AGRONOMIC AND PHYSIOLOGICAL APPROACHES TO IMPROVING PRODUCTIVITY OF SELECTED SWEET POTATO (IPOMOEA BATATAS L.) CULTIVARS IN KWAZULU–NATAL: A FOCUS ON DROUGHT TOLERANCE

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Submitted in fulfilment of the requirements for the degree of Doctor of Philosophy (CROP SCIENCE)

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DECLARATION

I, Nozipho Mgcibelo Motsa, declare that:

• The research reported in this thesis, except where otherwise indicated, is my original

work and has not been submitted for any degree or examination at any other

university.

• This thesis does not contain data, graphs or other information from other researchers,

unless specifically acknowledged as being sourced from other persons.

• This thesis does not contain other persons' writing, unless acknowledges as being

sourced from other researchers. Where other written sources have been quoted, then

their words have been re-written and duly referenced.

• This study was funded by the Swedish International Development Cooperation

(SIDA) Organization for Women in Science from the Developing World (OWSD) and

by Professor Albert Modi from his research funds.

Signed: Nozipho Mgcibelo Motsa

Date: 30 November 2014

Signed: Professor Albert T. Modi

Date: 30 November, 2014

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DEDICATION

To my daughter Wakhile N. Simelane

GENERAL ABSTRACT

Sweet potato (Ipomoea batatas L) is a resilient food security crop with wide adaptation characteristics and hence can fit well under smallholder production. Its importance as a food security crop in relation to drought is still underestimated and fails to attract sufficient attention from agricultural researchers. The adaptive responses of different sweet potato cultivars to different agro-ecological areas may vary, and sweet potato is an important crop for small-holder farmers in KwaZulu-Natal (KZN) of South Africa, which has diverse agroecological areas. Adaptive responses of sweet potato cultivars in KZN's ecological regions are not known. The possible varying adaptive responses may impact on the food and nutrition security role of sweet potato. This study evaluated the ecophysiology, growth, yield and nutritional composition of three locally bred sweet potato cultivars in response to a range of climates and soils from KZN, South Africa. With the help of smallholder farmers, field experiments were conducted at three sites located in three different agro-ecological areas (Deepdale, Umbumbulu and Richards Bay) of KZN. Agronomic, physiology and yield data were collected. Harvested roots were further analysed for selected nutrients (starch and βcarotene) and other metabolic responses to drought stress. A separate study on physiological and yield response of sweet potato to water stress was conducted under controlled environment. Metabolic analyses were conducted continuously during plant growth. Plant growth, physiological responses and yield were significantly ($P \le 0.05$) influenced by growth environment. Drought stress in Richards Bay resulted in poor plant growth, low yields and nutritional content (starch and β-carotene content). High temperatures evapotranspiration (ETo) were associated with drought stress. The other locations (Deepdale and Umbumbulu) where ETo was low showed increased plant growth, yields and nutritional content. The cultivars' ecophysiology, growth and yield were not suitable for the Richards Bay agro-ecology/bioresource group. They were more suitable for Umbumbulu and Deepdale agro-ecology/bioresource groups. Under controlled conditions, the cultivars adapted to water stress through reduced canopy size. When fully-irrigated, they increased vegetative growth than storage root growth, thus resulted in low storage root yield. This suggested that the cultivars were drought tolerant and suitable for production in marginal areas. Leaf phytochemical content was high in sweet potato leaves compared to other common leafy vegetables. It was even higher in leaves of water stressed plants. This indicated that both leaves and storage roots can be utilised for improved food and nutrition security. Under marginal areas where storage root yield is compromised, the leaves can contribute to food and nutrition security. In areas where rainfall is not limited, communities can benefit from both storage roots and leaves.

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CHAPTER 1

INTRODUCTION

1.1 Rationale for the research

Sweet potato (*Ipomoea batatas* L.) is one of the most important root crops globally. It is considered an important, versatile and underutilised food security crop (Kulembeka et al., 2004; Elameen et al., 2008; Lebot, 2009). It represents the second most important set of food crops in developing countries (Lebot, 2009). It is highly nutritive, and it outranks most carbohydrate foods in terms of vitamin, mineral, dietary fibre and protein content (Chattopadhyay et al., 2002; Mukhopadhyay et al., 2011). It is one of the crops that can be used to combat malnutrition in developing countries. A recent report by the FAO (2013) indicated that 24.8% of the population in Sub-Saharan Africa remained undernourished and agricultural projects (crops) that can deliver quick results were needed to meet the millennium goal number 1 (MGD1) targeted for 2015. Sweet potato can be used as a quick turnover crop due to its wide ecological adaptation, drought tolerance and a short maturity period of three to five months (Bovell-Benjamin 2007; Agili et al., 2012). It can also be harvested sequentially thus ensuring continuous food availability and access, an important dimension of food security.

Not until 1986, sweet potato was regarded as a low value, low status, and highly perishable commodity in Africa, especially in the Southern African Development Community (SADC) (Laurie et al., 1999). It received little research attention until the Southern African Root Crops Research Networks (SARRNET) intervened by distributing germplasm and encouraging demand—led research. This initiative prompted the release of several new cultivars within the SADC region. South Africa alone, through the Agricultural Research Council (ARC), has released about 12 cultivars during the period between 1959 and 1989. Laurie et al. (2004) reported that in the 2003/4 season alone, seven new (cream-fleshed) cultivars and one locally bred orange-fleshed cultivar (Serolane) were released for local production in South Africa. Current reports indicate that there are now nine orange-fleshed cultivars (OFVs) in South Africa (Laurie, 2010).

Orange–fleshed (and yellow-fleshed) cultivars have been recognised as good sources of β -carotene, a precursor for vitamin A (Osiru et al., 2009; Mukhopadhyay et al., 2011; Laurie and Van Heerden, 2012) and are thus being promoted across Sub-Saharan Africa. According to Laurie (2010), vitamin A deficiency (VAD) in South Africa is still a serious health problem. Studies by Labadarios et al. (2005) reported that 64% of children between the ages of 1-9 years and 27% of women at reproductive ages were vitamin A deficient in South Africa. The highest prevalence was concentrated in Limpopo (43.5%) and KwaZulu-Natal (38.9%) provinces (Wenhold and Faber, 2008).

1.2 Justification

In South Africa, sweet potato is grown throughout the tropical and warm regions including KwaZulu-Natal (KZN) (Laurie and Niederwieser, 2004; DAFF, 2011). Its consumption in general (irrespective of whether it is orange of white fleshed) is steadily increasing (in South Africa) despite relatively low production levels (DAFF, 2011). Average yields are ten times lower among small-scale farmers than those achieved by commercial growers with access to irrigation, fertilizers, and credit. Laurie and Magoro (2008) had earlier reported mean yields of 3.9 -9.5 t/ha on communal gardens as compared to 25.2 t/ha at experimental stations. This confirmed the huge yield gap that exists between communal farmers and better equipped farmers. The adaptive responses of different sweet potato cultivars to different agroecological areas may vary and moreover, adaptive responses of sweet potato cultivars in KZN's ecological regions are not known. Since sweet potato is considered an important source of food, particularly in the dry season (CIP, 2012b), its importance in improving food security cannot be over-emphasized. However, the possible varying adaptive responses may impact on the food and nutrition security role of sweet potato. Despite the reported low yields among smallholder farmers, the crop has flexible planting and harvesting schedules especially in frost free areas (Mukhopadhyay et al., 2011).

Sweet potato is a drought tolerant crop with potential to enhance food and nutrition security especially in rural South Africa (especially KZN which has the highest concentration of rural setup). Drought stress accounts for 25% total annual yield loss (Placide et al., 2013), compared to > 50% yield loss or complete failure in other crops such as maize under drought stress. KwaZulu-Natal boasts of diverse agro-ecological areas which some of them are prone to drought stress. Despite this advantage its importance as a food security crop in relation to drought is still underestimated and fails to attract sufficient attention from agricultural

researchers. Producing a crop like sweet potato with little loses due to drought stress means it is very capable of ensuring food security during times of drought and in areas that face water scarcity.

1.3 Aims and objectives

KwaZulu-Natal, provides a perfect adaptation assessment for sweet potato cultivars, given the diverse environments, undulating landscape and wide diversity in soil types and fertility. It is hypothesized that sweet potato will adapt in the different agro-ecological areas of KZN and therefore, plants grown in these areas will not differ in growth, yield and nutritional content.

The main aim of the study was to evaluate growth, development, yield and nutritional composition of different sweet potato cultivars in response to a range of climates and soils in KwaZulu-Natal, South Africa.

1.3.1 Specific objectives:

- To evaluate the physiology, growth and yield of different sweet potato cultivars in response to different growing seasons and agro-ecological locations in KwaZulu-Natal (KZN);
- To evaluate the nutritional composition of different sweet potato cultivars grown in different geographical locations and seasons in KZN, and
- To evaluate the growth and physiological responses of three different sweet potato cultivars to water stress.

CHAPTER 2

LITERATURE REVIEW

2.1 Sweet Potato as a traditional crop in Southern Africa

Sweet potato is among the oldest crops in the world, especially in the wet tropics, and was among the first staple crops before the introduction of cereals. Today it is counted among other root crops (cassava, sweet potato, yams and aroids) that represent the second most important set of food crops in developing countries, closely following cereal crops (Lebot, 2009). According to Gush (2003), sweet potato is usually the most misunderstood vegetable, often confused with yams. Portuguese explorers are believed to have introduced sweet potato to Africa in the 16th century (Woolfe, 1992; Allemann et al., 2004; Laurie, 2004; Lebot, 2009). From there it was taken to the eastern and western parts of the world in the form of storage roots by Spanish explorers. It was until it failed to yield in European countries due to low temperatures that it was brought back to Africa and was planted in the warm coastal regions of Africa where it spread rapidly (O'Brian, 1972; Lebot 2009).

According to Laurie (2004; 2010), sweet potato was introduced to South Africa in 1652 around the time when the Dutch, under the leadership of Jan van Riebeeck colonized the Cape. It is now referred to as one of the traditional crops (Laurie and Van Heerden, 2012) and plant of the past (Lebot 2009) because of the time which it arrived in Southern Africa. It has been around for a long time now such that it has become indigenised (Schippers, 2002; 2006) and classified as a neglected and underutilised crop species (Laurie et al., 1999, Ebregt et al., 2007). About 12 cultivars were released by the Agricultural Research Council (ARC) during the period between 1959 and 1989. Seven new (cream-fleshed) cultivars and one locally bred orange-fleshed cultivar (Serolane) were released for local production (in South Africa) during the 2003/4 season (Laurie et al., 2004). According to Laurie (2010), nine of the released cultivars to date, are orange-fleshed (OFVs). Production has also spread throughout the tropical and warm regions of the country and the main producing regions being the Northern Cape, Western Cape, Northeast Limpopo, Free State, Eastern Cape, Gauteng, Mpumalanga Lowveld, and Northern KwaZulu–Natal (Laurie and Niederwieser, 2004; DAFF 2011).

2.2 KwaZulu-Natal environment for sweet potato production

KwaZulu-Natal (KZN) is a subtropical region situated on the east coast of South Africa and boasts of diverse climate, soils and topography, virtually making it a 'world-in-one' (Joubert, 2012). It is a summer rainfall area (600 – 2000 mm/annum) with typical weather that ranges from extremely hot along the coast in summer to heavy snow in the mountainous midlands in winter (The Climate Group, 2004; Southafrica.info, 2012). According to Nqobo and Dladla (2002), KZN has two climate zones of which 44% subtropical and 55% temperate. The kind of environment exhibited in the tropical part of the province is regarded as suitable for sweet potato production throughout the year. Depending on whether farmers produce the OFSVs, the high prevalence of VAD in the province (KZN) could be controlled.

Variations in climatic zones of KZN are bound to bring about several environmental stresses including abiotic and biotic stress conditions. Abiotic stresses have been identified as a major threat to global food security in the 21st century (Battisti and Naylor, 2009). Underutilized crops such as sweet potato tend to possess characteristics adaptable to abiotic stress conditions such as heat and drought which are already causing significant agricultural yield losses. These conditions are predicted to become even more prevalent in the coming decades due to effects of climate change (Ortiz et al., 2008; Battisti and Naylor, 2009; Schulze, 2011). The current and predicted challenges call for an agricultural system that would be able to provide food crops that are capable of meeting current and future food and nutrition security. Such crops will have the effect of increasing resilience and lowering risk in vulnerable communities.

2.3 Importance of sweet potato

The most important plant parts of sweet potato are the starchy roots and immature leaves. These are used for human consumption, animal feed and, to some extent, for industrial purposes (Laurie et al., 2004; Mukhopadhyay et al., 2011). China, the world's leading sweet potato producer uses 40% of its produce for animal feed, while Brazil and Madagascar use 35% and 30%, respectively (Laurie et al., 2004).

Sweet potato is regarded as the most important root crop of the tropics because of its flexibility in a number of production aspects (Mukhopadhyay et al., 1990). It can be planted and harvested at any time of the year, especially in frost free areas. It has a short cropping season, uses non-edible parts as planting material, and has a non-trellising growth habit as

well as low requirement for soil nutrients (Martin, 1985). Woolfe (1992) also reported that it produces more edible energy than any other major food crop. It is more productive within short periods of time on marginal lands and plays an important role in the economy of poor households (Mukhopadhyay et al., 2011)

Laurie (2004) and Manamela (2009) reported that in some African countries, starchy crops such as sweet potato were the staple food while other countries use it as an additional or food security crop. The latter use concurs with earlier reports by Ebregt et al., (2007) that sweet potato storage roots were often kept in the ground and harvested when needed. This is an indication that sweet potato can provide a continuous food supply during the off season with no requirement for expensive storage infrastructure. It becomes important in the diet seasonally, typically in the month or two before the major grain harvest or when the grain stock from the previous year has been exhausted. It also provides a food reserve when the major grain crop fails due to drought and pests infestation.

2.4 Sweet potato utilization

As one of the most nutritious food security crops, both the storage root and leaves are edible to humans. Storage roots can be boiled, baked or roasted with some people preferring to eat them raw. The leaves can also be consumed as a green leafy vegetable (Laurie and Niederwieser, 2004). Sweet potato leaves may possibly have some medicinal properties due to the polyphenol-rich green extracts reported to play a role in reducing prostate cancer (Karna et al., 2011). Ross and Kasum (2002), also reported that flavonoid compounds of fruit and vegetables are considered as therapeutic agents since they have beneficial health effects such as their supposed protection against certain cancers, cardiovascular diseases and aging. This suggests that sweet potato may possibly have some nutraceutical benefits which are still an aspect that need further investigation in South Africa. It would also need to be investigated if and how drought alters these properties since phenols and flavonoids are reported to increase in response to stress.

Reports from China, the leading global sweet potato producer, indicate that they also use sweet potato for starch production and as a raw material for biofuel production (Woolfe, 1992; Liu, 2011). The plant sap (juice) of red sweet potato has also been reported to play a vital role in producing dye for cloth in South America. Another non-nutritive use of sweet potato is its aesthetic value due to its attractive foliage (Anon, 2011).

2.5 Nutritional value

The sweet potato storage root provides a balanced diet on its own. It can provide almost all the nutrients needed by the human body. Woolfe (1992), reported that it contained significant amounts of carbohydrates as compared to other starchy crops such as rice, maize and sorghum porridge, although the protein content is slightly lower than in potatoes and other grain crops. It contains almost all the macro - and micro - nutrients, substantial quantities of vitamin C, moderate amounts of vitamin B complex (Vitamin B1, B2, B5 and B6) and folic acid, as well as satisfactory amounts of vitamin E (Walter et al., 1983; Laurie, 2004).

Orange-fleshed sweet potato has been reported by several authors to contain high amounts of vitamin A and is currently being promoted by the Food and Agriculture Organization (FAO) and other in-country programs as a supplementary food to combat vitamin deficiencies in children (Kulembeka et al., 2004; Ebregt et al., 2007, Amagloh et al., 2011). According to Laurie (2004), South Africa is also using orange-fleshed sweet potato cultivars in food diversification programs to alleviate vitamin A deficiencies. Such programs include school and community garden projects within the integrated nutrition programme (Maduna, 2007; Sikhakhane, 2007).

