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1. General introduction

1.1 Importance of plantain

Plantains (Musa spp) remain one of the earliest crop plants to have been domesticated. They were originally adapted from the humid tropics to broad subtropical climatic conditions. Plantains continue to be the most important, but orphan and under researched, food crops in the world. Plantains provide a staple food for over 70 million people; particularly many ethnic groups in Africa, an area where the green revolution has had little influence (Ravi, et al., 2013) and the region identified as a climate change hotspot (Shepard, 2019). Plantains are important food security crops, providing a cheap and easy to produce source of carbohydrate. Plantains are known to be potassium-rich, boost immune system and regulate digestion. In addition, they are rich in certain minerals and vitamins A, C, and B6. Plantains are consumed in various forms, and consumption methods have evolved and been refined by humans over time. They are eaten cooked, baked, steamed, or fermented. Among some ethnic groups, the whole plant is exploited with uses for the leaves, pseudostem, medicinally rich plant sap or fiber. Plantains are grown for specific purposes apart from the edible fruit; the crop has been integrated into the culture and livelihoods of many societies (Ravi, 2013). Among the typical Akan tribe in Ghana, shortage of plantain on the market is a sign of famine. Although today plantains are best known as a food crop, almost every part of the plant can be used in one way or the other. Plantain is a tall (2-9 m) perennial monocotyledon (INIBAP, 2000). The whole trunk is a pseudo-stem and consists of concentric layers of leaf sheaths rolled into a cylinder of between 20 and 60 cm diameter (Pillay and Tripathi, 2007). The true stem is a large underground rhizome, which produces aerial shoots that arise from the lateral buds, which develop into buds and later suckers. The continuous vegetative growth of suckers perpetuates the rhizome's life and hence the perennial status of plantains. Plantains have adventitious roots which extend laterally to 5m around the plant. They are predominantly clustered in a mat composed of the parent plant and some followers. Plantains produce a leaf a week under irrigated conditions and a leaf in 12 days under rain-fed conditions. Plantains produce at least 42 leaves by the time of flowering and the last leaf forms the floral leaf. Flowering in plantain usually begins around six (6) (under irrigation) – nine (9) (under rain-fed) months after planting, depending on climatic conditions and type of soil (Augstburger et al., 2001). At emergence, the inflorescence is usually erect but points downward due to its weight (Robinson, 1996). Development of the bunch takes 3 - 4 months depending on the weather and cultivar. After full formation of the bunch, the parent plant dies off. The death of the parent crop after harvest weakens the base of the mat. This is due to decomposition of the base which results deposition of fibrous materials in the hole. New roots from daughter plants are not able to grow toward the centre where the parent crop was growing hence weakening the anchorage. Edible plantains do not have seeds but degenerated seeds are visible as blackened bands at the centre (Augstburger et al., 2001). Plantains grow well in soils rich in organic matter with good drainage and a pH range of 5.6 - 7.5. They also require considerable amounts of nitrogen (N) and potassium (K) to maintain high yields. In situations where the soil fertility is poor, organic and inorganic soil amendment could be incorporated. The temperature required for growth is 13 - 38° C with the optimal

Plantain production systems vary from backyard garden, subsistence and commercial plantation systems with the subsistence system being the most common in the tropics (Karamura et al., 1998). Under subsistence production, farm sizes are often 0.4 of a hectare. Plantation systems have high levels of management and reduced pest and disease incidence. This is often practiced by the International export companies in sub-Saharan Africa. In Ghana for example, there are three major international export companies producing on the average 700 hectares each. Their production systems are solely monocropping under irrigation unlike the subsistence system which is characterized by intercropping.

being 27° C and annual mean rainfall of 1500 - 2500 mm.

1.1.2 Plantain as food

Plantains can be boiled green, slightly ripe, ripe or over-ripe. It can also be processed into flour, fermented for production of vinegar, wine and beer (Pillay and Tripathi, 2007). It can also be fried green, ripe and over-ripe (Dzomeku et al., 2006; 2007). The trunk and leaves can be fed to livestock. Plantain leaves have a variety of uses. The leaves can be used as bio-plates for wrapping food (Dzomeku, et al., 2007), polish floors, as thatch and are also a good material for mulch. Fibres from the pseudostem and leaves are used for making cloth and ropes (Nelson et al., 2006). The sap from the pseudostem can be used as dye in the textile industry (Dzomeku and Boateng, 2012)

Plantains have been found to have medicinal properties (INIBAP, 2003); as the sap is used to stop bleeding of fresh wounds. The root sap can be used to treat mouth thrush in children and skin warts and plantain peel has been found to have antibiotic properties (Nelson et al., 2006). In the rural settings, the ash from the peel are used for cleaning the teeth and for soap making.

1.1.3 Field establishment

Plantain plantations are established on land that has been cleared manually or with herbicides. A plantain plant occupies a minimum space of six square meters. The recommended spacing is 3m x 2m or 3m by 3m between and within rows depending on the cultivar. On sloping land, the rows follow contours and terracing can be done to prevent soil erosion (Tushemereirwe et al., 2001). Planting of conventional sucker on slopes requires that the cut edge of the sucker be planted opposite the slope. This helps the plant during bunch carrying on the tree. Weeding is done to prevent competition and mulching to preserve moisture. Old dry leaves and surplus shoots are removed periodically from the plant. Plantain varieties with tall trunks are often propped using forked sticks to prevent the plant from falling due to the weight of the bunch. Bunches are harvested and transported carefully to prevent postharvest diseases (Augstburger et al., 2001).

1.1.4 Cultivated area and yield performance under optimal conditions

Plantains are part of the smallholder cropping systems and contribute significantly to the socioeconomy of the growing countries. Traditional plantains are grown as intercrops of smallholders and also provide shade for cocoa. With the exception of a few large farms, most farms are about 0.8 hectare in size. Plantains are grown in more than a hundred tropical and subtropical countries and provide staple food for hundreds of millions of people. Plantains are grown in about 120 countries around the world, exhibiting a spectacular production of 122.85 million tons (FAOSTAT, 2012). Four of the top eight plantain producing countries in the world are in West Africa (Ghana, Cameroon, Nigeria, and Côte d'Ivoire). African countries account for the majority of global production (about 62% of total plantain production) with Ghana as the largest world producer; growing over 3.5 million MT (\approx 13% of global production).

1.1.5 Plantain production constraints

Plantain production in Ghana has not reached its full potential. The average yields are about 11 tons per hectare as against a potential yield of up to 20 tons per hectare (MOFA-SRID, 2015). This is mostly due to poor soil fertility, pests and diseases that have increased considerably;

leading to reduced life span of plantations from 8 to 4 years. Scarcity of clean planting material has contributed to disease spread due to farmers using infected suckers. There are no formal standards to regulate plantain planting material production (Macharia et al., 2010) with many farmers using naturally regenerated suckers due to their availability and low cost. Also, there are limited seedling production facilities that provide quality plantain seedlings. Thus, there is the need for farmers to have access to certified, healthy and affordable planting material. In addition, rising population pressure on land has led to shortened fallow periods leading to decline in soil fertility. Limited opportunity for rotation also has led to planting in infected soil thus further compounding the problems. Low levels of farm inputs, lack of credit facilities and poor management due to lack of information by farmers result in poor yields. Other constraints include; poor marketing infrastructure, perishability, postharvest losses, competition with other crops for land, high cost of labour and capital, genetic erosion and climate variability (Tushemereirwe et al., 2001).

1.1.6 Environmental stress and plant responses

Water is one of the most important abiotic factors for plant growth. Plants require water for photosynthesis, nutrient uptake and transportation as well as cooling (Farooq et al., 2009). Plantains are sessile hence unable to move when the environment becomes unfavorable. Accordingly, plantains have to be able to respond and adapt to the local environmental changes. Since water is essential for their survival, the ability to tolerate water stress is crucial.

Water limitation is a major problem for plantain production globally, permanently affecting 28% of the world's soils with almost half of all soils intermittently limited because of shallowness, poor water holding capacity, and other factors (Dudal, 1977). Drought in agriculture is "shortage of water in the plant rhizosphere, resulting in decreased crop yield" (Salekdeh et al.,2009). Drought tolerance consists of drought avoidance and/or dehydration tolerance that are ultimately measured by the reproductive success of the species (Taylor et al., 2007). Drought avoidance strategies in plants include deep rooting, conservative use of available water to ensure fruit filling is completed, and lifecycle modifications to match rainfall. Dehydration tolerance involves the plants' ability to partially dehydrate but remain viable and grow again when rainfall resumes.

The effect of water stress on plantain growth is complex and they respond with many protective adaptations. Water stress causes the plantains to suffer from dehydration and overheating of its

cells and tissues. Hence, water stress resistance of the plantain includes the ability to withstand dehydration and ability to withstand overheating (heat-resistant). High heat-resistance is not always linked with high drought-resistant and there is no universal mechanism of adaptation of plants to drought. With severe water stress plantain experience photosystem collapse. Drought-resistance is a property which is formed and developed in the process of ontogenesis and is based on the whole preceding phylogeny of the plant. Based on the above, Henckel (1964) defined drought resistance as follows: "Drought-resistant plants are those which in the process of ontogenesis are able to adapt to the effect of drought and which can normally grow, develop, and reproduce under drought conditions because of a number of properties acquired in the process of evolution under the influence of environment."

Drought is one of the important abiotic constraints restricting plantain cultivation and its further adoption into non-conventional growing areas. About 80% of the plantain plant is water, hence during prolong drought periods under rain-fed systems, the crop experiences severe water loss leading to stem logging. It is therefore not recommended to carry out pruning of leaves during the dry seasons. Breeding for drought alone has not been focused among any of the global banana breeding programs but it has been an essential trait considered along with other important ones like Fusarium wilt (race 1, 2, and 4), Sigatoka leaf spot (*M. fijiensis, M. eumusae*, and *M. musicola*), etc. Recent issues of climate change have warranted the need for the development of new cultivars suitable for less water environments.

1.1.7 Plantain plant water relations

Plantains pose challenge to physiologists to measure indicators of water deficits, due to the presence of large air pockets within the leaves, and laticifers containing latex within the leaves, fruit, and corm that hinder the use of standard methods of measuring water relations (Turner and Thomas, 1998). Milburn et al. (1990), Kallarackal et al. (1990), Turner and Thomas (1998)), subsequently demonstrated different methods to measure a series of physiological indications in relation to drought tolerance, *viz.*, of water potential, the volumetric (relative leaf water content), or thermodynamic tissue water status (leaf water, osmotic, and pressure potentials) of a laticiferous plant like the plantain. The method described by Milburn et al. (1990), which is based on measurements of the refractive index of exuded latex, was preferred and its reliability subsequently confirmed by Thomas and Turner (2001). The water potential of well-watered plants was found to cycle diurnally within the remarkably narrow range of 0

to -0.35 MPa. In fact, the rate of extension of the youngest leaf may be the most sensitive indicator of plant water status (Kallarackal et al., 1990), provided it is not too hot (Thomas and Turner, 1998). Under hot, arid conditions, leaf folding is not considered to be a reliable plant-based indicator of when to irrigate (Thomas and Turner, 1998).

Water stress influences the phenology of plantain crop. Whereas plantain is a crop with permanent production, the harvesting periods could be altered by external factors such as strong winds and rainfall. These production movements or periods in turn cause upward and downward price trends according to supply and demand volumes. Plantains are scarce on the market from May to August. The scarcity is due to the prolonged drought period that dehydrates the plants and strong winds associated with the start of the rains. Plantains therefore experience stem logging hence affecting production in West Africa.

Plantain production constraints are dominated largely by biotic and abiotic stresses. However, while research on biotic stresses has drawn sufficient attention worldwide, abiotic stresses have gone unnoticed. Among the abiotic stresses, drought, salinity and heat are the most important. Drought has rarely been addressed in the past, but is gaining importance in the face of depleting natural resources. The results of successful cultivation, especially of the water loving Cavendish dessert bananas, in drought prone areas with protected irrigation have provided the required momentum to perform research on drought in bananas. In subtropical and semi-arid banana cultivation zones, where rainy days are limited and there is an uneven distribution of rainfall, new crop management practices in terms of varieties selected, soil improvement (in terms of physical properties and nutrient enrichment), water management, etc. are being adopted. Although a large amount of research has been carried out in the tropics including water management, drip irrigation, and fertilizer use, work on evaluation of plantain varieties under conditions of water deficit is still very limited, as is the availability of related information. Probable reasons could be that most genebanks and breeding programs actively involved in germplasm evaluation and development are located in the humid tropics with ample rainfall. Also, creating large-scale drought conditions for a crop like plantains that is large and of long duration (12–20 months), presents many practical difficulties.

Screening germplasm for the drought has been conducted in some breeding programs such as that of the International Institute for Tropical Agriculture (IITA), Nigeria, NRCB, India and the Centro de Investigación Científica del Yucatán (CICY), Mexico. IITA has planted a large amount of germplasm in semi-arid zones of Uganda. The materials that were screened for drought tolerance includes landraces, East-African highland bananas, plantains and their triploid and tetraploid hybrids (Ravi and Uma, 2009).

1.1.8 Responses of leaf stomatal density to water status and its relationship with photosynthesis

In an agricultural context, water deficit is one of the most important environmental factors constraining plant photosynthesis and productivity. Consequently, plant responses to drought have been extensively investigated from molecular, physiological, and individual plant to ecosystem levels (Bray, 1997; Chaves et al., 2003). Water stress may reduce leaf net photosynthetic assimilation (An) by both stomatal and metabolic limitations (Farquhar and Sharkey, 1982; Chaves et al., 2003; Ghannoum et al., 2003; Ripley et al., 2007). Also, many studies have reported that stomatal effects are major under moderate stresses, but biochemical limitations are quantitatively important during leaf ageing or during severe drought (Grassi and Magnani, 2005; Galle´ et al., 2007). An early response to water deficit is a reduction in leaf area and plant growth, which allows plants to reduce their transpiration, thus increasing water use efficiencies (WUE) (Xu and Zhou, 2005; Monclus et al., 2006; Aguirrezabal et al., 2006), and promoting interspecies competition capacity under drought (Xu et al., 2007).

Plant stomata, the vital gate between plant and the atmosphere plays a pivotal role in plant responses to environmental conditions, which have been and are being investigated from molecular and whole plant perspectives, as well as at ecosystem and global levels (Nilson and Assmann, 2007). Many researchers have reported stomatal density responses to various environmental factors, such as elevated CO₂ concentration (Woodward, 1987), heat stress (Beerling and Chaloner, 1993), salt stress (Zhao et al., 2006), drought (Lecoeur et al., 1995; Zhao et al., 2001; Galme's et al., 2007), precipitation change (Yang et al., 2007), and plant density (Zhang et al., 2003). Other studies have shown that water deficit can lead to increase in stomatal density (McCree and Davis, 1974; Cutler et al., 1977; Yang and Wang, 2001; Zhang et al., 2006), and a decrease in stomatal size (Cutler et al., 1977; Quarrie and Jones, 1977; Spence et al., 1986; Martinez et al., 2007).

Leaf morphological traits, including stomatal density and distribution, and epidermal features also influence gas exchange quite remarkably and their relationships with key environmental factors such as light, water status, and CO₂ levels (Woodward, 1987; Nilson and Assmann, 2007). It is reported that increasing the blue-light response of stomata through red light is closely associated with guard cell chloroplast activity (Roelfsema et al., 2006), indicating that leaf photosynthetic activity may involve stomatal movement and development (Shimazaki et

al., 2007). The balance between carbon gain through photosynthesis and water loss through transpiration could also affect instantaneous WUE. Unfortunately, how stomatal density affects gas exchange has received little attention.

1.1.9 Responses of stomatal parameters and SPAD values in plantain

Studies have shown that stomata may respond homogeneously or heterogeneously to environmental conditions. It is reported that when the widths of the stomatal pores are varied, plants are able to control the flux of CO_2 and H_2O between the leaf and the environment in response to changes in environmental factors. Nevertheless, not all leaf stomata are open at the same time at a particular moment. This physiological behaviour, named patchy stomatal closure, occurs when stomata of different locations on the leaf show different responses to similar perturbations (Terashima, 1992; Larcher, 1995; Mott et al., 1997; Haefner et al., 1997). Studies by Cardon et al. (1994) showed that during strong oscillatory behaviour or even at a steady-state stomatal conductance, a surprising number of different behaviors in the dynamics of the photosynthetic activity were found.

Heath (1950) in his study observed that reductions in CO_2 below ambient levels induced stomatal opening, an eco-physiological response of great interest, and that the site of CO_2 sensing was most probably in the sub-stomatal cavity and not the guard cells. Since then stomatal research has become critically important to crop production, biodiversity responses, and hydrology (particularly in terms of 'run-off') with respect to rising atmospheric CO_2 , changing water regimes, and growing populations (Matthew et al., 2011). Stomatal physiological research has brought to the fore the role of stomata in the evolution of terrestrial vegetation and development of the terrestrial landscape and atmospheric composition is becoming increasingly evident, alongside the use of fossil stomata as palaeo-proxies of past atmospheres (McElwain et al., 2004; Berry et al., 2010; Smith et al., 2010).

The stomatal control responses of plants consist of 'short term' stomatal aperture changes in response to availability of water, light, temperature, wind speed, and carbon dioxide, and also 'longer term' changes in stomatal density that set the limits for maximum stomatal conductance in response to atmospheric CO₂, light intensity/quality, and root-to-shoot signals of water availability (Davies et al., 2000; Casson et al., 2009). Stomatal control determines the water use efficiency (WUE) of a plant by optimizing water lost against carbon gained. Also, stomatal control mechanisms employed by a plant species will determine: the risk of xylem embolism

by reducing the probability of cavitation through stomatal closure during episodes of high transpirative demand (Brodribb and Jordan, 2008; Meinzer et al., 2009); leaf temperature and resistance to heat stress (Jones et al., 2002); tolerance to toxic atmospheric gases (Mansfield and Majernik, 1970); nutrient uptake via promotion of root mass flow (Van Vuuren et al., 1997); and the maximum rate of photosynthesis (Korner et al., 1979). It therefore implies that plants with more effective stomatal control will be expected to be more successful than those with less effective stomatal control.

1.1.10 Fruit developmental characteristics of Apantu (Local False Horn) plantain for harvest indices determination

Plantain (AAB subgroup) is one of the earliest crops domesticated by man and grown in organized agriculture in the humid forest zones of West and Central Africa. Plantain makes up the fourth most important world food item after cereals in terms of the gross value of production. They have been identified to be of great socio-economic and nutritional significance in growing regions (Dzomeku et al., 2011). In the developing world plantain is identified as the fourth most important food commodity after rice, wheat and maize (FAO, 2010, Wattanachaiyingcharoen and Boonyanuphap, 2003).

Despite the high value of plantain, poor soils, growing pest threats and disease pressures have affected production, the most notable being the fungal disease - Black Sigatoka (*Mycosphaerella fijiensis*) (IITA, 1992; Stover and Simmonds, 1987; Swennen, 1990). Yield losses due to the disease are highly significant ranging from 20 to 50%. Under very severe conditions yield losses may be as high as 80%. All the plantain landraces in Ghana are susceptible to black Sigatoka disease and other pests except cooking bananas (Dzomeku, et al., 2006). The fear that the disease could wipe out the susceptible cultivars, research efforts have been to introduce developed hybrids in readiness for any eventuality. New hybrids, resistant or tolerant to the Black Sigatoka disease were introduced into the country as a long term solution. A large deployment project was executed with four hybrids.

