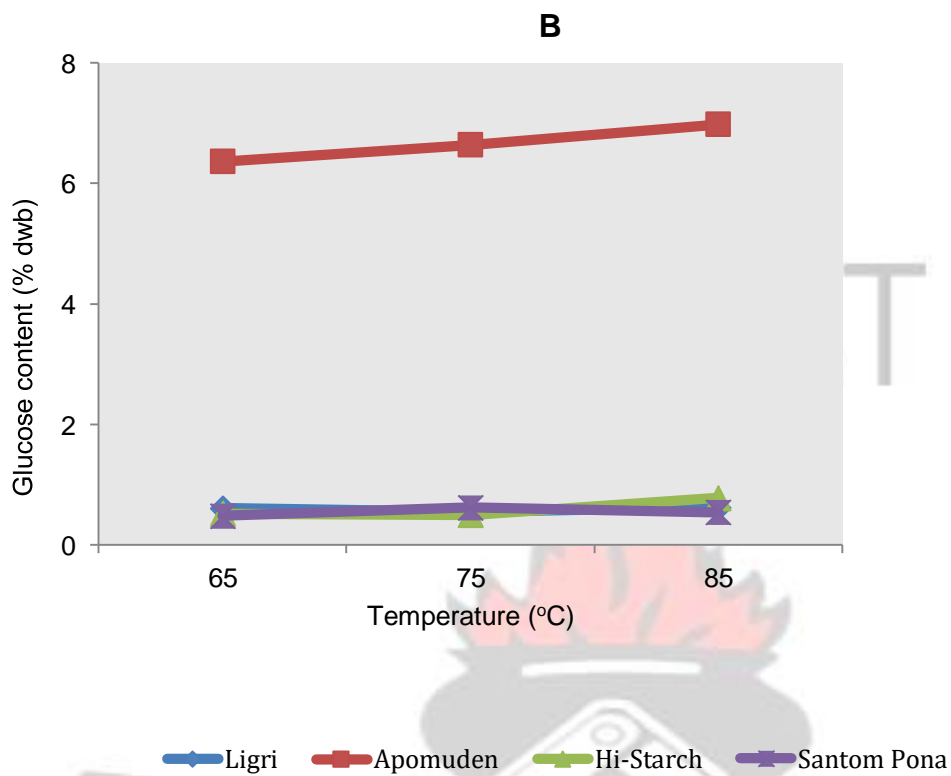
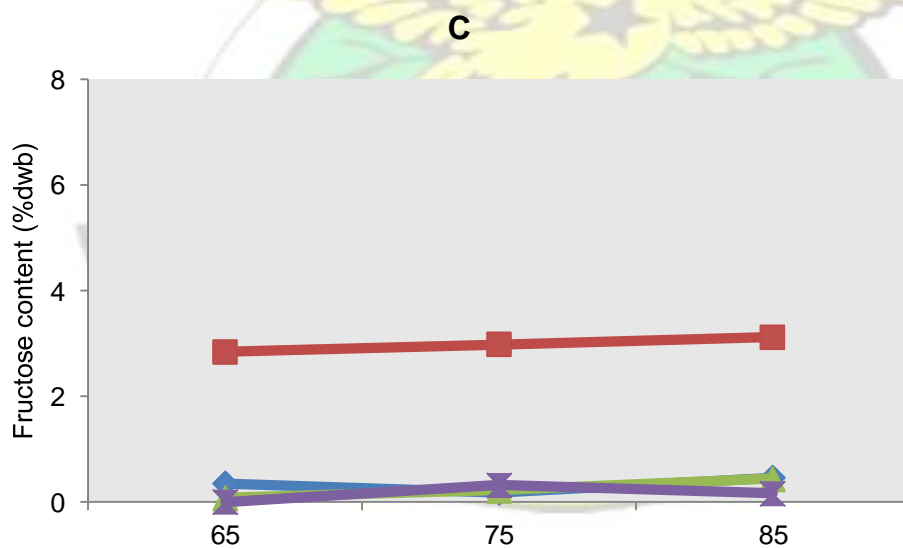


Fig 4.2. Changes in sucrose (A) and glucose (B) contents during heating of four sweetpotato varieties.



**D**

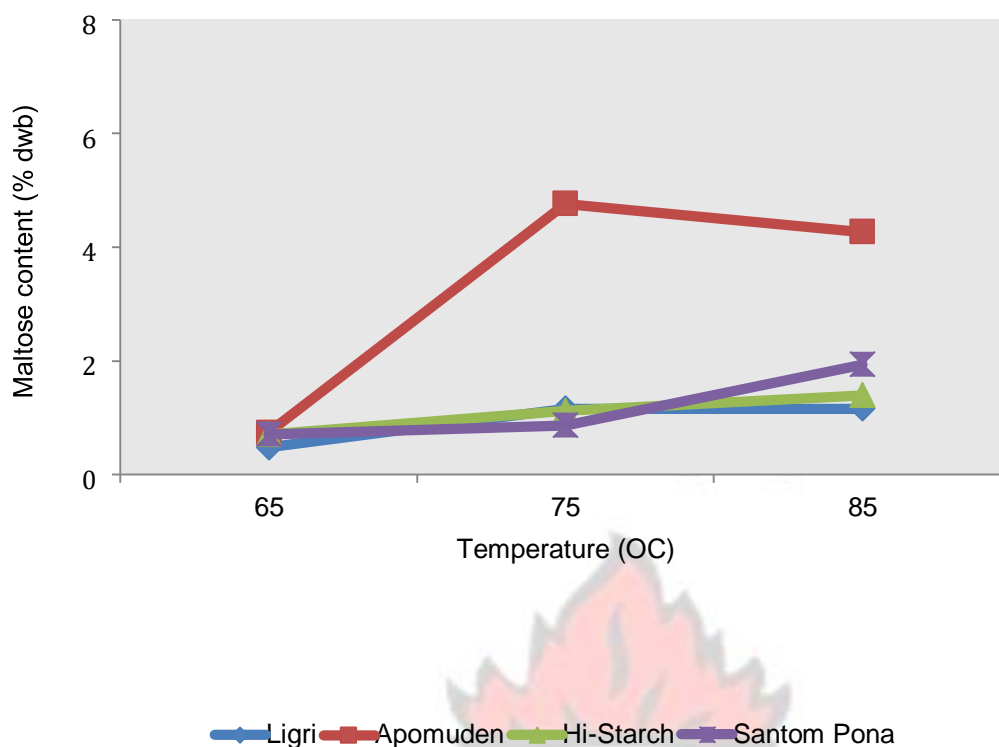


Fig 4.3. Changes in fructose (C) and maltose (D) contents during heating of four sweetpotato varieties.

Alpha-amylase activity increased from 65°C to 75°C and declined at 85°C in all the varieties. Apomuden and Hi-Starch had the highest activity at 65°C while Santom Pona had the lowest. The activities were however not significantly different among the varieties at 75°C except Ligri. Ligri exhibited the highest activity of 90U/g at 75°C, and also retained 75% of the activity at 85 °C. Alphaamylase had a higher optimum temperature (75°C) than  $\beta$ -amylase (65°C) demonstrating its ability to break down starch molecules at relatively higher temperature during processing. Variation in maltose content in this study resulted from the hydrolytic activity of both  $\alpha$ - and  $\beta$ -amylases.

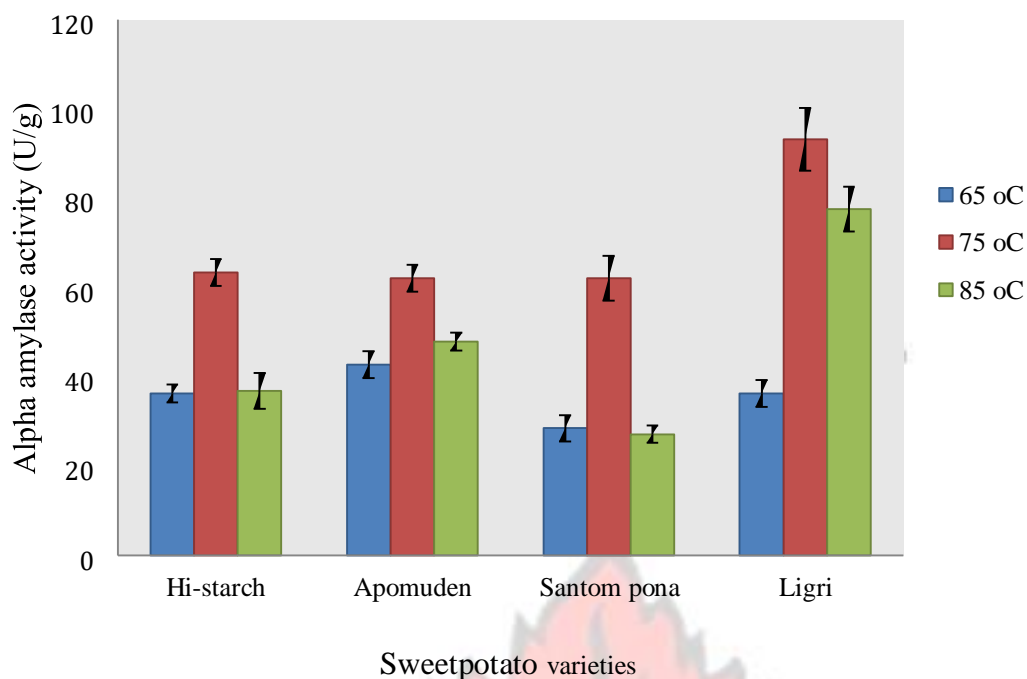


Fig 4.4 Alpha amylase activity of four sweetpotato varieties during heating at 65°C, 75°C and 85°C. Error bars represent standard deviations.

#### 4.4 Conclusion

The outcome of the study establishes that stability and hydrolytic power of  $\beta$ amylase in sweetpotato roots is significantly influenced by the genetic composition of the variety, temperature and period of heating. In all, temperature and genotype showed the highest variability of 50.31% and 25.94% respectively. Except for maltose, final concentration of sugars was virtually directly proportional to the initial levels. Sucrose remained the major sugar after heating up to 85°C, though maltose content increased drastically. The sweetpotato varieties contained substantial amounts of  $\alpha$  –amylase and its presence may have enhanced the conversion of starch into maltose at higher temperatures.