Sweet potato leaves are also nutritious (Table 2.1) and are a good source of proteins and vitamins A, C, and B₂ (riboflavin). According to Laurie (2004), sweet potato leaf protein content is twice as much as that from the storage roots. Research done by Khachatryan et al., (2003) indicated that the leaves were an excellent source of lutein. Lutein is responsible for central vision in the human eye and helps keep eyes safe from oxidative stress and high-energy photons of blue light. Sweet potato is also popular in low fat diets and is recommended as a low glutamate index (GI) food (Podsedek, 2007). Given the ailing nutritional status of South Africa, plus the fact that other food sources which typically provide these nutrients are not so drought tolerant, challenges their ability to sustainably supply these nutrients. The nutritional content of sweet potato and the role it can play in addressing the current low nutrition status in the South African communities therefore places sweet potato in a very important category of food production.

Table 2.1: Nutritional composition of sweet potato leaves compared to other common vegetables.

	Total	Beta-				Ascorbic
	protein	carotene	Calcium	Iron	Riboflavin	acid
		(μg 100 g ⁻				
Crop	(g 100 g ⁻¹)	1)	(mg 100 g ⁻¹)			
Sweet						
potato				1.8 –		
leaves	29	2700	75 – 183	3.9	0.35	32 - 136
Amaranth	28	6545	176	2.8	0.22	23
Cabbage	1.9	ND	44	0.4	ND	ND
Carrot	0.7	ND	48	0.6	ND	ND
Spinach	3.2	ND	93	3.1	ND	ND
Tomato	0.9	ND	13	0.4	ND	ND
Lettuce	1.0	1000	23	0.9	0.08	15

(Adopted from Woolfe, 1992). ND = No data.

2.5.1 Sweet potato as a food security crop

The world is reported to produce enough food to feed everybody (women, men and children) but there is still approximately 30% of Sub-Saharan Africa's population (218 million people) who suffer from chronic hunger and malnutrition (Mbabu and David, 2012). According to the World Health Organization (WHO, 2013), food security is built upon three pillars; food availability, food access and food use (utilization and stability). In trying to address the food availability pillar of food security, South Africa has identified small scale agricultural production as a potential contributor to food security. This is an area which still needs to be investigated since little effort has been made in channelling investment towards it. Rising food prices, particularly of maize and wheat which are the staple diet for the poor in South Africa, poses serious problems for urban and rural poor people since they are the net buyers of food (Altman et al., 2009).

Sweet potato, on the other hand, is considered an excellent food security crop since it often survives where other crops (e.g. maize) fail (Low et al., 2007). It contributes to food security by improving rural livelihoods (Yngve et al., 2009), especially those involved in small scale

agriculture. It contributes to food availability by providing high output per unit of land (yields about 3.9–9.5 t ha⁻¹ under communal agriculture compared to 3 t ha⁻¹ reported for maize under similar management) and can produce on marginal soils. The fact that it has higher energy (Woolfe, 1992) than maize derivatives, also adds more strength to its potential role as a food security crop. Moreover, the fact that new OFSVs also contribute significantly towards Vitamin A supplementation in rural households where VAD is more prevalent makes it a crop of choice. Good health is considered as an outcome indicator for food utilization in food security measurement (FAO, 2013), and the high nutritional status of sweet potato qualifies the crop as a food security alternative. Improved early maturing cultivars are often ready for harvest in 3 – 5 months and can be harvested as needed over several months (Mbabu and David, 2012). The benefit of harvesting early and consuming over several months implies that there is a quick turn-over and lasting source of food, therefore improved food access, availability and stability. The food security benefits of sweet potato mentioned above can also lead to the crop being removed from the 'underutilized' category since it would have caught the eye of the consumers and will be utilized accordingly.

Accessibility to sweet potato is also simple because the vines and roots can be easily stored or multiplied. In drought prone areas, storage roots can be sprouted to produce vines so that communities have access to planting material. According to Lebot (2009), sweet potato has often been a life-saver; it saved the Japanese nation when typhoons destroyed all their rice fields just before the First World War. During the early 1960s, China was attacked by famine and sweet potato saved millions of the population from starvation. When cassava was attacked and destroyed by an unknown virus in the 1990s, sweet potato provided food for the Ugandan rural communities (CIP, 2012a). This is evidence that sweet potato is indeed a food security crop as referred to by many authors (Ebregt et al., 2007; Amagloh et al., 2011; CIP.2012b).

2.5.2 Sweet potato for combating vitamin A deficiency

Its storage roots (raw and cooked) are highly nutritious (Woolfe, 1992; Kruger et al., 1998; Laurie, 2010). Moreover, orange-fleshed sweet potato is reported to contain high amounts of β -carotene, a precursor for vitamin A. The β -carotene content increases with the depth in orange colour of the sweet potato flesh (Laurie, 2004). Leighton (2007) (cited by Laurie, 2010) went further to test for β -carotene in different South African orange-fleshed sweet

potato cultivars (Table 2.2), and found that, indeed β -carotene concentration varied with depth of colour.

Low et al. (2007) reported that studies conducted in sub-Saharan Africa demonstrated that consumption of boiled orange-fleshed sweet potato improved vitamin A status of children. A separate study by Amagloh et al. (2011) confirmed that orange-fleshed sweet potato was high in vitamin A and could be used as a complementary food in infant feeding. To improve the consumption of this vitamin A rich type of sweet potato, Laurie et al. (2012) further prepared different food types (chips, doughnuts, juice and a green leafy dish from the leaves) from orange-fleshed sweet potato to determine consumer acceptability. They reported that 92% of the consumers liked the colour of the products while 88% indicated that they would buy such products or prepare them at home.

Table 2.2: Concentration of β -carotene on different cultivars of orange-fleshed sweet potato in South Africa.

		μ RAE*/100g	μ RAE/100g	
Cultivar	Flesh colour	(uncooked)	(Boiled root)	
Resisto	Dark orange	1371	1165	
Khano	Dark orange	1170	995	
2001-5-1	Dark orange	983	836	
W-119	Orange	872	741	
Beauregard	Dark orange	804	683	
	Dark orange -			
	Orange - Pale			
1999-1-7	orange	797	677	
	Orange,			
	yellow			
Excel	cortex	433	368	
	Orange with			
	yellow			
Serolane	ring	426	362	
	Orange - Pale			
Impilo	orange	424	360	

(Adopted from Laurie and Magoro, 2008 and Laurie, 2010). RAE: Retinol activity equivalents.

Sweet potato seems to cover a significant proportion of the food security aspect as a crop. The issue of stability and continuous production is still tricky since it depends mainly on prevailing environmental conditions regularly due to climate change. The crop's flexibility to adapt to various environments and its drought tolerance characteristics then needs to be factored in so that continuous supply is not hampered should there be erratic spells of drought or floods during the year.

2.6 Drought and water stress on plants

Drought is a meteorological term commonly defined as a period without significant rainfall. The crop physiologists' understanding of drought refers to when available water in the soil is reduced and atmospheric conditions cause continuous loss of water by transpiration and evaporation (Jaleel et al., 2009). Stress is defined by Gasper et al. (2002) as an alteration in plant physiology caused by factors that disrupt the equilibrium. Strain on the other hand is any physical and chemical change produced by stress. In this regard, drought stress is considered to be a moderate loss of water which leads to stomatal closure and limitation of gas exchange. Desiccation, on the other hand, is much more extensive loss of water which can potentially lead to gross disruption of metabolism and cell structure, eventually leading to the cessation of enzyme catalysed reactions (Smirnoff et al., 2001). It is characterized by reduction of soil water content, diminished leaf water potential and turgor loss, closure of stomata and inhibition of cell division and expansion (Jaleel et al., 2008). According to Glantz et al. (1997), drought is not only the lack of rain, it can also be defined by its impacts such as crop shortages and indirect impact such as food price increases. The authors (Glantz et al., 1997) further explained drought as an exogenous supply side-shock that causes marked declines in agricultural output, export earnings, employment and income levels. It affects all facets of economy due to close sectorial linkages.

The four categories of drought (meteorological, agricultural, hydrological and socio-economic) (Whilhite, 2000) have a direct impact on food security in the sense that they challenge the whole notion of food availability, accessibility, utilisation and stability. Drought single handedly famished South Africa in 1991/92 and to a lesser extent in 1997/98 and transformed agricultural landscapes (Mason and Tyson, 2000). The fact that already, South Africa is a drought prone nation gives reason to the focus on drought. Additionally, climate change predictions that there will be increased frequency and severity of such droughts (Wayne, 2008; Schulze, 2011) gives an even greater impetus to identify crops that are

resilient and can produce under such adverse conditions (Modi and Mabhaudhi, 2013). These crops need not be just drought tolerant, but also nutritious in order to supply families with adequate nutrition during periods of drought; this makes them central to any discussion on food security. It is within this context that sweet potato is being touted as a possible fit. However, before one can successfully promote the crop under these auspices, there is need to generate empirical information detailing the crop's responses to such conditions.

2.6.1 Effect of drought on plant growth

Plant roots are the primary sensors of depleting soil water. They interconnect the physiological and biochemical disturbances in stems and general pant growth to changes in plant nutrition, carbon dioxide balance and photosynthesis, water relations, respiration, translocation, ion uptake, carbohydrates, nutrient metabolism and growth promoters (Jaleel et al., 2009; Alireza et al., 2011). Water stress inhibits cell enlargement more than cell division (Jaleel et al., 2009)

A ramified root system is very important to support above ground dry mass accumulation, but there are observed differences in response to that among plant species. A prolific root system is, according to Jaleel et al. (2009), able to confer the advantage to support accelerated plant growth during early crop growth stages and extract water from shallow soil layers (Blum, 2009) that is otherwise easily lost by evaporation in legumes. Fuentes and Chujoy (2009), reported a similar root system for sweet potato and thus classified it under drought tolerant plants. The ramified root system, however, does not occur at planting as espoused by Jaleel et al. (2009) since sweet potato is vegetatively propagated from vines. Some supplementary irrigation may be required at the time of planting for proper sprouting and establishment (Herzra et al., 2011).

Plant morphological characteristics are affected whereby elongation and expansion growth is impaired (Shao et al., 2008). Studies on potato plants showed a significant reduction in stem length under water stress (Heuer and Nadler, 1995). Similar results were also observed by Specht et al. (2001) on soybean stem length. The inhibition of cell expansion and cell growth is mainly due to low turgor pressure under water stress conditions. Osmotic regulation can enable the maintenance of cell turgor for survival or to assist plant growth under severe drought conditions (Shao et al., 2008). Water stress reduces leaf growth which in turn reduces

leaf area of plants as reported in soybeans (Zhang et al., 2009) and several other plant species studied by Farooq et al. (2009).

Diminished biomass due to water stress was observed in almost all genotypes of sunflower, but some genotypes showed better stress tolerance than others (Tahir et al., 2002). Other plants that showed reduced biomass under water stress were soybeans (Specht et al., 2001), *Poncirus trifoliate* seedlings, common beans and green gram and *Petroselinum crispum* (Jaleel et al., 2009). The fact that water stress inhibits leaf expansion suggests that vine extension and general vegetative growth in sweet potato would be compromised. Since the canopy represents the only source of biomass for subsequent partitioning to the storage root, this would therefore, impose a source limitation to yield attainment.

Studies by Jaleel et al. (2009) indicated that water stress can generally reduce yield in many crop plants but different crops respond differently to water stress. Since the canopy represents the only source of biomass for subsequent partitioning to the storage root in sweet potato (Lebot, 2009; Lewthwaste and Triggs, 2009), if biomass is limited it would consequently impose a source limitation of assimilates to storage roots thus lowering final yield. There was scant literature detailing the extent to which sweet potato yield would be affected if vegetative growth was to be inhibited by water stress. According to Lebot (2009), attempts to demonstrate relationships between yields and particular morphological or physiological characteristics have so far been unsuccessful. However, cultivars with small canopies, short stem length and small leaves were reported by Wilson et al. (1989) to have the capability of giving higher yields than those with long stems and numerous broad leaves. The higher yields in small canopy cultivars might be a result of plants maximising storage root development and expansion over shoot extension. Given that different genotypes will respond differently, there is a need to evaluate available genotypes across the range of climates where sweet potato can be grown. This would allow for the formulation of better recommendations to assist farmers with cultivar selection.

2.6.2 Drought in the context of crop production

Plant roots have been shown to be the main plant part responsible for water uptake. Reduction in uptake and transpiration are usually associated with a reduction in water content of the shoots and stomatal aperture, suggesting that water stress has developed in the leaves (Alireza et al., 2011). Plants exposed to environmental stresses such as drought and heat stress respond

by showing ingenious adaptations at physiological level, sometimes this may be accompanied by changes in various gene expression(Ueda et al., 2001; Alireza et al., 2011). The biological response and tolerance of plants to drought stress such as physiological, biochemical and transgenic basis for drought tolerance need to be understood.

2.6.2.1 Physiological responses of plants to drought and water stress

2.6.2.1.1 Chlorophyll composition

Photosynthetic pigments are important for harvesting light and producing reducing powers in plants. It is also considered as the basis of all crop yields (Jaleel et al., 2009; Mabhaudhi, 2009). Chlorophylls 'a' and 'b' are reported to be susceptible to soil drying (Farooq et al., 2009). Foliar photosynthetic rate of higher plants is known to decrease as relative water content and leaf water potential decrease (Lawlor, 2002). Zulini et al. (2007) indicated that severe levels of drought were capable of causing irreversible damage to photosynthetic apparatus. Lawlor (2002), had earlier highlighted an area of confusion that still needed clarification as to whether drought stress mainly limits photosynthesis through stomatal closure or through metabolic impairment. According to Farooq et al. (2009), both stomatal and non-stomatal limitations were generally accepted to be the main determinants of reduced photosynthesis under drought stress conditions.

Two cultivars of okra responded to water stress by increasing chlorophyll b while chlorophyll a remained unaffected in both lines resulting to a low Chl a:b ratio. Alireza et al. (2011), reported a reduction in chlorophyll content at high water stress. Both chlorophyll a and b, and total chlorophyll were reduced to the lowest amount. This reduction was reported to be associated with an increase in electrolyte leakage caused by leaf senescence and reduced water use in plants.

C4 plants are reported to have developed a drought adaptation strategy to fix carbon dioxide (CO₂) for sugar production with minimum water loss (Xoconostle-Cazares et al., 2011). This is achieved through a particular leaf anatomy where the bundle sheath cells have chloroplasts besides the mesophyll cells just like in C3 metabolism. This allows photorespiration to take place in both the bundle sheath and mesophyll cells. This enables the plants to convert CO₂ to a 4-carbon organic acid which is later regenerated in the chloroplast of the bundle sheath cells. These cells use the CO₂ to synthesize carbohydrates for the plant via the normal C3

pathway. CAM/C4 plats that have adapted C3 mechanism open their stomata during the night when conditions are less stressful since their expense of water is very low as compared to other plants (Zhu et al., 2008; Xoconostle-Cazares et al., 2011). C3 plants of tropical origin such as sweet potato colonize easily in drought environment because they have high phenotypic plasticity. This refers to the ability of the plant to alter its characteristic in order to acclimate to variable environmental conditions (Pigliucci, 2001).

In sweet potato and other C3 plants, all photosynthetic cells are functionally equivalent thus allowing each cell to acclimate to new environments in a more autonomous manner than C4 plants. This allows for photosynthetic plasticity at cellular rather than tissue level thus resulting in greater acclimation ability of C3 plants in general (Sage and McKown, 2006). This is the type of metabolism that allows sweet potato to be classified as a drought tolerant plant. Earlier studies on C3 plants had also indicated that maximum rate of assimilation was strongly temperature dependant than water dependant and considerable photorespiration takes place in sunlit photosynthetic organs. Where light intensity is low, there would be 1% decrease in light use efficiency for every degree of temperature increase (Haxeltine and Prentice, 1996). This report emphasises the importance of light and temperature in sweet potato production. Increases in photosynthetic plasticity and assimilates results in higher yields. This may also explain why sweet potato possesses wide adaptability when compared to other common C4 staple crops such as maize.