Apantu, a False Horn plantain is the most popular plantain cultivar in Ghana. Apantu cultivation has become a feature of great socioeconomic importance in Ghana from the point of view of food security and job creation. It belongs to the non-traditional sector of the rural economy, where it is used mainly to shade cocoa and is also an essential component of the diet. Apantu is also very important sources of rural income (Ortiz, and Vuylsteke, 1996). More than

90% of the cultivated area in Ghana belongs to smallholder farming system. In the agricultural sector, plantain is ranked fourth in Ghana (FAO, 2010) and contributes about 13.1% to the Agricultural Gross Domestic Product (AGDP). Its per capita consumption of 84kg (SRID-MOFA, 2010) is higher than all other starchy staples. A total of 359,865 hectares of land area in Ghana is used to cultivate plantain producing an annual average of 3.7 million tonnes of fruits, of which more than 95% is sold on the domestic market (SRID-MOFA, 2010). Apantu production is concentrated in the three agro-ecological zones namely Rain forest, Moist semi-deciduous forest and Forest-savanna transition. The rainfall pattern is bi-modal from March to July as the major rainy season and August to November as the minor season.

Although False Horn (Apantu) plantain plays a significant role in the farming systems in Ghana, there has not been any conscious effort to develop any maturity indices for the crop. Maturity indices of plantain under rainfed production could be unreliable as they are often influenced by the environment. Physical, biochemical and physiological parameters are used to define the maturity stage for harvesting of fruits (Jha, et al., 2006). Huda, (2003) and Robinson and Saúco, (2010) described that the banana fruits having the stage of three quarters round was considered as the commercial maturity. Irtwange (2006) considered the parameters such as chronological age (elapsed days from bloom to harvest), size, shape, surface characteristics, colour, firmness, compositional factors (soluble solids), development of abscission layers, surface morphology, solidity, tenderness, sugars, starch, sugar-to acid ratio and oil content for the determination of maturity indices. The common maturity indices used by most plantain farmers for assessing for harvest in view of the absence of universal criteria, are any combination of the following: (i) by experience and judged largely by the visual appearance of the hanging bunch and particularly by the angularity of individual fingers (Palmer, 1971). (ii). fruits harvested when the fingers of the first hand on the bunch show signs of ripening or yellowing or when the finger tips turn black (Dadzie, 1994b,c). (iii). In commercial plantations, fruits destined for distant markets are harvested at a stage known as 'three quarters full', when the fingers are still clearly angular. For local markets fruits are often harvested when fingers are full or rounded (iv). Usually coloured ribbons are used in commercial plantations to provide information regarding bunch age. (v). fruit diameter (or caliper grade of fruit) and fruit length may be used as criteria to determine when to harvest (Dadzie and Orchard, 1997). Dadzie and Orchard (1997) stated again that while it is advantageous if the maturity index is non-destructive, (Reid, 2002) so that every fruit can be evaluated, it is important that the indices can be measured in a rapid, simple and inexpensive way. The method of evaluation should be simple and easily replicable, not requiring any complex scientific methodology so that farmers with basic literacy in mathematics can evaluate their plantain production and maturity time. In other jurisdictions, various instrument based techniques are used to measure maturity and ripeness. It is important to note that if the fruits are very mature at harvest, particularly following a heavy rain, peel splitting can occur.

It is evident that under rainfed conditions, the maturity indices are influenced by the environment. In the dry season angularity may be deceptive as an index of maturity. Also the soil nutrient and soil moisture could also influence the maturity indices.

1.1.11 Objectives and hypothesis

The objective of this study was to understand the climate change effects on the whole plant physiology of plantain. Plantain, though a food crop, is a long duration crop under subsistence production system and therefore any adverse effect of climate change affects the food security of the system. It was therefore critically important to evaluate the eco-physiological responses of the crop to climate variability and the characteristics of Apantu with maturity indices.

Abiotic stresses play a major role in influencing the growth and development of plantain. Abiotic stresses, drought and heat are the most important factors that affect plantain fruit development. Deficiency of any of these factors can affect the full developmental potential of the fruit. Assessing physiological responses of plantains and bananas to water deficits with a standard method is hindered by the presence of large air pockets within the leaves, and laticifers containing latex within the leaves, fruit, and corm (Turner and Thomas, 1998).

The specific objectives were:

- 1. to understand plantain response to on-farm technology of rapid multiplication of uniform and healthy planting materials
- 2. To assess the possibility of plantain yielding itself to natural mycorrhization
- 3. To evaluate the responses of the crop to drought stress
- 4. To elucidate the postharvest characteristics of plantain for harvest indices determination
- 5. To evaluate climate change effects on carotenoid pro-Vitamin A levels of plantain

1.1.12 Publications

This doctoral thesis generated five publications. These articles have been published (Article I, II, III, IV, V) or are submitted to peer reviewed academic journals. In the following, each publication is presented in one chapter. Within each chapter, the reference system, figure style and language style was applied according to the authors guidelines of the journal to which the publication was submitted. The copy right of these articles are with the corresponding author and the articles are allowed to be reproduced in my thesis.

Article I

Dzomeku, B.M.; Darkey, S.K.; Wünsche, J. and Bam, R.K., 2014. Response of selected local plantains to PIBS (Plants Issues de bourgeons secondaires) technique. J. of Plant Development. 21:117-123.

Article II

Dzomeku, B.M.; Darkey, S.K.; Bam R. K.; Sarkodie Addo, J. and Wünsche , J. 2016. Exploratory assessment of potential mycorrhization of two landraces of plantain. Journal of Basic and Applied Research International. 18 (3):146-151.

Article III

Dzomeku, B.M.; Sarkordie-Addo, J.; Darkey S.K.; Bam, R.K.; Wünsche, J. and Staver, C. 2016. Responses of Leaf Stomatal Parameters to Induced Water Stress and its Relationship with Stomatal Conductance in False Horn Plantain. International Journal of Plant & Soil Sciences. 12 (2):1-14.

Article IV

Dzomeku B. M.; Sarkordie-Addo J.; Darkey S. K.; Bam R.K and Wuensche J. 2016. Evaluating postharvest characteristics of Apantu (Local False Horn) plantain for harvest indices determination. International Journal of Plant Physiology and Biochemistry. 8(1):1-6. DOI: 10.5897/IJPPB2015.0235

Article V

Dzomeku, B.M.; Wald, J. P.; Wuenche, J. and Nohr; D. and Biesalski, H.K., 2020. Climate change enhanced Carotenoid pro-Vitamin A levels of selected plantain cultivars. Plants *9* (4), 541; https://doi.org/10.3390/plants9040541

2. Response of selected local plantains to PIBS (Plants Issues de bourgeons secondaires) technique

2.1. Abstract

One major constraint to plantain production has been inadequate healthy planting materials at the time of planting. Several technologies for multiplying healthy planting materials exist but could not meet farmers' demand. A study was conducted to assess the performance of various landraces plantain to plants issus de bourgeons secondaires (PIBS) technique. Five cultivars of Musa sapientum (Apantu (False Horn), Asamienu (True Horn), Oniaba (intermediate French plantain) and FHIA-21 (tetraploid hybrid plantain) were tested to determine their response to the PIBS technique. Sword suckers of each cultivar with weight of between 0.2-0.5 kg were prepared and buried in fine sawdust in a humidity chamber built using transparent polyethylene sheets. Results at harvest showed that removal of rooted sprouts started three weeks after planting and every week thereafter for eight weeks. The intermediate French plantain cultivar (Oniaba) produced the least average number (about 20) of healthy planting. Apantu (False Horn) produced an average of about 75 healthy planting materials. The hybrid FHIA-21 on the other hand generated an average of about 85 healthy planting materials. Asamienu (True Horn) produced the highest healthy seedlings of about 90 healthy planting materials. The results revealed that the leaf scar carries a primary bud at the intersection of each leaf sheath and several eyes along the entire length of the leaf sheath which could not have developed into suckers. However, with this technique the eyes could be activated to sprout as healthy planting materials. The technique proved as an efficient method of multiplying healthy planting materials for plantain and could thus be recommended for adoption not only by peasant farmers but also to others who could become commercial seed producers. But there will be a need for certification guidelines for seed growing systems.

Keywords: Musa, plantain, macro-propagation, planting material, PIBS

2.2 Introduction

Plantains and bananas are classified according to genome group. Majority of cultivated plantains are triploid (2n = 3x = 33), that are derived from intra-specific crosses within *Musa acuminata* Colla (A genome) and inter-specific crosses between *M. acuminata* and *Musa balbisiana* Colla (B genome). The remainder is mostly diploid, while tetraploid clones are naturally rare. The tetraploid plantains are often as a result of breeding programmes by

research. They are also classified as belonging to False Horn, True Horn and French plantain groups based on the morphology of the fruits. Local landraces of plantain a member of the AAB subgroup is among Africa's most important starchy food and cash crops [STOVER & SIMMONDS, 1987]. Nearly 30 million tons of plantain is produced yearly in Africa, mostly by small holders and consumed locally [FAO, 2010]. It is a crop suitable for the humid forest zones with high rainfall conditions. Despite the economic potential of plantain, farmers are confronted with high yield losses caused by pest and disease constraints such as nematodes [FOGAIN, 2000], banana weevils, and foliar diseases such as black leaf streak (Mycosphaerella fijiensis) and invasive weeds [ROBINSON, 1996] such as Chromolaena odorata, Panicum maxima, etc [HAUSER & MEKOA, 2009]. In the traditional low input systems, no pesticides are used and integrated control methods are still not user friendly for farmers. Plantains are a perennial tropical and subtropical crop, which grow in a wide range of environments. However, the plantain production systems can be divided into three broad categories depending on the number of cultivars grown and the intensity of management. Plantains are starchy even when ripe compared to banana and are only eaten when cooked. Though the average yield of local plantain in Ghana is 11.0 metric tons per hectare (mt/ha) [SRID-MOFA, 2011], the potential achievable yield of the landraces is 20.0 mt/ha. The yield gap of 9.0 mt/ha could be attributed to several factors. Nonetheless, the achievable yield potential of the crop could be attained if research efforts are geared toward using high-yielding landraces which are already tolerant to the adverse biotic and abiotic factors complemented with elite materials and other agronomic practices. Plantain as parthenocarpic (produces fruit without fertilization) and seedless, it is propagated traditionally by planting corms and suckers (daughter plants that grow from the rhizomes at the base of the mother plants). Due to the unavailability of disease- and pestfree or clean planting materials, farmers in sub-Saharan Africa traditionally plant suckers derived from their own plantations, most of which are affected with pests and diseases. The morphology of the crop shows that each leaf scar carries a bud [SWENNEN & ORTIZ, 1997]. The quality of the planting material is one of the major factors for successful crop production [TENKOUANO & al. 2006]. In plantain production, farmers use planting material from old plantain fields, irrespectively of the health status of the mother plant. Often planting materials derived from these infected mother stocks results in perpetuation of diseases (e.g. viruses such as banana bunchy top, banana streak) and pests (e.g., nematodes and weevils) leading to low yields and poor quality fruits. It is evident that farmers have no strong concept of infectious plant pests and diseases that are propagated by infested suckers. Poor sucker quality leads to high plant losses [HAUSER, 2000], and shortened

plantation longevity [GOCKOWSKI, 1997] with occasional complete failure of the ratoon crop [HAUSER, 2007]. These could subsequently lead to over 50% yield losses. The poor quality and inadequate planting materials is threatening plantain production. Unlike grains and legumes, plantain is vegetatively propagated. It is evident that quality planting material coupled with good agronomic practices could contribute to achieving the productive potential of plantain in Ghana. Bioversity International with their partners in Latin America, in their study reported that high quality planting material (genetic and phytosanitary) has been shown to contribute significant gains in productivity in smallholder systems in Latin America. Intravarietal variability is well known to occur naturally in plantains, but only recently is this being considered as an opportunity for selecting improved planting materials [CÔTE & al. 2008] Also, developing techniques for the rapid propagation of clean, healthy planting material through in vitro, which yield high performing and true-to-type plantlets through somatic embryogenesis, has been successful [CÔTE & al. 1993; VULSTEKE, 1998]. Simplified macro-propagation techniques and more traditional sanitation techniques of suckers have not been overlooked [AUBOIRON, 1997; KWA, 2002, 2003; TENKOUANO et al., 2006; HAUSER & MESSIGA, 2010]. However, the high cost and low availability of planting material, especially healthy, good quality material with varietal traceability, is viewed as a major constraint and key obstacle for improved plantain productivity [NKENDAH & AKYEAMPONG, 2003]. While traditionally heavy emphasis has been placed on breeding activities, agronomic and pest management constraints have also received significant attention [SWENNEN & VUYLSTEKE, 1993; ORTIZ & VUYLSTEKE, 1998; BAIYERI & TENKOUANO, 2008]. In field production of plantain, numerous types of planting materials exist. They are classified into sword, maiden, peeper, and water suckers. The sword and maiden suckers are generally considered the most productive planting materials. Nonetheless, any type of sucker could be used for planting. Furthermore, corms of harvested plants could also be cut into small pieces and planted. This, however, would lengthen the crop cycle of the plant crop. An important condition for the optimization of yield of any crop is the use of healthy planting material [DAS & BORA, 2000]. The planting material used in banana and plantain cultivation is mainly confined to its vegetatively propagated suckers because plantains and bananas are parthenocarpic and seeds are sterile. Five methods are commonly employed to obtain planting material for the establishment of new planting material of plantain: (i) suckers extracted from plantain fields which are in production; (ii) suckers reproduced in field sucker multiplication plots; (iii) plants from micro-corms grown out in a nurseries; (iv) plants originating from secondary buds (PIBS), produced in a humidity chamber, seedbeds and grown in nurseries; and

(v) tissue culture plants grown in two-phase nurseries [TEZENAS DU MONTCEL, 2005; FAO, 2010]. Tissue culture technique can produce large quantities of uniform disease-free healthy planting materials within a short time. The technique also requires small space. However, this is not accessible to farmers, as it requires sophisticated laboratory facilities. The microcorm grown and sucker produced in field sucker multiplication plots on the other hand could be used by farmers but require space. These techniques could produce about four (4) suckers from a medium size sucker of about 0.2-0.5 kg. Some buds are also destroyed by these techniques. Planting materials produced from these techniques also require paring before planting. They also pass through a lag phase during transplanting compared to tissue culturederived plants. The quality of planting materials produced from these processes is always of concern to the buyer and quality controllers. The PIBS is the latest in vivo technology developed to optimize sucker production [KWA, 2002]. Like all other plants each plantain leaf bears an axillary (primary) bud at the point of overlapping of the leaf sheath. However, the architecture of the plant is such that several secondary buds occur along the entire length of the base of a leaf sheath [KWA, 2002]. Most of these buds remain dormant and never become suckers in the lifetime of the plant. These dormant buds could be activated to produce healthy planting materials within a short time. The entire potential of the corms and suckers could thus be exploited to produce large quantities of healthy planting materials within a short period. However, information on the amount of healthy planting materials that could be produced from an average sucker is scanty. Demand for plantain suckers in large quantities is currently very high. The objective of this study was to evaluate the response of the various cultivars to the new technique (PIBS).

2.3 Materials and methods

Plantain (Musa spp. AAB group) are triploid (2n = 33 chromosomes) starchy bananas, whose seedless parthenocarpic fruits are eaten cooked because they are unpalatable when raw. The Apantu, Asamienu and Oniaba used in this study are all triploid plantains. The FHIA- 21 belongs to the genome group AAAB with a ploidy level of 4x. The hybrid is cross between AAB Plantain cv. AVP-67 (French Plantain) x SH-3142. Ten (10) sword suckers each of Apantu (False Horn), Asamienu (True Horn), Oniaba (Intermediate French) and FHIA-21 (Tetraploid hybrid) were removed, cleaned and pared. Suckers weighing between 0.2 kg and 0.5 kg were used for the experiment. The leaf sheaths were removed (de-sheathing) 2 mm above the collar till the apical meristem was exposed [KWA, 2002]. The materials generated (now called explants) were kept in a clean and cool environment until all the explants were ready. The apical dominance was destroyed with crosswise incision made to the collar of the first leaf from the base. The explants were planted 3cm deep in smooth redwood sawdust in a locally made humidity chamber. The experiment was set up in a Complete Block Design (CBD) and replicated three times and repeated three times. The column was watered regularly to maintain moist environment. Harvesting of sprouts began three weeks after planting (WAP) in sawdust. Harvesting of sprouts was done once a week from the third week to the eighth week after planting in the sawdust. The harvested sprouts were transplanted in polyethylene pots filled with sterile loamy soils and placed under 60% shade net. Data was collected on number of sprouts harvested, sources of sprouts, survival of sprouts in polyethylene bags, establishment six weeks after harvesting from sawdust. Data was analyzed using ANOVA.

2.4 Results and discussion

Sprouting was observed two weeks after planting in the sawdust. Harvesting of the proliferations started four weeks after planting and planted in polyethylene bags. The rooted plantlets and plantlets without roots were removed at weekly intervals for five weeks. Healthy rooted seedlings became ready for field planting after six weeks in the polybag.



Figure 2.1. Local plantain cultivar's response to PIF technique.

The PIBS technique is an on-farm macro-propagation approach developed for mass propagation of healthy planting materials of plantains and bananas. The technique is applicable to other vegetatively propagated crops like cocoyams and pineapples. The technique is applicable all-year round and farmers can schedule to raise their planting materials to meet the planting season. The results of the investigation revealed a significant effect of the chamber on healthy planting material production. Asamienu (True Horn) produced the largest number (92) of healthy suckers (Fig. 1). There was a significant increase in the number of sprouts harvested from week four to week six for Asamienu and Apantu and then declined after that. With regard to Oniaba, there was a sharp decline five weeks after planting in the chamber (Fig. 2.1). On the other hand, FHIA-21 produced constant numbers during the fourth and fifth weeks. Asamienu produced the largest number of healthy seedlings with the highest harvested during the sixth week. However, the number dropped drastically (Fig. 1). FHIA-21 (hybrid plantain) produced proliferations faster compared to all the other cultivars (Fig. 1). Average production per sucker was at 86±7 for FHIA-21. Apantu produced consistent planting materials for the fifth and sixth weeks (Fig 1). Sucker production by Oniaba was the least (24 ± 5) among the cultivars (Fig. 1). This result quite agrees with the study of SINGH & al. (2011) who estimates about 50 seedlings per sucker. In another study, [CTA & ISF, 2011] reported an average of 10 harvested sprouts

per sucker using this technique. The study discovered that each leaf scar on the corm, in addition to carrying a primary bud [SWENNEN & ORITZ, 1997], also has several latent secondary buds that will never have developed into daughter suckers. It was evident that a high percentage (65%) of the sprouts was produced from the apical meristematic region (Plate 1 (a) and (b)).



Figure 2.2. (a and b). Sprouting meristems.

Often it is only few of the primary buds that develop into daughter suckers when the apical dominance is removed at flowering. However, with this technique, several of these latent eyes could be activated to sprout as healthy seedlings for planting. The study further revealed that the ability of the secondary buds to sprout was also dependent on the removal of the leaf sheaths very close to the leaf collar (about 2 mm above the leaf collar). The ability of the eyes to sprout could be attributed to the high temperature (about 50°C) generated within the growth chamber. The harvested sprouts when planted in the direct sun got scorched. The seedlings had to be acclimatized under 60% shade (Figure 2.3). The study showed that the ability of the technology to exploit the full potential of the sucker planted in the sawdust was dependent on some key factors. Notable among them include (i) ability to remove the leaf sheath at two millimeters above the leaf collar. Inability to remove the leaf sheath at the appropriate level results in a situation where the sheaths continue to grow hence prevents the sprouting of the buds. (ii) ability to destroy the apical dominance. Inability to break the apical dominance results in the apical tissue continue to grow. Also inability to bury the explants in the sawdust enough (3 cm) exposes the surface leading to surface dryness with not sprouts. In a similar experiment, MANZUR MACIAS (2001) superimposed the technique on the suckers while still attached to the mother plant in the field and injected them with 4ml of benzylaminopurine (BAP). The results showed that second generation suckers were observed after three months. Under field conditions, Asamienu (True Horn) could produce several buds, however, only few could develop into healthy planting materials. Similar behavior was exhibited under the PIBS nonetheless the warm condition within the humidity chamber forced the buds to develop into healthy planting materials. Sucker production by Oniaba in the field was reflected in the PIBS. Under the field conditions, sucker production by Oniaba (intermediate French) is normally low. The results showed that the technology could exploit the entire potential of plantain to generate sufficient planting materials. In the field, False Horn plantain could produce about 39 leaves during its crop cycle; French plantain could produce over 50 leaves during the crop cycle. It presupposes that if each leaf produces one axillary bud, then the crop could generate several suckers, however, they produce only about 10 suckers during the crop cycle due to apical dominance. This technology could therefore break apical dominance hence activate all the dormant buds to become healthy planting material.