## CHAPTER FIVE

### 5.0 COOKING EFFECTS ON SUGAR PROFILE AND SWEETNESS OF ELEVEN-RELEASED SWEETPOTATO VARIETIES

#### 5.1 Introduction

Sweetness, derived from sugars in the raw sweetpotato root and maltose formed during cooking, is the predominant attribute controlling the taste of cooked sweetpotato products (Kays *et al.*, 2005; Wang and Kays, 2000). The level of sweetness in the root determines the type of product or formulation that can be developed. A number of factors including maturity period, storage, amylase potential, curing and baking treatment significantly influence sweetness/sugar content of sweetpotato roots (Adu-Kwarteng *et al.*, 2014; Dziedzoave *et al.*, 2010; Wang and Kays, 2000). Baking treatment and the amylolytic potential nonetheless have the greatest effect on sugar content of the final product (Chan *et al.*, 2012; Takahata *et al.*, 1994; Morrison *et al.*, (1993). Baking generally increases sugar content of sweetpotato roots (Sawai *et al.*, 2009; Kays *et al.*, 2005; Hagenimana *et al.*, 1996; Morrison *et al.*, 1993, Woolfe 1992). The heat produced during baking coupled with the degradative activity of the native amylases are responsible for the increased in the sugars. Increase in sugar content during baking can be dramatic, leading to a very sweet product (Chan *et al.*, 2012). Even though the effect of baking treatment on sugars of sweetpotato roots has been extensively investigated, limited data is available on other cooking treatment such as steaming and microwaving. Sweetpotato roots are cooked by different treatments including microwaving, baking, steaming and boiling prior to consumption with the aim to increase the culinary properties and enhance digestibility (Wang and Kays, 2000; Woolfe, 1992). Temperature, time and mode



of heat transfer differentiate these cooking methods. Conventional baking usually lasts for 60 – 90 min at 180 - 220 °C, depending on the genotype and root size (Chan *et al.*, 2012). Baking temperature as reported by Simkovic *et al.* (2003) and Chan *et al.* (2012) can however cause sucrose caramelisation, a phenomenon, which results in conversion of sucrose to oligomers and polymers. Microwave cooking employs a high temperature, short time heating mechanism to cook food products (Sun *et al.*, 1993). Heat is transferred by convection and conduction during baking whilst electromagnetic waves penetrate food materials causing agitation and friction to produce heat for cooking during microwaving (Wang and Kays, 2000). The effect of steaming on quality characteristics of sweetpotato root has not been widely reported.

Although effects of some cooking methods, especially baking, on quality attributes of sweetpotatoes have been evaluated (Chan *et al.*, 2012; Wang and Kays, 2000, Sun *et al.*, 1993), comparative studies with the view of understanding the effects of different cooking treatments on sugar profiles, sweetness and utilisation of sweetpotatoes are limited. Moreover, the influence of cooking treatments on sugars of eleven officially released sweetpotato varieties in Ghana has not been investigated. To better understand the effect of different cooking methods on sugar levels and sweetness of sweetpotato roots, individual sugar and sweetness levels of eleven released varieties in Ghana were determined.

## **5.2 Methodology**

### **5.2.1 Experimental design**

Triplicates of eleven sweetpotato varieties released (Table 5.1) by the Council for Scientific and Industrial Research (CSIR) – Crops Research Institute (CRI) were planted in a randomized complete block design on May 2014 at the CSIRCRI experimental station, Fumesua, Ghana. Harvesting was done four months after planting (September, 2014) and each plot was treated as a separate sample during laboratory evaluations. Harvested roots were stored for a week at room condition (25 to 30°C) prior to processing.

### **5.2.2 Sample preparation**

Four medium-size intact roots of each variety were washed with clean water, rinsed and air-dried. The clean roots were then quartered, rinsed with de-ionised water and dried using paper towels. Each quarter was sliced across its longitudinal axis to approximately 1.0 cm thickness and composite samples from each plot, divided into four groups of 50 g. One group was designated as raw and the rest were subjected to three different processing methods; baking, steaming and microwaving. For baking, one group of the sliced samples was wrapped in aluminium foil and placed in a forced air oven (Genlab MINI/50/DKG), which has been preheated to 205°C, for 30 mins at 205°C. For steaming, another group of root samples was placed in a Kitchen steamer with 1500 mL of boiling water and cooked for 10 min. The third group of the root samples was wrapped in a paper towel and moistened with about 5 mL of potable water and microwaved (Sharp microwave model R-228H) for 5 min inside a plastic microwaveable food container. Cooked samples were allowed to cool to room temperature for about

20 min, transferred to whirl-Pak polyethylene bags and frozen at – 20°C before drying using the freeze dryer (True Ten, Ind, YK18-50, Taiwan). Dried samples were milled and sieved as described in Chapter four (section 4.2.3) prior to sugars determination.

**Table 5.1. Phenotypic attributes and yield of the sweetpotato varieties used for assessment of changes in sugar content**

Variety	Skin Colour	Skin Shape	Flesh colour	Yield (t/ha)
Apomuden	Reddish brown	Obovate	Reddish orange	48.9
Bohye	Purple	Obovate	Pale orange	16.8
Dadanyuie	Dark purple	Round elliptic	White	10.5
Faara	Deep purple	Long elliptic	Cream	16.9
Hi-Starch	Creamy	Elliptic	Cream	14.7
Ligri	Cream	Round elliptic	Pale yellow	16.3
Okumkom	Cream	Long elliptic	Cream yellow	19.91
Ogyefo	Purple	Long elliptic	White	25.9
Otoo	Cream	Long elliptic	Light orange	30.7
Patron	Dark yellow	Long elliptic	Dark yellow	15.9
Sauti	Cream	Long elliptic	Yellow	15.4

Source: CSIR-CRI, 1998, 2005 and 2012.

### 5.2.3 Sugar determination.

Freeze-dried and milled sweetpotato samples were sent to the Quality Plant Product Laboratory (Department of Crop Science, University of Gottingen, Germany) for sugar analysis. Water extract of the freeze-dried sweetpotato samples (0.1 g in 100 mL) was used. The samples were incubated in a water bath at 60°C for 1 h and treated with 0.2 mL Carrez I and Carrez II solution to remove proteins. Samples were purified by centrifugation (Sorvall RC-5B Refrigerated

Superspeed, GMI, Ramsay, USA) at 10,000 rpm for 10 min at 20°C. Sugars were determined from the membrane-filtered supernatant (pores size 0.45 µm).

Glucose, fructose, sucrose, and maltose were separated using a LiChrospher 100 NH<sub>2</sub> (5 µm) 4 x 4 mm pre-column in combination with a LiChrospher 100 NH<sub>2</sub> (5 µm) 4 x 250 mm separation column (Merck KGaA, Darmstadt, Germany) and an acetonitrile: pure water solution (80:20 v/v) as mobile phase at a flow rate of 1.0 mL min<sup>-1</sup> at 20 °C and an injection volume of 20 µL. Sugars were detected with a Knauer differential refractometer 198.00 (Knauer, Berlin, Germany).

#### **5.2.4 Determination of amylase activity**

The 3,5-dinitrosalicylic acid (DNSA) method for reducing sugars was employed to determine the total amylase activity of the freeze-dried sweetpotato roots (Owusu-Mensah *et al.*, 2010; Osman, 2002) as described in section 3.2.7 of chapter three.

A unit (U) of amylase activity was defined as the amount of enzymes required to release reducing sugars equivalent to one µmole of maltose/min under the above stated conditions (Osman, 2002).

#### **5.2.5 Calculation of Sweetness level**

In order to ascertain and compare sweetness levels among the varieties, sweetness (sucrose equivalent) was calculated from the equation: Sucrose Equivalent (SE) = 1.2 fructose + 1 sucrose + 0.64 glucose + 0.43 maltose (Kays *et al.*, 2005 and Shallenberger, 1993). Based on the SE values obtained, the varieties were classified into four categories: non sweet (SE ≤ 12 g/100g dry weight); low sweet



(SE 13 – 20 g/100 g); moderate sweet (SE 21 – 28 g/100 g); and high sweet (SE29 – 37 g/100 g) (Kays *et al.*, 2005)

### **5.2.6 Statistical Analysis**

Experimental means were calculated from triplicate values of each variety per treatment. Data obtained were subjected to analysis of variance using Statistical Analysis System (SAS, 2007). Significant differences among means were assessed using Least Significant Difference (LSD) at probability level of 5%.

## **5.3 Results and Discussion. 5.3.1 Effect of cooking, genotype and interaction on sugars of cooked**

### **sweetpotato roots**

The effect of cooking, genotype and their interaction were significant on all sugars (maltose, sucrose, glucose and fructose), though the percentage contributions varied considerably (Table 5.2). Cooking treatment showed the highest effect of the total variance on the sugars except fructose. The effect was more profound on maltose content with percentage variance of 90.12%. Nearly 80% and 53% of the total variance in sucrose and glucose contents respectively of the cooked roots were due to the cooking treatment. Effect of genotype was highest on fructose relative to the other sugars. While 45.68% of the variation in fructose resulted from the genotypic composition of the roots, only 7.26% of the difference in maltose content was due to genotypic effect. Percentage variability resulting from genotypic effect on sucrose and glucose was 16.93% and 38.82% respectively. Overall variation from interactions between cooking treatment and genotype ranged from 2.60% to 11.47% of the entire differences noticed.

Although it was significant, it contributed the least of the total variation.

**Table 5.2. Percentage variance of cooking treatment, genotype, and interactions on sugars of cooked sweetpotato roots**

Source of Variation	*Variance (%)			
	Maltose	Sucrose	Glucose	Fructose
Genotype (G)	7.26**	16.93**	38.82**	45.68**
Cooking treatment (CT)	90.12**	79.04**	52.60**	43.12**
GxCT	2.60**	4.03**	8.65**	11.47**

\*\* Significant at  $p < 0.05$ . \* Calculated from sum of squares.

The results from the analysis of variance depict that changes in sugar concentrations during cooking are significantly dependent on cooking treatment, genotype and their interaction. Among these factors cooking treatment exerted the highest effect. Its effect was more profound on maltose content, which increased from 7.26% prior to cooking to 90.12% afterward. Cooking increases temperature intensity and penetration, and also facilitates breakdown of hydrolytic bonds holding starch granules (Wang and Kays, 2000). Such conditions enhance the activity of native amylase resulting in starch degradation and the production of sugars mainly maltose as observed in the study (Hagenimana and Simard, 1994; Morrison *et al.*, 1993). Apart from fructose, changes in individual sugars were remarkable. Response from fructose was higher for genotype effect relative to cooking treatment.

### 5.3.2 Effect of cooking treatment on sugars of sweetpotato roots

Table 5.3 shows the means and ranges in sugars as a result of the different cooking treatments. Wide variation existed among the sugars of the cooked sweetpotato roots, with maltose and sucrose showing the highest variability.

Maltose was hardly present in the raw form whilst sucrose (10.58%) predominated. This finding agrees with Morrison *et al.* (1993) and Sun *et al.* (1993) who reported that sucrose is the major sugar in raw forms and the most important sugar for predicting sweetness in sweetpotatoes (Chan *et al.*, 2012). Sucrose concentration, generally, increased slightly when baked, though it was not significantly different compared to the raw, but remained constant at microwaving and decreased significantly during steaming.