Other anatomical adaptation characteristics of drought tolerant plants include the development of spongy tissues that act as water reservoirs. Impaired growth and reduced folia area to limit water loss through transpiration (Chaves et al., 2002; Jaleel et al., 2009). Leaf rolling, floral abscission and cuticle permeability (Taiz and Zeiger, 1998). Some plants may have floral induction which is associated with long-distance movement of flowering Locus T (FT) proteins (Lin et al., 2007; Xoconostle-Cazares at al., 2011)a trait observed in sweet potato plants grown under water stress (Lebot, 2009).

2.6.2.1.2 Biochemical Responses to drought and water stress

According to Xoconostle-Cázares et al. (2011), plant adaptive responses to drought and water stress are based on complex changes mainly focused to maintain water potential in key tissues. Earlier reports by Bartels and Sunkar (2005) had indicated osmotic adjustment as a common biochemical adaptation strategy in plants since it results in newly-synthesized metabolites. These metabolites include hydrophilic highly soluble molecules that are able to produce a salvation surface that captures water molecules to be later available during water limitations. Examples of these osmolytes are proteins (amino acids), sugars and sugar alcohols. According to Chen and Murata (2002), some of the osmolytes have additional functions such as coping with oxidative stress by suppressing reactive oxygen species. This section will only discuss protein synthesis and selected phytochemicals produced by plants as a response mechanism to drought stress.

2.6.2.1.3 Protein synthesis

Protein expression, accumulation and synthesis is modified in plants exposed to drought stress (Cheng et al., 2004). According to Bray (2002), these drought-induced proteins are important for physiological adaptation of plants to water stress. The drought-induced proteins were earlier referred to as dehydrins and belong to the group II late embryogenesis abundant (LEA) proteins (Close et al., 1993). Studies performed on plants (wild watermelon and maize) under mild water stress reported more than sixteen proteins involved in water stress responses (Riccardi et al., 1998; Kawasaki et al., 2005). Among the proteins identified in wild watermelon was citrulline, a scavenger of hydroxyl radicals (Kawasaki et al., 2005). Studies on mild drought stress in rice leaves showed that drought-induced changes in about 42 proteins were reversed completely within ten days of re-watering. Of the 42 proteins identified in leaves of water stressed rice there were those involved in basic metabolic pathways such as ATP production, photosynthesis, protein synthesis, oxidative stress tolerance and cytoskeleton reorganisation (Selekdeh et al., 2002).

Proline is one of the proteinogenic amino acid with exceptional conformational rigidity and is essential for primary metabolism in plants (Szabados and Savoure, 2009). It is involved in the synthesis of proteins that are necessary for stress response (Khedr et al., 2003). This amino acid is of particular interest in stressed plants and is usually considered as an osmoprotectant

and accumulates in cells to obtain suitable conditions for water uptake under limited water conditions in the roots. It is also involved in reducing oxidative damage by scavenging and/or reducing free radicals (Ozgur and Mithat, 2008). Findings by Vendruscolo et al. (2007) indicated that proline was involved in tolerance mechanisms against oxidative stress and that this was the main strategy by which plants avoided detrimental effects of water stress. This contradicted earlier reports by Zlatev and Stoyanov (2005) who suggested that proline accumulation could only be useful as a possible drought injury sensor instead of its role in stress tolerance mechanism. A study conducted on wheat to find out the influence of water stress on proline accumulation concluded that increases in proline levels helped protect cell membranes from oxidation instead of osmotic regulation as an initial response to water stress (Ozgur and Mithat, 2008).

According to Szabados and Savoure (2009), proline accumulation can influence stress tolerance in multiple ways since it functions as a molecular chaperone able to protect protein integrity and enhance the activities of different enzymes. While there are two schools of thoughts in as far as proline accumulation is concerned, the bottom line is that it will accumulate in response to environmental stresses. Whether or not its accumulation is a sign of stress tolerance or rather a symptom of the severity of the stress is still debatable. This is an avenue that still needs to be explored especially in food security crops like sweet potato. Modi and Mabhaudhi (2013) suggested that, despite the controversy surrounding its exact role, proline accumulation could still be a useful index in drought tolerance. However, they went on to state that its value as an index could only be fully understood if other stress metabolites were also quantified alongside it.

2.6.2.1.4 Selected phytochemical responses

Phytochemicals are secondary metabolites that are widespread in the plant kingdom (Rispail et al., 2005). They protect plants against bacteria, viruses and fungi invasion (Islam, 2006). The different compounds constituting phytochemicals are considered to have biological significance but are not established as essential nutrients in human or animal diets (Bacchetti et al., 2013). They have been however, reported to possess bioactive properties with potential health benefits (Okarter and Liu, 2010). Phytochemicals such as antioxidants, including carotenoids, anthocyanins and flavonoids can be classified into different categories depending

on their structure and have been associated with flavour and colour characteristics of fruits and vegetables (Podsedek, 2007; Jahangir et al., 2009). Antioxidants such as ascorbic acid and glutathione commonly found in chloroplasts and other cellular compartments in plants are very crucial for plant defence against oxidative stress and have been reported to be indicators of plants' hypersensitivity to stress conditions (Mittler, 2002). Earlier findings by Sairam and Saxena (2000) had indicated that enzymatic and non-enzymatic antioxidants in plant cells played an important role in avoiding detrimental effects of free radicals. In addition to their high antioxidant and antiradical activities in humans, the health beneficial properties have also been attributed to many other mechanisms such as anti-inflammatory properties, inhibition of enzymes and induction of detoxification enzymes (Lopez-Martinez et al., 2011). Podsedek (2007) also reported antioxidants as vital in protection against cancer and cardiovascular diseases in humans.

Carotenoids are plant pigments that can partially help plants withstand adversities of drought (Jaleel et al., 2009). They form a key part of the plant's antioxidant defence system (Wahid, 2007; Farooq et al., 2009). Carotenoids are divided into carotenes and xanthophylls (Jaleel et al., 2008). Beta-carotene is present in all chloroplasts of green plants and is exclusively bound to the complexes of photosystems one (PSI) and two (PSII). It functions as an accessory pigment and as an effective antioxidant and plays a unique role in protecting and sustaining photochemical processes (Jaleel et al., 2009). Farooq et al. (2009) identified the protective role of β -carotene in photosynthetic tissue as achieved through direct quenching of triplet chlorophyll thus preventing the generation of singlet oxygen and protection from oxidative damage.

As a phytochemical, carotenoids were reported to protect against eye disease of macular degeneration (Mozaffarieh et al., 2003). Beta-carotene is of particular importance because it is a precursor for vitamin A (Leigton, 2007). Vitamin A is important in human nutrition because of the role it plays in improving vision, bone growth, reproduction, maintenance of epithelia and overall growth (Tee, 1995). The OFV of sweet potato contain high amounts of β -carotene and are being promoted in vitamin A deficiency alleviation programmes in South Africa and other developing countries (Kaguongo et al., 2012; Laurie et al., 2012). The fact that sweet potato can synthesize β -carotene to protect itself from photosensitized oxidation and further store the β -carotene in both leaves and storage roots is an indication that it is an important

food security crop which can withstand drought stress conditions and still alleviate chronic malnutrition diseases associated with vitamin A deficiencies.

2.6.2.2 Genes expressed during drought stress

A lot of responses to water stress are controlled by a number of genes with different functions. Many regulatory processes are initiated the minute water is lost from plant cells, to help adjust cellular metabolism. These adjustments will result in changes of gene expression (Bray, 2002). Changes in expression patterns have been monitored on the plant when drought stress is perceived. These changes range from genes whose products are involved in early responses such as signal transduction, transcription and translation factors; to late response genes, such as water transport, osmotic balance, oxidative stress and damage repair (Xoconostle-Cazares et al., 2011). Dure (1989) had earlier indicated that these genes were mainly located in the cytoplasm. Whether these water-stress-induced genes perform an adaptive role or not is still to be investigated.

Genetic engineers have tried to develop transgenic sweet potato plants by gene coding for sperimidine synthase which is used to improve environmental stress. The transgenic sweet potato from this genetic coding showed high tolerance to drought, salt, chilling and heat stresses (Kasukabe et al., 2006). Recent reports by Fan et al. (2012) indicate that transgenic sweet potato containing the gene from *Spinacia oleracea* (betaine aldehyde dehydrogenase) which is important for osmoregulation and abiotic stress have showed tolerance to multiple environmental stresses such as protection against cell damage, improved photosynthetic activity and increased activity of free radical scavenging enzymes. These reports are a proof of good progress in genetic engineering in as far as sweet potato drought tolerance is concerned but there is still a shortage of successful screening methods and multidisciplinary approaches and genotype by environment interaction (Placide et al., 2013). It is therefore believed that with the existence of these strength-enhanced drought tolerant cultivars, food security will improve as long as the genotype by environment interaction selection is carried out in the environment in which the cultivars will be released and grown (Mwanga et al., 2007).

2.7 Agronomic practices associated with sweet potato production

Smallholder production of sweet potato is commonly practised on marginal lands with limited inputs. Thus, yields are lower than the potential of the crop especially in developing countries (Ebregt et al., 2007). According to Lebot (2009), yields can be increased by adopting suitable cultivation techniques, selecting sites with high sunlight, an average temperature of at least 25°C, receiving a well–distributed annual rainfall (1000 – 2000 mm) and selecting good sweet potato cultivars (Woolfe, 1992; Laurie and Niederwieser, 2004).

2.7.1 Environmental requirements

Adequate water supply through rainfall or irrigation is essential for the optimum development of storage roots but excess water on the other hand results in poor aeration. Water shortage at 50-60 days after planting, when roots begin to bulk, was reported by Mohankumar (2000) to drastically reduce sweet potato yields. According to Woolfe (1992), sweet potato yields relatively better in tropical areas as compared to other crops. Growth and development takes place at temperatures not lower than 17.5° C. At temperatures below 15° C, little growth occurs and at below 10° C growth is severely retarded (Laurie and Niederwieser, 2004).

Temperature of the soil also affects storage root development, storage root development occurs when soil temperatures reaches 25°C, while temperatures of 15°C or 35°C inhibit their development (DAFF, 2011). Lebot (2009) further emphasized that sweet potato produces high storage root mass when grown over a constant soil temperature of 30°C, combined with an air temperature of 25°C during the night. Frost occurrence destroys the above ground material completely (Mukhopadhyay et al., 2011). According to Ngeve (1993), frost damage of sweet potato restricts production to areas with minimum frost-free period of 4 to 6 months in the temperatures should be relatively high during much of the growing period.

Solar radiation also plays a vital role in plant development and yield, it has an effect on flowering, storage root formation and yield (Mortely et al., 1990). Sweet potato is a heliophylous, sun-loving species. Lebot (2009) reported that it grows well under high light intensities and can only grow under low light intensity while still attached to the storage root. Experiments by Burrell et al. (1993) had shown that a combination of longer photoperiod and

low light intensity increased sweet potato yield more than a combination of shorter photoperiod and high light intensity. This response was further explained by the authors as non-genotype-dependant. Field experiments in lowland tropics also concurred with the findings of Burrell et al. (1993) where sweet potato was reported to give higher yields during the short days of the cool season than during the long hot days of the summer season (Lebot, 2009). Carvalho et al. (2010) studied photoperiod on leaf colour and flavonoids content and their results indicated that plants grown under long day conditions displayed enhanced red colour compared to yellow/green leaves when plants were grown under short day photoperiod; this accounted for the high flavonoid content in plants grown under long day conditions.

The probability of getting the environmental requirements suitable for sweet potato production outlined in this section in KZN province is relatively high. According to Smith (2006), there are 23 identified vegetation and climate types (bioresource groups) in the KZN province, George26% of the bioresource groups are located along the coastline where it is warm enough (annual average of 21°C) with good amount of sunshine (light intensity and photoperiod)to sustain sweet potato growth throughout the year. The remaining 74% of the province is the inland region which is characterised by hot and humid summers and mild winters. This part of the province can sustain sweet potato growth only during the summer season (October – April) (The Climate Group, 2004; Southafrica.info, 2012). The province also receives an average annual rainfall of 1000 mm (Southafrica.info, 2012) which is twice the annual average (500 mm) of the country. This puts it in a good position to support rain-fed sweet potato cultivation. The potential for improving food security through sweet potato availability and access throughout the year in this province is promising, given these environmental conditions.

2.7.2 Soil and soil nutrient requirements

Sweet potato is generally not a very demanding crop with regards to soil type (Mukhtar et al., 2010). However, soil structure plays an important role on quality and appearance of storage roots (Van den Berg and Laurie, 2004). For good storage root quality and appearance, it was recommended that sweet potato should be grown on sandy-loam, loam and clayey-loam soils. Heavy clay soils were reported to restrict storage root development causing them to become long and cylindrical, clay soils may also cause poor aeration and lifting problems at

harvesting (Mukhopadhyay et al., 2011). Plants grown in waterlogged conditions such as clay soils also fail to produce storage roots probably due to inadequate oxygen within the root zone (Lebot, 2009). Clayey soils and waterlogged conditions increases as you move into the midlands of KZN. Farmers located in such soils conditions will need to improve soil aeration by adding organic matter and improving drainage. While drought tolerance in sweet potato plants has been reported (Hartemink, 2003; Laurie, 2004; Lin et al., 2007; Kaguongo et al., 2012; Iheagwara, 2013), flooding tolerance is yet to be reported. Under water logging and flooding conditions, yield may be improved remarkably by ridging or planting in moulds (Lebot, 2009; Githunguri et al., 2006).

Although sweet potato is often described as adapted to poor soils (Belehu, 2003), Hartemink (2003) reported that sandy-loam soils resulted in unfavourable growth conditions for storage root production if nitrogen (N) application was delayed. According to Mukhtar et al. (2010), sweet potato readily produces adventitious roots and colonizes in marginal soils. The colonization is due to the fact that it can fix atmospheric nitrogen (N₂) through association with symbiotic non-nodulating bacteria in the soil (Hartemink, 2003). This symbiotic association of sweet potato and the non-nodulating bacteria might probably be the reason why storage root yields are sometimes depressed in fertile or heavily fertilized soils (Belehu, 2003). Nevertheless, potassium (K) nutrition is required in higher quantities compared to the other two main nutrients (N and P) (George et al., 2002; Belehu, 2003). Potassium is the main nutrient that influences storage root yield by increasing photosynthetic efficiency, leaf area duration, suppressing leaf growth and decreasing stem yield. Uwah et al. (2013), also reported high yield responses of sweet potato to K application in continuously (sweet potato) cropped land.

Soils in KZN province are mostly deficient of P, while levels of K may vary from low to very high. Parts of northern KZN (Makhathini and Tugela valley) contain enough K for good crop growth and areas with low levels of K in the province are associated with high rainfall (KZNDAE, 2013). The K status of KZN soils is an added advantage to sweet potato production since K is the main nutrient influencing yield. Moreover, the fact that sweet potato has a low N requirement, which is usually the limiting nutrient in crop production, makes it very suitable for low input agricultural systems practised by local subsistence farmers. According to Belehu (2003), sweet potato farmers generally would not apply fertilizer under

smallholder or subsistence production systems. The two main reasons for that practice being: (i) the response of sweet potato cultivars to different types of fertilizers has not been clearly established, and (ii) the crop is sold at a relatively low price such that it cannot recover the costs of fertilizers. Sensory evaluation studies by Ossom et al. (2011), further indicated that storage roots fertilized with inorganic fertilizer were the least preferred by evaluators.

Sweet potato is vegetatively propagated. Stem cuttings (30 to 40 cm long) from the tip of the vine are the best planting material. Cuttings from the middle and the base of the vine can also be used, but they will often produce lower yields. Cuttings can be taken from an established crop or from nurseries specifically prepared for propagule preservation (Lebot, 2009). Sweet potato is usually grown in pure stands but can be intercropped with beans and maize under land population pressure (Ebregt et al., 2007; Lebot, 2009). Lack of appropriate agronomic practices such as plant spacing, land preparation (ridging) and difficulty in management activities for different cultivars (long and short vines) was reported as the underlying problem affecting sweet potato yield in Ethiopia (Teshome et al., 2011).