Figure 2.3. Acclimatization of plantain Seedlings from growth chamber.

2.5. Conclusions

The technique is an effective on-farm method that could generate large quantities of healthy planting materials of plantain from any type of sucker. Latent eyes that would not have sprouted could be activated to generate healthy planting materials. The number of suckers produced using the technology and time period showed that the technique was efficient. The technology does not require any sophisticated equipment for its application but only skills; hence could be used by anyone for sufficient healthy planting material production.

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3. Exploratory assessment of potential mycorrhization of two landraces of plantain

3.1 Abstract

Arbuscular mycorrhizal fungi play a crucial role in ecosystem services which are beneficial to humankind. Mycorrhizal association is known to have several beneficial effects on plant growth and development. An exploratory assessment was conducted to assess the natural mycorrhization of plantain roots. Plantain seedlings of two cultivars belonging to the subgroup AAB (False Horn and Giant French) raised using macropropagation technique were used. The mycorrhizal colonisation was estimated using root staining and microscopy. Data were collected on frequency of mycorrhiza and intensity of mycorrhizal colonisation. Soil samples collected from the rhizosphere of plantain growing under nursery soil were also analysed. Results showed that False Horn plantain had a higher frequency and intensity of mycorrhizal colonisation than French plantain in all root samples. The mycorrhizal spores observed were almost of the same diameter and could be assigned to the genus Glomus. The abundance of arbuscules in the root system of Apantu was μ =30.89 % ± 6.40 % with some root pieces completely mycorrhized and a dense arbuscule development. Different structures of arbuscules were observed, as well as few vesicles and spores. Arbuscule abundance in the root system of French was very low with μ =1.15 % ± 1.13 %. AMF frequency and intensity was low, while the abundance of unidentified microbiota was high and diverse. The presence of nonmycorrhizal fungal structures (not analysed), was higher for French than False Horn plantain. Mycorrhizal spores with size <125 µm were counted in the soil sample from the root rhizosphere of the seedlings. The study revealed the potential of mycorrhizal colonization of plantain roots which could lead to nutrient and water use efficiency in plantain production.

Keywords: Plantain; false horn; giant French; Arbuscular Mycorrhiza Fungi (AMF); root rhizosphere.

3.2 Introduction

Mycorrhizal association is known to have several beneficial effects on plant growth and development. Wheat inoculated with mycorrhiza showed that mycorrhizal plants used less water to produce one unit of shoot of Dry matter (WUE-Water Use Efficiency) than non-mycorrhizal plants, but water-stressed and well-watered plants did not differ in Water Use Efficiency. Also, these plants had higher shoot and root dry matter than non-mycorrhizal plants

regardless of water stress level [1]. In a study determine the effects of water stress vs. no water stress and the arbuscular mycorrhizal (AM) fungi Glomus monosporus on growth, acquisition of phosphorus (P), zinc (Zn), copper (Cu), manganese (Mn), and iron (Fe), and water use in two wheat cultivars exhibiting differences in resistance to water stressed (WS), [2] observed that shoot and root dry matter, leaf areas, total root length and root colonization with AM for plants grown under non-WS were higher than for plants grown under WS. Much of the reduction in dry matter was overcome by AM plants grown under WS. Nutrients contents were greater under WS than under non-WS conditions. The AM plants had higher water efficiency values than non-AM plants when grown under WS. Their results of this study indicated that AM plants had greater tolerance to drought stress than non- AM plants. relationship between some soil fungi with the plant roots [3]. In this mutual association, the plant can benefit from these fungi by efficient transfer of inorganic nutrients like P, N, K, Ca, S, Fe, Mn, Cu and Zn, as well as water. In turn, the fungi receive plant carbohydrates [4,5], but some exceptions exist [6]. Others characteristics are intra-radical structures (the vesicles), constituting generally storage organs, which are also able to support colonization of new plants as propagules [4]. Hyphae grow intra- and extra-radical, while the latter exploits the soil matrix beyond the nutrient depletion zone of the plant and therefore improves nutrient supply for the plant.

AMF play a crucial role in ecosystem services - which are benefits to humankind provided by ecosystems, including supporting (nutrient cycling, soil formation), provisioning (food and water supply), regulating (climate and disease control) and cultural (recreational and spiritual benefits) services [7] - mainly within the supporting category. A focus has been on its role in plant nutrition, mainly due to nutrient transport to the plant. AMF can increase nutrient content and growth parameters of banana plants [8], and improve soil structure by releasing glomalin, when hyphae die and decay hence improves soil stability and increases water retention [9]. Also, AMF are known to increase plant tolerance against biotic and abiotic stresses in a large variety of cases. Mineral content and secondary metabolites in plants are affected by the symbiosis, which can result in improved nutritional and medicinal status of plants for human health [10].

Many studies on banana and plantain (*Musa* sp.) showed the beneficial effects of AM on plant tolerance against abiotic and biotic stresses. It was shown that AMF can increase banana tolerance to salt stress, generally related to increased plant growth [11] and to aluminium toxicity, also with positive growth effects of inoculated plants but without significant difference in water and nutrient uptake of inoculated and non-inoculated plants if no Al was applied [12]. AMF are assumed to induce systemic resistance against plant parasitic nematodes [13] and to

increase root branching, thereby affecting nematodes which prefer primary roots [14,15]. Nematode built up was either not affected [16] or negatively affected by AMF [17,18,19,20]. Furthermore, the disease *Cylindrocladium spathiphylli*, causing root rot in banana plants, was negatively affected by AMF [21]. Experimental set-up and plant genotype – AMF species – environment interaction is reported to play an important role in the functioning of this symbiosis. This study therefore was an exploratory assessment of mycorrhization of plantain roots (False Horn and Giant French).

3.3 Materials and methods

Plantain seedlings of False Horn (Apantu) and Giant French (Apem) were raised using macropropagation technique and planted in polybags at Fumesua. Black soils for the nursery bags were collected from a backyard with no agricultural activities. The soil was thoroughly mixed before being dispensed to polybags. The seedlings were grown under 60% shade net acclimatization screen house. The polybags were arranged randomly in the screen house. The seedlings were irrigated regularly with tap water. Seedlings were allowed to grow for nine weeks in the black polyethylene polybag in unsterile soil. Root samples from the seedlings growing under unsterile nursery soil were analysed. Root samples were washed under running tap and stored in filter paper. Samples were transported to Germany for analysis. Analysis was done in the laboratory of INOQ GmbH, Solkau, Germany. On arrival in the laboratory, the root samples were quite dry, so they were left to soak in tap water. Roots were stained for analysis. The roots were cleaned by soaking them with KOH 10 % (100 g KOH dissolved in 1 L of tap water) in a beaker and heating the sample in a microwave until boiling. The roots were then rinsed with tap water to remove the KOH. China black ink solution (China black ink 5 %, Acetic acid 5 % in tap water) was used for staining [22]. Roots were soaked in the ink solution in a beaker and heated in a microwave until boiling. They were rinsed with tap water for distaining. 30 root pieces of ~ 1 cm were put on a microscope slide and covered with glycerol for conservation. The mycorrhizal colonisation was estimated according to [23] using a microscope for observation of mycorrhizal structures.

Frequency (F%) of mycorrhiza in the root system was estimated by the number of root pieces, which contain mycorrhizal structures divided by the total number of root pieces. The intensity of mycorrhizal colonisation in the root system (M%) estimates the amount of root area colonised by AMF. Similarly, the intensity of mycorrhizal colonisation can be estimated only

for the colonised root pieces, not taking into account non-colonised pieces (m%). Accordingly, the abundance of arbuscules can be estimated for the root system (A%) and for the mycorrhized parts of the roots only (a%).

Soil samples collected from the root rhizosphere of plantain growing under nursery soil were also analysed. 5 g of soil were placed into a beaker with tap water and soil aggregates were crushed. Wet sieving was applied through sieves of following mesh sizes: 425 μ m, 200 μ m, 125 μ m, 75 μ m, 40 μ m. The content of each sieve was transferred into a Petri dish with tap water. Spores were counted under a binocular microscope.

3.4 Results and discussion

Plantain, as a giant monocotyledonous herb could show great potentials to establish mycorrhizal symbiosis [24,25]. This symbiosis has been studied to promote growth and nutrient uptake in micro-propagated bananas. The study showed that the roots of False Horn (Apantu) expressed a higher frequency and intensity of mycorrhizal colonisation than the Giant French (Apem) in all root samples (Table 3.1).

The abundance of arbuscules in the root system of Apantu was μ =30.89 % ± 6.40 % with some root pieces being completely mycorrhized and a dense arbuscule development. Different structures of arbuscules were observed, as well as few vesicles and spores. Despite unidentified microbiota in the roots of Apantu their incidence was low.

Arbuscule abundance in the root system of Apem was very low with μ =1.15 % ± 1.13 %. AMF frequency and intensity was low, while the abundance of unidentified microbiota was high and diverse. The presence of non-mycorrhizal fungal structures (not analysed), was higher for Apem than Apantu.

Roots of Giant French appeared a bit thicker and less branched, with shorter secondary roots, than roots of False Horn. Eight (8) spores were counted in 5 g of soil. No spores with a size > 125μ m were found. Six (6) spores between 40-75 μ m and two (2) spores between 76-125 μ m were counted. No nematodes were observed.

False Horn roots were well-colonised by AMF and did not show high incidence of other microbiota. Contrary to False Horn, the Giant French roots were poorly colonised by AMF with a rather high incidence and diversity of other microbiota, maybe also pathogens. It is believed in some case that AMF can reduce root infection by pathogens, which might lead to increased fresh weight and yield of plants, compared to infected non-mycorrhizal plants

[10,26]. We noticed that the Giant French roots were containing more non mycorrhizal fungal structures.

Table 3.1. Frequency (F%) of mycorrhizal structures in root pieces, intensity (M%) of mycorrhizal colonisation in the root system and in the mycorrhized root pieces (m%) and abundance of arbuscules in the root system (A%) and in the mycorrhizal parts of the roots (a%) are given for each sample, as well as the mean (\overline{x}) and standard deviation (SD) for each cultivar.

Cultivar	Sample	F%	М%	m%	A%	a%	\overline{x} (F%) ± SD	\overline{x} (M%) ± SD
Apantu	1	80.00	37.70	47.13	25.37	67.29	89.17 ± 7.39	50.33 ± 8.77
	2	93.33	53.70	57.54	33.43	62.26		
	3	86.67	57.87	66.77	38.77	66.99		
	4	96.67	52.03	53.83	26.00	49.97		
Apem	1	6.67	4.67	70.00	1.17	25.00	10.08 ± 5.71	3.88 ± 1.24
	2	16.67	4.53	27.20	2.27	50.00		
	3	6.90	2.45	35.50	0.02	0.70		



Figure 3.1. Stained root fragments of A) False Horn, containing arbuscular mycorrhizal structures and B) Giant French, containing non-mycorrhizal fungal structures (unidentified).


Figure 3.2. Mycorrhizal structures in root fragments of Apantu. A) Dense colonisation with arbuscules and hyphae. B) Vesicules (black arrows), arbuscules and hyphae.

those studies were conducted under controlled growth conditions with sterilized substrate [21, 15,14,12]. As the False Horn and the Giant French plants were growing in the same substrate and were of equal age it seems that False Horn is more receptive to indigenous AMF species than Giant French, pointing perhaps to AMF - plantain genotype interaction.

Despite only few spores counted in the soil sample, the mycorrhizal colonisation of False Horn roots was very good and high in comparison to most literature values, while the Giant French roots were poorly colonised.

3.5 Conclusion

This exploratory result showed that indigenous mycorrhizal fungi symbiotic relationship with plantain exist in Ghanaian soils. This association could be exploited commercially for water and nutrient use efficiency in plantain production. It is also likely that mychorhizal- plantain interaction could be cultivar- specific. The exploitation of the ability of soil micro-organism-plantain symbiosis could enhance the environment. This need further investigation to substantiate the particular species of arbuscular fungi involved in the association. False Horn is also likely to have the potential for AMF-association for sustainable growth and development. The study has revealed interesting potentials of ecosystem services rendered in the rhizosphere. The study also exposes the possible of dauciform rooting in False Horn, a characteristic associated with plants growing in nutrient deficient soils.

3.6 References

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4. Responses of leaf stomatal parameters to induced water stress and its relationship with stomatal conductance in false horn plantain

4.1 Abstract

Water deficit is one of the most important environmental factors affecting agricultural production especially in rain-fed cropping systems. To understand how morphological and physiological factors interact, a study was conducted to assess the responses of leaf stomatal parameters to water status and its relationship with chlorophyll content in False Horn plantain. Each plant was grown in 120kg soil in bowls and subjected to varying watering regimes (10ml, 15ml, 20ml, 25ml, 30ml and control) without allowing natural rain into the bowls except the control. The result showed that False Horn plantain was sensitive to soil moisture stress; and this was reflected in changes in reduced growth through reduced plant height. The study also showed a systematic growth of the leaf with time. The patterns of leaf length growth remain similar under the different watering regimes. There was a significant difference in the leaf growth between the control and the varying amount of watering. The undulating pattern, however, showed that other factors could influence the leaf length. The large leaf area used for transpiration in Apantu is likely to be affected by soil water deficit. There was no correlation, however, between the leaf length to width ratio and the soil moisture content. The study further revealed that leaf length and width in False Horn plantain increases with growth, however, leaf sizes started to reduce when the plant approaches the flower primordial stage of growth. Watering regime influenced stomatal opening and closure in False Horn plantain and chlorophyll content. Leaf chlorophyll content was not highly influenced by the varying watering regimes; indicating that the presence of laticifer cells may be a factor for water stress tolerance in plantain.

Keywords: Plantain; physiology; stomata; water stress; chlorophyll content.

4.2 Introduction

Plantains are an important staple starch food and cash crop in humid sub-Saharan Africa. Plantains have been cultivated in the region over centuries but there is little information on their physiology and growth patterns. Knowledge on plantain crop physiology and growth patterns is important to establish the potential of the crop, explore the possibility of extending the crop to other areas and improve yields by resolving major yield constraints. Water deficit is one of the most important environmental factors affecting agricultural production especially in rain-fed cropping systems. The consequences of water deficit include its adverse effects on plant phenology, development, assimilate partitioning, carbon assimilation, growth, and plant reproduction processes. Consequently, plant responses to drought have been extensively investigated from molecular, physiological, and whole plant to ecosystem levels [1]. Physiological responses to soil water deficit are the features that are most likely to determine the response of a crop to irrigation. Water stress could reduce leaf net photosynthetic assimilation (*A*n) by both stomatal and metabolic limitations [2,1,3,4]. Also, many studies have reported that stomatal effects are significant under moderate stresses, but biochemical limitations are quantitatively important during leaf ageing or during severe drought [5,6]. An early response to water deficit in a plant is a reduction in leaf area and plant growth, which allows plants to reduce their transpiration, thus increasing water use efficiencies [7-9] and promoting interspecific competition capacity under drought [10].

Plants subjected to abiotic stress use various defence mechanisms to cope with the stress. A common strategy is the synthesis and accumulation of osmoprotectants or compatible solutes like proline, glycine betaine, polyamines or trehalose. Tolerance to abiotic stresses can be acquired by pre-treatment with such a protective compound [11].

Several factors are known to influence stomatal opening and closure. Notable among them are light (intensity, duration and quality) internal water status of the leaf, carbon dioxide concentration of the surrounding air, and the leaf temperature. Soil water deficit coupled with other environmental factor create a complex mechanism that is very difficult to understand when not studied under control environment.

Also it becomes complex to interrelate all these factors at the same time. Wind speed, soil moisture, solar radiation leaf temperature and relative humidity all contribute to influence the physiological responses. Plant stomata are crucial gateways between plant and atmosphere and play a central role in plant/vegetation responses to environmental conditions. Several studies have been conducted and continue to be conducted from molecular and whole plant perspectives, as well as at ecosystem and global levels [12,4,13]. Studies carried out have shown stomatal density responses to various environmental factors, such as elevated CO₂ concentration [13], heat stress [14], salt stress [15], drought [16,17,18], precipitation change [19], and plant density [20].

Other studies have shown that water deficit leads to an increase in stomatal density [21,22], and a decrease in stomatal size [23,24,25] indicating could influence adaptation of plant to drought [23,25,26]. While studies have shown that some selected abiotic factors influence

physiological responses, others like wind speed, relative humidity, solar radiation, temperature and CO_2 also play a significant role in shaping the physiological responses.

Several authors have analyzed plant responses to water deficit in relations to various plant traits. While some authors related stress to stomatal conductance [27] others linked responses to photosynthesis [1]. Others also linked plant responses to environmental factors as a type of homeostasis [28]. A study showed that in some plants, stomata on the same leaf may respond homogeneously for heterogeneously to environmental conditions [29,27,30,31]. This behavior, named patchy stomatal closure, occurs when stomata at different locations on the same leaf show different responses to similar perturbations [32,33,30,31]. Some plants are known to rhythmically vary the width of the stomatal pores, to control the flux of CO_2 and H_2O between the leaf and the surround air in response to changes in environmental factors [32,33]. Although the stomatal response to the environment involves complex mechanisms influenced by many factors, stomatal movements appear to be governed by two principal control circuits, one involving gradients of CO_2 and the other involving gradients of H_2O vapour [34].

Patchwise stomatal closure is especially pronounced under stress situations, such as water deficit [35,33]. According to Cardon [36], during strong oscillatory behavior or even at a steady-state stomatal conductance, a surprising number of different behaviors in the dynamics of the photosynthetic activity may be found. Leaf morphological traits, including stomatal density and distribution, and epidermal features are known to affect gas exchange quite remarkably and their relationships with key environmental factors such as light, water status, and CO₂ levels [13,12]. It is reported that increasing the blue-light response of stomata through red light is closely associated with guard cell chloroplast activity [37,38,39], indicating that leaf photosynthetic activity may involve stomatal movement and development. The balance between carbon gain through photosynthesis and water loss through transpiration was reported by Iyyakkutty [40] to also affect instantaneous water use efficiency (WUE [37,38]. Unfortunately, how stomatal density affects gas exchange has received little attention over the years.

Plantain as a giant monocot pose challenge to physiologists to measure indicators of water deficits, due to the presence of large air pockets within the leaves, and laticifers containing latex within the leaves, fruit, and corm that hinder the use of standard methods of measuring water relations [41]. Several methods have been used to measure physiological indicators of response [42,43,41]. However, none of these methods have been accepted as the only standard to be used, though [44] confirmed the reliability of method by [42] that is dependent on the refractive index of exuded latex. Although some authors [44,42] used leaf folding as an

indicator of response to water deficit, in plantain under hot, arid conditions, leaf folding is not considered to be a reliable plant-based indicator of when to irrigate [45]. While physiological mechanisms of stomatal responses are complex and are not yet fully understood, it is even worse with False Horn plantain as study describing how stomatal parameters respond to different water stresses, and their relationships with physiological processes are limited. The purpose of this study was to determine the stomatal response patterns to different water stratus, and to develop the relationship of stomatal parameters and photosynthetic processes.