**Table 5.3. Means and ranges of individual sugars in raw and cooked sweetpotato roots**

Individual Sugars (% DM)	Raw	Cooking Treatment		
		Baking	Microwaving	Steaming
Sucrose	10.58 (9-23) <sup>a</sup>	11.01 (6 - 20) <sup>a</sup>	10.72 (7-16) <sup>a</sup>	4.30 (0-8) <sup>b</sup>
Glucose	1.63 (0.4-5) <sup>b</sup>	1.55 (0-5) <sup>b</sup>	0.84 (0 - 2) <sup>a</sup>	0.92 (0-2) <sup>a</sup>
Fructose				0.95 (0-4) <sup>a</sup>
Maltose	0.63 (0-1) <sup>a</sup>	20.13 (5 -36) <sup>b</sup>	5.07 (2-15) <sup>c</sup>	14.35 (2-27) <sup>d</sup>

Ranges of means are presented in brackets. Figures in rows with the same superscripts are not significantly different (p<0.05)

Glucose and fructose contents were not significantly affected by the different cooking treatments, although the levels were generally lower compared to that of raw roots. Maltose content rose from 0.63% before cooking to 20.13%, 14.35% and 5.07% after baking, steaming and microwaving respectively. It became the principal sugar following baking and steaming. Increase in maltose content following cooking has been observed in several sweetpotato varieties (Takahata *et al.*, 1994, Morrison *et al.*, 1993, Sun *et al.*, 1993). Changes in maltose and sucrose (the major sugars) concentrations per variety during cooking were also assessed and results presented in Fig 5.1 and Fig 5.2 respectively.

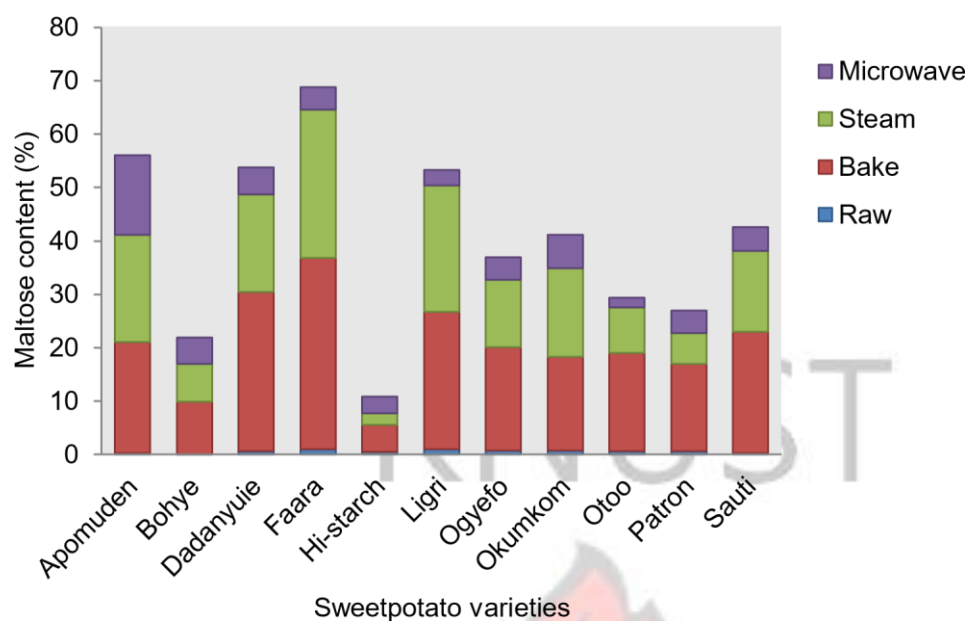


Fig 5.1. Changes in maltose content of sweetpotato roots as affected by different cooking treatments; microwaving, steaming and baking.

Maltose, which was not detected in most of the varieties prior to cooking increased dramatically after baking and steaming (Fig 5.1). Faara, Dadanyuie, Ligri, Sauti and Apomuden had the highest increase and Hi-Starch the lowest in maltose content following baking and steaming. Though the effect of microwave cooking was also positive and significant on maltose content for all the varieties, it was comparatively much lower compared to both baking and steaming.



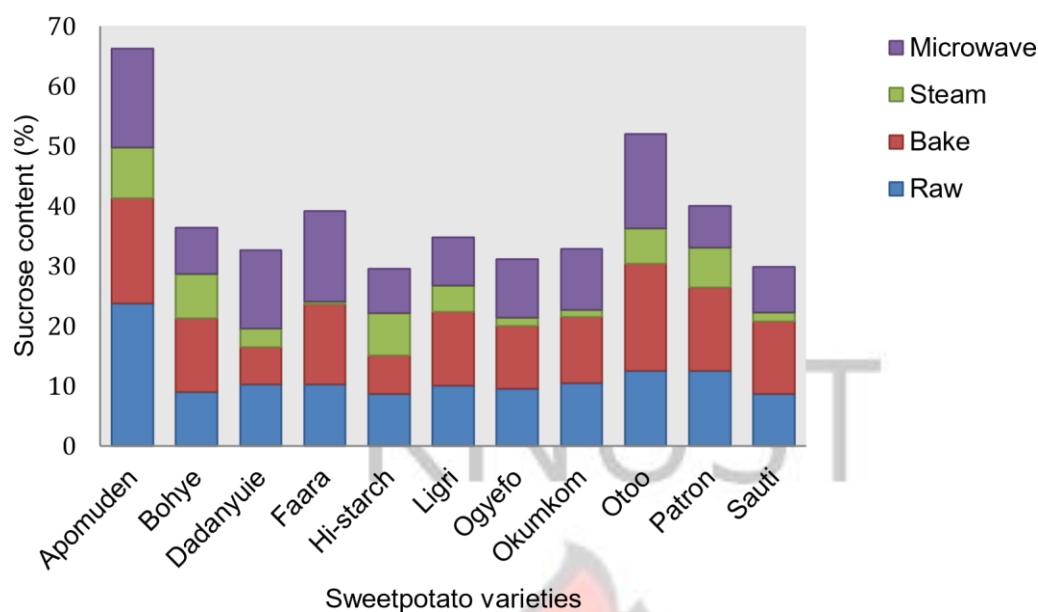


Fig 5.2. Changes in sucrose content of sweetpotato roots as influenced by three cooking treatments; microwaving, steaming and baking. LSD=0.86

In contrast, sucrose content decreased in some of the varieties while increasing slightly or remaining the same in others during cooking (Fig 5.2). Apomuden, Dadanyuie, and Hi-starch recorded a decrease whilst Bohye, Faara, Otoo, Sauti and Ligri showed an increase after baking. Sucrose contents in Ogyefo, Okumkom and Patron were not significantly affected by baking treatment. Steaming reduced sucrose content in all the varieties. The magnitude of reduction was extremely high in Faara, which lost almost 96% of its sucrose content. Effect of microwave treatment on sucrose was similar to that of baking.

While negatively affecting sucrose content in Apomuden, Bohye, Hi-Starch, Ligri, Patron, and Sauti, microwaving enhanced sucrose levels in Dadanyuie, Faara, and Otoo. Sucrose content in Ogyefo, and Okumkom were not significantly affected.

Concentration of sugars in sweetpotato roots varies significantly during cooking, (Takahata *et al.*, 1994; Sun *et al.*, 1993; Morrison *et al.*, 1993) with the extent of

variability being highly dependent on; 1) initial sugar concentration, 2) amylase activity and 3) cooking method employed. The impact of cooking treatment on sugar content is related to temperature, time, and mode of heat transfer. Baking treatment resulted in the highest sugar (maltose) formation mainly due to the long cooking period (30 min) and the nature of the heat transfer mechanism. Moreover, there was no direct contact between the sample and the heating medium, a system that prevented possible leaching of soluble sugars, during baking. Heat is transferred from the periphery to the centre of the root by conduction in baking as compared to microwaving for instance where electromagnetic radiation penetrates the entire root causing agitation and friction to produce heat for cooking instantaneously (Wang and Kays, 2000). Hence baking utilises more time, a system that allows adequate starch gelatinisation and subsequent conversion to maltose by amylases (Sawai *et al.*, 2009; Lewthwaite *et al.*, 1997). It has been demonstrated that increasing heating temperature over a time frame increases starch degradation and maltose production (Takahata *et al.*, 1994; Hashimoto *et al.*, 1994; Sun *et al.*, 1993). Baking treatment at higher temperatures can however cause sucrose caramelisation, a phenomenon, which results in conversion of sucrose to oligomers and polymers as reported by Simkovic *et al.* (2003) and Chan *et al.* (2012). Hence the reduction in sucrose content of some of the varieties (Fig 5.2) may be attributed to this effect. This finding corresponds with Chan *et al.* (2012) and Morrison *et al.* (1993) who reported a decrease in sucrose content of several sweetpotato cultivars during baking.

The rapid heating mechanism of microwaving deactivated the native amylases responsible for maltose formation, and consequently the reduction in its levels (Wang and Kays, 2000; Sun *et al.*, 1993). Moreover, the short heating period of

microwaving does not enhance starch gelatinisation, a rate-determining step in the initial stages of hydrolysis (Sawai *et al.*, 2009; Takahata *et al.*, 1994). Whereas baking resulted in a dramatic increase in maltose content of Jewel, microwaving inhibited its formation, reducing the total sugar content of the cooked product (Sun *et al.*, 1993). Microwave cooking can therefore be an ideal method for food preparations where high sugar content is not a desirable attribute. In regions like Sub-Sahara Africa where less sweet sweetpotato varieties are perceived to be the preferred choice (Tumwegamire *et al.* 2011), microwave cooking could be the recommended choice.

Steaming treatment resulted in an increase in maltose content in all the varieties. On the contrary, it caused a reduction in sucrose content in all the varieties relative to the raw roots. The wet heat transfer mechanism of steaming treatment allowed direct contact between the roots and the heat source. Such heat exchange technique allows movement of soluble substances; where solutes move from high concentration to low concentration. Sucrose, which was initially high in the raw roots, may have consequently moved from the roots to the water vapour. Hence the reduction in sucrose content observed in the roots after steaming.

Increase in sugars, particularly maltose, in sweetpotato root can also be attributed to the hydrolytic ability of native amylases present in the uncooked roots (Dziedzoave *et al.*, 2010; Hagenimana *et al.*, 1996; Morrison *et al.*, 1993; Takahata *et al.*, 1994). Sweetpotato roots contain high levels of amylases, mainly  $\alpha$ - and  $\beta$ -amylase, which significantly influence levels of sugar in processed sweetpotatoes (Nabubuya *et al.*, 2012). Amylases hydrolyse gelatinised starch into maltose and short-chain branched oligosaccharides (limit dextrins) during cooking resulting in a sweet taste (Lewthwaite *et al.*, 1997; Morrison *et al.*, 1993).

However, it should be noted that amylases in sweetpotato roots are denatured above 80°C (Hagenimama *et al.*, 1994). The amylase activity of the varieties was therefore determined to ascertain the general hypothesis that amylases are also responsible for the increase in sugar content.