According to Lebot (2009), sweet potato requires little field management once it has established; this eliminates repeated weeding as is the case with other staple crops. This bodes well for subsistence farming systems where labour is usually a limiting factor. Occasional hilling-up is still necessary for storage root expansion. The KwaZulu–Natal province through the Department of Agriculture and Environmental Affairs (KZNDAE) provides support through extension work and publication of information on basic agronomy of different crops. Sweet potato is among those crops that are promoted and information on basic agronomy and management is being provided (KZNDAE, 2013). However, there is a gap in accessing and utilising this information by small scale farmers. Suitability of existing sweet potato cultivars to specific environments in KZN is unknown.

2.7.3 Planting dates

Seasonal planting is practised in temperate areas while the opposite is true in the sub-tropical and tropical areas (Belehu, 2003; Lebot, 2009). It is possible to grow two crops of sweet potato per year in the humid tropics (Belehu, 2003). In the Wolaita area of Ethiopia sweet potato is grown in the two cropping seasons called Belg and Meher. Belg is the short rainy season while Meher is the long rainy season (Getahun, 1993). Given the climatic conditions

of KZN, it might be possible to grow two crops of sweet potato along the coastline where temperatures do not drop beyond 20°C. However, unlike Ethiopia, KZN does not have a bimodal rainfall season hence issues related to drought tolerance in sweet potato become central. In the midlands of KZN, only one crop can be grown during the summer season since there are significant decreases in temperature during winter season.

Planting dates of crops in general can change over time due to climate change and variability as well as changes in technological and socio-economic factors (Kucharik, 2006). According to Sacks et al. (2010), planting dates are governed by temperature changes and seasonality of precipitation in some regions. It was, however, assumed that a temperature limitation takes priority since farmers have more means to alleviate precipitation limitation (e.g. irrigation) than temperature limitations. However, in practice, most subsistence farmers do not have access to irrigation and water is also a limiting factor in the areas where they reside. Bondeau et al. (2007) had earlier found that planting decisions were often driven by factors that were much more complex than those assumed in existing crop models.

Summer rains seems to be the main determinant of planting dates in KZN especially in the midlands where temperatures become very low in winter. Summer planting tends to be the norm among small scale farmers since they are highly dependent on rain-fed agriculture. This is a situation that highlights a gap among local farmers on how to select suitable planting dates that can allow for sweet potato harvesting throughout the year. What is evident is that in rain–fed production systems planting date selection is often mostly influenced by rainfall. Farmers will often want to plant in late spring before the onset of the rains or after the first rains. Their planting date selection, to a limited extent, is also influenced by trying to avoid known dry spells during the season. Already, climate change is resulting in seasonal shifts, and farmers' inability to respond has resulted in crop failures (Mabhaudhi, 2012). This further highlights the need for drought tolerant germplasm to be made available to farmers in order to guarantee yields and food security.

Sacks et al. (2010) suggested that planting dates may be chosen to ensure a favourable climate later in the growing season, such as during flowering, than to ensure an optimal climate early in the crop's growth; for example, if there is potential for drought stress during the summer season. For example, Ekanayake et al. (1990) had earlier reported that formation of sweet potato roots was stimulated by drought stress occurring sometime during the growth cycle, so aligning planting dates in such a way that the mid-summer drought stress coincides with that

particular growth stage would increase sweet potato yields. Mohankumar (2000) suggested that deficit irrigation strategies such as irrigating sweet potato plants once they had depleted about 40% of available soil water tended to give higher yields. Withholding water until 60 days after planting (DAP) was also recommended in order to provide translocation of more photosynthates to storage roots and the development of storage roots (Ekanayeke et al., 1990).

Other researchers have also used heat units for crops planted on different dates during the season or under different microclimates (Wolfe et al., 1989). The use of growing degree days (GDD) in combination with climate-based predictor variables measured within 20 days after transplanting was studied on sweet potato and it provided high predictive accuracy for harvesting dates (Villordon et al., 2009). Knowing when to harvest can also play a vital role in improving food security since farmers will be in a position to know the waiting period before food supply is increased. That information can help in food budgeting.

2.8 Crop management for drought mitigation

Increased productivity in rain-fed agriculture reduces pressure on the limited arable land and water resources (Lansigan, 2002). However, due to climate change variability, water availability for agriculture in terms of its temporal and spatial distribution has become extremely vulnerable. This is continuously threatening human livelihoods and environmental systems including rain-fed agriculture in many tropical regions (Lansigan, 2002; Mall et al., 2006). Das (2005), suggested two distinct phases in which the application of the knowledge of weather and climate can reduce the impact of drought on communities. The first is long term planning in which strategies can be devised, and precautions taken to reduce impact.

Climate risk and vulnerability of rain-fed crop production systems to climate variability and change can now be evaluated using scientific systems research tools such as simulation models, optimization techniques, geographic information systems and use of databases (Smith, 2000; Lansigan, 2002) Combined use of simulation models with temperature gradient tunnel (TGT) experiments have greatly facilitated better understanding of the effects of change in the climatic environments on crop growth, development and crop yield (Horie et al., 1995). Therefore, according to Rosenzweng and Tubiello (2007), farmers may successfully adapt to these changes in climates by applying a cultivar of agronomic techniques that already

work well under current climates. These techniques including adjusting planting dates (or timing of planting) and harvesting, substituting cultivars and, where necessary, farmers can modify or change their cropping system altogether. In the case of sweet potato production in KZN, adjusting planting dates so as to allow yield maximization could be a suitable management strategy. The ARC has also created models for sweet potato and other crops. These models still need to be promoted and utilised in a way that will allow farmers to benefit from their capacity.

The second phase which the application of the knowledge of weather and climate can reduce the impact of drought is the action taken during the onset of the event to reduce adverse effects (Das, 2005). This may include, but not limited to, adopting proper crop management strategies such as water conservation strategies, manipulation of plant population, water recycling, rainfall harvesting into ponds or village tanks and even mid-season corrections (Mekhora, 2000; Das, 2005). Farooq et al. (2009) further recommended adoption of strategies such as mass screening and breeding, marker-assisted selection and exogenous application of hormones and osmoprotectants to seed or growing plants, as well as engineering for drought tolerance.

Adaptation to future changes which are likely to increase with regard to climate change will require farmers and the whole agriculture sector to pay more attention to stability and resilience of production (Rosenzweig and Tubiello, 2007). Sweet potato is known to be one of the resilient crops (Oggema et al., 2007; Agili et al., 2012; Iheagwara. 2013) when compared with the staple crop (maize) in the country. Introducing and promoting this crop would mitigate drought in terms of guaranteeing yield when the main crop fails. Crop management and cropping systems have also evolved to provide stability of production and thus steady income in situations of uncertain weather. According to Lal et al. (1999), positive manipulation in soil management increases the equilibrium soil carbon pool by increasing carbon inputs into the soil or by slowing decay rates of soil organic matter.

Conservation tillage is another practice aimed at enhancing sustainability and resilience of agriculture systems (Fischer et al., 2001). Other best practices include cover crops, nitrogen fixing crops, judicious use of fertilizers and organic amendments and improved cultivars with

high biomass production (Rosenzweig and Tubiello, 2007). Sweet potato crop management matches all the characteristics of best practice as explained in the agronomy section of this review.

The two phases suggested by Das (2005), of using weather and climate to mitigate and plan for drought; adaptation of agriculture to stability and resilience of production (Rosenzweig and Tubiello, 2007) and conservation tillage and other best practices all form part of climate-smart agriculture (CSA). Climate-smart agriculture was defined by FAO as an approach contributing to the achievement of sustainable development goals. It integrates the three dimensions of sustainable development (economic, social and environmental) by jointly addressing food security and climate challenges (FAO, 2014). According to DAFF (2014), CSA seeks to increase productivity in an environmentally and socially sustainable way, strengthen farmer's resilience to climate change, and reduce agriculture's contribution to climate change by reducing greenhouse gas emissions and increasing carbon storage of farmland.

2.9 Conclusions

Sweet potato remains an important root crop with an inherent ability to produce more edible energy than most major food crops. It is suitable for production on marginal lands and can play an important role in the economy of rural households as a food security crop. The importance of orange-fleshed sweet potato cultivars (OFSV) in supplementing vitamin A deficiency cannot be over emphasized. Production of the OFSVs in areas with prevalence of vitamin A deficiency, like KwaZulu–Natal should be promoted. This is the case in KwaZulu–Natal whereby some parts of the province can even support crop production throughout the year. This will strengthen food security in the province.

Rain-fed crops are always subjected to water stress during the course of growth due to the unpredictability of rainfall in subtropical environments. Other abiotic stresses associated with drought cannot be excluded as contributing factors to the decline in crop productivity. These conditions affect plant physiology and consequently dry matter synthesis and partitioning. Plants are reported to respond differently to such conditions, some tend to synthesise particular nutrients and elements at the expense of the other and sweet potato is one of the crops that produces a number of secondary metabolites to help in drought stress adaptation. It has proven to be a relatively resilient crop in as far as stressful conditions are concerned. Its management involves several practices that can enhance the sustainability of agricultural systems, and thus could be used in drought mitigation and food security programmes in KZN and other sub-tropical areas.

2.10 Structure of the experimentation part of the thesis

Field and controlled environment experiments were conducted during seasons 2012/13 and 2013/14. Metabolic analyses of sweet potato plant material were also conducted. The experimentation part of the thesis is reported in four (4) research chapters. An over-arching general discussion is provided for the research chapters as well as over-arching conclusions and recommendations:

Chapter 3 reports on field trials conducted at three locations (Deepdale, Umbumbulu and Richards Bay) across KwaZulu-Natal each located in a different bioresource/ agro-ecological area. It answers the objective about adaptability of sweet potato cultivars to different environmental areas under low-input agricultural systems. It describes results of field trials such as stomatal conductance, chlorophyll content index, vine length, leaf number, number of branches, yield and yield components of the three sweet potato cultivars used in the study.

Chapter 4 reports on results of experiments conducted under controlled environment to evaluate growth, physiological response and yield of sweet potato cultivars under simulated water stress. This experiment was linked to the first objective of the study. It also reports on stomatal conductance, chlorophyll content index, relative water content, vine length, leaf number, and number of branches, yield and yield components of the sweet potato cultivars.

Chapter 5 reports on the nutritional content of sweet potato cultivars when grown in different bioresource/agro-ecological areas of KZN. It reports on starch content, β -carotene content and antioxidant activity in the storage roots. It also reports on carotenoids and chlorophyll content and antioxidant activity of the leaves.

Chapter 6 reports on phytochemical content of sequentially harvested leaves of sweet potato plants produced in experiments explained under chapter 4. It addresses the same objective as chapter 5, but focuses on sweet potato leaves. Phytochemicals measured were carotenoids content, antioxidant activity, chlorophyll content and proline content.

The general discussion forms the last and final chapter of the study. It provides a holistic discussion, encompassing all the separate studies reported in this thesis. It also highlights on major findings, outcomes and implications of the study. This section winds up with concluding statements to the thesis as well as recommendations for future studies.

References from all chapters are listed at the end of the thesis.

CHAPTER 3

GROWTH, PHYSIOLOGICAL AND YIELD RESPONSES OF SWEET POTATO CULTIVARS TO DIFFERENT NATURAL ENVIRONMENTS

Abstract

The need to address food insecurity, in terms of energy and micronutrient deficiencies, has led to the development of new sweet potato cultivars. These cultivars have not been widely tested for production by target communities who practise smallholder farming. The objective of this study was to evaluate the physiology, growth and yield of three sweet potato cultivars (A40, A45 and 199062.1) under low-input agricultural systems in response to different seasons (winter and summer) and agro-ecological areas/bioresource groups (BRGs) of KwaZulu-Natal (KZN). Field planting trials were done at three locations located indifferent BRGs of KZN (Deepdale, Richards Bay and Umbumbulu) during winter and summer seasons over two planting seasons (2012/13 and 2013/14). At each location, the experimental design was a one way experiment arranged in a randomized complete block design with three replications. Data collected included stomatal conductance (SC), chlorophyll content index (CCI), vine length, leaf and branch number, yield (biomass and storage root) and harvest index. The sweet potato plants were able to survive and grew in winter at Richards Bay and Umbumbulu while in Deepdale the crop failed. Growth (vine length, leaf and branch number) was stunted during winter. Winter yield (0.51 t ha⁻¹) was only recorded in Richards Bay during the 2012/13 planting season. Summer trials showed that sweet potato planted at Richards Bay had low (P ≤ 0.05) SC and CCI as well as stunted growth. Richards Bay is characterised by high evapotranspiration (ETo), high temperatures and sandy soils; this exposed the crop to water stress. At Deepdale and Umbumbulu sweet potato showed better ($P \le 0.05$) growth. Water stress was also less pronounced at these locations. For the summer trials, Richards Bay recorded low (P \leq 0.05) storage root yield (5.4 and 5.0 t ha⁻¹) across both planting seasons, while Deepdale recorded higher yields (42.0 t ha⁻¹) during 2012/13; yields were 67% lower (13.6 t ha⁻¹) during the 2013/14 planting season. Storage root yields from Umbumbulu were stable across both planting seasons (29.4 and 28 t ha⁻¹ during 2012/13 and 2013/14, respectively). Adding fertilizer only improved storage root yield in Richards Bay. The A45 orange-fleshed sweet potato cultivar showed good environmental plasticity. This indicated its suitability for use in food security programmes under low-input agricultural systems.

Keywords: locations, low-input, planting seasons

3.1 Introduction

Sweet potato is an important tropical root crop (Bovell-Benjamin, 2007; Iheagwara, 2013) and accounts for about one-third of root and tuber crop production in developing countries (ENV/JM/MONO, 2010). In Sub-Saharan Africa, its cultivation is predominantly done by resource-constrained farmers(Agili et al. 2012), mainly women (CIP, 1985; Ebregt et al., 2007) in small plots for subsistence and food security(high caloric content and vitamin A nutrition) (Laurie et al., 2012). Here, sweet potato is the third most important root crop after cassava and yam (Ewell and Mutuura, 1994). Previously, sweet potato, although commonly referred to as the 'poor man's crop', has received little research attention. The growing demand for food security crops and sweet potato's adaptability to a wide range of environmental conditions (Oggema et al., 2007; Agili et al., 2012; Iheagwara, 2013) have since created renewed attention on the crop (Agili et al., 2012).

The KwaZulu-Natal (KZN) province of South Africa is a subtropical region situated on the east coast of the country, with summer rainfall of 600 - 2 000 mm/annum. The weather ranges from extremely hot summers along the coast to heavy snow in the mountains in winter. It is a 'world-in-one' (Joubert, 2012) with diverse climate, soils and topography. The midlands of KZN are drier than the coast and can be very cold in winter (The climate group 2004; SouthAfrica.info 2012). Although sweet potato is reported to have wide adaptability to a range of environments, several reports also suggest that it may be sensitive to varying environmental conditions (Abidin et al., 2005). In Uganda, Osiru et al. (2009) and Mcharo et al. (2001) reported variations in yield of sweet potato genotypes within each environment and season. In Ethiopia, Teshome et al. (2011) reported that lack of appropriate agronomic practices among smallholder farmers also affected sweet potato yield. These reports suggest that a combination of environmental conditions and agronomic practices limits sweet potato yields. There have been limited studies evaluating suitable cultivars for the varying environmental conditions in KZN. In addition, it seems no studies have been conducted to identify suitable agronomic practices (especially fertilizer application) for these different environments. This may result in farmers cultivating sweet potato cultivars that are not suited to their particular environments.

The contribution of agricultural production to household nutritional status in KwaZulu-Natal indicated that among other crops produced by subsistence farmers, sweet potato (irrespective of the flesh colour) was only produced by 9.8% of the sample population (Kristen et al.,

1998). As a strategy to promote sweet potato production, especially the orange-fleshed cultivars, Laurie and Magoro (2008) conducted participatory sweet potato cultivar selection trials for low input agricultural systems in KZN and other parts of the country. Their aim was to evaluate adaptability and acceptability of the crop, especially the orange-fleshed sweet potato cultivars (OFSV). Orange-fleshed sweet potato cultivars are among the few crops that are rich in beta-carotene and can contribute to improved nutritional status of poor communities (Laurie and van Heerden, 2012). These authors (Laurie and van Heerden, 2012) also reported good acceptance of OFSV by farmers despite it being a new crop. The renewed attention on sweet potato has led to new cultivars being developed, with particular interest on the OFSVs. Desclaux (2005) reported that very few of the new cultivars usually meet the needs of smallholder farmers because they are mostly dependent on rainfall for irrigation and practice low input agriculture. This type of agriculture is usually accompanied by shortcomings such as poor soil fertility and ineffective crop management (Dawson et al., 2007; Laurie and Magoro, 2008; Teshome et al., 2011). It was however reported that low input systems may have high soil fertility and high yields due to other cultural practices such as crop rotation, fallowing and green manures (Dawson et al., 2007). Involvement of smallholder farmers in varietal selection was reported to improve their livelihoods (Desclaux, 2005).