4.3 Materials and methods

4.3.1 Study area

The study was conducted at the Crops Research Institute plantain orchard in Kumasi. The study area is located on latitude 06° 42'740"N; and longitude 001° 31' 827"W) with altitude of 300m above sea level. The first set of experiments was conducted between April 2013 and July 2013. The experiments were repeated between April 2014 and July 2014.

4.3.2 Plant materials used

Planting materials of (False Horn plantain AAB subgroup) were generated using micropropagation technique [46]. Healthy seedlings of uniform sizes with six leaves were selected and used for the study. The plantain plants were subjected to different water regimes for three months. Each plant was grown in 120 kg of sterile soil in large plastic bowls. The bases of the bowls were perforated to avoid water logging.

The surfaces of the bowls were covered with black polyethylene sheets to prevented external rain water from entering. The soils were initially soaked to field capacity at planting. The bowls were then subjected to various water regimes- 10 ml, 15 ml, 20 ml, 25 ml, 30 ml per week. The surfaces of the bowls with control plants were uncovered to receive natural rain. Irrigation was done with a large syringe through a pore at the base of the pseudostem. Fertilizer (NPK) was applied weekly at 15 g per plant. Fertilizer application was at the time of watering.

Each treatment had 15 plants replicated four times in a randomized complete block design and repeated three times. The Data were taken weekly from the first week to the 12th week on leaf

emergence, leaf length, leaf width petiole length of fully opened new leaf. Plants were released from the stress condition after the 12^{th} week. The plants were subjected to one-month water stress again from the 37^{th} to the 42^{nd} (flowering) leaf stage of growth. Data was collected on the leaf length and leaf width. Data were analysed using Analysis of variance (ANOVA) (P<0.05).

4.3.3 Leaf stomatal density and guard cell size

The impression approach was used to determine leaf stomatal density, which was expressed as the number of stomata per unit leaf area [47]. The leaves selected were those for which chlorophyll content was also measured. Micromorphological observations were carried out 12 weeks after planting using a digital microscope (Amscope, USA). The stomatal density (SD) stomatal length (SL) and stomatal width (SW) were determined from the underside (abaxial surface) of each leaf using prints made with nail varnish. SL was measured between the junctions of the guard cells at each end of the stoma as defined by [48,49]. The SW was measured perpendicularly to maximum width, which represents the maximum potential opening of the stomatal pore, but not the aperture of opening that actually occurs. SD (number of stomata per mm²) was determined as described by [47]. Stomatal surface (SS) stomatal shape coefficient (SSC), potential conductance index (PCI) and relative stomatal surface (RSS) was obtained using equations 1 to 4 (equations from Wang et al. [50] with some modifications). $SS = (SL * SW * \pi)/4$ (1)

SSC = 100* SW/SL (2) PCI = $(SL)^2 * SD* 10^{-4}$ (3)

 $RSS = SPS * SD * 100 \tag{4}$

The SL and SW values are measured as micrometers (μm) , SS in $(\mu m)^2$ and RSS in percentages. Stomatal conductance = (stomatal aperture length)² *Total stomatal density *10⁻⁴ [51]. The number of guard cells was estimated by doubling the number of counted stomata in the same leaf area [47]. Stomatal size was defined as the length in micrometres between the junctions of the guard cells at each end of the stoma, and may indicate the maximum potential opening of the stomatal pore, but not the aperture of opening that actually occurs [48,49].

4.3.4 SPAD value

Chlorophyll content was determined from intact leaves using a chlorophyll meter (SPAD 502 Manitol) Reading of SPAD values was done on weekly basis. Three measurements were made per plant, three leaves were chosen from each plant (lower, middle and upper leaves of a plant), and three different regions of each leaf (middle and two ends of leaf) were used for tests. The chlorophyll meter was used to estimate the nitrogen status of the crops. The instrument measures transmission of red light at 650 nm, at which chlorophyll absorbs light, and transmission of infrared light at 940 nm, at which no absorption occurs. One the basis of these two transmission values the instrument calculates a SPAD value that is quite well correlated with chlorophyll content [52,53].

4.4 Results and discussion

The study area experiences bimodal rainy season with the major rainy season starting from March to July and the minor reason season from September to November. Subjecting False Horn to water stress affected physiological morphological and biochemical processes. The average temperature of the study area was 26°C and the relative humidity was 82% (Table 1).

 Table 4.1. Weather conditions at the experimental location (2012-2014).

Months	Temp (^o C)	Rainfall(mm)	RH (%)	Wind speed ((m/s) Solar Radiation (W/m^2)
January	26.5	8.0	56.6	0.7	155.8
February	28.1	17.8	65.8	0.9	165.3
March	27.2	82.4	81.5	1.0	172.5
April	27.0	152.6	80.9	0.9	185.9
May	26.4	169.4	87.2	0.7	162.1
June	25.7	199.2	89.1	0.9	147.5
July	24.6	43.6	89.1	1.0	117.8
August	23.9	7.4	90.1	1.1	101.5
September	23.7	255.4	90.1	128.9	121.4
October	24.7	215.2	90.0	139.4	154.9
November	26.2	41.4	85.2	0.6	156.0
December	25.9	40.8	80.1	0.6	149 1

The minimum and maximum annual temperatures recorded during the period were 24°C and 28°C respectively (Table 1). The lowest temperatures occurred in the months between the major and the minor rainy seasons (Table 1); whereas the maximum was observed during the

peak (February) of the dry season. This deviates widely from the notion that April is the hottest month in the middle belt of Ghana. In April, the relative humidity is often high coupled with the high temperature, the weather becomes humid and hot; unlike in February when the weather is dry hazy with the north-west westerly winds. The lowest relative humidity was recorded in the dry season (January). This is not surprising as the weather is often dry and hazy in January with the north-west westerly winds bringing in a lot of dust from the Sahara Desert. It is interesting to observe the strongest winds experienced in September and October.

However, the heaviest lodging in plantain is experienced between March and April when the rains are starting after a long drought. The beginning of the rains comes along with strong winds; and because the plants are dehydrated as a result of the long drought there is severe lodging. Strong winds are known to cause stem and root lodging in plantain but not at tender ages. However, strong wind coupled with low relative humidity could influence water loss from the leaves through stomatal opening and closure to conserve water in the tissues.

Water stress adversely affected plantain growth and development. Leaf emergence was highly influenced by water stress [41]. The control maintained regular leaf emergence compared to the other treatments (Table 2). Under 25 ml and 30 ml of water per week regimes, False Horn produced one leaf in every seven days; however, as the watering reduced, leaf emergence was delayed to a leaf in 12 days.

Treatment	Time (weeks)										
	1	2	3	4	5	6	7	8	9	10	11_
Control	7.0	7.0	7.0	7.0	7.0	7.0	6.0	6.0	5.0	5.0	5.0
10ml	6.0	7.0	6.0	5.0	5.0	5.0	5.0	5.0	5.0	4.0	4.0
15ml	7.0	7.0	7.0	5.0	6.0	6.0	6.0	6.0	4.0	4.0	4.0
20ml	7.0	7.0	8.0	5.0	6.0	6.0	6.0	6.0	5.0	4.0	4.0
25ml	7.0	7.0	7.0	5.0	6.0	5.0	5.0	5.0	5.0	5.0	5.0
30ml	7.0	9.0	9.0	8.0	5.0	5.0	4.0	4.0	4.0	4.0	4.0_
SE	0.4	0.4	0.4	0.5	0.4	0.5	0.3	0.4	0.5	0.3	0.4
CV%	5.7	5.1	6.7	9.5	6.8	7.4	5.9	7.0	10.4	6.0	10.1
LSD (5%)	1.2	1.2	0.8	0.8	0.8	0.8	0.7	1.1	1.0	1.2	1.4

Table 4.2. Analysis of variance of leaf emergence of False Horn plantain under water stress.

SE= Standard Error; CV= Coefficient of variation; LSD= Least Significant difference.

Though there was no sign of dehydration of the leaves, it was observed that leaf necrosis was very high. The numbers reduced with time especially after week three. This agrees with Kallarackal, et al. [43] and Turner and Thomas [41] that plantain response to water deficit by stomatal closure and that as the soil water decreases, the plants remain highly hydrated.

However, weekly leaf emergence is affected by water stress. This prolongs the crop cycle. False Horn plantain, as a determinate plant is expected to produce a fixed number of leaves (about 42) leaves before flowering. Under good irrigation conditions coupled with soil nutrient flowering starts after six months and harvesting at the ninth month. However, with new leaf emergence at fortnightly intervals, flowering would start after 12 months and harvesting at the fifteenth month. This situation would contribute to high cost of production.

Treatment			Plant	height(cm) wit	h Time	(weeks)			
	1	2	3	4	5	6	7	8	9	10	11
Control	22.0	23.0	32.0	36.0	39.0	47.0	60.0	63.0	65.0	66.0	79.0
10ml	17.0	20.0	24.3	27.8	28.8	33.3	42.3	43.0	43.7	43.7	48.5
15ml	18.1	19.9	24.8	27.5	29.0	29.8	37.2	43.8	44.2	44.5	45.5
20ml	15.6	19.7	25.8	29.2	30.8	33.3	42.8	42.8	46.2	46.5	50.3
25ml	18.0	21.2	28.7	31.3	33.7	34.7	41.7	42.5	45.0	47.8	51.8
30ml	18.9	21.6	26.8	26.9	28.9	31.7	37.5	38.9	42.1	43.9	44.8
SE	2.3	2.3	3.1	2.7	2.9	1.3	1.7	1.4	1.6	1.4	2.7
CV%	6.0	4.9	3.5	2.6	2.6	3.6	3.8	3.0	3.3	3.0	5.1
LSD (P<0.5)	3.5	4.7	4.7	4.2	4.4	5.1	7.5	8.4	8.7	8.9	12.5

Table 4.3. Analysis of variance of plant height of False Horn plantain under water stress.

The sensitivity of False Horn plantain to soil moisture stress is reflected in changes in reduced growth through reduced plant height (Table 4.3). There was a significant difference (p<0.5) in plant height between the treatments (Table 4.3). Soil water stress affected plant growth. While plant height was increasing at a weekly average rate of 5.7 cm in the control, it was an average of 3.1 cm in the 10 ml, 2.9 cm in 15 ml, 3.1 cm in 20 ml, 3.3 cm in 25 ml and 3.0 cm in 30 ml of water per week. The results showed that False Horn plantains need more than 30 ml of water per week to grow and achieve its optimum potential. Though there was no sign of dehydration in the pseudostem the results showed that plantain has an internal mechanism for water stress endurance. Growth is determined by multiplication of cells and expansion of cells, hence any environmental factor that can hinder water loss can influence growth. In their study, [43,41] reported that, plantains are sensitive to soil water deficits; expanding tissues such as emerging leaves and growing fruit are among the first to be affected. They also observed that as the soil begins to dry, stomata close; however, leaves remain highly hydrated. Turner, [54] observed that stomatal closure under soil water deficits conditions may likely be linked to a signal from the roots rather than water deficit in the leaves. In another experiment [55] reported that drying part of root system of banana had no effect on leaf water status but did close the stomata.

However, severing the roots on the dry side caused the stomata to reopen. These observations therefore support the view that the roots produce a signal that is transported to the leaves. This mechanism conserves the plant's water, but reduces carbon assimilation and productivity. The concept that plantains use large amounts of water could not have a strong physiological basis as the plants remain hydrated under severe soil moisture deficit.

Treatment					Time	(weeks))				
	1	2	3	4	5	6	7	8	9	10	11
Control	25.0	28.0	44.0	46.0	47.0	51.0	64.0	68.0	65.0	71.0	72.0
10ml	20.1	21.8	31.3	34.0	35.2	34.8	41.8	44.5	47.8	47.0	51.5
15ml	20.9	20.9	29.2	34.0	35.5	36.0	43.0	43.8	40.9	46.0	49.0
20ml	19.9	20.0	32.2	35.6	37.0	39.3	48.3	51.3	51.3	53.8	57.2
25ml	20.2	22.9	32.3	34.2	38.0	37.8	46.3	47.8	47.7	50.5	53.7
<u>30ml</u>	21.0	23.6	32.5	37.8	37.3	38.7	43.9	52.8	42.6	51.9	52.2
SE	1.37	3.09	0.54	0.93	0.82	0.34	1.22	1.58	1.82	1.2	1.4
CV%	6.5	3.7	1.6	2.5	2.2	0.9	2.6	3.1	3.7	2.3	2.6
LSD (P<0.5)	2.7	4.7	7.5	6.2	5.8	5.2	6.8	7.6	8.8	7.0	8.2

Table 4.4. Analysis of variance of leaf length of False Horn plantain under water stress.

The new leaves produced continue to increase in length with severe soil water deficit. There was a significant difference between the control and the treatments (Table 4.4). There was no significant difference between in the leaf length between treatments 10 ml, 15 ml, 20 ml and 25ml at week 4 (Table 4.4). This showed that other factors would also be relevant to the growth of the plant. It is observed that under thick and prolonged cloud (about 2-6 weeks) plantains and banana cease to growth. This has been a major challenge for commercial banana growers in Ghana. It is observed that during a period when day and night temperature and almost the same, there is lack of rains, but with high humidity and thick cloud cover, fruits cease to grow irrespective of the amount of water and nutrient applied. This often occurs between March and April every year in Ghana.

Watering Regime (ml)	Mean leaf length (cm) and SD	Mean leaf width (cm) and SD	Leaf length: width ratio
10	51.9±4	30.2 ±6	1.7
15	42.2 ± 5	21.9 ± 7	1.9
20	55.8 ± 7	32.1 ±4	1.7
25	63.4 ± 5	35.1 ±6	1.8
30	53.5 ± 4	31.4 ±3	1.7
Control	71.7 ± 10	38.2 ± 8	1.9
<u>LSD (P<0.05)</u>	<u>5.5</u>	2.4	

Table 4.5. Leaf length-width ratio of False Horn plantain under varying water regimes.

 Table 4.6. Analysis of variance of leaf width of False Horn plantain under varying water regimes.

Treatment						Time	(weeks)			
	1	2	3	4	5	6	7	8	9	10	11
Control	13.0	14.0	23.0	25.0	22.0	28.0	34.0	38.0	68.0	40.0	39.0
10ml	8.2	9.2	14.0	15.3	15.4	16.9	22.2	23.7	27.7	25.8	28.0
15ml	8.9	9.6	12.8	15.3	15.0	15.6	22.8	24.3	24.8	23.3	27.4
20ml	9.0	10.0	14.5	16.4	17.6	18.6	24.7	27.8	28.8	29.9	31.0
25ml	9.6	11.5	14.3	14.8	17.3	18.7	28.3	24.8	44.0	26.3	29.2
30ml	10.6	12.0	14.6	17.6	20.0	20.7	25.1	27.8	53.9	29.6	29.9
SE	0.7	0.6	0.7	0.8	0.5	0.7	2.6	1.6	2.3	1.3	1.1
CV%	7.0	5.2	4.7	4.7	2.8	3.7	9.9	5.7	5.6	4.4	3.5
LSD (P<5%)	1.8	3.1	3.8	4.4	3.8	3.6	6.7	5.1	8.1	5.8	5.4

Table 4.7. Analysis of variance of petiole length of False Horn plantain under water stress.

Treatment						Time	(weeks	5)			
	1	2	3	4	5	6	7	8	9	10	11
Control	3.8	5.0	5.0	5.2	5.7	8.0	8.7	10.3	9.0	10.2	14.2
10ml	3.0	4.5	4.1	4.1	3.8	5.5	6.5	6.8	7.2	7.6	7.5
15ml	3.4	3.5	4.1	4.3	4.4	4.7	6.5	6.5	7.0	7.1	7.1
20ml	3.3	3.9	4.3	4.6	4.3	6.2	6.8	7.0	7.8	8.1	8.0
25ml	3.4	4.8	4.2	4.1	5.5	5.5	7.5	7.0	7.5	6.8	9.4
<u>30ml</u>	3.9	4.2	4.1	4.1	4.6	4.7	6.3	7.0	7.2	7.0	7.4
SE	0.2	0.2	0.3	0.2	0.3	0.5	0.5	0.3	0.4	0.5	0.5
CV%	6.2	3.9	6.5	5.2	7.1	9.3	6.6	4.1	4.7	6.9	5.9
LSD (P<0.5)	1.0	0.8	0.8	0.5	0.7	0.9	1.3	1.1	1.0	1.2	2.1

Petiole length was not significantly influenced by soil water stress (Table 4.7). However, there was a reduction growth between weeks 3 and 6 as a result of other weather conditions. The control however, had a frog leap in length after week 10. The length of the petiole has an influence the leaf lamina as it the base that holds the lamina and exposes to the sun. The length would contribute to the orientation of the lamina for effective photosynthesis.

The study showed a systematic growth of the leaf with time (Table 4.5). However, the pattern of the leaf length shows moderately undulating patterns. The patterns of leaf length growth remain similar under the different watering regimes (Table 4.5). There was a significant difference between the control and the varying amount of water application. The undulating pattern, however, showed that other factors could influence the leaf length growth. The large leaf area used for transpiration in False Horn is likely to be affected by soil water deficit. The large leaf surface area coupled with the long crop cycle under rain-fed conditions, False Horn plantain must therefore develop an appropriate mechanism for withstand soil water deficit.

There was no significant difference between the leaf length to width ratio and the soil moisture content. There was no correlation between watering regimes and the leaf length: leaf width ratio. This could be linked to the study by Turner and Thomas [41] in which they stated that plants remain hydrated under severe soil moisture deficit. Thus water-use efficiency in plantain could come from a closer match between plant water use and the amount of water applied.

Leaf Number	Mean leaf length (cm)	Mean leaf width (cm)	leaf length:width ratio
37 th	189 ±14	82 ±12	2.3
38 th	171 ± 11	70 ±13	2.4
39 th	153 ± 14	61 ±11	2.5
40 th	142 ± 11	57 ± 16	2.4
41 st	118 ± 12	49 ±10	2.4
LSD (P<0.05)	6.0	7.0	-

Table 4.8. Flower primordial growth stage leaf length-width ratio of False Horn plantain under water stress.

False Horn produces on the average 42 leaves before flowering. It was observed that leaf length and width in False Horn plantain increases with growth, however, these features start to reduce when the plant approaches the flower primordial stage of growth (Table 4.8). Leaf length and width reduction starts from 39th leaf to the 42nd (flag leaf). The leaf length: width ratio did not

change much with growth (Table 4.8) when the plants were subjected to water stress for one month before flowering.

Table 4.9. Flower primordial growth stage leaf length: width ratio of False Horn plantain under well watering regime.

Week	Leaf length (cm)	Leaf width (cm)	Ratio	
37 th	194 ±12	86 ± 19	2.3	
38 th	187 ± 14	77 ±13	2.4	
39 th	163 ± 18	68 ±16	2.4	
40 th	142 ± 11	60 ± 14	2.4	
41 st	120 ± 19	51 ±11	2.4	
LSD (P<0.05)	5.6	4.7		

Under the control experiment, the leaf length and width also reduces as the plant approached the flower primordial stage of growth (Table 4.9). However, there was no difference between the leaf length: width ratio during growth and development. The reduction in leaf length and width proportionately could be an innate characteristic of the crop. It showed that ripeness to flower primordial initiation results in energy storage hence reduction in vegetative growth.

The study revealed a homogeneous closure of stomata in response soil water deficit in False Horn. However, under severe soil moisture deficit, stomata closure is pronounced (Plate 4.1a). The water regime influenced stomatal opening and closure. Under the 10ml of water per week regime, the stoma closes as compared to 20ml and the control (Plates 4.1, 4.2 and 4.4). Soil water deficit is known to reduce stomatal conductance and leaf size [43] increased leaf senescence [54].