**Table 5. 4. Means and levels of amylases in sweetpotato varieties**

Sweetpotato varieties	Total amylase activity	Groupings
Ligri	927.14(40.56)	Very high
Dadanyuie	882.05 (26.82)	“
Sauti	809.24 (30.45)	“
Ogyefo	804.10 (30.67)	“
Okumkom	779.25 (37.76)	“
Faara	687.32 (50.34)	High
Otoo	650.67 (20.45)	“
Patron	489.81 (15.56)	Moderate
Apomuden	454.10 (21.56)	“
Bohye	414.26 (13.24)	“
Hi-starch	387.06 (25.67)	“

Grouping was based on ranges of amylase activity found: Very High ( $\geq 750$ ), High (749-550), moderate (549- 350), low ( $\leq 349$ ). Standard deviations are presented in brackets. LSD = 14.45

Table 5.4 presents amylase activity of the sweetpotato varieties investigated. It ranged from 927.14 U/g in Ligri to 387.06 U/g in Hi-starch. Based upon levels of activity found, Ligri, Dadanyuie, Sauti, Ogyefo and Okumkom were grouped as very high amylase varieties. Faara and Otoo are high category varieties whilst Patron, Apomuden, Bohye and Hi-Starch are considered moderate types. The level of amylase activity related positively with the formation of maltose after cooking (Fig 5.1). Most of the high amylase varieties including Dadanyuie, Ligri, and Faara of low initial total sugar content (Fig 5.1) showed very high increase in maltose content after baking and steaming. Similarly, Hi-starch with a lower



amylase activity but similar initial sugar content as that of Ligri for instance produced little extra maltose, and was not significantly different from the uncooked roots. Apomuden with moderate amylase potential produced moderate maltose content though it had the highest content prior to cooking. This result supports previous findings that maltose content in cooked sweetpotato is a function of amylase activity of the roots (Takahata *et al.*, 1994; Morrison *et al.*, 1993). However, it should be noted that different cooking treatments produced significantly different effects on maltose content of the cooked roots (Fig 5.1). From this study however, baking treatment resulted in the highest final sugar contents.

### **5.3.3 Baking treatment and sweetness of sweetpotato roots**

To study the effect of cooking treatment on sweetness levels of the varieties, baking treatment, which resulted in the highest increase in sugars, was selected. Individual sugars in raw and baked roots were first converted to sucrose equivalent (SE) based on sweetness factors (Shallenberger, 1993). Such conversion allows easy comparison of sweetness among sweetpotato varieties. Kays *et al.* (2005) employed this method to evaluate the sweetness levels of 272 baked sweetpotato clones and categorised the clones into five main groupings based on SE: Very high  $\geq 38$ ; high 29 – 37; moderate 21 – 28; low 13 – 20 and non sweet  $\leq 12$  g per 100g dry mass.

Sweetness among the sweetpotato varieties prior to and after baking is presented in Fig 5.3. The levels increased significantly after baking in majority of the varieties, and the effect was more pronounced in the high amylase types (Table 5.4); Faara, Ligri, Otoo and Sauti. The increase also corresponded well with the

maltose content after baking (Fig 5.1). Apomuden had the highest sweetness value of 29.79 SE, and Hi-Starch the lowest of 10.79 SE prior to baking (Fig 5.3). The other varieties had values in the range of 12 to 16 SE. Based upon Kays *et al.* (2005) classification, the varieties fell under the following classes prior to baking: Apomuden – High sweet; Bohye, Dadanyuie, Faara, Ligri, Okumkom, Otoo, Patron and Sauti – Low sweet; and Hi-starch, Ogyefo and Sauti – non sweet. However the levels of sweetness and subsequently the sweetness categories of the varieties changed significantly following baking. Whereas Apomuden dropped from high sweet category (29.79 SE) to moderate sweet (28 SE), majority of the varieties including Dadanyuie, Faara, Ligri, Otoo and Patron moved from low sweet to moderately sweet category. The increase in SE of Bohye and Okumkom were not significant enough to place them in the moderate class. Whilst Ogyefo and Sauti increased in SE values and were categorised as low and moderate sweet respectively, Hi-starch, remained in the same non-sweet category following baking.

Sweetness in sweetpotatoes is a function of cultivar, amylase activity, storage condition, and cooking treatment (Chan *et al.*, 2012; Kays *et al.*, 2005; Wang and Kays, 2000; Sun *et al.*, 1993; Morrison *et al.*, 1993). Nonetheless, amylase activity, initial sugar concentration and maltose formed during cooking are the most critical in determining the final sweet sensation of cooked root (Kays *et al.*, 2005; Morrison *et al.*, 1993). These factors can completely change the sweetness status of a variety as observed in Dadanyuie, Faara, Ligri, Sauti, Otoo, Patron and Ogyefo (Fig 5.3) which were low or non-sweet prior to cooking, but changed to moderate sweet when baked.

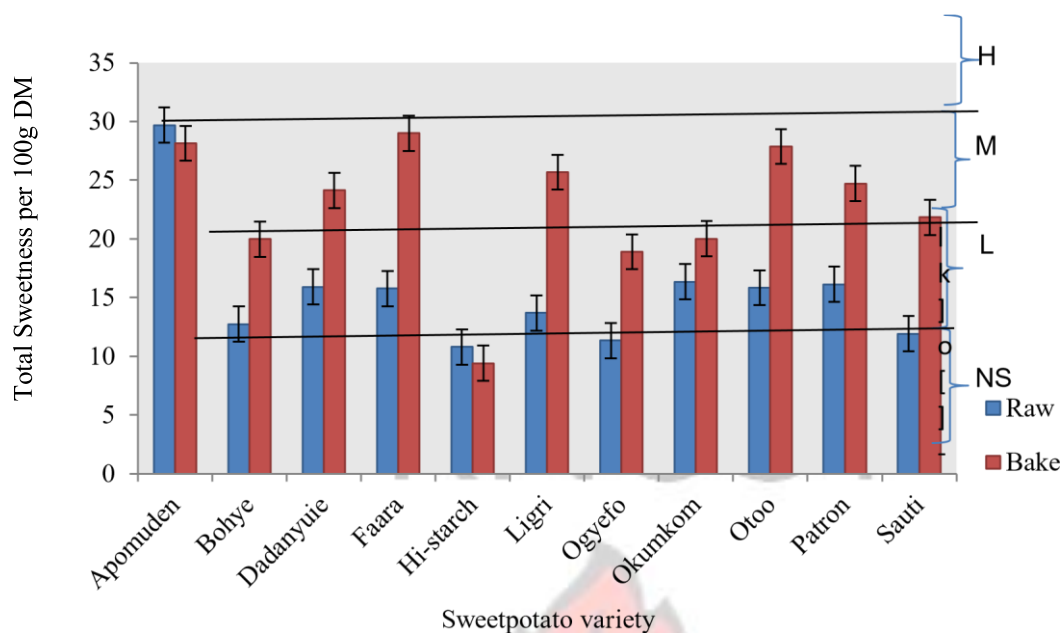


Fig 5.3. Changes in sweetness levels of sweetpotato roots after baking.

H- High sweet, M-Moderately Sweet, L- Low sweet, NS- Non sweet  
Error bars show significant differences

The sweetpotato varieties in this study were also classified into four general groups based on initial sucrose equivalent (SE) and starch hydrolytic potential (Morrison *et al.*, 1993). These are low initial SE/low starch hydrolysis; Low initial SE/high starch hydrolysis; High initial SE/low starch hydrolysis and High initial SE/high starch hydrolysis. Fig 5.4 shows the classification of the sweetpotato varieties assessed under this grouping.

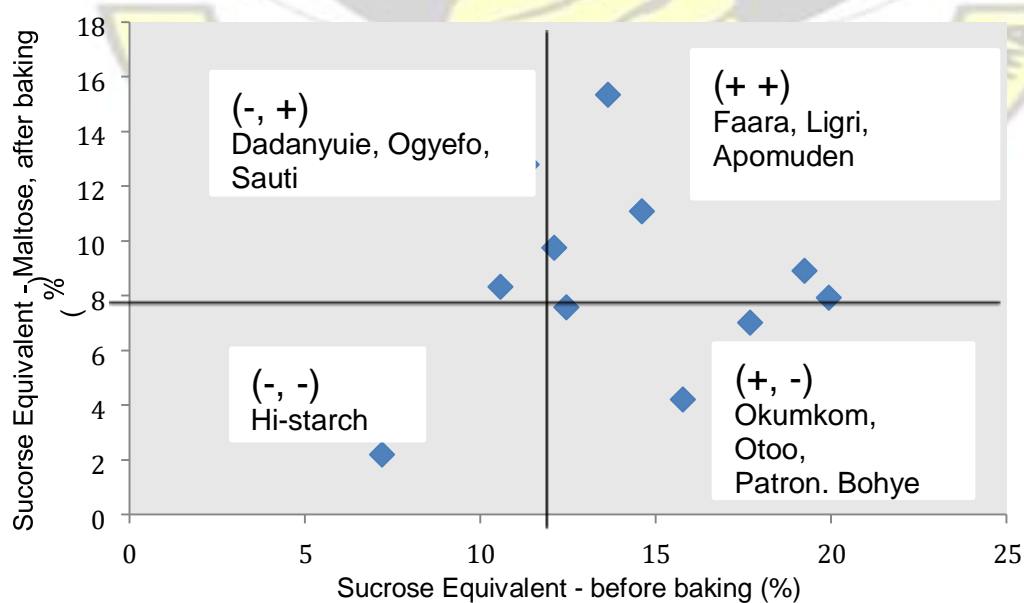


Fig 5.4. Classification of eleven sweetpotato varieties based on sucrose equivalent (SE) derived from starch hydrolysis (using maltose as indicator) during baking and endogenous sugars (sucrose, glucose and fructose).

(-, -) – Low initial SE/low starch hydrolysis; (-, +) – Low initial SE/high starch hydrolysis; (+, -) – High initial SE/low starch hydrolysis; (+, +) – High initial sugar/High starch hydrolysis (Kays *et al.*, 2005; Morrison *et al.*, 1993)

Hi-starch was the only variety belonging to the class of low initial SE content coupled with low starch hydrolysis (-, -). It produced small amount of maltose upon cooking (Fig 5.1) as a result of its low amylase activity (Table 5.4). Natural inhibitors and starch-based structural resistance to hydrolysis are also probably inhibitory mechanisms for the low starch hydrolysis (Morrison *et al.*, 1993). Kumagai *et al.*, (1990) also attributed this lack of activity to a recessive allele called  $\beta$ -amy for which the variety Satsumahikari was homozygous. Amylase activity in this variety was detected in vitro, but apparently was below the threshold required for effective hydrolysis during baking. Dadanyuie, Ogyefo and Sauti had low initial SE but produced significant amounts of maltose when baked (-, +) whilst Okumkom, Otoo, Patron and Bohye had moderate to high initial sugar content and produced low levels of maltose upon baking (+, -). The last group, Faara, Ligri and Apomuden, had relatively high initial SE and moderate to high starch hydrolytic (+, +) potential following baking. The outcome of this investigation establishes that final sweetness of cooked sweetpotato roots is a function of initial sugar content and amylase potential of the raw root. Hence it would be unreliable to classify sweetpotato clones in terms of sweetness prior to cooking.