Uptake of these cultivars has not been widespread across KZN. This may be because the involvement of local communities and their crop management knowledge have previously not been given much consideration in evaluation trials. Consequently, there has been a lack of ownership with regard to uptake of new cultivars. It was hypothesised that involvement of local communities and utilization of their local knowledge in evaluations of locally bred sweet potato cultivars would improve acceptability and production of the crop by the communities. Therefore, the objective of this study was to evaluate the physiology, growth and yield of different sweet potato cultivars grown under low-input cropping system in response to different growing seasons and bioresource groups/ agro-ecological locations of KwaZulu-Natal (KZN).

3.2 Materials and methods

3.2.1 Planting material

Three sweet potato cultivars (A40, A45 and 199062.1) were sourced from the University of KwaZulu-Natal's (UKZN) Plant Breeding Department. Two of them (A40 and A45) were

locally bred at UKZN while the third (199062.1) was originally obtained from the International Potato Centre (CIP) and multiplied at UKZN. Two of the sweet potato cultivars (A45 and 199062.1) were orange-fleshed, while the third (A40) was white-fleshed. Planting vines (30 cm long) were cut from the tip of mother plants which were raised in a typical warm subtropical nursery at UKZN. Planting vines were defoliated to one top fully expanded leaf to reduce photosynthetic demand during crop establishment.

3.3.2 Description of experimental sites

The study was carried out on three small-scale farms located in three different locations (Deepdale, Umbumbulu and Richards Bay) of KwaZulu-Natal (KZN), South Africa. The three locations selected for the study were representative of three distinct agro-ecologies commonly referred to as bio-resource groups (BRGs) of KZN (Table 3.1). Bio-resource groups are vegetation types primarily based on climate and vegetation using dominant indicator species (Smith, 2006).

3.3.3 Experimental designs

Field trials were conducted at three locations (Deepdale, Umbumbulu and Richards Bay) during the winter and summer seasons spanning 2012/13 and 2013/14. Winter season is between June, July and August while summer season is during December, January and February. For both seasons, experiments were planned such that data collection coincided with the actual months of the targeted season. Winter planting date was in April, while summer planting was in November. Winter and summer trials were repeated twice (2012/13 and 2013/14).

Each location had a one way experiment evaluating three sweet potato cultivar/cultivars (A40, A45 and 199062.1). The experiments were laid out in a randomised complete block design (RCBD) replicated three times. Experimental design and layout were consistent across locations and seasons. The total plot area was 105 m² with an individual plot size of 6 m²ateach location. Plant density was kept at 33 333 ha⁻¹ with a plant spacing of 1 m x 0.3 m. Experiments were strictly rainfed, no supplementary irrigation was applied. Total rainfall (Table 3.2) received during the corresponding seasons were obtained from nearby automatic weather stations.

Table 3.1: Experimental site description for Deepdale, Richards Bay and Umbumbulu.

	Deepdale	Richards Bay	Umbumbulu		
Geographical location	28°01'S; 28°99'E	28°19'S; 32°06E	29°98'S; 30°70'E		
yAltitude (m a.s.l.)x	998	30	632		
		Moist coast	Moist coast		
Bio-resource group/ agro-	Coast hinterland	forest, thorn and	hinterland and		
ecological zone	thornveld	palmveld	ngongoni veld		
Annual rainfall	750 – 850 mm	820 – 1423 mm	800 – 1160 mm		
Average temperature	18.4°C	22°C	17.9°C		
Frost occurrence	Moderate	None	Light and occasional		
*Soil texture class	Clay	Sand	Clay		
Clay content	53%	< 5%	>60%		
	Jonkersberg form				
Soil types	(Jk)	Inhoek form (Ik)	Hutton form (Hu)		
Effective rooting depth					
(cm)	60	100	50		
^z Field capacity (%)	45.2	10.9	45.1		
^z Permanent wilting point					
(%)	34.7	6.2	34.5		
^z Saturation (%)	50	47.1	51		
Previous crop	Fallow (3 years)	Fallow (1 year)	Fallow (2 years)		
*Soil Classification, a Taxonomic	Contain for Couth Africa 1	1001. VM-tura al-arra ar-	11 7 Volume of an 11		

^{*}Soil Classification, a Taxonomic System for South Africa 1991; ^y Metres above sea level. ^z Values of soil water content are in percentage volumetric water content.

Table 3.2: Total rainfall received during experimentation seasons.

		Total rainfall received (mm)		
Year	Locations	Winter	Summer	
7	Deepdale	141.5	466.9	
2012/	Umbumbulu	102.2	557.4	
Ä	Richards Bay	25.2	563.2	
- 3/	Deepdale	177.4	461.5	
2013	Umbumbulu	194.0	442.5	
Ø	Richards Bay	98.8	460.7	

3.3.4 Agronomic practices

Sweet potato vines were planted on ridges of ~30 cm high. A minimum of three vine nodes were inserted into the ridge at planting. Land preparation at Deepdale and Umbumbulu was initially done using a tractor-mounted mouldboard plough (one pass), after which ridges were prepared by hand. At Richards Bay, land preparation was done using hand-hoes. Local farmers provided assistance with guidance on how to plant sweet potatoes at their respective locations. At all locations, farmers indicated that sweet potatoes were normally planted on land that had been fallow for at-least a year (Table 3.1).

For all four seasons, soil samples were taken prior to planting and analysed for fertility and textural characteristics. According to the small-scale famers' local knowledge, no fertilizer, pesticides or supplementary irrigation were required for the duration of crop growth at all experimental locations. A fertilized trial was only planted adjacent to the experimental plots during 2013/14 planting season for purposes of comparison. Details of weather parameters (maximum and minimum temperatures, reference evapotranspiration (ETo) and rainfall) were obtained from the Agricultural Research Council – Institute for Soil, Climate and Water's network of automatic weather stations.

3.3.5 Data collection

Sweet potato plants were allowed one month for establishment before data collection commenced. Plant growth and physiology data collected included vine length, leaf and branch number, stomatal conductance (SC) and chlorophyll content index (CCI). Stomatal conductance was measured using a steady state leaf porometer (Model SC-1, Decagon Devices, USA) and CCI was measured using the CCM200 Plus chlorophyll meter (Opti-

Sciences USA). Measurements of SC and CCI were respectively taken from the abaxial and adaxial surfaces of the third youngest, fully expanded and fully exposed leaf. Stomatal density in sweet potato leaves is generally higher on the abaxial than adaxial surface and the stomata are also evenly distributed across the leaf.

Soil water content in the field was monitored using PR2/6 Profile Probe connected to an HH-2 Handheld moisture meter (Delta-T Devices, UK). Sweet potato plants were harvested at 120 days (4 months) after planting. Measurements recorded included whole plant, above ground and below ground biomass, as well as harvest index (HI) and mass of marketable storage roots. Considered marketable storage roots were whole (undamaged) and weighed between 0.1 –1.4 kg and without harvest wounds, pest and disease damage (Ossom and Rhykerd, 2007). Yield was recorded in tonnes per hectare (t ha⁻¹).

3.3.6 Statistical analyses

Data were subjected to statistical analyses of variance (ANOVA) using GenStat® version 14 (VSN International, Hemel Hempstead, UK 2011). Tukey's test was used to separate means at the 5 % level of significance.

3.3. Results

3.3.1 Winter experiment

3.3.1.1 Plant physiology: Chlorophyll content index and stomatal conductance

Results of stomatal conductance (SC) were strongly influenced by environmental conditions. Highly significant differences ($P \leq 0.001$) were recorded among locations (Figures 3.1). Cultivar A40 recorded significantly higher SC values than the 199062.1 and A45 cultivars (Figure2). When comparing locations, Umbumbulu recorded higher SC in winter than Richards Bay. The constantly high SC was maintained throughout the growth period in Umbumbulu while in Richards Bay, SC values started to increase towards the end of data collection. Temperatures in Richards Bay were higher than at the Umbumbulu (Table 3.3) during 2012 but both locations experienced almost similar temperatures in 2013.Umbumbulu received more winter rainfall than Richards Bay. Soil water deficit (ETo outstripped rainfall); this was evidenced by the low values of SC observed during the study.

Chlorophyll content index results showed highly significant ($P \le 0.001$) differences among cultivars. Interactions between locations, seasons and cultivars were also significant ($P \le 0.05$). Planting seasons (2012 vs. 2013) and locations did not have significant influence on CCI. Cultivar 199062.1 had significantly higher ($P \le 0.001$) CCI followed by cultivars A45andA40 (Figure 3.1). Cultivar A45 managed to record constant CCI throughout the data collection period even after a cold spell that reduced CCI in the other cultivars during 2012 winter season. Richards Bay recorded higher CCI than Umbumbulu although there were no significant differences.

3.3.1.2 Plant growth

Planting sweet potatoes in winter resulted in plants failing to branch. Number of leaves and vine length varied significantly ($P \le 0.001$) across locations and among cultivars (Figures 3.3 & 3.4). Interactions between locations, planting season and cultivars had a significant ($P \le 0.05$) effect on vine length (Figure 3.3), while no significant interaction was recorded for leaf numbers (Figure 3.4). In both locations, cultivar A40 had significantly ($P \le 0.001$) fewer leaves than cultivars 199062.1 and A45 (Figure 3.4). Cultivar 199062.1 had significantly ($P \le 0.05$) higher number of leaves than cultivar A45. Cultivar A40 had longer vines than the other two cultivars. Vine length recorded no significant difference between cultivar A45 and 199062.1. Both parameters (vine length and number of leaves) recorded higher values in Richards Bay than at Umbumbulu in both growing season (2013 and 2013).

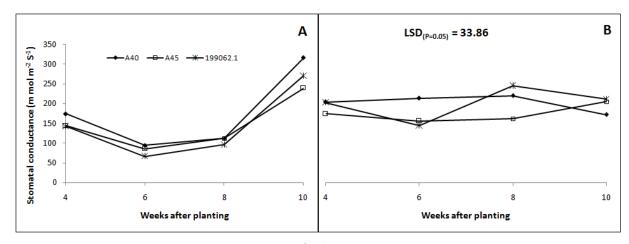


Figure 3.1: Stomatal conductance (mmol m⁻² s⁻¹) of three sweet potato cultivars grown in two different locations (A: Richards Bay and B: Umbumbulu) during winter season of 2012.

Table 3.3: Climatic data for the three experimental locations during winter season of 2012 and 2013.

			Richards Bay						Umbuml	oulu		
		Temp	(°C)		(mm)		Temp (°C)			(mm)		
Season	Months	Max	Min	Rain	ЕТо	Deficit	Max	Min	Rain	ЕТо	Deficit	
	April	27.5	15.2	6.3	88.0	81.7	22.8	12.0	20.3	77.3	57.0	
7	May	28.1	15.1	0.5	76.9	76.1	23.1	11.9	15.5	62.2	46.8	
201	June	25.4	12.4	20.3	61.7	41.4	20.8	8.9	7.8	55.9	48.1	
2	July	24.9	11.7	3.9	69.1	65.2	20.4	8.3	11.7	66.7	55.0	
	August	26.9	12.5	4.1	84.6	80.5	22.3	10.5	107.5	74.4	-33.1	
				35.1*	380.3*				162.8	336.5		
	April	27.2	15.8	22.8	111.0	88.2	21.3	15.0	117.5	102.9	-14.6	
	May	25.8	13.8	22.7	82.8	60.1	23.1	15.5	69.8	97.0	27.2	
13	June	25.8	12.4	16.7	102.6	86.3	26.2	17.3	119.5	117.8	-1.7	
201	July	24.1	13.1	16.3	84.5	68.2	26.2	17.3	50.7	107.6	56.9	
	August	26.0	13.7	20.3	148.0	127.7	25.7	16.2	85.0	100.5	15.5	
				98.8*	528.9*				442.5*	525.8*		

^{*}Rainfall and ET_o were not averaged but totalled.

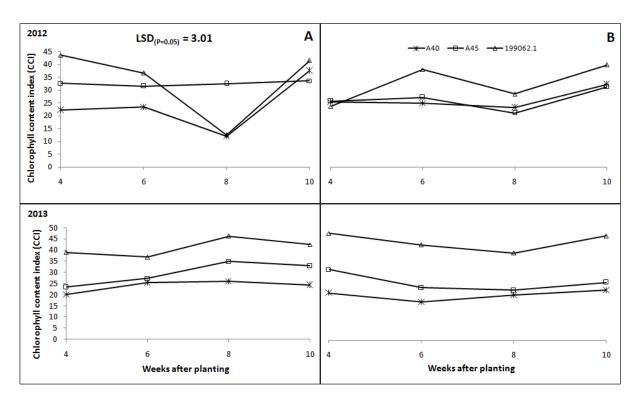


Figure 3.2: Chlorophyll content index (CCI) of three sweet potato cultivars grown in two different locations (A: Richards Bay and B: Umbumbulu) during winter seasons of 2012 and 2013.

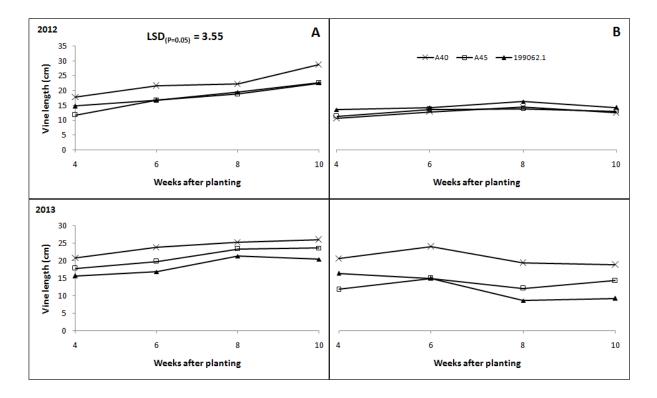


Figure 3.3: Vine length of three sweet potato cultivars grown in two different locations (A: Richards Bay and B: Umbumbulu) during winter seasons of 2012 and 2013.

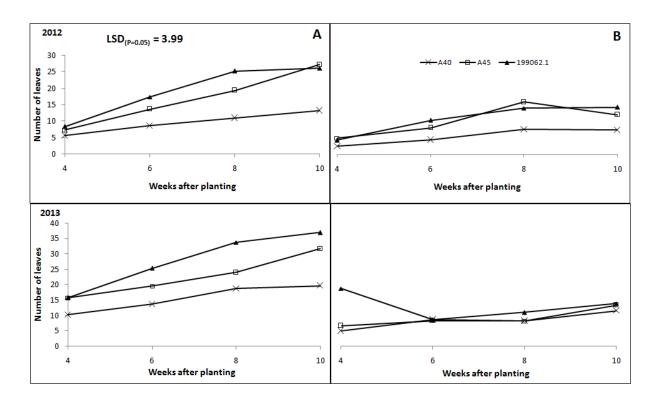


Figure 3.4: Leaf number of three sweet potato cultivars grown in two different locations (A: Richards Bay and B: Umbumbulu) during winter seasons of 2012 and 2013.

Table 3.4: Yield components of sweet potato cultivars (A40, A45 and 199062.1) grown in Richards Bay during winter season of 2012.

	Biomass	Storage roots	HI*
Cultivars	(t. ha ⁻¹)	(t. ha ⁻¹)	(%)
A40	0.92a	0.58a	23.7a
A45	1.11a	0.62a	21.5a
199062.1	0.73a	0.32a	19.0a
Mean	0.92a	0.51a	21.4a
LSD _(P=0.05) cultivar	0.47	0.59	10.8

^{*}HI = harvest index. Values in the same column sharing the same latter are not significantly different at LSD $_{(P=0.05)}$.