There was a significant difference in leaf width between the control and the other treatments from week one to week eleven (Table 4.6). Leaf width experienced reduced rate of expansion between week 3 to week 6 for all treatments. The leaf surface area is the main photosynthetic organ and reduction affects the photosynthetic process of the plant. Plantain with broad leaf surface and the large number of stomatal pores would contribute to the physiological processes. However, water stress is known to reduce stomatal development, size and density; and hence affect photosynthetic activities [39,56]. This could be attributed to other environmental factors. The weather could be contributing to the reduced growth. The high humidity, thick cloud cover without rains and the narrow difference between day and night temperatures could have contributed to the reduced growth as photosynthesis ceases.



Figure 4.1. (a) Stomatal shape under 10ml water regime. (b) Stomatal shape under 20ml water regime.



(c) Stomatal shape under 30ml water regime.

(d) Stomatal shape under the control.



(e) Stomatal shape under 15ml water regime.

Plantains (*Musa* spp.) rarely attain their full genetic potential for yield due to limitations imposed by abiotic factors which limit the plant's photosynthesis. Plantains are reported to be particularly sensitive to changes in the environment [57]. This thus affect the productivity of

the crop because there is often early closure of stomata to conserve water and hence affect photosynthetic activities.

Leaf folding is on characteristic feature of plants to response to water deficit. Leaf folding results in reduction in surface area. In their study soil water deficit and banana responses, [41] reported that there was a link between leaf folding and water deficit. In another study Lu et al. [58] did not find any link between leaf folding and soil water deficit as they compared sap flow system with gravimetric system to measure drought resistance in banana. In this leaf folding was not observed as a characteristic in response to water deficit. It was observed that leaves of False Horn plantain remained hydrated throughout. Leaf drying and delayed leaf emergence were rather observed during the experimental period.

Table 4.10. Changes in guard cell length, stomatal density and potential conductance index of False Horn plantain with varying water regimes.

Watering regimes	Guard cell length (µm) with SD	Stomatal density (count/mm ²) with SD	Potential Conductance Index (PCI) with SD
10ml	151.4 ± 0.6	464 ± 0.8	106.4 ± 0.5
15ml	$225.0 \pm \ 0.4$	352 ± 0.5	178.2 ± 0.3
20ml	167.3 ± 0.6	304 ± 0.3	85.1 ± 0.4
25ml	178.2 ± 0.3	$320~\pm~0.6$	85.1 ± 0.6
30ml	209.7 ± 0.4	304 ± 0.2	$63.5~\pm~0.5$
Control	$155.2\pm~0.3$	304 ± 0.8	85.1 ± 0.4
LSD (P<0.05)	5.3	2.7	5.4

Treatment			Chlor	ophyll o	conducta	nce wit	th Time	(week	s)		
	1	2	3	4	5	6	7	8	9	10	11_
Control	49.7	49.3	32.5	45.2	44.7	49.5	50.1	42.7	43.6	46.8	45.5
10ml	42.8	51.1	45.0	44.9	45.7	53.6	53.6	51.6	48.5	49.1	49.9
15ml	41.3	49.3	44.7	55.6	46.4	52.7	53.5	59.3	55.8	49.6	46.4
20ml	44.9	48.7	43.8	49.1	45.5	51.6	50.3	45.4	47.0	44.9	42.3
25ml	48.0	50.8	41.8	46.6	45.1	49.6	48.6	46.6	48.5	47.6	44.7
<u>30ml</u>	50.3	47.9	36.5	42.2	49.7	50.6	60.4	41.6	38.6	42.1	45.3_
SE	1.7	1.7	3.4	1.9	2.3	3.3	3.4	5.7	3.7	3.1	1.4
CV%	3.7	3.4	8.4	3.9	5.6	6.3	6.5	12.0	7.8	6.5	3.0
LSD (P<5%)	6.0	5.6	6.9	9.0	4.9	6.1	6.7	15.3	8.5	8.4	6.2_

Table 4.11. Analysis of variance of SPAD chlorophyll conductance of False Horn plantain

 before routine application of water.

All the plants recovered after three months of water stress. The result therefore showed that water stress at early stages of growth has little influence on the final growth and yield of crops with long gestation.

The chlorophyll content did not show any significant trend in response to watering regime. The chlorophyll content showed undulating characteristics indicating that there was no relationship between soil moisture stress and chlorophyll content. Studies showed that the presence of large air pockets within the leaves, and laticifers containing latex within the leaves, fruit, and corm that hinder the use of standard methods of measuring water relations [41]. This peculiar characteristic of plantains and bananas make them adjust quickly to water stress without showing significant physiological changes. In a study with two tobacco species, there was a significant relationship between the heavy metal concentration and stomatal parameters [29].

Watering regime	Stomatal density	Stomatal length (um)	PCI
10ml	464 ±8	151.4±6	106.4
15ml	352 ±7	225.0 ±8	178.4
20ml	304 ±8	167.3±5	85.1
25ml	304 ±6	178.3±7	85.1
30ml	320 ±5	209.7±6	63.5
Control	304 ±7	141.0±9	85.1
P<0.05	2.3	3.4	4.7

Table 4.12. Stomatal conductance index (PCI) of False Horn plantain under varying watering.

Stomatal conductance is linked to stomatal density and guard cell length. The study has not shown any straightforward link between water deficit and physiological responses with growth in False Horn (Table 4.12). There was no correlation between stomatal density and length stomatal conductance index (PCI) in False Horn. It is known that stomatal densities and stomatal lengths have a correlation with stomatal conductance index [51]. These parameters are also influenced by water stress; however, in some plants like plantain the presence of large air pockets within the leaves, and laticifers containing latex within the leaves, fruit, and corm influence physiological responses. Stomatal conductance is known to be a function of stomatal density, size and the degree of opening of the stomatal pores [59]. However, in their study, Lawson and Blatt, [59] reported that stomatal density and size can be negatively correlated as the stomatal opening is induced by abiotic factors.

Variation in the abiotic factors (water, temperature, light, wind speed and CO2) affect stomatal aperture within minutes and can also induce long term changes in stomatal density, hence determine the limits for maximum stomatal conductance [60]. The world-wide water shortage and uneven distribution of rainfall as a result of climate variability makes drought resistance important factor to consider in physiological studies. The physiological changes within leaves such as changes in stomatal aperture or leaf folding or leaf elongation in response to soil water deficit did not correspond with changes in leaf water status measured. False Horn plantain productivity is greatly affected by environmental stresses such as drought, however, the crop responds and adaption to these stresses to survive could be at the molecular and cellular levels as well as at the physiological and biochemical levels.

4.5 Conclusion

The False Horn plant was sensitive to soil moisture stress. This was reflected in reduced growth through reduced plant height. Response to water stress reflects equally on the leaf length and width hence the leaf length: Leaf width ratio. The study has shown that False Horn plantain for that matter plantain genotypes have different inbuilt mechanisms for resistance to drought stress. Physiological responses of plantain to stress require that each parameter be studied under control environment without other having influence.

4.6 Competing Interests

Authors have declared that no competing interests exist.

4.7. References

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5. Evaluating postharvest characteristics of Apantu (Local False Horn) plantain for harvest indices determination

5.1 Abstract

Plantain (Musa sp., genome AAB) is a major locally grown starchy staple food highly consumed by Ghanaians with a per capita consumption of 84.4 kg. However, the maturity index of the fruit has been dependent on various characteristics including angularity, fruit tip drying, etc. A physiological study was conducted on the fruit characteristics of Apantu at harvest to determine the appropriate maturity index to be used by smallholders. The results were subjected to correlative analysis and revealed a strong positive correlation between age at harvest and pulp to peel ratio. The number of leaves declined with age while pulp to peel ratio, bunch weight, peel weight and fruit weight showed positive correlation with age. The low correlations observed between the age at harvest and peel thickness, peel weight, density of fruit and pulp thickness makes them unreliable indices, possibly due to the difference in environmental conditions. Generally, this is true since smallholders rely upon rainfed irrigation and physiological characteristics are greatly influenced by the watering component of the environmental. It is therefore recommended that farmers use bunch ages to determine harvest maturity. This could be done through tagging of plants at flower emergence due to the strong correlation it had with pulp:peel ratio. Further testing on each variety is needed to make tangible conclusions on some of the maturity indices to be adopted or must be backed with other variables.

Keywords: Musa, Apantu, physiology, postharvest, maturity indices

5.2. Introduction

Plantain (AAB subgroup) is one of the earliest crops domesticated by man and grown in organized agriculture in the humid forest zones of West and Central Africa. Plantain makes up the fourth most important world food item after cereals in terms of the gross value of production. They have been identified to be of great socio-economic and nutritional significance in growing regions (Dzomeku et al., 2011). In the developing world plantain is identified as the fourth most important food commodity after rice, wheat and maize (FAO, 2010, Wattanachaiyingcharoen and Boonyanuphap, 2003).

Despite the high value of plantain, poor soils, growing pest threats and disease pressures have affected production, the most notable being the fungal disease - Black Sigatoka

(*Mycosphaerella fijiensis*) (IITA, 1992;Stover and Simmonds, 1987; Swennen, 1990). Yield losses due to the disease are highly significant ranging from 20 to 50%. Under very severe conditions yield losses may be as high as 80%. All the plantain landraces in Ghana are susceptible to black Sigatoka disease and other pests except cooking bananas (Dzomeku et al., 2006). The fear that the disease could wipe out the susceptible cultivars, research efforts have been to introduce developed hybrids in readiness for any eventuality. New hybrids, resistant or tolerant to the Black Sigatoka disease were introduced into the country as a long term solution. A large deployment project was executed with four hybrids.

Apantu, a False Horn plantain is the most popular plantain cultivar in Ghana. Apantu cultivation has become a feature of great socioeconomic importance in Ghana from the point of view of food security and job creation. It belongs to the non-traditional sector of the rural economy, where it is used mainly to shade cocoa and is also an essential component of the diet. Apantu is also very important sources of rural income (Ortiz and Vuylsteke, 1996). More than 90% of the cultivated area in Ghana belongs to smallholder farming system. In the agricultural sector, plantain is ranked fourth in Ghana (FAO, 2010) and contributes about 13.1% to the Agricultural Gross Domestic Product (AGDP). Its per capita consumption of 84 kg (SRID-MOFA, 2010) is higher than all other starchy staples. A total of 359,865 ha of land area in Ghana is used to cultivate plantain producing an annual average of 3.7 million tonnes of fruits, of which more than 95% is sold on the domestic market (SRID-MOFA, 2010). Apantu production is concentrated in the three agro-ecological zones namely rain forest, moist semideciduous forest and forest-savanna transition. The rainfall pattern is bi-modal from March to July as the major rainy season and August to November as the minor season. Although Apantu plays a significant role in the farming systems in Ghana, there has not been any conscious effort to develop any maturity indices for the crop. Maturity indices of plantain under rainfed production could be unreliable as they are often influenced by the environment. Physical, biochemical and physiological parameters are used to define the maturity stage for harvesting of fruits (Jha et al., 2006). Huda (2003) and Robinson and Saúco (2010) described that the banana fruits having the stage of three quarters round was considered as commercial maturity. Irtwange (2006) considered the parameters such as chronological age (elapsed days from bloom to harvest), size, shape, surface characteristics, colour, firmness, compositional factors (soluble solids), development of abscission layers, surface morphology, solidity, tenderness, sugars, starch, sugar-to acid ratio and oil content for the determination of maturity indices. The common maturity indices used by most plantain farmers for assessing for harvest in view of the absence of universal criteria, are any combination of the following: (i) By experience and

judged largely by the visual appearance of the hanging bunch and particularly by the angularity of individual fingers (Palmer, 1971). (ii) Fruits harvested when the fingers of the first hand on the bunch show signs of ripening or yellowing or when the finger tips turn black (Dadzie, 1994b, c). (iii) In commercial plantations, fruits destined for distant markets are harvested at a stage known as 'three quarters full', when the fingers are still clearly angular. For local markets fruits are often harvested when fingers are full or rounded (iv) Usually coloured ribbons are used in commercial plantations to provide information regarding bunch age. (v) Fruit diameter (or caliper grade of fruit) and fruit length may be used as criteria to determine when to harvest (Dadzie and Orchard, 1997). Dadzie and Orchard (1997) stated again that while it is advantageous if the maturity index is non-destructive so that every fruit can be evaluated, it is important that the indices can be measured in a rapid, simple and inexpensive way (Reid, 2002). The method of evaluation should be simple and easily replicable, not requiring any complex scientific methodology so that farmers with basic literacy in mathematics can evaluate their plantain production and maturity time. In other jurisdictions, various instrument based techniques are used to measure maturity and ripeness. It is important to note that if the fruits are very mature at harvest, particularly following a heavy rain, peel splitting can occur. It is evident that under rain-fed conditions, the maturity indices are influenced by the environment. In the dry season angularity may be deceptive as an index of maturity. Also the soil nutrient and soil moisture could also influence the maturity indices. The objective of this study therefore was to critically evaluate the physiological characteristics of Apantu with maturity indices.

5.3 Materials and methods

Field samples of Apantu (False-horn) plantain were harvested at 5- day intervals of 70 to 105 days after flower emergence. The fruits were harvested from the Crops Research Institute plantain orchard at Fumesua near Kumasi in the rainy and dry seasons. Data was taken on 20 plants and replicated three times. In all, a total of 480 plants were harvested from the orchard. The field is located at 1° 31'W and 60 43'N. The soil type is the sandy-loam (Aeronosols). The annual rainfall ranges between 1400 and 1700 mm. Annual temperature range of 22 to 33°C can be recorded on this field all year round.

In this study, 60 plants per maturity were tagged at flower emergence to determine their age after flowering. At sampling ages fresh samples were harvested in the cool of the morning to conform to postharvest standards of keeping the fruit at a minimum achievable temperature to preserve harvest quality before reaching the pack house, market or the consumer. This study

was carried out on the field right after harvesting and continued in the laboratory before any deterioration. Studies were carried out within 48 h of harvesting. The methods of Dadzie and Orchard (1997) and Gowen (1995) were used. All measurements were limited to the fingers of the second hand on each bunch sampled. However, when there were not enough samples, the third hand was included. Some agronomic data were recorded prior to harvesting to assess any correlation between them and observations at harvest. Some of these were; (a) plant height; (b) pseudostem girth; (c) number of functional leaves; (d) bunch weight; (e). total number of fingers, and (f) number of hands.

At harvest, bunch weight was determined by weighing individual bunches with a balance. Number of hands was obtained by counting the number of hands on each bunch. Number of fingers was obtained by simply counting the number of fingers (per hand) on each bunch. Fruit weight was determined by weighing individual fruit on a weighing scale. Fruit length was determined by measuring the outer curve of individual fruit with a tape measure from the distal end to the point at the proximal end where the pulp is judged to terminate. Fruit girth or circumference was determined by measuring individual fruit with a tape measure at the widest midpoint of each fruit. Volume of fruit was obtained by direct volume displacement or by weighing fruit under water (Kushman and Pope, 1968; Kushman et al., 1966). Pulp and peel weight were determined after fingers have been hand-peeled and peel and pulp weighed separately with Mettler electronic balance usually to 2 decimal places). Pulp and peel were separated, weighed individually and expressed as pulp to peel ratio. Hand peeling of each fruit after cutting transversely at the midpoint was done, and the peel and pulp separately measured with a pair of calipers.

The data were subjected to ANOVA and the standard deviation was emphasized to reveal or reduce significant errors between duplication.

5.4 Results and Discussion

Maturity indices must meet two requirements for producers, locations and seasons (years): (i) Minimum acceptable eating quality, and (ii) Long storage life (Dadzie and Orchard, 1997). The False Horn plantain fruit diameter recorded throughout the year ranged between 33.4 and 44.2 mm. They all fall within the minimum acceptable limits indicating that all were mature for consumption and compliant to African standards. With visual observation of fruit angularity,

the ages at which fruits were harvested displayed significant change from angular, at 70 days after flower emergence (AFE) to rounded, at 95 days AFE, a visual indication of maturity.



Figure 5.1. Relationship of pulp: peel ratio of Apantu with bunch age.

There was a positive correlation between pulp-to peel ratio and bunch age (Figure 1). This corroborated well with the results of Dadzie and Orchard, (1997) on hybrid cooking bananas as the ratio of pulp to peel increases with age at 1.27 (70 days) to 1.63 (95 days); a strong indicator of increasing maturity and readiness for harvest. The only challenge with the use of this index is the destructive approach. A finger must be removed from the bunch to determine if the requirement is met. This approach could serve as an entry point for pathogens as a wound would be created in the process of removal of the finger. However, smearing of petroleum jelly to prevent entry of pathogens through the wounded portion is recommended. There was a positive correlation between pulp-to peel ratio and bunch age (Figure 1). This corroborated well with the results of Dadzie and Orchard (1997) on hybrid cooking bananas as the ratio of pulp to peel increases with age at 1.27 (70 days) to 1.63 (95 days); a strong indicator of increasing maturity and readiness for harvest. The only challenge with the use of this index is the destructive approach. A finger must be removed from the bunch to determine if the requirement is met. This approach could serve as an entry point for pathogens as a wound

would be created in the process of removal of the finger. However, smearing of petroleum jelly to prevent entry of pathogens through the wounded portion is recommended.



Figure 5.2. Relationship of functional leaves of Apantu with maturity.

There was a decline in the number of functional leaves as the fruit approached maturity (Figure 2). This was not surprising as this is directly linked to translocation of nutrients as into fruits during fruit -filling as well as the high susceptibility of the landrace to diseases. The most destructive of these diseases being Black Sigatoka caused by *M. fijiiensis* as indicated by Dzomeku et al. (2006), IITA (1992).



Figure 5.3. Relationship of fruit weight to bunch age of Apantu.

The relationship between age and the weight of the fruit (Figure 3), showed a clear increase possibly due to the fruit filling. Plantain is a determinate plant and after flowering, the physiological process of nutrient translocation is for fruit filling. There is also translocation of photosynthate into the fruit without any new leaf production. Evidence shows that fruit filling is a critical stage of the plantain crop when abiotic factors can influence the yield. Turner and Thomas (1998) reported that, plantains are sensitive to soil water deficits; expanding tissues such as growing fruit are among the first to be affected.



Figure 5.4. Relationship between number of fingers and bunch weight of Apantu.

Bunch weight weakly correlated to number of fingers (Figure 5.4). The varying growth and developmental differences of the various fingers translated into physiological difference manifested in the weights. The environmental conditions under rainfed systems often show variations. It has been observed that the number of hands on a bunch as well as the number of fingers could be influenced by soil nutrient and rainfall.



Figure 5.5. Correlation between bunch maturity and peel thickness of Apantu.

There was a low correlation observed between the maturity at harvest and peel thickness (Figure 5.5). This correlation does not make the parameter a reliable maturity index for Apantu. There is normally a decline in fruit density due to the reduction of its displacement power (weight) and this was reflected in the test.



Figure 5.6. The relationship between bunch maturity and peel weight of Apantu.

The low correlations observed between the age at harvest and peel thickness, peel weight, density of fruit and pulp thickness makes them unreliable indices. They can, however; be indicators of other environmental conditions within which the fruit grew. Most of these parameters are cultivar dependent and as such cannot be blindly compared to other cultivars in literature.