#### 5.4 Conclusion and recommendations

The findings of this study indicate that cooking method, genotype and their interactions significantly influences sugars and sweetness of sweetpotato root. Among these factors cooking treatment showed the highest variability. Baking which lasted for a longer time resulted in the highest maltose formation. Maltose was barely present in raw roots but increased considerably after cooking. The amount of maltose synthesized was however dependent on the level of amylase present in the raw root. Activity of amylases was facilitated by temperature, time, and mode of heat penetration by the cooking method. While baking conditions enhanced hydrolysis, electromagnetic radiation generated by microwave cooking deactivates amylases, suppressing maltose formation and rendering the product less sweet. Sweetness was found to be dependent on initial sugar content, amylase activity and cooking method. Cooking treatment should therefore be considered as a key criterion when evaluating quality attributes of sweetpotatoes for appropriate utilization.

## CHAPTER SIX

### 6.0 DEVELOPMENT OF CALIBRATION MODEL FOR EVALUATING INDIVIDUAL SUGARS AND TOTAL STARCH CONTENT IN COOKED SWEETPOTATO ROOTS USING NEAR-INFRARED REFLECTANCE

#### SPECTROSCOPY (NIRS)

##### 6.1 Introduction

Eating and processing qualities of sweetpotatoes are mainly controlled by sugars and dry matter of the roots (Lewthwaite *et al.*, 1997). Sugars regulate the final flavour of the cooked products (Kays *et al.*, 2005; Morrison *et al.*, 1993, Wang and Kays, 2000) while starch, the major component of the dry matter, influences the cooking and textural properties of the roots (Kays *et al.*, 2005; Collado *et al.*, 1999). Selection of desired clones in many sweetpotato breeding programmes including the “Sweetpotato Action for Security and Health in Africa” (SASHA) is based on these traits, which control acceptability, marketing and production of sweetpotatoes (Lu *et al.*, 2006; Wang and Kays, 2000). However, the high cost and the long time needed for wet chemical analytical methods such as High Performance Liquid Chromatography (HPLC) for sugars and calorimetric method for starch do not permit screening of large numbers of early generational sweetpotato clones. These wet chemical methods though accurate have many limitations with respect to large numbers of routine analysis (Lu *et al.*, 2006); they are too laborious, complex, expensive and time consuming. Due to these limitations, nutritional evaluations of breeding materials are conducted at advanced trails with few clones. By then, breeding clones with preferred eating and processing qualities might have been discarded.

Recent efforts have therefore been directed toward developing a rapid and inexpensive, but sufficiently accurate technique, which allows simultaneous determination of traits in a timely fashion. Among the most efficient, long established methods for measuring individual sugars and total starch in sweet potatoes is the Near-Infrared Reflectance Spectroscopy (NIRS). NIRS calibrations with moderate to high accuracy ( $R^2 = 0.91-0.74$ ) have been developed for total starch, individual sugars, total protein, beta carotene and with limited success for trace minerals (Lebot *et al.*, 2011; zum Felde *et al.*, 2009; Lu *et al.*, 2006). However, NIRS calibration developed for screening quality attributes of sweet potato roots so far are based on raw roots only. Nevertheless, quality attributes, mainly sugars, varied significantly when sweet potato roots are cooked (as established in chapter five of this study). This current study therefore sought to develop NIRS calibration models for determining individual sugar profiles and starch content in cooked sweetpotato roots.

## **6.2 Methodology**

### **6.2.1 Experimental design**

In order to develop robust NIRS calibration models, diverse sources of variation affecting concentrations of sugars and starch of sweetpotato roots were considered; genotypes, storage conditions, cooking methods, time and temperature regimes (Nabubuya *et al.*, 2012; Morrison *et al.*, 1993; Woolfe, 1992). Freshly harvested roots were stored for one month under ambient conditions (Temp: 25 - 27°C; %RH: 65-85%). Stored and freshly harvested roots were further exposed to three different cooking types (baking, microwaving and

steaming). A total of 108 processed sweetpotato samples were employed. Each test was conducted in triplicates.

### 6.2.2 Materials

Twelve officially released sweet potato varieties in Ghana were used for the study. Triplicates of each variety were cultivated at the Ohawu experimental field of the Crops Research Institute (CRI) and harvested at four months maturity. Ohawu is in the coastal savannah agro-ecological zone. Table 6.1 shows the varieties and their quality attributes before cooking. Wide variations in individual sugars and total starch content among the selected varieties were found.

**Table 6.1 Sugar profiles and starch content of raw sweet potato roots**

<b>Sweet potato Variety</b>	<b>Fructose (%)</b>	<b>Glucose (%)</b>	<b>Sucrose (%)</b>	<b>Total Sugars (%)</b>	<b>Total Starch (%)</b>
Apomuden	5.32	8.86	23.20	37.38	44.11
Bohye	1.94	3.56	11.92	17.42	64.90
Dadanyuie	2.21	3.96	10.17	16.34	68.50
Faara	2.04	3.76	9.67	15.47	66.91
Hi-Starch	0.86	2.21	8.70	11.77	73.00
Ligri	1.47	3.03	11.93	16.43	68.22
Ogyefo	0.34	1.55	9.34	11.23	72.33
Okumkom	3.23	5.17	11.39	19.79	63.07
Patron	1.25	2.64	14.37	18.26	65.82
Santom Pona	1.68	3.03	10.66	15.37	67.15
Sauti	1.13	2.57	8.50	12.20	69.08
Otoo	1.78	2.89	8.34	13.01	66.56

*Source: Table 3.3 and 3.4 (Chapter Three)*



### **6.2.3 Methods**

#### **6.2.3.1 Cooking treatment**

Four undamaged roots were selected from each triplicate. After washing and rinsing with clean tap water, the roots were air-dried. The clean roots were then quartered and opposite pair of each root composited to represent the sample for each variety. The samples were sliced into approximately 0.5cm thickness, divided into four portions and were subjected to three different cooking methods; baking, steaming and microwaving.

Baking: One portion of each sliced root sample was wrapped in aluminium foil and baked in forced a air conventional oven (Genlab MINI/50/DKG) at 205°C for 30min. The oven was pre heated to 205 °C before the baking process. After cooling to room temperature, the samples were peeled and frozen. The frozen samples were then freeze-dried, milled and kept at -20°C prior to analysis.

Steaming: This preparation method was conducted for 10min for each sample. The samples were allowed to cool to room temperature, peeled and stored at 20°C prior to analysis.

Microwaving: The sliced samples were wrapped in paper towel moistened with about 5 ml of potable water and placed in a microwave for 3 min. After microwaving the samples were allowed to cool to room temperature, peeled and stored at -20°C prior to analysis.

#### **6.2.3.2 Heating treatment**

Freshly harvested roots of the twelve varieties were used for this experiment.

Four medium size intact roots of each variety were washed, rinsed and air-dried. The roots were then quartered, rinsed with de-ionised water and dried with tissue papers. Each quarter was sliced across its longitudinal axis into 1.0cm thickness and composite samples within each variety divided into three groups of 50g. Each set was tightly wrapped in aluminium foil and placed in a forced air temperature controlled oven (Genlab MINI/50/DKG). The samples were heated to 65°C, 75°C and 85°C for 20min at each temperature setting. Termination of reaction was aided by submerging sweet potato root samples in an ice bath for 20min. The samples were then frozen at -25°C, and freeze dried (Trueten Biotech, YK-118/50, Taiwan) for 72 hours.

#### **6.2.3.3 Reference analysis of sweetpotato samples**

##### ***Starch determination***

Starch determination was carried out at the Quality of Plant Products Division, Department of Crop Science, Göttingen University, Germany. The polarimetric method using hydrochloric acid dissociation was employed for the starch determination (ICC No. 123/1, 1994).

##### ***Sugar determination***

Sugar analysis was also conducted at the Quality of Plant Products Division, Department of Crop Science, Göttingen University, Germany.. Water extract of the freeze-dried samples (0.1 g in 100 mL) was incubated in a water bath at 60°C for 1 h and treated with 0.2 mL Carrez I and Carrez II solution to remove proteins. The mixture was purified by centrifugation (Sorvall RC-5B Refrigerated Superspeed, GMI, Ramsay, USA) for 10 min at 10000 rpm. Total sugars were determined from the membrane-filtered supernatant (pores size 0.45 µm), and

sucrose, glucose, fructose, and maltose were separated using a LiChrospher 100 NH<sub>2</sub> (5 µm) 4 x 4 mm pre-column in combination with a LiChrospher 100 NH<sub>2</sub> (5 µm) 4 x 250 mm separation column (Merck KGaA, Darmstadt, Germany) and an acetonitrile - pure water solution (80:20 v/v) as mobile phase (flow rate 1.0 mL min<sup>-1</sup>) at 20°C and an injection volume of 20 µL. Sugars were detected with a Knauer differential refractometer 198.00 (Knauer, Berlin, Germany).

#### **6.2.3.4 NIRS Analysis**

NIRS analyses of the cooked and freeze dried sweet potato samples were performed following standard procedures, which included scanning of samples, selection of calibration and validation spectral sets as described in WinISI II software version 1.5 (NIRSystems, Inc., Silver Springs, MD, USA).

##### ***Scanning of samples***

Approximately 3 g of each cooked, freeze-dried and milled sweet potato sample was placed in a small ring cup. They were then scanned by NIRS monochromator model XDS (NIRSystems, Inc., Silver Springs, MD, USA) using the small ring cups, and their spectra collected between 400 and 2498nm, registering the absorbance values log 1/R at 0.5 nm intervals for each sample. Each sample was scanned twice (each scan produces 32 spectra) by refilling the ring cup cell to minimize effect of particle size and temperature on the sample. The average spectrum of each cooked sample was stored and used for calibration development.

##### ***Calibration and Validation***

Calibration and validation were performed according to the WinISI II software procedure. The spectra obtained from the scanned samples were separated into two sets, calibration set (89) and validation set (12) through random selection of

samples. Spectra of seven samples were deleted during the selection procedure due to the wide variation in their duplicate spectrum. Full spectra were used for the calibration equation development and cross validation procedure. Spectra in the calibration file, and reference values obtained from HPLC for sugars and polarimetric method for starch were used to establish multivariate equations. Spectra in the validation set were used to validate the calibration equation (Lu *et al.*, 2006). To optimize the calibration equations, combinations of 1<sup>st</sup> and 2<sup>nd</sup> derivatives, gap, smoothness and scatter correction were employed. Modified partial least squares (MPLS) regression with mathematical treatment 1,4,4,1 (The first number is the derivative, the second the gap, and the third and fourth numbers are the smooth) and scatter correction of standard normal variance and de-trend (SNVD) were employed for the development of optimum equations (Lu *et al.*, 2006). The results of the calibration equations were scrutinised by three types of outliers; critical T > 2.5, critical GH > 4 and critical X > 8 (Boneirbale *et al.*, 2008). The accuracy potential of the calibration equation was characterized by the standard error of calibration (SEC) and cross validation (SECV). The best calibration equation was measured by the highest coefficient of determination, and the lowest SEC and SECV (Wu *et al.*, 2002).