3.3.1.3 Biomass and storage root yield

Winter biomass and storage roots yield was only recorded in Richards Bay in 2012. Plants in both locations were unable to form storage roots in 2013. There were no significant (P<0.05) differences in biomass, storage roots yield and HI across all three cultivars in Richards Bay (Table 3.4). All yield parameters recorded very low figures indicating a direct link to the poor plant growth (vine length, number of leaves and poor branching) recorded earlier.

3.3.2 Summer experiment

3.3.2.1 Plant physiology: Chlorophyll content index and stomatal conductance

Measurements of stomatal conductance (SC) were only done during the 2012/13 growing season. Results were strongly influenced ($P \le 0.001$) by locations (Figure 3.5). Deepdale recorded significantly higher SC followed by Umbumbulu, then Richards Bay. The juvenile stage of the crop was characterized by high SC for all sweet potato cultivars at Deepdale. It (SC) later decreased as plants started to increase the number of leaves and branches (vegetative growth stage). This period coincided with increased demand for water by the crop as there were now more leaves transpiring thus the decline in SC over time. At the other two locations (Umbumbulu and Richards Bay), SC was initially low and remained relatively constant throughout the growing season. Soil water deficit was also high (Table 3.5) in these locations thus had a negative impact on SC. Significant differences between sweet potato cultivars were also observed where cultivar A40 recorded significantly higher ($P \le 0.05$) SC values than A45. Sweet potato cultivar 199062.1 was not significantly different from the two (Figure 3.5).

Chlorophyll content index (CCI) was also affected ($P \le 0.001$) by locations and cultivars. Significant interactions ($P \le 0.05$) were observed between locations and cultivars and between locations and seasons. Umbumbulu, recorded highest CCI followed by Deepdale then Richards Bay (Figure 3.6).Sweet potato cultivar199062.1 had significantly higher ($P \le 0.001$) CCI than A40 and A45 (Figure 3.6).The high CCI recorded for sweet potato cultivar 199062.1 did not however, translate to higher storage root yield as expected (Tables 3.6 & 3.7).

 Table 3.5: Climatic data for the three experimental locations during the summer season.

		Richards Bay					Umbum	bulu		Deepdale						
		Temp	o (°C)		(m	m)	Temp	Temp (°C) (mm)		m)	Temp(°C)		(mm)			
Season	Months	Max	Min	Rain	ETo	Deficit	Max	Min	Rain	ЕТо	Deficit	Max	Min	Rain	ETo	Deficit
	November	27.0	17.2	66.9	95.7	28.8	21.3	13.9	165.2	70.6	-94.6	24.2	13.6	117.1	2.9	-114.2
13	December	29.7	9.2	214.7	127.6	-87.1	25.5	16.6	64.7	99.8	54.1	28.5	16.2	76.4	3.8	-72.6
2012/13	January	30.6	19.3	145.4	121.1	-24.3	25.5	16.9	105.6	98.8	-6.8	28.5	16.5	146.9	3.7	-143.2
20	February	30.6	19.2	60.8	119.8	59.0	25.9	16.6	84.5	90.4	5.9	29.8	16.2	89.3	3.8	-85.5
	March	29.3	18.2	75.4	103.1	27.7	24.6	15.6	137.4	89.6	-47.8	28.1	15.4	37.2	3.0	-34.2
				563.2*	567.2*				557.4*	449.2*				466.9*	17.2*	
	November	28.6	18.0	134.0	192.8	58.8	21.3	15.0	117.5	102.0	-15.5	28.0	14.0	84.0	117.5	33.5
/14	December	27.2	18.5	114.0	121.7	7.7	23.1	15.5	69.8	97.0	27.2	26.2	15.2	84.4	102.6	18.2
2013/14	January	30.9	20.6	24.8	164.4	139.6	26.2	17.3	119.5	117.8	-1.7	30.2	16.8	108.9	135.5	26.6
20	February	31.7	20.7	30.4	150.2	119.8	26.2	17.3	50.7	107.6	56.9	30.8	16.9	49.1	115.6	66.5
	March	29.4	19.9	157.5	119.6	-37.9	25.7	16.2	85.0	100.5	15.5	29.0	15.7	135.1	99.7	-35.4
				460.7	748	3.7*			442.5*	524	1.9*			461.5*	57	0.9*

^{*}Rainfall and ET_o were not averaged but totalled.

3.3.2.2 Plant growth

Plant branching and number of leaves was significantly ($P \le 0.001$) influenced by location, growing season and the interaction of the two ($P \le 0.05$). Vine length on the other hand varied significantly ($P \le 0.001$) across locations, among sweet potato cultivars and over the two growing seasons. The interaction of the latter and former was also significant ($P \le 0.05$). Cultivar 199062.1 was shorter than both A40 and A45 (Figure 3.7) mainly because it is a non–twining, semi–erect cultivar. The number of leaves however, was statistically similar across all sweet potato cultivars (Figure 3.8). Growth parameters tended to increase with plant growth, with the exception of leaves which later decreased due to leaf senescence and abscission as plants started ageing.

When comparing locations, Deepdale and Umbumbulu were more conducive for sweet potato growth (leaves, branches and vine length). Plants grown in Richards Bay recorded poorest growth (significantly ($P \le 0.05$) shorter vines, fewer branches and low number of leaves) across all three cultivars when compared to the other locations. The first growing season (2012/13) showed superior plant growth when compared to the second growing season 2013/14 (Figures 3.7, 3.8 & 3.9).

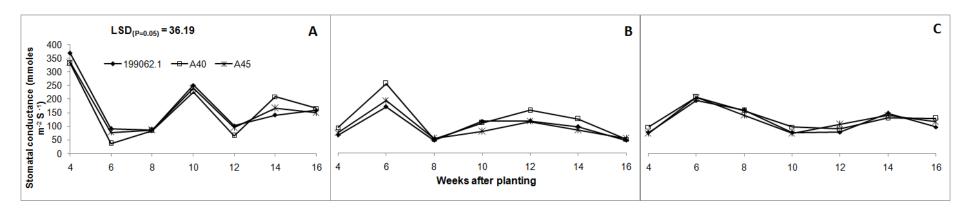


Figure 3.5: Stomatal conductance (mmol m⁻² s⁻¹) of sweet potato cultivars (A40, A45 and 199062.1) grown during the winter season in different locations of KZN.

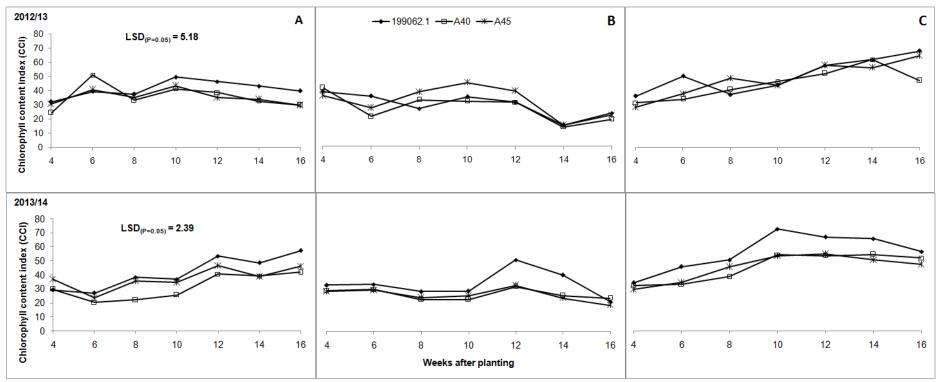


Figure 3.6: Chlorophyll content (CCI) of sweet potato cultivars (A40, A45 and 199062.1) grown in different locations (A: Deepdale, B: Richards Bay and C: Umbumbulu) of KZN.

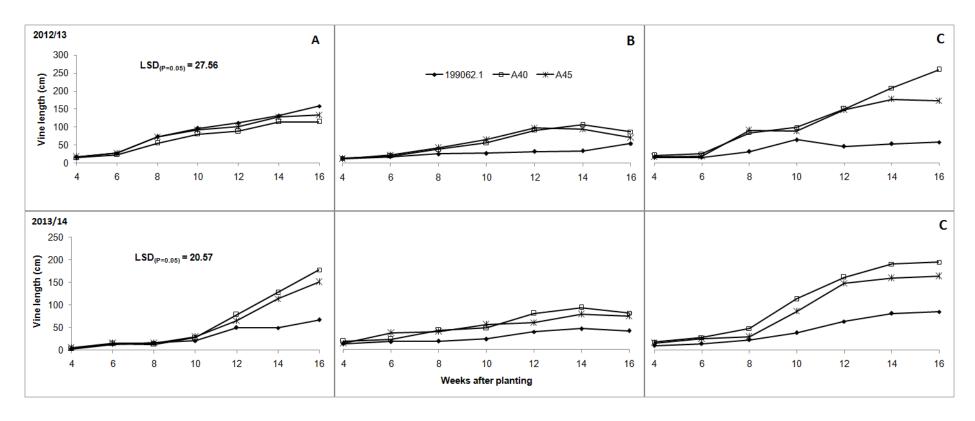


Figure 3.7: Vine length of sweet potato cultivars (A40, A45 and 199062.1) grown in different locations (A: Deepdale, B: Richards Bay and C: Umbumbulu) of KZN.

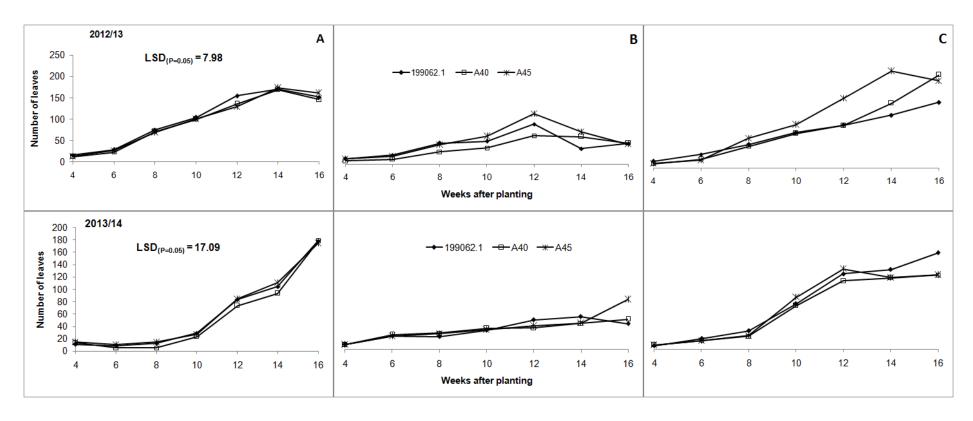


Figure 3.8: Number of leaves of sweet potato cultivars (A40, A45 and 199062.1) grown in different locations (A: Deepdale, B: Richards Bay and C: Umbumbulu) of KZN.

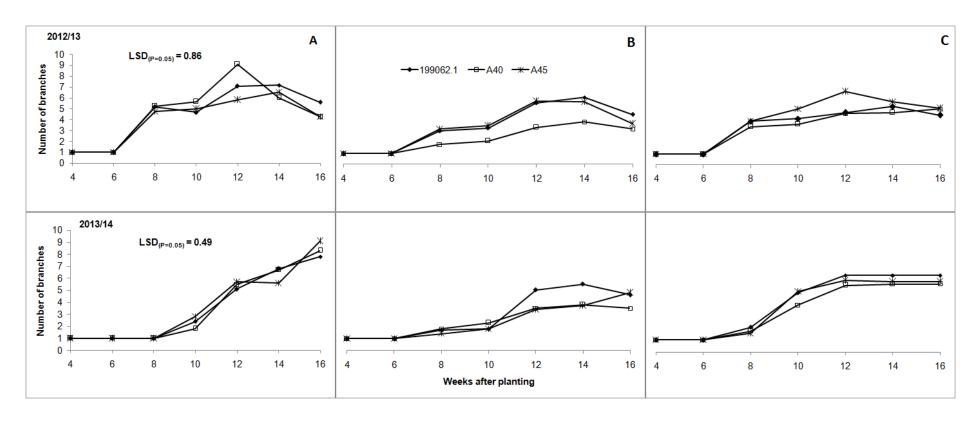


Figure 3.9: Branch numbers of sweet potato cultivars (A40, A45 and 199062.1) grown in different locations (A: Deepdale, B: Richards Bay and C: Umbumbulu) of KZN.

3.3.2.3 Biomass and storage root yield

Locations had a highly significant ($P \le 0.001$) effect on biomass and storage root yield. Seasons had a highly significant ($P \le 0.001$) effect on storage roots yield only. Interaction of location and season was also significantly effective. Umbumbulu and Deepdale showed highly significant ($P \le 0.001$) differences for both yield parameters when compared with Richards Bay (Table 3.6 & 3.7). This was directly linked to observed plant growth and physiological responses in these locations. No significant differences were observed among sweet potato cultivars, but cultivar A45 recorded slightly higher biomass and storage root yield than the other two (Table 3.6 & 3.7). Correlations between biomass and storage roots yield showed that biomass contributed (r = 0.781; $P \le 0.05$) significantly to storage root yield. The first growing season (2012/13) recorded significantly higher ($P \le 0.05$) storage root yield than the second season (2013/14) at Deepdale. There were no significant differences in storage root yields from Richards Bay and Umbumbulu during both growing seasons.

Deepdale recorded the highest storage root yield (42.0 t ha⁻¹) during 2012/13 growing season and lower yield (13.6 t ha⁻¹) during 2013/14 growing season, which was 67% reduction in yield. Storage root yield from Umbumbulu and Richards Bay did not fluctuate that much during both seasons. Yield from Umbumbulu was 29.4 and 28.0 t ha⁻¹ while it was 5.4 and 5.0 t ha⁻¹ in Richards Bay during 2012/13 and 2013/14 growing seasons, respectively. A greater percentage of storage roots from Richards Bay weighed less than 0.1 kg thus classified as non-marketable. Comparison of fertilized and non-fertilized sweet potato trials conducted during 2013/14 season (Table 3.8) only showed significantly higher (P \leq 0.05) storage root yield (19.6 t ha⁻¹) from fertilized trials at Richards Bay. Applying fertilizer increased storage roots yield by 76% in Richards Bay when compared to the non-fertilized crop.

Table 3.6: Yield components of sweet potato cultivars (A40, A45 and 199062.1) grown in three different locations of KwaZulu-Natal, South Africa during 2012/2013 summer season.

	Deepdale			J	J mbumbulu		Rie	chards Bay		
		Storage			Storage			Storage		
	Biomass	root	HI*	Biomass	root	HI*	Biomass	root	HI*	
Cultivars	(t.ha ⁻¹)	(t. ha ⁻¹)	(%)	(t.ha ⁻¹)	(t. ha ⁻¹)	(%)	(t.ha ⁻¹)	(t. ha ⁻¹)	(%)	
A40	19.1 ^a	37.5 ^a	34.0 ^a	25.9 ^a	29.7 ^b	46.7 ^a	5.7 ^b	7.4°	48.0 ^a	
A45	25.7 ^a	45.3 ^a	35.4^{a}	25.6 ^a	38.3 ^b	38.9^{a}	3.8 ^b	2.4°	60.0^{a}	
199062.1	18.7ª	43.1 ^a	30.1 ^a	9.2 ^b	20.2^{b}	30.9^{a}	4.3 ^b	6.3°	41.1 ^a	
Location mean	21.2a	42.0 ^a	33.2a	20.2a	29.4 ^b	38.8 ^a	4.6 ^b	5.4 ^c	49.7 ^a	
LSD _(P=0.05) Cultivar	7.33	9.38	13.70				L			
LSD _(P=0.05) location	7.33	9.38	13.70							
$LSD_{(P=0.05)}location \times$										
cultivar	12.70	16.25	23.73							

^{*}HI = harvest index. Values in the same column sharing the same latter are not significantly different at LSD (P=0.05). Significant differences for location mean are presented on specified rows.

Table 3.7: Yield components of cultivars grown in three different locations of KwaZulu-Natal, South Africa during 2013/14 summer season.