The study has shown that rainfed plantain production cannot in anyway bring about the full potential of the crop. Productivity is affected because of the erratic nature of the rain coupled with the soil nutrient. The critical stage of growth of the crop is at flowering. Water deficit at flowering severely affects fruit filling. Plantains (Musa spp.) rarely attain their full genetic potential for yield due to limitations imposed by water limiting the plants photosynthesis. Plantains are known to be particularly sensitive to changes in the environment (. Plantain growth and yield is known to decrease drastically when the interval between watering was increased with the soil moisture falling below 66% of total available soil moisture (Robinson and Bower, 1998). Under severe soil moisture stress, physiological and biochemical processes occur. Physiologically stomata closes and this affects photosynthesis. It is worth noting that if the fruit is too matured at harvest, particularly following a heavy rain, peel splitting can occur. In the commercial banana industry, estimating the maturity is through fruit diameter differs with the standards used the world over differ; they may be expressed in the following ways: (a) Total thirty-seconds of an inch (e.g. grade 42 which is 110/32 inch); (b) The number of thirty-seconds of an inch above thirty-two (e.g. grade 10 which is 110/32 inch); (c) Millimeters (e.g. grade 42 is 33 mm using 0.794 for each thirty-second of an inch). In Central and South America grade is expressed as the number of thirty-seconds of an inch above 1in, whereas millimeters are used in the Caribbean and Africa (Stover and Simmonds, 1987). It is evident that under rainfed conditions these parameters may vary according to the seasons. During the major harvesting seasons (June to February) angularity would be a very pronounced index unlike in the harvesting months of March to May.

5.5 Conclusions

The peel to pulp is the singular most important indicator of fruit maturity that can be used on the field. However due to its destructive nature, it cannot be extensively used. In combination with the other parameters measured, the age of bunch is the most reliable stand-alone parameter and is recommended for extensive promotion and use by smallholder farmers.
5.6 Conflict of Interests

The authors have not declared any conflict of interests.

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6.0 Climate change enhanced Carotenoid pro-Vitamin A levels of selected plantain cultivars

6.1 Abstract

Diet diversification and the exploitation of traditional, micronutrient-rich germplasm of staple crops are generally regarded as sustainable and low-cost approaches to increase the micronutrient intake of resource-poor people. Sun's UV index was collected daily throughout the year. The study assessed the seasonality of provitamin A carotenoids in three plantain cultivars in response to climatic condition. Fruits were harvested at three maturities and freezedried before analysis. The results showed that there were high levels of the sun's UV-B radiations throughout the year with the highest occurring from November to May when the area experienced clear skies with minimal cloud cover. These high levels of the sun's UV-B index occurred between 9.00 h GMT and 17.00 h GMT. The study also showed that α-carotene content increased with maturity in "Apantu" during the rainy seasons ranging from 95 to 172µg/100g of dry pulp. Similar trends were observed during the dry season with a range of 28 to $489\mu g/100g$. The α -carotene contents were very high in the periods of high sun's UV-B radiations compared to the periods of low sun's UV-B radiations. The α-carotene levels in the giant French plantains showed similar trends. Intermediate French "Oniaba" and False Horn "Apantu" plantain cultivar showed the highest content of β -carotene during the dry season. The high provitamin A carotenoid levels in the cultivars coincided with the high levels of the sun's UV index.

Keywords: UV-B radiation; micronutrient; carotenoids; climate variability; Musa spp

6.2 Introduction

Diet diversification and the exploitation of traditional, micronutrient-rich germplasm of staple crops are generally regarded as sustainable and low-cost approaches to increase the micronutrient intake of resource-poor people [1]. Micronutrient deficiencies were reported to reach clinical levels in many countries in sub-Saharan Africa [2]; [3], including Ghana [4,5]. In most of these countries, plantains are a major staple crop, often grown in association with other food crops, like cassava, sweet potato and vegetables.

Micronutrient deficiencies are known to afflict millions of people in the developing world. Studies have shown that diet rich in carotenoids is associated with reduced risk of heart disease and cancer [1-3, 6]. Though micronutrients are readily available in fruits, vegetables and animal products, they are often not easily accessible to the poor. Often, the micronutrient-rich food products are undervalued by those who need them most. The diets of these vulnerable consist mainly of carbohydrates such as roots and tubers and cereals.

Reports from WHO/World bank indicate that around two billion people worldwide suffer from micronutrient malnutrition [7]. Vitamin A deficiency (VAD) is reported to be the leading cause of preventable blindness in children and increases the risk of disease and death from severe infections. In pregnant women VAD causes night blindness and may increase the risk of maternal mortality [5,8]. Vitamin A (vit A) deficiency is reported to be a public health problem in 118 countries, affecting an estimated 250 million preschool children [8]. Up to 500,000 children become blind every year, and 50% die within 1 year of becoming blind [9]. Reports indicate that close to 20 million pregnant women are also vitamin A vit A deficient. Iron (Fe) deficiency is reported to be the most common micronutrient deficiency in the world [9]. The WHO estimates that up to 1/3 of the world's population is Fe deficient [10–12]. In addition, close to 60% of the population in developing countries is thought to also be at risk of Zinc (Zn) deficiency [9]. Micronutrient deficiency in developing countries therefore is alarming; though all the crops rich in these nutrients abound [13]. Several indigenous vegetables are very rich in micronutrients compared to the introduced varieties and fruits in these countries; however, their low consumption is mostly due to stigmatization. It is often as a result of the mindset and the lack of information on the nutritional values of the crops.

Reducing vitamin and mineral deficiencies is an essential part of the overall effort to fight "hidden hunger" and malnutrition. Supplementing micronutrients in the form of pills or syrups, fortifying processed foods with micronutrients and breeding crops for increased micronutrients levels are three commonly applied options to increase micronutrients intake [14]. However, to reach resource-poor people, who often have limited access to health-care systems and/or formal markets, diet diversification and the exploitation of traditional, micronutrient-rich germplasm of staple crops are generally regarded as more sustainable and low-cost approaches, that fit well in traditional food systems [1,15].

It is reported that certain vitamin concentrations in some fruits and vegetables may be affected by irradiation; however, there is naturally a large variation of these vitamins in fruits and vegetables. These variations are also dependent on the plant cultivar, growing conditions, maturity of the edible portion, post-harvest handling and storage conditions [16]. Studies have demonstrated that there is a high level of variation in both carotene and vitamin C content in tropical fruits, with the greatest variation coming from cultivar type [17,18]. Growing season, location and harvest time also influence nutrient composition of fruits [19]. Plantains (*Musa* spp) are among the most popular starchy staple grown in the humid tropics where micronutrient deficiency is prevalent. It comprises the world's fourth most important food crop, with an annual production of about 100 million tonnes. They constitute a starchy staple across some of the poorest parts of the world, including sub-Saharan Africa (with per capita consumption up to 400 kg) [20]. Plantains are a good source of income for millions of rural households in vulnerable developing countries with about 25 million tonnes produced annually [21]. Various cultivars of plantain are consumed at the green or half-ripe stages as cooked starchy carbohydrate or when ripe as dessert banana. For millions of rural poor in West Africa, plantains are not only a primary source of energy but also an important source of dietary minerals and vitamins. As such, the promotion and increased production of micronutrient-rich cultivars [1] has the potential to have a significant long-term beneficial impact on the incidence of micronutrient deficiencies.

The occurrence of orange-fleshed Musa cultivars with exceptionally high provit A carotenoids (pVACs) contents has been described [22–26] and results of large-scale screening activities showed that there is a high degree of genetic variability in the fruit pVACs contents of Musa genotypes, with values ranging from 0 to as high as 11,337 μ g/gdw [27]. Plantains, a major staple in West African countries, seem to have higher pVACs contents than dessert bananas. However, even within the plantain subgroup, substantial variation exists, suggesting that gains could be achieved by promoting more pVACs-rich cultivars over traditionally consumed cultivars. An ex-ante impact assessment using household data from plantain-growing regions of Ghana indicated that substituting Musa cultivars with high-pVACs content could reduce the burden of vitamin A deficiency-related illness by up to 17% and be more cost effective than other health-nutrition interventions [28,29].

Several studies have reported the nutritional composition of plantains [30–32]. Nutritional Analysis of within-fruit, within-hand and within-plant as well as the between-plant of plantains from West and Central Africa showed that significant variations exist in both provitamin A carotenoids (pVACs) and mineral micronutrient (Fe, Zn) contents across all sample groups [33].

In this era of climate change there are speculations of its effect on crop nutrition and nutrient composition in crops. Several studies have shown climate change and its effect on micronutrients in crops; and other studies have also reported an increase in CO₂ concentration

in the atmosphere could alter the levels of proteins, B vitamins and zinc [34,35]. Nutrient reduction is projected to be particularly severer in sub-Saharan Africa, where levels of undernutrition are already higher and diets are more vulnerable to direct impacts of climatic parameters associated with climate change.

Climate change is known to impact on the accumulation of minerals and proteins in crops, with elevated CO₂ being the underlying factor of most of the reported changes. These reports indicated that the effects are dependent on the type, intensity and duration of the imposed stress, plant genotype and developmental stages. Strong interactions (both positive and negative) are known to be found between individual climatic factors and soil available nitrogen (N), potassium (K), iron (Fe) and phosphorus (P). Some authors have proposed that future interventions to ensure plentiful, safe and nutritious food for the world's population may need to rely on breeding for nutrients under the context of climate change, including legumes in cropping system, better farm management practices and utilization of microbial inoculants that enhance nutrient availability [36]. Nevertheless, evidence shows that under seminatural field conditions UV-B radiation is not as detrimental for plant growth and physiology, as previously believed [37]. Furthermore, UV-B radiation effects are species specific and depend on interactions with other environmental parameters [38-40]. Studies under controlled environment showed that some species exposure to UV radiation had increased carotenoid concentration, in others there was a reduction. However, the effect of UV radiation on plant is also reported to be influenced by life forms [41]. Several studies on the effect of UV radiation on carotenoids in plants species showed that its effects could be variety dependent [42-44] [42]; [43]; [44] as each plant species behaves differently to adapt to the environment.

This study was therefore conducted to assess the pro-vitamin A active carotenoid levels of three selected plantain cultivars with season and maturity.

6.3 Materials and Methods

Weather data were collected from flowering to harvest using AccuWeather application using Huawei phone collaborated with Davis Vantage Pro2 Plus weather station. Weather data were collected daily at hourly intervals throughout the study period. Weather data (temperature, relative humidity and sun's UV index) were taken hourly from 9.00 Hours GMT to 17.00 Hours GMT for two conservative years (2014–2015).

Fruit samples were collected from a plantain orchard of the Crops Research Institute at Fumesua, Kumasi. The experimental design was Randomised Complete Block (RCBD) with four replications. The plant spacing was $3m \times 2m$ between and within rows giving plant population of 1667 plants per hectare. Each plot contained 25 plants. Planting was staggered (April and September) for fruiting to coincide with the rainy (June to October) and dry (November to May) seasons. At bolting, plants were tagged and monitored. Field maintenance was carried out by slashing as and when needed and no pesticide application was carried out. No chemical fertilizer was applied to the fields. However, pruning of dried leaves was done regularly.

Three plantain cultivars - "Apantu" (False horn), "Apem" (Giant French) and "Oniaba" (medium French) -were tagged at flowering in the Crops Research Institute plantain orchard at Fumesua, Kumasi. Two bunches from each plot were harvested from the middle portion of each plot at each maturity date (70, 80 and 90 days after flowering). This was done for dry and rainy seasons. These were pooled together and three bunches were randomly selected and sent to the International Sweetpotato center (CIP) laboratory for freeze-drying and milling. In the laboratory, samples for freeze drying were taken from the second and third hands. Four fingers were taken, peeled and sliced longitudinally for freeze drying. Samples were milled after drying and packaged in black polyethylene bags and stored at -20 °C for five days before shipment. Milled samples were carried in black polyethylene bags and sent to Institut für Biologische Chemie und Ernährungswissenschaft, University of Hohenheim, Germany for provitamin A analysis.

6.4 Carotenoid Analysis

Samples were extracted and analyzed as previously described [45]; however, minor adaptations in the extraction process as well as in the chromatographic settings were made. Briefly, about 100 mg sample were extracted in 2 mL tubes using a mixture of methanol and hexane (containing 1 mmol BHA and BHT as well as internal standard: apocarotenal; 30 min at room temperatures). Phase separation was enhanced using saturated sodium chloride solution and centrifugation (1 min, 13.200 rpm). The nonpolar layer was removed and stored separately. The remaining suspension was washed twice with hexane, thus pooling the organic phases that were evaporated and disolved in isopropanol, subsequently. After membrane filtration (PTFE, 0.20 μ m, 13 mm), solutions were injected into the Shimadzu HPLC system (CMB-20A

communication module, SIL-20AC HT autosampler, LC-20AT liquid chromatography pumps, SPD-M20A diode array detector). For chromatographic separation, a Prontosil 200-3-C30 column (Bischoff) was used, however, flow rate was reduced to 1.5 mL/min that led to gradient adaptations (min/%A: 0/100; 18/40; 22/0; 25/0). Provitamin A active carotenoids were detected using PDA (450 nm). Data were analysed using analysis of variance (ANOVA).

6.5 Results and Discussion

6.5.1 Carotenoid Contents Depending on Maturation and Seasonality

The sun UV radiation index was studied for two consecutive years using AccuWeather. The study showed that the sun's UV radiations were higher from November to May, which coincided with the dry and beginning of the rainy seasons. (Table 1). The sun's UV radiation index was relatively lower during the rainy seasons (June to October) (Table 1). The period of November to April is characterized by clear atmosphere with strong sunlight. The average minimum and maximum temperatures during dry season and rainy seasons range between 27 and 39 °C and 25 and 28 °C respectively. The relative humidity during the dry periods were between 25% and 35% whereas the wet periods experienced relative humidity of between 75% and 85%.

Months												
Year	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
2014	12 ±	12 ±	12 ±	12 ±	12 ±	7 ±	7 ±	8 ±	8 ±	8 ±	9±	12 ±
	0.1	0.1	0.01	0.1	0.1	0.01	0.01	0.01	0.01	0.01	0.01	0.01
2015	12 ±	12 ±	12 ±	12 ±	12 ±	7 ±	7 ±	7 ±	8 ±	8 ±	9±	12 ±
	1.0	0.1	0.02	0.1	0.1	0.01	0.01	0.01	0.01	0.01	0.01	0.01

Table 6.1. Monthly Means values of sun's UV-B Index for 2014 and 2015.

Provitamin A active carotenoids in three different plantain cultivars were determined (Tables 2, 3 and 4). Whereas cryptoxanthin was not detected in any of the plantain samples, substantial amounts of α - and β -carotenes were observed. Intermediate French "Oniaba" was the plantain cultivar with the highest content of provitamin A investigated in this study, followed by False Horn "Apantu" and Giant French "Apem" respectively (Figure 1). Despite minor deviations,

provitamin A contents in each plantain cultivar increased with higher degree of maturity. Our study revealed that periods of high levels of the sun's UV-B index coincided with high levels of provitamin carotenoids in plantain (Tables 1 and 4). The seasonality of the levels of carotenoid could be attributed to sun's UV radiation index having a significant impact on the enhanced contents. The results further showed that β -carotene content was higher than α carotene in the False Horn "Apantu" cultivar (Table 4). However, the trend was different in the Giant French "Apem" and the intermediate French "Oniaba". Overall, there were significant differences (p < 0.05) in carotenoid levels across cultivars and maturities. The results corroborated well with that of Shen et al., [46], Mark and Tevini, [47] and Sullivan and Teramuta, [38]. In their study, Shen et al. [46] showed an 8% increase in leaf carotenoid content even at low UV-B exposure (+9.75 μ W/cm²) from 2 to 8 days exposure, compared to nonexposed plants. However, at high UV-B exposure ($\pm 20.76 \,\mu\text{W/cm}^2$), the carotenoid content increased rapidly after a day's exposure (10.41% higher than the control). In a similar study on tomato fruits exposed to UV-B, Pérez, et al., [48] observed that exposure of tomato fruits to UV-B before harvest resulted in accumulation of lycopene and β -carotene content. They also observed that the highest accumulation of lycopene and β -carotene was induced by a UV-B dosage of 0.075 Wh m⁻² after 22 h of exposure. It was concluded that carotenoids synthesis was promoted by moderate UV-B radiation before harvest, nevertheless, the time and duration of exposure were paramount to a plant compound-specific response. Hu, et al.; [49] showed that LED and UV irradiations significantly accelerated ripening in orange and also caused changes in the soluble sugar, organic acid and carotenoid contents. Unlike fruit subjected to dark shade (DS) treatment, UV-treated (UVA, UVB, and UVC) fruits experienced significant increase in total soluble sugar, fructose and glucose contents.

Table 6.2.	Provitamin A	A active	carotenoid	contents	of	three	different	plantain	cultivars	at
maturation	and seasonal	ity.								

Cultivar	Maturity (Days) during the Rainy Season I									
Cultimor	α -carotene Le	e vels μg/100 g	edible pulp)	β-carotene Levels μg/100 g edible pulp)						
Cultivar	at T	Three Maturiti	es	at Three Maturities						
	70	80	90	70	80	90				
"Apantu"	95.7 ± 9.11a	108 ± 13.8a	171 ± 32.2a	151 + 15.7a	142 + 9.66	220 ± 29.2a				
"Apem"	$78.6 \pm 5.34b$	$128 \pm 4.31b$	$291 \pm 7.43b$	$108 \pm 9.3b$	148 + 9.25	$386 \pm 4.09b$				
"Oniaba"	83.7 ± 12.4ab	-	328 ± 57.7c	$102 \pm 22.1b$	-	$400\pm86.4\mathrm{b}$				
CV	12.4	10.4	30.6	10.7	11.8	30.1				
LSD(p<0.05)	12.0	13.2	20.2	8.9	10.9	12.3				

Table 6.3. Provitamin A active carotenoid levels $\mu g/100$ g edible pulp) three different plantain cultivars at maturation and seasonality.

Cultivar	Maturity (Days) During Rainy Season 11								
	α -carotene C	ontents at Thr	ee Maturities	β-carotene Content at Three Maturities					
	70	80	90	70	80	90			
"Apantu"	297 ± 38.1a	254 ± 36.3a	$474 \pm 45.0a$	362 ± 20.7a	$288 \pm 29.4a$	567 ± 59.7a			
"Apem"	$310 \pm 3.26b$	356 ± 5.79b	$379 \pm 2.12b$	$221 \pm 42.6b$	279 ± 39.3a	$210\pm44.5\mathrm{b}$			
"Oniaba"	160 ± 9.77c	$533 \pm 60.4c$	$435 \pm 4.70 \mathrm{c}$	$209 \pm 11.8b$	$558 \pm 65.4b$	$451 \pm 3.89c$			
CV	15.1	20.4	40.6	10.7	11.8	45.1			
LSD (p <0.05)	9.3	15.2	12.2	12.9	11.9	15.3			

Table 6.4. Provitamin A active carotenoid levels $\mu g/100$ g edible pulp of three different plantain cultivars at maturation and seasonality.

Maturity Date (Days) during the Dry Season										
Cultivar	α -carotene C	ontents at Thr	ee Maturities	β -carotene Content at Three Maturities						
	70	80	90	70	80	90				
"Apantu"	283 ± 5.08a	$420 \pm 2.80a$	$489 \pm 7.57a$	$358 \pm 5.24a$	577 ± 4.09a	573 ± 7.79a				
"Apem"	310 ± 3.26a	356 ± 5.79b	379 ± 2.12b	299 ± 2.31b	371 ± 5.09b	386 ± 4.09b				
"Oniaba"	719 ± 6.93b	793 ± 8.39c	$542 \pm 0.36c$	685 ± 7.63c	744 ± 10.3c	577 ± 2.75a				
CV	18.1	20.4	40.6	10.7	31.8	50.1				
LSD(<i>p</i> < 0.05)	30.3	15.2	12.2	12.9	21.9	16.3				

Intermediate French plantain "Oniaba" showed higher levels of α -carotene than β -carotene at early fruit filling stages in the dry season and reduced level at physiologically matured stages (Table 3). This period of reduction also coincided with the lower level of the sun's UV index in the field (Table 1). Studies have shown a wide inter- and intraspecific differences in response to UV-B irradiation with respect to growth, production of dry matter and physiological and biochemical changes in plants [50–52]. This phenomenon was also observed by Lidon and Ramalho [53] when leaves of rice were exposed to UV-B stress, the chlorophyll apparatus was

observed to increase; however, there was recovery 14 days after the stress. Studies showed that UV irradiation influenced accelerated orange ripening and also caused significant changes in the soluble sugar, organic acid and carotenoid content [49]. While in some species exposure to UV radiation could increase carotenoid concentration, in others there may be a reduction. However, the effect of UV radiation on plant physiological responses is also reported to be influenced by life forms [41]. It is also recorded to be influenced by abiotic factors like air temperature [47], atmospheric carbon dioxide concentrations [54] and soil nitrogen [55]; [56] and moisture content [38, 57]. The high carotenoid levels in our study also coincided with the periods of high ambient temperature levels.