## **6.3 Results and discussion**

### **6.3.1 Diversity in individual sugar profile and total starch content of cooked sweet potato roots for NIRS calibration.**

Means, ranges, and standard deviations of sugar profile (sucrose, glucose, fructose and maltose), and total starch of cooked (baked, microwaved and steamed) sweet potato roots at harvest and storage are presented in Tables 6.2 and 6.3. Changes as influenced by different heating temperatures are also shown in Table 6.4. Wide



variation existed among the quality attributes of the freshly harvested and cooked samples, and stored and cooked samples. Whilst concentrations of sucrose, glucose, fructose, and starch generally decreased following cooking, maltose and total sugar contents increased dramatically. The increase in maltose content was primarily attributed to the degradative action of native amylases on starch during cooking as reported in Chapter Five in this study. Starch was nonetheless the predominant carbohydrate constituent of the cooked roots, constituting 40 to 70% of the dry matter content. The effect of different cooking treatments was also substantial. Baking caused the highest increased in sugars, followed by steaming and microwaving. The reasons for the highest effect by the baking treatment is described extensively in Chapter five, page 79.

**Table 6.2. Individual sugars, total sugar, and total starch of freshly harvested and cooked, and raw sweet potato roots**

Traits	Cooking treatments			Raw
	Baking	Microwaving	Steaming	
<b>Sucrose</b>	12.33(6 - 20)	10.72 (7-16)	8.30 (0-8)	11.80 (9-23)
<b>Glucose</b>	1.10 (0 - 3)	1.63 (0-5)	1.55 (0-5)	2.31 (1-9)
<b>Fructose</b>	0.84 (0 - 2)	0.92 (0-2)	0.95 (0-4)	1.95 (0-5)
<b>Maltose</b>	20.13 (5 -36)	5.07 (2-15)	14.35 (2-27)	0.63 (0-1)
<b>*Total Sugar</b>	33.64 (12-43)	18.05 (11-33)	24.93(10-34)	17.43 (11-37)
<b>Starch content</b>	50.40 (42-70)	57.99 (44-72)	57.76(43-72)	59.45 (44-73)

Ranges of attributes are presented in brackets

\*Total sugar was calculated from the individual sugars (sucrose, glucose, fructose and maltose)

**Table 6.3 Individual sugars, total sugars and total starch of stored and cooked, and raw sweet potato roots**

Traits	Cooking treatment			Raw
	Baking	Microwaving	Steaming	
<b>Sucrose</b>	11.28(1-10)	10.72 (8-14)	10.04 (1-19)	11.80 (9-23)
<b>Glucose</b>	0.43 (0-2)	0.40(0-2)	0.6 (0-2)	2.31 (1-9)
<b>Fructose</b>	0.20 (0-1)	0.23 (0-0.8)	0.3 (0-1)	1.95 (0-5)
<b>Maltose</b>	22.03(13-25)	8.53 (2-19)	16.81(13-30)	0.63 (0-1)
<b>*Total Sugar</b>	32.95(15-34)	19.88(10-33)	27.75 (26-49)	17.43 (11-37)
<b>Starch content</b>	54.24 (43-70)	58.95 (44-72)	52.92 (42-67)	59.45(44-73)

Ranges of attributes are presented in brackets

\*Total sugar was calculated from the individual sugars (sucrose, glucose, fructose and maltose)

Variability in quality traits during the heating experiment was comparatively lower than in the cooking treatments (Table 6.4). Changes in sucrose, glucose, fructose and starch content were not extensive. Increase in maltose and total sugar contents were however significant after heating. Storage treatment also induces formation of sugars especially maltose. Mean maltose content was higher in stored and cooked roots than freshly harvested and cooked. The outcome of this investigation depicts that sweetpotato roots undergo significant changes in chemical composition during storage and processing, and such variation should be considered during characterization and prediction of its end uses.

**Table 6.4. Mean values for individual sugars, total sugars and total starch of**

### heated sweetpotato genotypes

Traits (%DM)	Heating temperature (°C)			Raw
	65	75	85	
<b>Sucrose</b>	12.83 (12 - 14)	13.55(13-16)	12.52 (12-14)	11.80(9-23)
<b>Glucose</b>	1.99 (1.7- 2.3)	2.07 (1.4-2.7)	2.23 (1.8- 2.9)	2.31(1-9)
<b>Fructose</b>	0.82 (0.7 -12)	0.92 (0.6-1.2)	1.05 (0.8 -1.3)	1.95 (0-5)
<b>Maltose</b>	0.66 (0.6 -0.8)	4.04 (2-6)	6.07 (2-8)	0.63 (0-1)
<b>*Total Sugar</b>	17.31(16-18)	20.03 (18-21)	21.58 (17-21)	17.43 (11-37)
<b>Starch</b>	56.27 (43-72)	55.83 (43-72)	55.81(42-71)	59.45(44-73)

Ranges of attributes are presented in brackets

### 6.3.2 NIRS calibration development

NIRS calibration developed on the basis of 108 cooked sweetpotato samples for sugars (sucrose, maltose fructose, and glucose) and starch, are shown in Table 6.5. Statistically the calibration showed very high coefficient of determination for the calibration,  $R_c^2$ , (0.88 – 0.96). Starch content recorded the highest  $R_c^2$  of 0.96 and sucrose lowest of 0.88. The standard error of calibration (SEC) was very low, ranging from 0.25 to 1.89%. Fructose content showed the lowest SEC of 0.25% and maltose the higher of 1.89%. Validation results gave high coefficient of determination,  $R_{cv}^2$ , (0.80 – 0.92) and a small standard error of cross validation, SECV, (0.32 - 1.96%).

**Table 6.5. NIRS calibration and cross-validation statistics for cooked**

### sweetpotato root attributes

Attribute	Calibration		Cross Validation	
	$R_c^2$	SEC (%)	$R_{cv}^2$	SECV (%)
Fructose	0.91	0.25	0.90	1.07
Glucose	0.89	0.54	0.80	0.40
Sucrose	0.88	1.05	0.85	0.32
Maltose	0.94	1.89	0.92	0.64
Total Sugar	0.95	1.78	0.89	1.96
Starch	0.96	1.67	0.92	1.90

$R_c^2$  = Coefficient of determination in calibration. SEC = Standard error of calibration.  $R_{cv}^2$  = Coefficient of determination in cross validation. SECV = Standard error cross validation.

Lu *et al.* reported similar findings, in 2006 when a precision model was developed for rapid assessment of starch physicochemical and pasting properties in sweetpotato roots. With high coefficient of determination ( $RSQ = 0.84-0.92$ ) and low standard errors of prediction, NIRS technology was recommended as an effective and accurate tool for screening large numbers of starch samples in food and sweetpotato breeding programmes. In 2009, zum Felde *et al.* expanded the NIRS calibration model for sweetpotato quality attributes estimation to include; sugars (sucrose, glucose, maltose and fructose), beta-carotene, total carotenoids and total proteins. Again, high coefficients of determination ( $0.80-0.96$ ) and lower standard error of cross-validation ( $0.61-1.58$ ) were obtained. Results from Lebot *et al.* (2011) also confirmed earlier conclusions that NIRS calibration model has higher prediction accuracy for sugars and starch than minerals of sweetpotato roots. But these prediction models were developed on raw unprocessed sweetpotato root samples. Nevertheless, sweetpotato roots are usually cooked before consumption and quality attributes varied subsequently. This current study

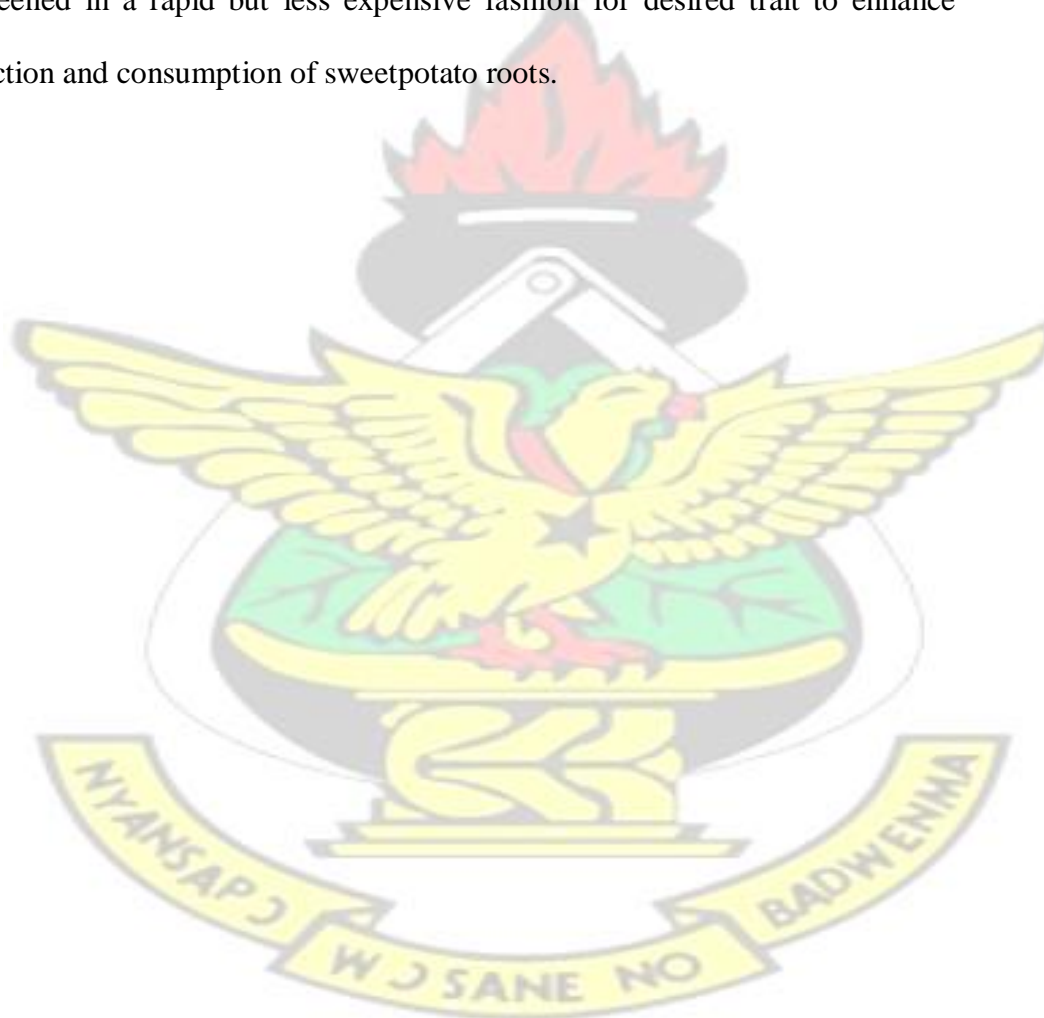


has established that NIRS calibration model has high precision for quantifying sugars and starches in cooked sweetpotato roots. Since cooking treatments differ in temperature, time and mode of heat transfer (Wang and Kays, 2000; Sun *et al.*, 1993), three different cooking treatments were considered and their contribution effects included. With this new development, determination of chemical composition in cooked sweetpotato samples, which significantly influences preferences and acceptability of sweetpotato materials, can be rapidly and efficiently evaluated. By so doing, desired eating and processing traits can be screened at the early stages of the sweetpotato value chain development, thereby ensuring effective selection of varieties with attributes likely to be demanded by customers. Where large numbers of materials are involved, a simple but accurate and less expensive method will be of great importance and NIRS is an obvious choice. NIRS also permits simultaneous estimation of multiple traits within a few seconds as compared to HPLC. Moreover, HPLC employs sophisticated pre-treatment procedures such as extraction with expensive solvents resulting in high cost of operation. Using NIRS to evaluate sugars in sweetpotato breeding programs would have benefits, particularly in West Africa where sugar content is a key attribute for consumer acceptability. Consumers in West Africa prefer less sweet types than traditional sweet varieties (Akoroda *et al.*, 2009) whilst in the United States sweeter, low dry matter dessert types are preferred (Kays *et al.*, 2005; Takahata *et al.*, 1994).