Deepdale			Umbumbuli	1		Richards Bay			
	Storage			Storage			Storage		
Biomass (t.	root (t. ha ⁻		Biomass (t.	root (t. ha		Biomass (t.	root (t.		
ha-1)	1)	HI* (%)	ha ⁻¹)	1)	HI* (%)	ha ⁻¹)	ha ⁻¹)	HI*(%)	
45.4a	6.9b	14.9c	68.7a	31.0a	45.2b	12.8a	5.8a	44.6a	
50.5a	15.3a	29.7b	52.7a	24.1b	45.8b	8.9a	5.9a	38.7a	
45.8a	18.5a	40.8a	44.6b	28.9a	65.1a	6.7a	3.1a	46.4a	
47.2a	13.6b	29.7b	55.4a	28.0a	52.0a	9.5b	5.2c	43.3a	
7.55	4.29	9.82							
7.55	4.29	9.82							
			1						
13.08	7.3	17.01							
	Biomass (t. ha ⁻¹) 45.4a 50.5a 45.8a 47.2a 7.55	Storage Biomass (t. ha ⁻¹) root (t. ha ⁻¹) 45.4a 6.9b 50.5a 15.3a 45.8a 18.5a 47.2a 13.6b 7.55 4.29 7.55 4.29	Storage Biomass (t. root (t. ha ⁻ ha ⁻¹) HI* (%) 45.4a 6.9b 14.9c 50.5a 15.3a 29.7b 45.8a 18.5a 40.8a 47.2a 13.6b 29.7b 7.55 4.29 9.82 7.55 4.29 9.82	Storage Biomass (t. root (t. ha ⁻ ha ⁻¹) Biomass (t. ha ⁻¹) 45.4a 6.9b 14.9c 68.7a 50.5a 15.3a 29.7b 52.7a 45.8a 18.5a 40.8a 44.6b 47.2a 13.6b 29.7b 55.4a 7.55 4.29 9.82 7.55 4.29 9.82	Storage Storage Biomass (t. ha ⁻¹) root (t. ha ⁻¹) HI* (%) Biomass (t. ha ⁻¹) root (t. ha ⁻¹) 45.4a 6.9b 14.9c 68.7a 31.0a 50.5a 15.3a 29.7b 52.7a 24.1b 45.8a 18.5a 40.8a 44.6b 28.9a 47.2a 13.6b 29.7b 55.4a 28.0a 7.55 4.29 9.82	Storage Biomass (t. root (t. ha ⁻¹ ha ⁻¹) HI* (%) Biomass (t. root (t. ha ⁻¹ ha ⁻¹) HI* (%) 45.4a 6.9b 14.9c 68.7a 31.0a 45.2b 50.5a 15.3a 29.7b 52.7a 24.1b 45.8b 45.8a 18.5a 40.8a 44.6b 28.9a 65.1a 47.2a 13.6b 29.7b 55.4a 28.0a 52.0a 7.55 4.29 9.82 7.55 4.29 9.82	Storage Storage Biomass (t. root (t. ha ⁻ ha ⁻¹) Biomass (t. root (t. ha ⁻ ha ⁻¹) Biomass (t. root (t. ha ⁻ ha ⁻¹) Biomass (t. ha ⁻ ha ⁻¹) HI* (%) ha ⁻¹) Biomass (t. ha ⁻ ha ⁻¹) Biomass (t. ha ⁻¹) HI* (%) ha ⁻¹) Biomass (t. ha ⁻¹) HI* (%) ha ⁻¹) Biomass (t. ha ⁻¹) HI* (%) ha ⁻¹) Biomass (t. ha ⁻¹) HI* (%) ha ⁻¹ Biomass (t. ha ⁻¹) HI* (%) ha ⁻¹ HI* (%) ha ⁻¹ HI* (%) ha ⁻¹ HI* (%) <th colspan<="" td=""><td>Storage Storage Storage Biomass (t. root (t. ha¹) HI* (%) Ha¹) HI* (%) HI* (%) Biomass (t. root (t. ha²) Ha¹) ha¹) ha¹) HI* (%) ha¹) ha²¹) ha²² ha²¹) ha²²<!--</td--></td></th>	<td>Storage Storage Storage Biomass (t. root (t. ha¹) HI* (%) Ha¹) HI* (%) HI* (%) Biomass (t. root (t. ha²) Ha¹) ha¹) ha¹) HI* (%) ha¹) ha²¹) ha²² ha²¹) ha²²<!--</td--></td>	Storage Storage Storage Biomass (t. root (t. ha¹) HI* (%) Ha¹) HI* (%) HI* (%) Biomass (t. root (t. ha²) Ha¹) ha¹) ha¹) HI* (%) ha¹) ha²¹) ha²² ha²¹) ha²² </td

^{*}HI = harvest index. Values in the same column sharing the same latter are not significantly different at LSD (P=0.05). Significant differences for location mean are presented on specified rows.

Even so, the yield was still lower than non-fertilized crop at Umbumbulu during that season. Fertilizer application at Deepdale and Umbumbulu did not increase storage root yield, in-fact, it gave lower yield than the non-fertilized crop. Soil analysis results taken before planting did not indicate changes in soil chemical content (Table 3.9).

Harvest index (HI) was significantly ($P \le 0.05$) affected by seasons, locations and cultivars. The 2012/13 growing season recorded higher ($P \le 0.05$) HI than 2013/14 growing season. While all locations recorded none significant differences during the 2012/13 growing season, Deepdale recorded the lowest (28.5%) HI during 2013/14 season. Umbumbulu and Richards Bay recorded similar HI values (52.0 and 43.0%, respectively) during the same season. Cultivar199062.1 recorded a significantly higher ($P \le 0.05$) HI than the other two cultivars.

Table 3.8: Fertilized and non-fertilized storage root yield from three different locations of KwaZulu-Natal during 2013/14 growing season.

	De	eepdale	Richard	ds Bay	Umbumbulu		
		Non-fertilized storage	Fertilized	Non-fertilized	Fertilized	Non-fertilized	
	Fertilized storage	roots	storage roots (t	storage roots	storage roots (t	storage roots	
Cultivars	roots (t ha ⁻¹)	(t ha ⁻¹)	ha ⁻¹)	(t ha ⁻¹)	ha ⁻¹)	(t ha ⁻¹)	
A40	8.9a	6.9b	19.7a	5.8a	14.2a	31.0a	
A45	2.3c	15.3a	18.5a	5.9a	13.3ab	24.1b	
199062.1	4.8b	18.5a	20.5a	3.1a	16.7a	28.9a	
Location mean	5.3d	13.6c	19.6b	5.2d	14.7c	28.0a	
LSD _(P=0.05) Fertilizer	2.25						
LSD (P=0.05) Location	2.76						

Values in the same column sharing the same latter are not significantly different at LSD $_{(P=0.05)}$. Significant differences for location mean are presented on specified rows.

LSD (P=0.05) Location

X Fertilizer

3.90

Table 3.9: Soil analysis results for the three experimental locations during summer seasons.

	Experimental locations								
Soil nutrient	Richar	ds Bay	Umbu	ımbulu	Deepdale				
parameters	2012/13	2013/14	2012/13	2013/14	2012/13	2013/14			
Nitrogen (%)	0.1	0.08	0.3	0.02	0.4	0.33			
Soil pH (KCl)	4.2	5.0	4.2	4.25	4.8	4.48			
Phosphorus (g/kg)	4.1	20.7	1.0	8.1	5.6	11.1			
Potassium (g/kg)	12.4	20.0	63.3	72.7	392.1	294.4			
Magnesium (g/kg)	0.0	47.6	252.0	344.4	471.9	603.3			
Calcium (g/kg)	71.0	244.8	1092.9	1238.4	1570.8	2173.3			
Manganese (g/kg)	2.8	6.9	13.3	80.8	22.5	68.9			
Copper (g/kg)	0.3	0.4	8.6	10.3	4.7	5.6			
Zinc (g/kg)	0.4	1.4	1.2	1.4	2.5	3.9			
Exch. Acidity cmol/l	0.3	0.07	0.3	0.36	0.1	0.19			
Total cations (cmole/l)	0.9	2.48	7.8	9.47	11.4	15.1			
Acid saturation (%)	37.0	3.0	4.0	4.0	1.0	1.0			
Organic carbon (%)	<0.5	1.1	3.4	3.3	4.4	4.4			
Clay (%)	6.0	11.0	58.0	55.0	49.0	50.0			

3.4 Discussion and conclusions

Varying environmental conditions at the three locations had a significant effect on plant ecophysiology, growth and yield. During the juvenile stage, SC was high. It started to decrease as the crop moved into the vegetative growth stage. This period coincided with increased demand for water by the crop as there were now more leaves transpiring. However, since there was a deficit (ETo outstripped rainfall), the crop had to adjust by closing its stomata. This would explain the pattern of declining SC over time. Reports by Chaves et al. (2002) and Mabhaudhi et al. (2013) indicated that stomatal closure is a drought avoidance mechanism. By closing the stomata, not only does the plant limit water loss through transpiration, it also reduces intracellular carbon dioxide (CO₂) availability (Mabhaudhi et al., 2013; Zhao et al., 2014). This affects photosynthesis since CO₂ is the chief substrate; hence resulting in biomass reduction (Lawlor, 2002; Blum, 2009). Some of the metabolic changes affected by CO₂ are themselves are consequence of photosynthetic apparatus resistance to dehydration (Cornic, 2000). According to Chaves et al. (2002), the changes are capable of maintaining an osmotic pressure in the photosynthetic cells by increasing nitrate concentration and decreasing carbohydrate export. This was evident in plants grown in Richards Bay where relatively low values of SC were recorded throughout the growing season and this translated into very low biomass in both winter and summer experiments.

Soil profiling revealed that Richards Bay was the only location without an impeding layer in the first 100cm depth. An observation that further explained the low SC and CCI recorded in that location. Not only did the location lose soil water through ETo, it also lost water through the porous soil profile. This may have forced the crop to adjust by closing its stomata thus reducing SC, CCI, growth parameters and consequently storage root yield. Moreover, the water deficit may have been caused by poor water retaining capacity of the sandy soil (Zhao et al., 2014) in Richards Bay (Table 3.1). Such soils are more porous and characterized by very low field capacity (FC) and permanent wilting point (PWP) (Table 3.1). Crops grown in such soils can easily experience periods of water deficit even if there has been significant rainfall since much of the water is drained away from the root zone. The low SC confirms earlier reports that stomatal responses are closely linked to soil water content than to leaf water status (Gowing et al., 1990; Chaves et al., 2002). The bioresource group (Table 3.1) where Richards Bay is located may have contributed to the drought stress conditions in that area.

There other two locations (Deepdale and Umbumbulu) had clayey soils (Table 3. 1) and an impending layer at 50cm depth. These characteristics promote good water retention capacity and may have limited soil water loss through soil profile. The preserved water is believed to have supplemented the plants metabolic activities after ETo losses. The bioresources groups where the two locations are situated also experience misty conditions which may have reduced the length of exposure to water deficit during the summer season. Winters in these areas are characterised with no mist and less rains thus may have hampered growth and vital metabolic activities. Temperatures below 15°C (Table 3.3) were reported by Laurie and Niederwiser (2004) to cause little growth and below 10°C causes severe retard ingrowth. This may have been the main reason for the retarded plant growth during the winter season.

Decreases in chlorophyll content under drought stress conditions have been associated with oxidative stress and may be the result of pigment photo-oxidation and chlorophyll degradation (Jaleel et al., 2009; Anjum et al., 2011). The fact that SC and CCI were low in Richards Bay implies that photosynthesis was both substrate and energy limited. Consequently leaves showed rapid signs of chlorosis and later abscission. In the other locations, photosynthesis was not limited by neither chlorophyll degradation nor water deficit and CO₂ flow. This implied that there was sufficient soil water to meet plant growth and evaporative demands (Table 3.3 & 3.5). Cultivar A40 recorded low CCI but high SC values during both summer seasons suggesting that it might not respond to water stress by closing stomata to control water loss and limit CO₂. Instead, it responded by chlorophyll degradation and photo-oxidation (Figures 3.2 & 3.6).

Plant growth parameters, CCI and SC tended to have a link and tended to follow a particular trend. Low values of CCI and SC resulted in short vines, fewer leaves and fewer branches. This was another indication of drought stress and plant coping mechanisms as explained by Chaves et al. (2002) and Jaleel et al. (2009). Vine length was influenced by both cultivar growth habit within a location and environmental variations across the three locations. This observation was contrary to reports by Oggema et al. (2007) that growth habits of sweet potato plants were more related to varietal than environmental differences. Interesting observations were also made on the growth parameters. These cultivars tended to limit their

vine extension, branching and number of leaves under harsh environmental conditions like Richards Bay. Reduced plant size (height/length, leaf area and leaf area index) were reported by Blum (2005) as the major mechanism for moderating water use and reducing injury under drought stress conditions. These results, therefore, indicated that Richards Bay was characterised by constant water stress irrespective of the season. Observations of rainfall and ETo (Table 3.3 & 3.5) did indicate water deficit. It is therefore suspected that besides the high ETo, the prevalent sandy soils and high temperatures in Richards Bay may have accelerated soil water loss through rapid infiltration since it (soil) had very low clay content (<5%). It was further noted that ETo alone could not have caused such a high impact on soil water loss since it was relatively similar (Table3.3 & 3.5) across the locations in all seasons (with the exception of Deepdale in 2012/13 summer season).

Richards Bay's poor soil physical and chemical properties (Table 3.9) and poor water holding capacity renders crops grown on such soils prone to frequent episodes of water stress. To curb the situation and to avoid plants being blown away by wind, ridging and addition of compost was conducted at planting. The two agronomic practices were expected to conserve soil water (Everson et al., 2011). Instead, plants developed more fibrous roots to help scavenge for water and nutrients at the expense of developing storage roots. Under such stressful environments, even high yielding cultivars will produce low yields (Blum, 2005).

The tendency of sweet potato cultivars to branch in one location and not branch in another but prefer to extend in length or increase leaf number can be attributed to specific environmental adaptation. This study observed that the three sweet potato cultivars exhibited very poor growth in Richards Bay even in winter where it was expected to do better than Umbumbulu. Richards Bay is a coastal area with low elevation (30 m a.s.l) and classified as a "Moist coast forest, thorn and palm-veld" (Table 3.1) (Smith, 2006). Hartemik et al. (2000) also reported vine growth decline in lowland soils with low fertility, a similar condition as that in Richards Bay (Table 3.9). Growth improved as altitude increased and the best plant growth was recorded in the mid-elevation location (Umbumbulu, 632 m a.s.l) classified as a "Moist coast hinterland and ngongoni veld". It was also noted that the highest elevation area (Deepdale 998 m a.s.l) was characterised by profuse branching tendencies across all three cultivars in summer. Sweet potato cultivar A45 showed the greatest environmental plasticity with regards to showing wide adaptation to all three environments especially in summer. It was able to

grow, branch, produce leaves and higher yields across all locations. This study proves the concept of wide adaptation, indicating that adaptation has more of geographical than environmental meaning and it reduces genetic diversity while increasing genetic vulnerability (Ceccarelli, 1994).

Soil available potassium (K) was higher at Deepdale (Table 3.9) was sufficient to meet the large demand for K in sweet potato as reported by George et al. (2002). This may explain the high storage root yield recorded from this location during that season. According to Bourke (1985), storage K influences root yield by increasing dry matter allocation to the storage roots. The drastic reduction (of 67%) in storage root yield during 2013/14 may have been caused by lower temperatures (minimum temperatures) experienced during planting and establishment period (November 2013) (Table 3.5) which consequently delayed plant establishment and vegetative growth. The delayed plant growth further affected "storage roots filling" such that harvesting (after 120 days) time was fell due before storage roots could reach maximum expansion and filling. Dry matter allocation to storage roots at Umbumbulu was not limited by K levels during both growing seasons (2012/3 & 2013/14). The constantly warm temperatures at Umbumbulu during planting period favoured and enhanced crop establishment and vegetative growth. This allowed "root filling" to begin on time thus crop was able to give higher yields (at 120 days). This observation emphasises the vital role played by prevailing temperatures at planting stage of sweet potato crop (DAFF, 2011).It is an example of variations brought about by climate change. This further show why it is important for smallholder farmers to stay informed about future weather conditions before commencement of planting seasons so that they can adjust their planting calendar according to prevailing environmental conditions. According to Lebot (2009), low temperatures of 15°C and below, inhibit storage root development, which was the case at Deepdale (Table 3.5).