Figure 6.1. Maturation and season dependent changes in Retinol Activity Equivalents (conversion factors: 1:12 for β -carotene, 1:24 for other provitamin A active carotenoids) in plantain cultivars (A) "Apantu", (B) "Apem" and (C) "Oniaba". Samples were harvested after 70, 80 and 90 days after flowering in dry and rainy seasons.

The study further showed that α -carotene content increased with maturity in "Apantu" (Table 3) during the rainy season ranging from 95 to $172\mu g/100g$ of dry pulp. Similar trends were observed during the dry season with a range of 28 to $489\mu g/100g$. The α -carotene contents were very high in the minor rainy and dry seasons compared to the major rainy season. The α -carotene levels in the French plantains showed similar trends (Table 3).

 β -carotene levels were also observed to be low in the major rainy season and very high during the minor rainy and dry seasons when the sun's UV radiations were high. The carotenoid levels were also high with maturity. The major rainy season is often between March and July and the minor season between September and November each year. One major characteristic of the major rainy season is thick cloud cover with reduced sunlight. The minor season is associated with clear atmosphere with strong sunlight and intermittent rains.

It is believed that plantains may be responding to high levels of sun's UV-B, either by stimulating protection mechanisms or by activating repair mechanisms to cope with the different types of stress. Plantains are known to contain high levels of phenolic compounds which are commonly used by plants in response to UV-B and also to attenuate the penetration of the UV-B range of the sun's spectrum into deeper layers of the tissue.

These high α -carotene levels with seasonality could be attributable to the high incidence of the sun's UV index. It was observed that during the time of the study, high levels of the sun's UV indices were observed (Tables 6.1 and 6.2). Sun's UV index were observed between 9.00 GMT and 17.00hours GMT daily from November to May.

The results confirm the assertions of Lu et al. [17]; Dhuique-Mayer et al. [18] and González-Molina et al. [19]; Lee and Kader, [58] that the nutrient composition of tropical fruits are influenced by growing season, location and harvest time of fruits. This could be good news for addressing micronutrient deficiency in developing countries as food-based strategy; however, the challenge could be the bioavailability of the carotenoids and their conversion to retinol for use by the body [59]. Climatic conditions including light and average temperature have a strong influence on the chemical composition of horticultural crops [60]. It is reported that phenolic compounds in fruits are influenced by UV-B [61–66]. However, the plantain plant has laticifers containing latex full of phenolic compounds within the leaves and fruits that could be potential adaptation mechanism for high solar UV radiations. Studies showed that UV-B radiation had a positive effect on the flavour of melons; carotenoid content slightly increased in older leaves in plants in response to UV-B radiation [67]. In a study of exposing *Malva parviflora* L., *Plantago major* L., *Rumex vesicarius* L. and *Sismbrium erysimoids* to two weeks of enhanced UV.B radiation, the carotenoid levels were increased [68]. While previous studies recorded substantial variability of fruit carotenoid content in Musa spp. suggesting possibilities for breeding, our studies have added a new dimension of sun's UV-B, being that it influences the provitamin content of plantain pulp. In their study it was observed that carotenoids are precursors for norisoprenoid compounds in grapes [69], hence the suggestion that UV-radiation may also affect grape and wine flavour. Understanding the mechanism(s) by which physiological processes are damaged, repaired and/or protected is therefore important for elucidating the eco-physiological role of UV-B radiation in enhancing carotenoid levels in fruits.

6.6 Conclusion

UV-B radiation continues to be relevant in plant physiology especially with the impact of enhanced UV-B in sunlight resulting from stratospheric ozone depletion. The current high incidence of the sun's UV-B and its effects on secondary metabolites in food crops calls for concern. Our results shows that increases in the sun's UV-B can cause photomorphogenic as well as genetic and physiological changes in plants. The study revealed that provitamin A carotenoids content of plantain cultivars vary with seasonality and with maturity. The seasonality of the UV-B radiation from the sun should trigger new research approaches of interactions between abiotic and biotic stresses and physiological responses of plants, especially metabolites. Variation in the carotenoid levels in plantain could be influenced by sun's UV-B index. Climate change with its accompanying complexities therefore could have a positive effect on some plant secondary metabolites and improve nutritional content of some crops.

6.7 Author Contributions

For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used "conceptualization, B.M.D. and J.N.W.; methodology, B.M.D and J.P.W.; software, D.N.; validation, H.K.B., J.N.W. and D.N.; formal analysis, J.P.W.; investigation, B.M.D.; resources, B.M.D. ;J.N.W data curation, J.P.W.; writing—original draft preparation, B.M.D.; writing—review and editing, B.M.D.; J.N.W.; visualization, H.K.B.; supervision, J.N.W.; D.N.; project.; funding acquisition, J.N.W.; D.N.", please turn to the <u>CRediT taxonomy</u> for the term explanation. Authorship must be limited to those who have contributed substantially to the work reported.

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7.0 General Discussion

7.1 Drought effect on plant growth and development

Water deficit is a hazardous natural phenomenon that affects plant growth and development. Water deficit differs from aridity; which is normal condition of severe water unavailability in a specific region. Drought or water deficit is a deviation from normal climatic conditions in which there is lack of precipitation for a long period of time leading to shortage of soil moisture with its negative consequences. While all living organisms will suffer from water deficit in agriculture production could lead to famine in some cases. It is therefore imperative to gain proper understanding of the phenomenon and its effects on agriculture production so as to develop strategies for sustainable production. It is reported that climate change effects will continue to cause havoc on agriculture production and plant ecosystems (da Silva et al., 2013).

At the plant level the negative effects of drought can induce interesting adaptations in plants that allow them to survive and reproduce. These adaptations can lead to the emergence of new functional groups in a given ecosystem or serve as an important tool for improving agricultural practices and plant breeding programs. We need to study the physiological strategies used by plants to control water status, avoid oxidative stress and maintain vital functions in an attempt to understand the morphological and physiological changes plants undergo to ensure their survival under different environmental conditions.

A large amount of water and nutrients is required by plants throughout their life cycle as all aspects of their development is affected by reduced soil moisture content. This reduction in the amount of soil moisture leads to changes in the physical environment, which subsequently affect physiological and biochemical processes in plants (Sarker, et al., 2005; Sircelj, et al., 2005; Silva et al., 2009). Soil water deficit is known to cause nutrient deficiencies, even in fertile soils, as a result of reduced mobility and absorbance of individual nutrients, leading to a lower rate of mineral diffusion from the soil matrix to the roots (Silva et al., 2011). Thus, drought is doubtlessly the most important stress factor limiting plant life.

7.2 Plantain production and planting material

Plantain is a cheap starchy staple of great economic importance for over 120 million smallholders in West Africa. Its production is faced with a myriad of challenges from planting material availability at the time of planting through to post-harvest (Dzomeku et al., 2007). At the start of the rainy season (March/April) after a prolonged drought, which also marks the beginning of the planting season farmers struggle to acquire adequate and healthy planting materials to establish their farms. Plantains are mainly parthenocarpic due to their triploid nature and have to be propagated through vegetative means. They are propagated traditionally by planting naturally regenerated suckers and also use of seedlings propagated through tissue culture and macro- propagation. Natural regeneration is where farmers select planting material from existing plantations to start or expand plantations. Farmers make decisions about the cultivar, age and size of the sucker (Staver et al., 2010). This method is affordable because obtaining the suckers is relatively easy. It is used widely but has a risk of spreading diseases and pests, though disease spread can be reduced through treatment of suckers. Also, suckers do not have uniformity and the numbers are low and a high possibility exists of farmers not being sure of the variety (Sheela and Ramachandran, 2001) especially when suckers are purchased from the open market. These factors can lead to inconsistency in yield.

The prolonged drought destroys the young peepers and potential buds that could develop into sword suckers in the field. Tissue culture techniques are not easily affordable to the smallholder farmers. It is the on-farm techniques that could help, however, adoption of these techniques are also low coupled with lack of reliable water source to initiate rapid multiplication.

Plantain production is strongly influenced by abiotic and biotic factor; and in this era of climate change the impact on its production will be great. Cultivation under this scenario faces increased stress because of natural and anthropogenic factors leading to productivity stagnation. It has been very difficult to develop efficacious, low-cost, and easy-to-apply methods in abiotic stress management. Recent climate change has aggravated the situation reducing average yields by over 50%. While the potential of the crop is 20t/ha, the achievable yield has been hovering around 11t/ha. The most affected are the smallholder farmers in developing countries whose production systems are rain-fed. The crop had long gestation period (crop cycle) of between 12-14 months under rain-fed conditions resulting in drastic

reduction in yield especially between February and June in West Africa where the drought is very severe.

7.3 Environmental Stress and plantain responses

Water is one of the most important abiotic factors for plantain growth. Plantains require water for all the physiological processes like photosynthesis, nutrient uptake and transportation as well as cooling (Farooq et al., 2009). Plantains are sessile just as other plants; hence unable to move when the environment becomes unfavorable. Accordingly, they have to be able to respond and adapt to the local environmental changes. Since water is essential for their survival, the ability to tolerate water stress is crucial. Water limitation is a major problem for plantain production especially from November to March because of the prolonged drought. The period between November and February is the season of plantain glut and the price volatility is very high hence lean season starts just at the start of the rains.

The effect of water stress on plantain growth is complex and they respond with many protective adaptations. Water stress causes the plant to suffer from dehydration and overheating of its cells and tissues. Hence, water stress resistance of the plant includes the ability to withstand dehydration and ability to withstand overheating (heat-resistant). High heat-resistance is not always linked with high drought-resistant and there is no universal mechanism of adaptation of plantains to drought. Drought-resistance is a property which is formed and developed in the process of ontogenesis and is based on the whole preceding phylogeny of the plant. Based on the above, Henckel (1964) defined drought resistance as follows: "Drought-resistant plants are those which in the process of ontogenesis are able to adapt to the effect of drought and which can normally grow, develop, and reproduce under drought conditions because of a number of properties acquired in the process of evolution under the influence of environment."

Drought stress influences the process of cell growth mainly as a result of loss of turgor (Taiz and Zeiger, 2006). Water as a limiting factor results in impairment of cell elongation as there is poor water flow from xylem to the nearby cells (Nonami, 1998)). Plantains under irrigation produce a leaf in every seven days while under rain-fed conditions produce a leaf in 9 to 12 days. Since plantain produces 42 leaves before flowering, under supplementary irrigation plantain flowers six (6) months after planting while under rain-fed conditions it flowers at 9 to 11 months.

Although a large amount of research has been carried out in the tropics including water management, drip irrigation, and fertilizer use, work on evaluation of plantain varieties under conditions of water deficit is still very limited, as is the availability of related information. Probable reasons could be that most genebanks and breeding programs actively involved in germplasm evaluation and development are located in the humid tropics with ample rainfall. Also, creating large-scale drought conditions for a crop like plantains that is large and of long duration (12–20 months), presents many practical difficulties.



Figure 7.1. Photograph of water stress effect on leaf numbers of plantain.

The study has shown that rainfed plantain production cannot in anyway bring about the full potential of the crop. Productivity is affected because of the erratic nature of the rain coupled with the soil nutrient. The critical stage of growth of the crop is at flowering. Water deficit at flowering severely affects fruit filling. Plantains (*Musa* spp.) rarely attain their full genetic potential for yield due to limitations imposed by water limiting the plants photosynthesis. Plantains are known to be particularly sensitive to changes in the environment (Ravi et al., 2013) Plantain growth and yield is known to decrease drastically when the interval between watering was increased with the soil moisture falling below 66% of total available soil moisture (Robinson and Bower, 1998). Under severe soil moisture stress, physiological and biochemical processes occur.



Figure 7.2. Photograph of drought devastated a plantain field at Fumesua near Kumasi.

Drought as a complex environmental factor varies over location and time. This makes it difficult for the researcher to create a standard for drought when the crop is challenged under field conditions. These real time unpredictable situations are entirely different from screening for drought under controlled conditions (Ravi et al., 20130. Under soil water deficit the first physiological response in plantain is stomatal closure and as this occurs it affects photosynthesis.

Drought stress is observed to affect the morphology of plantains. Water stress affects number of leaves and the size of individual leaf are also reduced under the drought conditions. Leaf expansion normally depends on the turgor pressure and the supply of assimilates. When there is reduction in turgor pressure and slow rate of photosynthesis under drought conditions there is associated reduction in leaf expansion (Rucker et al., 1995). There is significant reduction in fresh and dry weights under water limiting conditions (Zhao et al., 2006). Other studies in maize showed significant reduction in plant height, leaf size, and the stem girth under the water limiting conditions (Khan et al., 2015). In a study, Kamara et al., (2003) reported that biomass accumulation in maize was significantly reduced under drought conditions imposed at various growth stages. However, in long duration cycle crops like plantain, early stage drought stress does not affect the final yield. There is a recovery from these early growth stage abiotic stresses; however; water limiting factors influence yield when it occurs just before flower emergence.

Climate change affects plantain growth in diverse ways. Plantain fruit growth and development is often hindered during prolonged cloud cover. It has been observed in the field that when there is continuous cloud cover of over 70% for about a month or more which is often associated with the rainy season, there is stagnation in the increase in size of plantain fruits. This phenomenon adversely affects export markets. The fruits fail to increase in size to mature and also to meet market standards.

In the commercial industry, estimating the maturity is through fruit diameter differs with the standards used the world over differ; they may be expressed in the following ways: (a) Total thirty-seconds of an inch (e.g. grade 42 which is 110/32 inch); (b) The number of thirty-seconds of an inch above thirty-two (e.g. grade 10 which is 110/32 inch); (c) Millimeters (e.g. grade 42 is 33 mm using 0.794 for each thirty-second of an inch). In Central and South America grade is expressed as the number of thirty-seconds of an inch above 1in, whereas millimeters are used in the Caribbean and Africa (Stover and Simmonds, 1987).

Maturity indices of plantain under rainfed production could be unreliable as they are often influenced by the environment. Physical, biochemical and physiological parameters are used to define the maturity stage for harvesting of fruits (Jha, et al., 2006). Huda, (2003) and Robinson and Saúco, (2010) described that the banana fruits having the stage of three quarters round was considered as the commercial maturity. Irtwange (2006) considered the parameters such as chronological age (elapsed days from bloom to harvest), size, shape, surface characteristics, colour, firmness, compositional factors (soluble solids), development of abscission layers, surface morphology, solidity, tenderness, sugars, starch, sugar-to acid ratio and oil content for the determination of maturity indices. The common maturity indices used by most plantain farmers for assessing for harvest in view of the absence of universal criteria, are any combination of the following: (i) by experience and judged largely by the visual appearance of the hanging bunch and particularly by the angularity of individual fingers (Palmer, 1971). (ii). fruits harvested when the fingers of the first hand on the bunch show signs of ripening or yellowing or when the finger tips turn black (Dadzie, 1994). (iii). In commercial plantations, fruits destined for distant markets are harvested at a stage known as 'three quarters full', when the fingers are still clearly angular. For local markets fruits are often harvested when fingers are full or rounded (iv). Usually coloured ribbons are used in commercial plantations to provide information regarding bunch age. (v). fruit diameter (or caliper grade of fruit) and fruit length may be used as criteria to determine when to harvest (Dadzie and Orchard, 1997). Dadzie and Orchard (1997) stated again that while it is advantageous if the maturity index is non-destructive, (Reid, 2002) so that every fruit can be evaluated, it is important that the indices can be measured in a rapid, simple and inexpensive way. The method of evaluation should be simple and easily replicable, not requiring any complex scientific methodology so that farmers

with basic literacy in mathematics can evaluate their plantain production and maturity time. In other jurisdictions, various instrument based techniques are used to measure maturity and ripeness. It is important to note that if the fruits are very mature at harvest, particularly following a heavy rain, peel splitting can occur.

It is evident that under rainfed conditions these parameters may vary according to the seasons. During the major harvesting seasons (November to February) angularity would be a very pronounced index unlike in the harvesting months of March to May.

7.4 Microbes and sustainable plantain production

Africa is currently home to 1.2 billion people up from just 477 million in 1980 and is projected to reach 2.5 billion people by 2050, which highlights the need for sustainable food production strategies. However, plantain productivity is low with a yield gap of 8% between the actual production and attainable potential in sub-Saharan Africa (SSA) (Sanchez, 2002; Bekunda et al., 2010). Poor soil fertility (Kadiata and Lumpungu, 2003; Tening et al., 2013) and susceptibility to crop pests and diseases (Gunes et al., 2007; Tavares et al., 2010) constitute major crop production constraints in sub-Saharan Africa (SSA). Plant growth promoting and protecting microbes have demonstrated the potential to enhance sustainable crop production gained popularity (Brock et al., 2018; Ngamau et al., 2012;). Plant growth promoting rhizobacteria (PGPR) can suppress plant diseases by competitive colonization, improving nutrient uptake, or stimulate phytohormone production (Berger et al., 2015, Tchuisseu et al., 2018). These have been used to successfully improve plant growth and protection under various ecological conditions such as flooding, drought, salt, flower wilting, metals, organic contaminants, and bacterial, nematode and fungal pathogens (Ruppel, et al., 2006; Shaharoona, et al., 2008; Perez-Montano, et al., 2014; Xue, et al., 2015; Karthika, et al., 2017; Liu et al., 2017; Berger et al. 2015). Plantain research has not taken advantage of these ecosystem services to improve production. It is therefore imperative to exploit the benefits of these microbes by identifying, isolating and formulating these beneficial microbes for plantain production, to improve plant nutrition and protection within the framework of integrated soil fertility management (Vanlauwe, et al., 2010; Bello, et al., 2017; Mukhongo, et al., 2017). Recent studies have provided a collection of native bacteria and fungi (e.g. Rhizobia, Bacillus, Mycorrhiza, and Beauvaria) associated with the rhizosphere of maize and legume plants in sub-

Saharan Africa (Fankem, et al. 2008; Tchuisseu, et al. 2018). These native microbes have

shown plant growth-promoting ability such as nitrogen fixation, phosphate solubilization, phytohormones and siderophores production, salinity tolerance, and rhizosphere colonization potential that enhance plant growth (Fankem et al., 2014; Fankem et al., 2015).

The role of microbes in biotic and abiotic stress management has been focused on more in recent times. Soil supports plant growth through complex and dynamic systems. Plantain growth and development are affected by some stresses which are major constraints for sustainable plantain production in the soil environment. Biotic stresses include plant pathogens and pests (viruses, bacteria, fungi, insects, and nematodes). Abiotic stresses are salinity, drought, flooding, heavy metals, temperature, gases, and deficiency of nutrients or excessive nutrients. Abiotic stresses cause yield reduction, and their intensity changes according to the soil types and plant factors. Some studies conducted recently have reported that microorganisms could help crops fight against abiotic stress. It has long been recognized that microorganisms have effects on plant growth (Verbon and Liberman, 2016), nutrient management (van der Heijden et al., 2008), and disease control (Mendes et al., 2013). Natural mycorrhizal colonization of plantain roots has never been documented in Ghana. Our study has therefore given the clue of the potential of exploring the use of arbuscule mycorrhizae

study has therefore given the clue of the potential of exploring the use of arbuscule mycorrhizae fungi (AMF) as an ecosystem service provisioning for sustainable production of plantain. What is left is to identify the fungal strains that are associated with these roots using molecular tools and hence develop culture for application in the field.