Actors along the value chain can give precise information on quality attributes of sweetpotato roots to the end user. Breeders can also release varieties with exact concentration of attributes of interest to farmers and consumers.

#### 6.4 Conclusion

NIRS calibration model has been developed for accurate estimation of sugar profile (fructose, glucose, sucrose, and maltose), and total starch content in cooked sweetpotato roots. The equation showed high coefficient of determination for calibration (0.88 – 0.96) and for cross validation (0.80 – 0.92). The standard errors reported were very low ranging from 0.25 – 1.98% justifying the efficiency of the equation. Large numbers of sweetpotato materials can thus be screened in a rapid but less expensive fashion for desired trait to enhance production and consumption of sweetpotato roots.



## CHAPTER SEVEN

### 7.0 GENERAL CONCLUSION AND RECOMMENDATIONS

#### 7.1 General Conclusion

The study sought to evaluate the carbohydrate composition (dry matter, starch and sugars) and hydrolytic power of the amylases in eleven officially released sweetpotato varieties by Crops Research Institute (CRI) under the Council for Scientific and Industrial research (CSIR), Ghana. Effects of the different production environments and processing conditions on the quality attributes were also investigated. In addition a calibration model was developed for screening the carbohydrate composition of cooked samples in a very fast but accurate fashion using Near Infrared Reflectance Spectroscopic (NIRS) technique.

In all, the study established that dry matter and starch content of the varieties evaluated were generally very high, with average value of dry matter content exceeding the world's mean. Sugar content in raw uncooked form was relatively low except in Apomuden which was classified as very sweet. Nevertheless the amylase potential, which indicates the hydrolytic power of the native amylases on gelatinised starch granules during processing, was high. Results from the different production environments, where sweetpotatoes are cultivated in Ghana, reveals significant levels with varied degree of variability on the carbohydrates and amylase activity. However genotypic influence was the highest compared to environmental. Effect of GxE was comparatively high on amylase activity than the other attributes evaluated. Some of the varieties showed high affinity for a specific environment in terms of a particular characteristic. Though environmental

control was a small fraction of the genotypic effect, it was significant for all the attributes. Thus concentration of dry matter, starch, sugars and amylase activity during development of sweetpotato roots is highly dependent on genotype, environment and interactions.

The present studies also revealed that  $\beta$ -amylase in sweetpotato roots are denatured at temperatures above 75°C. Most of the sweetpotato varieties evaluated completely lost their  $\beta$ -amylase activity at 85°C for longer holding time (40min). Genotype, temperature and their interaction significantly influences  $\beta$ -amylase during processing. Temperature and genotype showed the highest percentage of the total variability. The study has unveiled the biochemical processing potential of the sweetpotato varieties released in Ghana. With this scientific data, selection of varieties for specific industry application will be enhanced. The high amylolytic potential of sweetpotato varieties, in general, makes them a complete raw material for brewing, and syrup production.

Sugar profile and sweetness levels in sweetpotato root should be determined after cooking. The work revealed high significant differences between sugar concentrations in raw and cooked roots, as a result of starch conversion and formation of maltose during processing via the action of native amylases. Sugar levels in majority of the varieties evaluated, though low prior to cooking, became high following cooking. Nevertheless, effect of microwave treatment was minimal and was not significant in some of the varieties. To produce a less-sweet sweetpotato product, microwave cooking was recommended as an ideal method for consumers who preferred bland staples.

Moreover, NIRS calibration model was developed for estimating sugars and starch in cooked sweetpotato roots. The high precision of the calibration model shows



its accuracy in determining quality attributes especially sugars in cooked roots. By this technology, the utilisation potential of sweetpotatoes can be predicted in early generational breeding lines. The NIRS technique will also permit the selection of sweetpotato lines with superior quality attributes in addition to good agronomic traits.

## **7.2 Recommendations**

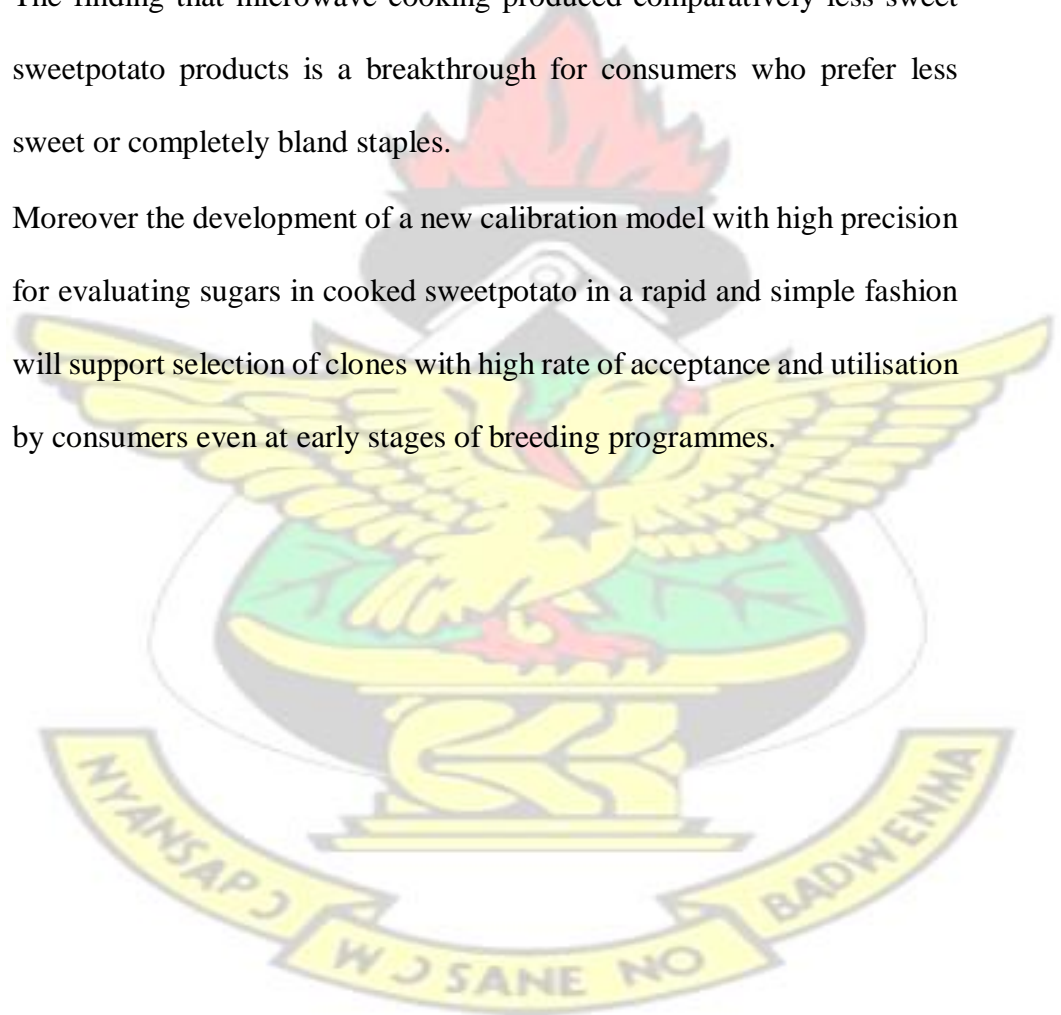
The consistency of the genotype by environment interaction effects over several years requires further studies. Data obtained should be used to test the viability of the current results. Moreover the possibility of applying the NIRS calibration model developed for cooked roots to estimate biochemical characteristic in raw roots or vice versa will enhance fast estimation of total quality attributes for selection of desired clones.

### **Contributions to knowledge**

The study has made significant contribution to knowledge by which food scientists and food industries can take advantage of;

- The work was successful in establishing the impact and significant contribution of genotype by environment (GxE) effects on amylase activity of sweetpotato roots. This current work was the first to analysis the effect of GxE on amylase activity using additive main effects and multiplicative interaction (AMMI) to apportion variety to its favourable environment(s). The amylase activity of the officially released sweetpotato varieties in the diverse production environments is known and hence actors along the sweetpotato value chain can select the highest or lowest amylase varieties per location for specific application.

- The complete profiling of sugars in cooked form will benefit both breeders and food scientists in selecting desired varieties as parents for further breeding programmes and defining clones suitable for the sweetpotato food industry respectively. Knowledge on sugar concentrations especially in cooked roots will also guide consumers and food processors in their choices of varieties for product development and food preparations.
- The finding that microwave cooking produced comparatively less sweet sweetpotato products is a breakthrough for consumers who prefer less sweet or completely bland staples.
- Moreover the development of a new calibration model with high precision for evaluating sugars in cooked sweetpotato in a rapid and simple fashion will support selection of clones with high rate of acceptance and utilisation by consumers even at early stages of breeding programmes.



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## APPENDIX

### 1.0 Preparation of solutions

#### 1.1 Betamyl-3 substrate

The substrate containing p-nitrophenyl- $\beta$ -D-maltotrioside (PNP $\beta$ -G3) plus  $\beta$ glucosidase (50 U) and stabilisers was dissolved in 10mL of boiled and cooled distilled water. The resultant solution was divided into four sized aliquots and stored in polypropylene tubes at -20°C between use. The unused reagents were stored on ice bath prior to use (Megazyme international, K-BETA, 2012).

#### 1.2 Tris/HCL extraction buffer

2.5ml of bottle containing Tris/HCL buffer plus disodium EDTA (20mM), BSA 10mg/mL and sodium azide (0.20% w/v) was diluted to 50mL with distilled water. Prior to use 0.88g of cysteine HCL was added and the final pH adjusted to 8.0 with 4 M NaOH. The solution was stored in refrigerator at 4°C

#### 1.3 MES buffer

48mL of MES buffer containing disodium EDTA (20mM), BSA 10mg/mL and sodium azide (0.20% w/v) was diluted to 500mL with distilled water.