Plantain production involves long term land use hence soil and crop management contributes to its productivity. Since production goes through several cycles of ratooning, smallholder farmers neglect the management of the mats, and the soil fertility after planting of the crop plant. The impact of different tillage and fertilization practices in plantain production influence fungal communities in a long-term. With the high cost of labour as a result of rural-urban migration thus shortage of youth manpower in the rural areas, there is a heavy use of herbicide application in plantain production.

7.5 Plantain mycorrhizal associations

Mycorrhizal association is a beneficial relationship between some soil fungi with the plant roots (Smith and Read, 2008). In this mutual association, the plant can benefit from these fungi by a better transfer of inorganic nutrients like P, N, K, Ca, S, Fe, Mn, Cu and Zn, as well as

water. In turn, the fungi receive plant carbohydrates (Habte and Osorio, 2001; Parniske, 2008), but some exceptions exist (Brundrett, 2004). There are different types of mycorrhizal associations with arbuscular mycorrhizas (AM) being widespread in the plant kingdom. It is estimated that around 74 % of all plant species are able to establish a symbiosis with arbuscular mycorrhizal fungi (AMF) (van der Heijden *et al.*, 2015), a group of fungi belonging to the phylum *Glomeromycota* (Berruti *et al.*, 2014). AMF are obligate biotrophs; which implies that they cannot complete their life cycle without a host plant (Berruti *et al.*, 2014). A distinctive feature of AMF is the formation of arbuscules, which are tree-shaped structures of branched fine hyphae developing inside the root cortical cells (Berruti *et al.*, 2014). Arbuscules are thought to be the main site of nutrient exchange interface between the plant and the fungus. Others characteristics are intra-radical structures (the vesicles), constituting generally storage organs, which are also able to support colonization of new plants as propagules (Habte and Osorio, 2001). Hyphae grow intra- and extra-radical, while the latter exploits the soil matrix beyond the nutrient depletion zone of the plant and therefore improves nutrient supply for the plant.

AMF play a crucial role in ecosystem services - which are benefits to humankind provided by ecosystems, including supporting (nutrient cycling, soil formation), provisioning (food and water supply), regulating (climate and disease control) and cultural (recreational and spiritual benefits) services (Millennium Ecosystem Assessment, 2005) - mainly within the supporting category. A focus has been on its role in plant nutrition, mainly due to nutrient transport to the plant. AMF can increase nutrient content and growth parameters of banana plants (Jaizme-Vega and Azcón, 1995), and improve soil structure by secreting glomalin, which improves soil stability and increases water retention (Gianinazzi et al., 2010). Also, AMF are known to increase plant tolerance against biotic and abiotic stresses in a large variety of cases. Mineral content and secondary metabolites in plants are affected by the symbiosis, which can result in improved nutritional and medicinal status of plants for human health (Gianinazzi et al., 2010). Many studies on plantain (Musa sp.) showed the beneficial effects of AM on plant tolerance against abiotic and biotic stresses. It was shown that AMF can increase plantain tolerance to salt stress, generally related to increased plant growth (Yano-Melo et al., 2003) and to aluminum toxicity, also with positive growth effects of inoculated plants but without significant difference in water and nutrient uptake of inoculated and non-inoculated plants if no Al was applied (Rufyikiri et al., 2000). AMF are assumed to induce systemic resistance against plant parasitic nematodes (Elsen et al., 2008) and to increase root branching, thereby affecting nematodes which prefer primary roots (Elsen, et al., 2003, Elsen, et al., 2003). Nematode built up was either not affected (Pinochet *et al.*, 1997) or negatively affected by AMF (Jaizme-Vega *et al.*, 1997, Koffi *et al.*, 2013, Olaniyi and Osuloye, 2014). Furthermore, the disease *Cylindrocladium spathiphylli*, causing root rot in banana plants, was negatively affected by AMF (Declerck *et al.*, 2002). Experimental set-up and plant genotype – AMF species – environment interaction is reported to play an important role in the functioning of this symbiosis.

7.6 General conclusion and recommendations

Plantain eco-physiology remains a complex phenomenon to study. The plant architecture does not yield itself for shielded production. As over 70% of this large herbaceous crop is water coupled with the shallow root system, any severe water deficit could cause dehydration. Under drought stress, the plant experiences toppling (stem logging) especially when carrying fruits.

7.6.1 General recommendations

It is evident that yield gaps in plantain-based production systems have increased over the years due to biotic and abiotic factors, drought has played a major factor in the reduction in yields at the farm level. As a crop for smallholders, application of irrigation though required for sustainable production could not be affordable. However, with irrigation and appropriate good farm management, plantain production could be very lucrative to feed the ever increasing population in the growing regions. The phenology of the crop is such that without proper farm management, glut periods remain between November and February. With proper sucker management, production can be all year round under supplementary irrigation.

Agroecological practices, such as the integration of ecosystem services- soil microbes, shadeand drought- tolerant crops, nitrogen-fixing and cover crops could potentially improve soil fertility and moisture retention, narrow yield gaps and overall farm productivity.

7.6.2 Recommendations specific for the study area

The West African subregion while identified as one of the hotspots of climate change, it is also a region of rapid population growth; hence more mouths to feed. Plantain, uniquely remains one of the cheapest starchy staple that can feed the population. The climatic conditions of the region are harsh for sustainable production. The farming system does not augur well for mechanization, however, irrigation and nutrient are needed to close the yield gap and sustainably produce to feed the population. Sustainable intensification in plantain production requires integration of readily available natural resources for production. Sustainable plantain production in West Africa requires among others, sucker management, irrigation and soil nutrient management.

7.7 General References

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8. Summary

West Africa suffers from climate uncertainty, high levels of variability, lack of access to realtime and future climate information, and poor predictive capacity are common barriers to adaptation though the region is identified as climate-change hotspot. The region's vulnerability is heightened by its overdependence on rain-fed agriculture, with its sensitivity to climate change and variability. Rain-fed agriculture contributes 30% of GDP and employs about 70% of the population, and it is the main safety net of the rural poor.

Drought affects plantain production in West Africa resulting in high level of food insecurity among the vulnerable. Under rain-fed production, achievable yield of plantain landraces are 11.0t/ha while the potential yield is 20t/ha. Plantain breeding is limited due to the complex nature of the breeding process. Plantain plant itself is a giant herbaceous plant occupying 6m² of land hence only 1667 could occupy a hectare of land. The production system is faced with a myriad of challenges from inadequate healthy planting materials at the time of plantain through the production system to post-harvest. Unlike bananas which are mainly produce by multinational companies under irrigation, plantain production is mainly by smallholder farmers under rain-fed agriculture.

Whereas banana production under irrigation is efficient, plantain production under rainfed production is haphazard and unsustainable. Farm sizes range between 0.4 to 5ha. Major limiting factors of the rainfed system is drought resulting in low yields and economic loses. Our study seeks to understand the effects of drought on the physiological responses of plantain crop under rainfed production. Different experiments were conducted to study the responses of various cultivars to on-farm rapid production of healthy planting materials. The results revealed that drought seriously affects natural regeneration of plantain planting materials as such new approaches need to be used for sufficient production. As each plantain plant produces averagely 42 leaves before flowering and each leaf has at least one axillary bud, it presupposes that 42 suckers should be produced at harvest. However, at harvest only 12 healthy suckers are produced around each plantain plant. The approach is able to exploit the full potential of every sucker to generate healthy planting materials. This technique therefore could be used to set up commercial propagation system for plantain planting material production. The study revealed Apantu and hybrid plantain FHIA-21 to show same responses. Plantain farmers can easily produce in large quantities healthy plantain planting materials using sawdust and building their own humidity chambers near water source. The study also showed that there is the potential

for farmers to use this technique for establishing commercial propagation centres to generate healthy planting materials. We also studied the physiological responses of Apantu crop to water regimes and natural mycorrhization of plantains. Also, study was conducted on the fruit maturity index and the effect of climate variability on fruit micronutrient content. The natural mycorrhization of plantain roots was the first to be reported on plantain in Ghana. During the study it was observed that plantain roots were naturally colonised by mycorrhizae. However, this phenomenon was cultivar specific; with Apantu roots more colonised compared to Apem. Sustainable intensification of plantain production could be achieved through the use of beneficial soil microbes in production. Conventional production systems, however, do not promote the survival of these microbes. The study further revealed that plantains respond to water stress by reduction in stomatal density, movement and reduction in leaf area. In addition, early stages of water stress had little effect on the final yield of the crop. The anatomical and physiological studies were challenges faced in laticiferous plant like the plantain. However, drought stress at advance stage of growth of plantain adversely affect yields. Our study also showed that fruit maturity index could vary with seasons. The various maturity indices used in plantain production could not be used under rain-fed conditions; especially angularity. The angularity index used was observed not to be appropriate in the dry season. The study also revealed high α -carotene levels with seasonality and maturity in plantain. The high levels of provitamin A in plantain fruits during the dry season coincided with high incidence of sun's UV index. This finding could play a significant role in the plantain industry as a food security crop the vulnerable who could not afford the high provitamin A foods especially for children under five years. However, the retention and bioavailability of the carotenoid after cooking need to be studied.

In conclusion all the chapters showed clear understanding of the behaviour of plantain under severe adverse environmental conditions and conclusions drawn to guide future production of the crop. Under climate change with its complexities, further studies on plantain is needed to improve productivity to achieve food security in West Africa.

9. Zusammenfassung

Westafrika leidet unter Klimaunsicherheit, hoher Veränderlichkeit, fehlendem Zugang zu Echtzeit- und zukünftigen Klimainformationen und mangelnder Vorhersagefähigkeit. Dies sind häufige Hindernisse für die Anpassung, obwohl die Region als Hotspot des Klimawandels identifiziert wurde. Die Anfälligkeit der Region wird durch ihre übermäßige Abhängigkeit von der Regenfeldbau-Landwirtschaft mit ihrer Empfindlichkeit gegenüber dem Klimawandel und ihrer Unbeständigkeit noch verstärkt. Regenfeldbau trägt 30% zum BIP bei und beschäftigt etwa 70% der Bevölkerung, und er ist das wichtigste Sicherheitsnetz der armen Landbevölkerung.

Die Dürre beeinträchtigt die Kochbananenproduktion in Westafrika und führt zu einem hohen Maß an Ernährungsunsicherheit unter den verwundbaren Bevölkerungsgruppen. Bei Regenfeldbau liegt der erzielbare Ertrag von Kochbananen-Landrassen bei 11,0 t/ha, während der potenzielle Ertrag bei 20 t/ha liegt. Die Kochbananenzüchtung ist aufgrund der komplexen Natur des Züchtungsverfahrens begrenzt. Die Kochbananenpflanze selbst ist eine riesige krautartige Pflanze, die 6m2 Land einnimmt, so dass nur 1667 einen Hektar Land einnehmen könnten.Das Produktionssystem ist mit unzähligen Herausforderungen konfrontiert, von unzureichendem, gesundem Pflanzmaterial zum Zeitpunkt der Pflanzung über das Produktionssystem bis hin zur Nachernte. Im Gegensatz zu Bananen, die hauptsächlich von Konzernen unter Bewässerung produziert wird multinationalen werden. die Bananenproduktion hauptsächlich von Kleinbauern im Regenfeldbau betrieben.

Bananenproduktion unter Bewässerung Während die effizient ist. ist die Kochbananenproduktion unter Regenfeldbau ungezielt und nicht nachhaltig. Die Betriebsgrößen liegen zwischen 0,4 und 5 ha. Ein wichtiger limitierender Faktor des Regenfeldsystems ist die Trockenheit, die zu niedrigen Erträgen und wirtschaftlichen Verlusten führt. Unsere Studie versucht, die Auswirkungen von Dürre auf die physiologischen Reaktionen der Kochbananenproduktion im Regenfeldbau zu verstehen. Es wurden verschiedene Experimente durchgeführt, um die Reaktionen verschiedener Sorten auf die schnelle Produktion von gesundem Pflanzmaterial im Betrieb zu untersuchen. Die Ergebnisse zeigten, dass Dürre die natürliche Regeneration von Kochbananenpflanzmaterial ernsthaft beeinträchtigt. Solche neuen Ansätze sollen für eine ausreichende Produktion genutzt werden. Da jede Kochbananenpflanze vor der Blüte durchschnittlich 42 Blätter produziert und jedes Blatt mindestens eine axillare Knospe hat, setzt dies voraus, dass bei der Ernte 42 Ableger produziert werden sollten. Bei der Ernte werden jedoch nur 12 gesunde Ableger um jede

Kochbananenpflanze herum produziert. Dieser Ansatz ist in der Lage, das volle Potenzial jedes einzelnen Ablegers auszuschöpfen, um gesundes Pflanzmaterial zu erzeugen. Diese Technik könnte daher zur Einrichtung eines kommerziellen Vermehrungssystems für die Kochbananenpflanzmaterialproduktion genutzt werden. Die Studie ergab, dass Apantu und die Hybridsorte FHIA-21 dieselben Reaktionen zeigen. Kochbananen-Bauern können problemlos gesundes Kochbananen-Pflanzmaterial in großen Mengen produzieren, indem sie Sägemehl verwenden und ihre eigenen Feuchtigkeitskammern in der Nähe von Wasserquellen bauen. Die Studie zeigte auch, dass es ein Potenzial für Landwirte gibt, diese Technik für den Aufbau kommerzieller Vermehrungszentren zu nutzen, um gesundes Pflanzmaterial zu erzeugen. Wir untersuchten auch die physiologischen Reaktionen der Apantu-Pflanze auf Wasserregime und die natürliche Mykorrhizierung von Kochbananen Ausserdem wurden der Fruchtreife-Index und die Auswirkung der Klimavariabilität auf den Mikronährstoffgehalt von Früchten untersucht. Die natürliche Mykorrhizierung von Kochbananenwurzeln war die erste, über die über Kochbananen in die Ghana berichtet wurde. Während der Studie wurde beobachtet, dass Kochbananenwurzeln auf natürliche Weise von Mykorrhizapilzen besiedelt werden. Dieses Phänomen war jedoch kulturspezifisch, wobei Apantu-Wurzeln im Vergleich zu Apem stärker kolonisiert waren. Eine nachhaltige Intensivierung der Kochbananenproduktion könnte durch den Einsatz von nützlichen Bodenmikroben in der Produktion erreicht werden. Herkömmliche Produktionssysteme fördern jedoch nicht das Überleben dieser Mikroben. Die Studie ergab ferner, dass Kochbananen auf Wasserstress durch Verringerung der Stomataldichte, Bewegung und Verringerung der Blattfläche reagieren. Darüber hinaus hatten frühe Stadien von Wasserstress wenig Einfluss auf den Endertrag der Pflanze. Die anatomischen und physiologischen Studien stellten Herausforderungen dar, denen sich latiziferische Pflanzen wie der Kochbanane stellen mussten. Dürrestress im frühen Wachstumsstadium des Wegerichs beeinträchtigte jedoch die Erträge. Unsere Studie zeigte auch, dass der Fruchtreife-Index mit Jahreszeiten variieren kann. Die verschiedenen Reifeindizes, die bei den der Kochbananenproduktion verwendet werden, konnten unter regenreichen Bedingungen nicht verwendet werden; insbesondere die Winkligkeit. Es wurde festgestellt, dass der verwendete Winkligkeitsindex in der Trockenzeit nicht geeignet ist. Die Studie zeigte auch hohe α -Carotin-Gehalte mit Saisonalität und Reife bei Kochbananen. Die hohen Provitamin-A-Gehalte in Kochbananenfrüchten während der Trockenzeit fielen mit dem hohen Auftreten des UV-Index der Sonne zusammen. Dieser Fund könnte eine bedeutende Rolle in der Kochbananenindustrie als Nahrungsmittelsicherheitskultur für bedürftige Menschen spielen, die sich die Nahrungsmittel mit hohem Provitamin-A-Gehalt nicht leisten konnten,

insbesondere für Kinder unter fünf Jahren. Allerdings muss die Rückhaltung und Bioverfügbarkeit des Carotinoids nach dem Kochen untersucht werden.

Zusammenfassend lässt sich sagen, dass alle Kapitel ein klares Verständnis des Verhaltens von Kochbananen unter ernsthaft widrigen Umweltbedingungen und Schlussfolgerungen für die zukünftige Produktion der Kulturpflanze aufzeigten. Angesichts des Klimawandels und seiner Komplexität sind weitere Studien über Kochbananen erforderlich, um die Produktivität zu verbessern und die Ernährungssicherheit in Westafrika zu erreichen.

10. Author's declaration

I hereby declare that this doctoral thesis is a result of my own work and that no other than the indicated aids have been used for its completion. All quotations and statements that have been used are indicated. I did not accept the assistance from commercial agency or consulting companies. Furthermore, I assure that the work has not been used, neither completely or in parts, for achieving any other academic degrees.

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The competitive project grants won

Year	Project Title	Donor	Amount
2015	Developing innovation platform in Offinso Humidtropics action site	FARA	10,000
2013	Clonal selection of plantain landraces for superior characteristics and higher productivity in Ghana	Bioversity International	25,000
2013	Opportunities for agroecological intensification and the increased use of agrobiodiversity in banana and cocoa growing communities	Bioversity International	42,500
2010	Improving Productivity of plantain in West and Central Africa	CORAF Competitive grant	\$800,000
2010	Installation of supplementary irrigation facility at Crops Research Institute for Improved crop and technology development in Ghana	Alliance for Green Revolution for Africa	\$289,580
2009	Assessing the molecular marker tools for the establishment of plantain genetic diversity in Ghana	International Fellowship for Agricultural Research (IFAR) Professional Development	\$11,000
2007	Measuring the impact of IARC and NARS development projects for clean seed multiplication and new cultivars of banana and plantain: Improving impact pathways for public goods	International Fellowship for Agricultural Research (IFAR) Professional Development	\$11,000
2007	Workshops for planning and reviewing results of impact assessment of new cultivars and clean seed multiplication promoted by TARGET project	Bioversity International	\$7,000
2005	Agronomical research in Ghana in the Field of Tree and Food Crops Cultivation	France Government	184,000 Euros

2004	Training of Extension Agents and farmers in	INIBAP	\$2000
	Kwabre District on Rapid Sucker Multiplication		
2002	Improving the delivery of superior (Disease	Cataby Charitabla	£217620
2005	Improving the derivery of superior (Disease-		£217029
	resistant plantain and banana hybrids and	Foundation, U.K.	
	associated technologies to smallholders in Ghana		
	(Phase II)		
2002	Peri-urban vegetable production: Soil	AgSSIP-CARGS	\$10750
	improvement and nematode control using		
	Crotalaria		
2002	Boost to food security in Ghana: Plantain Pilot	AgSSIP Priority	\$8000
	project for poverty alleviation	Research	
		programme	
2000	Evaluate of some cooking bananas in Ghana.	(INIBAP)	\$4000
1996	Environmental education in part of the Volta	IUCN, The	\$30000
	region of Ghana.	Netherlands	
1996	Train rural women in snail farming and also	Sophie Danforth	\$1500
	provide beehives for a community in the Volta	Conservation	
	region of Ghana	Biology Fund,	
		U.S.A	

Abomeku

Beloved Mensah Dzomeku

Kumasi, 03/08/2020