Table A1: ANOVA for dry matter, starch, and sugars of 11 sweetpotato genotypes at five environments

Source of Variation	Variance %					
	Dry matter	Starch	Fructose	Glucose	Sucrose	Amylase activity
<b>Genotype</b>	92.44**	88.26**	83.07 **	84.23 **	88.34 **	65.89 **
<b>Environment</b>	4.46**	5.84**	13.67 **	11.96 **	2.80 **	10.01 **
<b>Genotype X Environment</b>	3.09**	5.90**	3.26 ns	3.81ns	8.45 **	24.06 **

\*\* Significant at  $p < 0.05$ . ns = not significant. \* calculated from sum Sq.

Table A2: ANOVA for dry matter in 11 sweetpotato genotypes at five environments

Source of Variation	Df	Sum Sq	Mean Sq	F value	Variance	% Variance
Genotype	10	5520.19	552.02	96.55 **	36.42	92.44
Environment	4	255.08	63.77	31.24 **	1.76	4.46
Genotype X Environment	40	228.69	5.72	2.77 **	1.22	3.09
Residuals	100	206.57	2.07			
CV	4.14					

\*\* Significant at the 0.05 probability level, CV – coefficient of variation.

Table A3: AMMI analysis of variance in dry matter content of 11 sweetpotato genotypes grown at 5 environments

Source of Variance	Df	Sum Sq	Mean Sq	F value	% Variance *
PC1	13	156.47	12.04	5.83 **	68.42
PC2	11	38.19	3.47	1.68 ns	16.70
PC3	9	21.64	2.40	1.16 ns	9.46
PC4	7	12.40	1.77	0.86 ns	5.42

\*\* Significant at  $p < 0.05$ . ns = not significant. \* calculated from sum Sq. PC = principal component

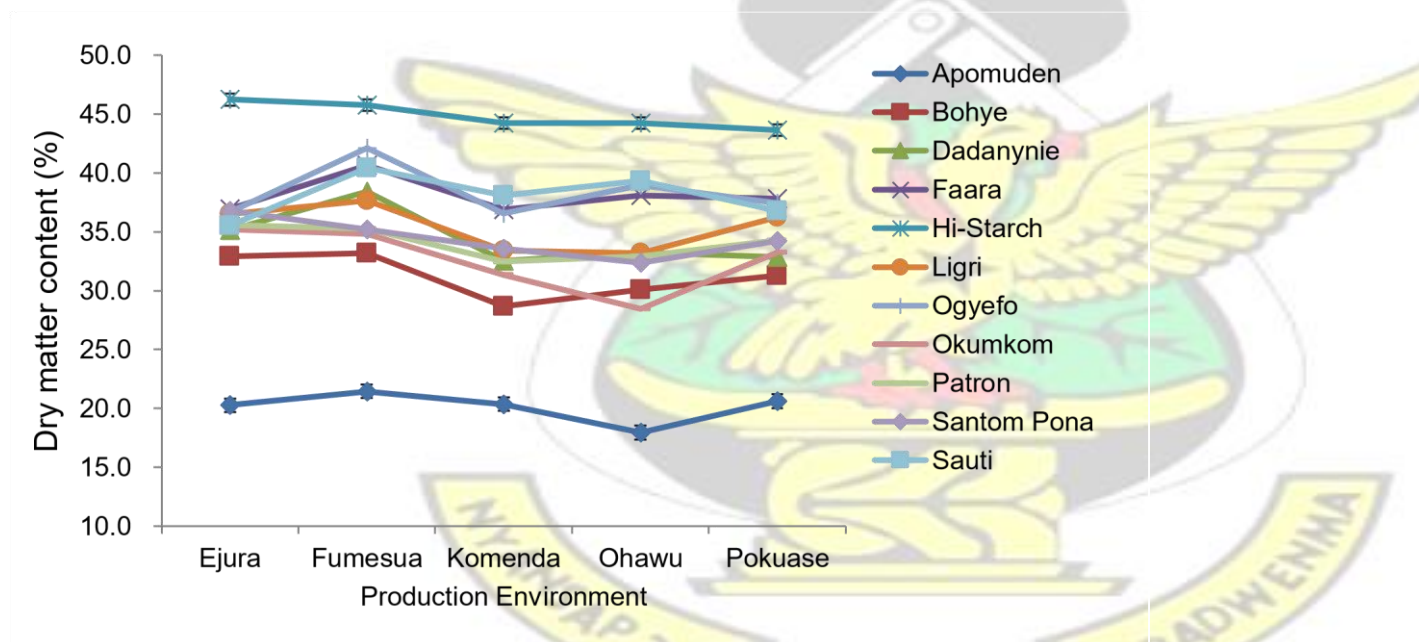


Fig A1. Storage root dry matter content of 11 sweetpotato genotypes across five environments. Error bars (Apomuden and Hi-Starch) represent significant differences between environments at  $p=0.05$

Table A4: ANOVA for starch content in 11 sweetpotato genotypes at five environments

Source of Variation	DF	Sum Sq	Mean Sq	F value	Variance	Variance %
<b>Genotype</b>	10	9003.15	900.32	252.19 **	58.99	88.26
<b>Environment</b>	4	587.43	146.86	23.72 **	3.90	5.84
<b>Genotype X Environment</b>	40	616.26	15.41	4.3 **	3.95	5.90
<b>Residuals</b>	100	357.00	3.57			
CV	2.87					

\*\* Significant at the 0.05 probability level, CV – coefficient of variation,

Table A5: AMMI analysis of variance in starch content of 11 sweetpotato genotypes grown at 5 environments

Source of Variation	DF	Sum Sq	Mean Sq	F value	% Variance*
<b>PC1</b>	13	328.68	25.28	7.08**	53.34
<b>PC2</b>	11	174.54	15.87	4.44**	28.32
<b>PC3</b>	9	72.77	8.09	2.26	11.81
<b>PC4</b>	7	40.27	5.75	1.61	5.53

\*\* Significant at  $p < 0.05$ . ns = not significant. \* calculated from sum Sq. PC = principal component

Table A6: ANOVA for endogenous sugars in 11 sweetpotato genotypes at five environments

Source of variance	% Variance		
	Fructose	Glucose	Sucrose
Genotype	83.07 **	84.23 **	88.34 **
Environment	13.67 **	11.96 **	2.80 **
G X E	3.26 ns	3.81ns	8.45 **

\*\* Significant at  $p < 0.05$ . ns = not significant. \* calculated from sum Sq. PC = principal component

Table A7: ANOVA for amylase activity in 11 sweetpotato genotypes at five environments

Source of Variation	DF	Sum Sq	Mean Sq	F value	Variance	% Variance
Genotype	10	5197979.99	519798.00	1689.53 **	32267.91	65.89
Environment	4	790447.22	197611.81	333.57 **	4895.803	10.00
G X E	40	1426565.24	35664.13	115.92 **	11782.78	24.056
Residuals	94	28919.86	307.66			

CV 2.74

Table A8: AMMI analysis of variance in amylase activity of 11 sweetpotato genotypes grown at 5 environment environments



Source of Variation	DF	Sum Sq	Mean Sq	F value	% Variance
<b>PC1</b>	13	857013.09	65924.08	227.95 **	60.07
<b>PC2</b>	11	373800.83	33981.89	117.50 **	26.20
<b>PC3</b>	9	114046.69	12671.85	43.82 **	7.99
<b>PC4</b>	7	81704.62	11672.09	40.36 **	5.73

Table A9: Sugar profile of 11 sweetpotato varieties

Variety	Fructose (%)	Glucose (%)	Sucrose (%)	Total Sugar (%)	Starch (%)
<b>Apomuden</b>	5.32	8.86	23.20	37.38(3.82)	44.11 (4.68)
<b>Bohye</b>	1.94	3.56	11.92	17.42(3.07)	64.90 (3.01)
<b>Dadanynie</b>	2.21	3.96	10.17	16.35(2.43)	68.50 (2.06)
<b>Faara</b>	2.04	3.76	9.67	15.47(2.37)	66.91 (2.93)
<b>Hi-Starch</b>	0.86	2.21	8.70	11.77(1.43)	73.00 (2.70)
<b>Ligri</b>	1.47	3.03	11.93	16.43(2.51)	68.22 (0.80)
<b>Ogyefo</b>	0.34	1.55	9.34	11.23(0.96)	72.33 (1.52)
<b>Okumkom</b>	3.23	5.17	11.39	19.79(3.40)	63.07 (3.22)
<b>Patron</b>	1.25	2.64	14.37	18.27(4.22)	65.82 (3.16)
<b>Santom Pona</b>	1.68	3.03	10.66	15.38(2.07)	67.15 (1.85)
<b>Sauti</b>	1.13	2.57	8.50	12.20(1.51)	69.08 (1.57)

Table A10: Soil analysis of five growing environment

Growing Environment	Agro-ecological zone	Soil parameters			
		Organic %	Carbon	Total Nitrogen %	Organic Matter %
<b>Ohawu</b>	Coastal Savannah	0.62		0.06	1.07
<b>Komenda</b>	Coastal Savannah	0.62		0.06	1.07
<b>Pokuase</b>	Coastal Savannah	0.62		0.06	1.07
<b>Ejura</b>	Forest Transition	0.62		0.06	1.07
<b>Fumesua</b>	Forest	0.69		0.07	1.19
<b>Mean</b>		0.63		0.06	1.09

## Soil analysis

Organic carbon, total nitrogen and organic matter were determined by method employed by Hesse (1971)

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## List of Publications

### Peer-review international journals

1. Owusu Mensah, E., Oduro, I., Ellis, W.O., Derry, K.E., and Carey, E.E (2016). COOKING TREATMENT EFFECTS ON SUGAR PROFILE AND SWEETNESS OF ELEVEN-RELEASED SWEETPOTATO VARIETIES. *J Food Process Technol* 7:580. doi:10.4172/2157-7110.1000580
2. Owusu Mensah, E., Oduro, I., Ellis, W.O., Derry, K.E., and Carey, E.E (2016). THERMAL STABILITY OF  $\beta$ -AMYLASE ACTIVITY AND SUGAR PROFILE OF SWEETPOTATO VARIETIES DURING PROCESSING. *Journal of Nutrition & Food Sciences*. Manuscript no. LifeSci-16-88

### Peer-review international conferences

1. Owusu-Mensah, E., Ibok, I., Ellis, O.W., Obeng-Bioh, E., Carrey, E.E Multi-locational assessment of some physicochemical attributes and amylase activity of sweetpotato varieties and elite materials in Ghana at the 16<sup>th</sup> Triennial Symposium of International Society for Tropical Root Crops (ISTRC), Abeokota, Nigeria, 2012.
2. Owusu-Mensah, E., Oduro, I., Ellis, W.O., and Carey, E.E. (2013). Temperature-time effects on  $\beta$ -amylase activity of sweetpotato roots during processing at 9<sup>th</sup> triennial conference of the Africa Potato Association, Naivasha, Kenya.



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