Fertilizer application during 2013/14 summer season did not have an effect on storage root yield in the already fertile soils (at Umbumbulu and Deepdale). Significant effects were only recorded in Richards Bay. According to Mukhtar et al. (2010), non-significant response of sweet potato to fertilizer application should be blamed on the innate quality of sweet potato and the fact that it colonises easily on marginal soils. Cultural practices such as fallowing being practised at Deepdale and Umbumbulu may have contributed to improved soil fertility and consequently to high storage root yields. This is an indication that small-scale farmers understand and appreciate the principles and benefits of low-input cropping systems. Ridging

also contributed to good yields since it conserves soil water (Everson et al., 2011) and allows for root expansion. The storage root yields achieved during summer season creates a scenario where small-scale farmers under low-input cropping systems would continue to ignore fertilizer recommendations if they can still get the best yield and improved food security just from applying their cost effective (Dawson et al., 2007) cultural practices alone. Their only concern would be the inability to produce sweet potato throughout the year since the crop failed completely under low temperature conditions. Where it does grow in winter (i.e. Richards Bay), it fails to give satisfactory yields. This situation would therefore cause episodes of food insecurity during the course of the year, but this can be avoided by applying simple sweet potato preservation techniques such as piecemeal harvesting, crisping and flour processing. Furthermore, farmers can sell the surplus since sweet potato has a very short shelf-life and use the income generated from the sales to buy other food crops that have a long shelf-life.

It can be concluded therefore, that environmental conditions can suppress the growth habits of sweet potato cultivars through limiting plant growth patterns thus affecting subsequent yield. Environmental conditions such as high temperatures, ETo and low water retention capacity of sandy soils create a drought stress scenario which results in plants responding by lowering SC and CCI. This therefore, decreases photosynthetic rates and assimilation. Consequently, the number of leaves which are the main source of assimilates utilized for plant growth and increasing storage root dry matter allocation are decreased resulting in poor plant growth and low yields.

Sweet potato cultivars selected for the study performed well under the low-input cropping system in Deepdale and Umbumbulu. This suggests that the three sweet potato cultivars can be produced using cost effective agricultural methods, a trait desirable to resource-constrained small-scale farmers and can improve the food security status of these farmers. Application of good cultural and agronomic practices, such as fallowing and ridging, contributed to the increased yields. Ridging conserved water in the soil and allowed enough room for storage root expansion while fallowing improved the soil nutrient status.

Fertilizer application did not improve storage roots yield when applied to naturally fertile soils. The innate quality and ability of sweet potato to colonise easily on marginal soils requires fertilizer application only in continuously cropped and exhausted soils. Sweet potato cultivar A45, an orange-fleshed cultivar, showed more environmental plasticity than the other two sweet potato cultivars (A40 and 199062.1). This indicates that its adoption can contribute towards improved food and nutrition security for resource-constrained and small-scale farmers since it can also supplement for Vitamin A deficiencies.

The Richards Bay environment was not suited for the sweet potato cultivars evaluated in this study. All the three cultivars experienced limitations to fully expressing their genetic potential. In order to improve sweet potato yields in Richards Bay (where temperatures allows year round production), farmers should be encouraged to improve soil water conservation strategies such as increasing the size of ridges, to apply recommended fertilizer amounts, to use more compost at planting in order to improve soil structure and soil water holding capacity.

It is also important to advise farmers to add fertilizers, especially those high in K, so as to improve storage root yield. Agronomic practices, such as using disease-free planting material, controlling pests and diseases and good crop management, should be encouraged at all the studied locations. It is envisaged that participation of farmers in future studies will encourage them to take-up these new cultivars and contribute towards strengthening household food security and nutrition.

CHAPTER 4

RESPONSE OF SWEET POTATO (IPOMOEA BATATAS L.) CULTIVARS TO WATER STRESS UNDER A CONTROLLED ENVIRONMENT

Abstract

Climate change and variability are predicted to result in more frequent droughts in sub-Saharan Africa. Versatile crops like sweet potato may be able to withstand such conditions. An experiment to evaluate sweet potato's adaptability under controlled water stress conditions was conducted. Three locally bred sweet potato cultivars A40, A45 and 199062.1 were grown under two water regimes (30% ETc and 100% ETc) in a split-plot design. Results showed that sweet potato cultivars were tolerant to drought. They responded to water stress by closing stomata to limit transpirational losses and maintaining high tissue water content (relative water content) under both water regimes. Low stomatal conductance was an indication that plants resorted to photorespiration in order to sustain their metabolic functions. Under water stress, plants limited growth to a smaller canopy size as a drought avoidance mechanism. This response gave higher yields than plants with long stems and numerous broad leaves grown under optimum water. Plants grown under optimum water resulted in rank growth syndrome, a characteristic that favours vegetative growth over storage root growth. The studies cultivars are suitable for marginal areas. In areas where there is optimum water supply, planting should be planned such that there is a short period of water deficit during the growing season so as to improve storage root yield. Cultivar 199062.1 was the most tolerant to drought conditions.

Keywords: drought, physiology, yield, water stress

4.1 Introduction

Agricultural production activities are the most sensitive and vulnerable to climate change and variability (Wreford et al., 2010). Impacts of climate change bring about changes in variability, seasonality, mean precipitation and water availability as well as emergence of new pathogens and diseases (Fischlin et al., 2007). Developing countries like South Africa are more vulnerable to climate change due to low capital and technological flexibility to adapt. Most of these countries are already in hot climates that are expected to get hotter in the future (Mendelson et al., 2000) thus further disrupting their agricultural production systems. Periodic drought conditions are common in these areas (Grayson, 2013). According to Flexas et al. (2006), drought is regarded as the main environmental factor limiting plant growth and productivity on a global scale.

Drought is defined by Jaleel et al. (2009) as a period without significant rainfall. It occurs when available water in the soil is reduced and there is continuous water loss to the atmosphere caused by atmospheric processes such as transpiration and evaporation. Drought causes water stress which is considered as the major cause of abiotic stress in plants (Jaleel et al., 2009; Alizera et al., 2011). According to Grayson (2013), research is playing a big role in developing new crop cultivars with high yield potential that can tolerate high temperatures and require less water to grow.

Sweet potato is one of the crops considered adaptive to water stress conditions (Woolfe, 1992; Agili et al., 2012), but selection for water stress tolerance is reported as a complex physiological trait. According to Ekanayake and Collins (2004), selection is often impeded by lack of appropriate field screening techniques. Studies on water stress tolerance have concentrated mostly on the sink organ (storage root) than the source (leaves) (Haimeirong and Kubota, 2003), but researchers later realized that the sink activity in sweet potato was restricted by source function (van Heerden and Laurie, 2008). Some workers have argued that water stress limits photosynthesis through diffusive (stomatal and mesophyll conductance) means (van Heerden and Laurie 2008; Jaleel et al., 2009) while others claim that it is through metabolic reactions (Flexas et al., 2006; Alizera et al., 2011). Blum (2009) reported that water stress reduced plant growth by limiting elongation and expansion of plant parts. All of these reports show the ingenious adaptation of sweet potato at physiological levels (Alizera et al., 2011) and all have an effect on the subsequent yield. This is an indication that water

availability is the major limiting factor in crop production and is threatening food security in developing countries.

The adaptation traits of sweet potato to water stress need to be investigated in order to select for cultivars suitable for such conditions. It is also important to link the physiological responses to water stress occurring in the source (leaves) to the ultimate storage root yield. It was hypothesized that physiological responses of locally bred sweet potato cultivars to water stress will improve storage root yield. This study was therefore aimed at measuring physiological, growth and yield responses of locally bred sweet potato cultivars to water stress.

4.2 Materials and methods

4.2.1 Planting material

Three sweet potato cultivars, A40, A45 and 199062.1 were sourced from the University of KwaZulu-Natal's (UKZN) Plant Breeding Department. Two of them, A40 and A45 were bred at UKZN while the third, 199062.1 was originally obtained from the International Potato Centre (CIP) and multiplied at UKZN during the 2011/12 planting season. Two of the sweet potato cultivars, A45 and 199062.1 were orange-fleshed, while the third, A40, was white-fleshed. Planting vines (30 cm long) were cut from the tip of mother plants which were raised in a typical warm subtropical nursery at UKZN. Planting vines were defoliated to one top fully expanded leaf to reduce photosynthetic demand at crop establishment stage.

4.2.2 Controlled environment description

The experiment was conducted under simulated drought conditions in beds in a growth tunnel at the University of KwaZulu-Natal. The environment in the tunnels is not fully controlled but is designed to mimic sub-tropical conditions (~18/33°C day/night temperatures and 60-80% relative humidity) (Modi 2007). Average temperature (T_{av}) and relative humidity (% RH) inside the tunnel were monitored electronically using HOBO 2K Loggers (Onset Computer Corporation, Bourne, USA). The HOBO loggers were programmed to measure T_{av} and % RH at hourly intervals. These were later computed to provide more representative daily averages.

4.2.3 Experimental design

The experimental layout was a spit-plot design with two factors; water regimes (30% and 100% crop water requirement (ETc)) as the main plot and cultivars (A40, A45 and 199062.1) as sub-plots. Cultivars were replicated four times. Irrigation scheduling was based on reference evapotranspiration (ET_o) and a crop factor (K_c) (Allen *et al.*, 1998). Drip irrigation was used to apply water in the beds. The system consisted of solenoid valves, a control box and online drippers. The system was set to have a maximum operating pressure of 200 kPa and an average discharge rate of 2 L/hr. Drip lines were placed according to plant spacing (0.6 m x 0.3 m).

4.2.4 Data collection

4.2.4.1 Growth and physiology

Sweet potato plants were allowed two weeks for establishment before data collection commenced. Plant growth and physiology data collected included vine length, leaf and branch number, stomatal conductance (SC), chlorophyll content index (CCI) and relative water content (RWC). Stomatal conductance was measured using a steady state leaf porometer (Model SC-1, Decagon Devices, USA) and CCI was measured using the CCM 200-*Plus* chlorophyll meter (Opti-Sciences USA). Measurements of SC and CCI were respectively taken from the abaxial and adaxial surfaces of the third youngest, fully expanded and fully exposed leaf. Stomatal density in sweet potato leaves is generally higher on the abaxial than adaxial surface and stomata are also evenly distributed across the leaf. Soil water content was monitored electronically using a Theta probe (ML2X) connected to a hand held HH-2 moisture meter (Delta-T, UK).

Relative water content was measured using the method of Yamasaki and Dillenburg (1999). Leaf samples were collected from the third youngest leaf using tweezers. A sharp razor blade was used to cut a 1cm x 1cm piece of the leaf lamina which was immediately weighed (fresh mass (FM)). The samples were then floated in distilled water inside a closed petri dish kept at room temperature. The imbibition period lasted for 4 hours before samples were weighed again to obtain turgid mass(TM) and placed in a pre-heated oven (80°C) for 48 hours. After oven drying samples were weighed to obtain dry mass (DM). Values of fresh, turgid and dry mass were used to calculate relative water content as follows:

RWC (%) = $\{(FM - DM)/(TM - DM)\}$ x 100

Equation 4.1

where: RWC = Relative water content,

FM = fresh mass,

DM = dry mass, and

TM = turgid mass.

4.2.4.2 Yield and yield components

Sweet potato plants were harvested at 120 days (4 months) after planting. Measurements recorded included whole plant, above ground and below ground biomass, harvest index (HI) and mass of marketable storage roots. Measurement of root length and diameter was also recorded for marketable roots. Considered marketable storage roots were whole (undamaged) and weighed between 0.1 - 1.4 kg and without harvest wounds, pest and disease damage (Ossom and Rhykerd, 2007). Yield was recorded in tonnes per hectare (t ha⁻¹).

4.2.5 Statistical analysis

Data were subjected to analysis of variance (ANOVA) using GenStat® version 16 (VSN International, Hemel Hempstead, UK 2011). Tukey's HSD test was used to separate means at the 5% level of significance.

4.3 Results

4.3.1 Physiological responses

Different water levels and different cultivars had a significant ($P \le 0.05$) effect on sweet potato plant physiology. Plants irrigated at 30% ETc recorded lower stomatal conductance (SC) than plants irrigated at 100% ETc (Figure 4.1). The interaction between water and cultivar was also significant ($P \le 0.05$) for SC. Cultivar A40 recorded higher ($P \le 0.05$) SC than cultivar A45 and 199062.1. The latter cultivars recorded SC that was not significantly ($P \le 0.05$) different from each other. Higher values of stomatal conductance for cultivar A40 were observed at 100% ETc but tended to decrease towards harvesting time (Figure 4.1).

Chlorophyll content index (CCI) varied significantly ($P \le 0.05$) among sweet potato cultivars (Figure 4.2). Water regimes and the interactions between the former and cultivars were not significant. Cultivars 199062.1 and A45 recorded higher ($P \le 0.05$) CCI than cultivar A40 irrespective of the water regime (Figure 4.2). Although there were no significant differences between the water regimes, it is worth noting that plants grown at 30% ETc recorded generally higher CCI than plants grown under 100% ETc.

Relative water content recorded no significant ($P \le 0.05$) differences for both water regimes and cultivars. Interaction of the two factors was not significant either. However, the recorded values of RWC were high (Figure 4.3) in both water regimes.

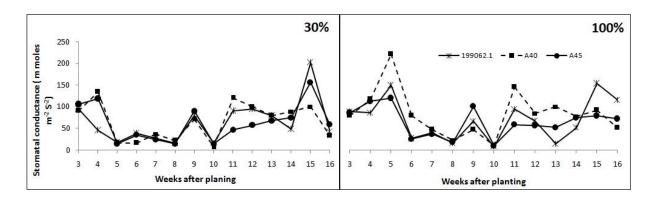


Figure 4.1: Stomatal conductance of three sweet potato cultivars grown under different water regimes.

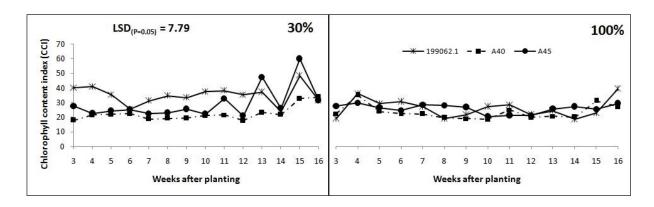


Figure 4.2: Chlorophyll content index of three sweet potato cultivars grown under different water regimes.

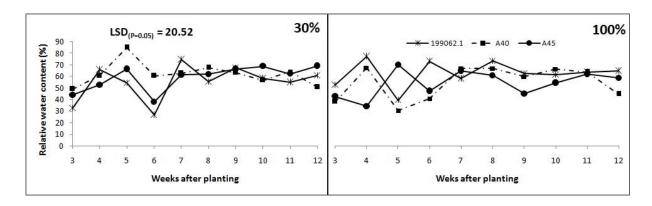


Figure 4.3: Relative water content of three sweet potato cultivars grown under different water regimes.

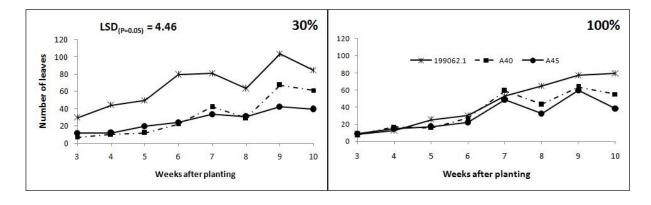


Figure 4.4: Leaf number of three sweet potato cultivars grown under different water regimes.

4.3.2 Plant growth

Vine length and number of branches varied significantly ($P \le 0.05$) among cultivars. The interaction between water regimes and cultivar was significant ($P \le 0.05$). Leaf number, on the other hand, was highly significantly ($P \le 0.001$) influenced by both water regime and cultivar. The interaction between the two factors also had a significant ($P \le 0.001$) effect on number of leaves. Growing sweet potato at 30% ETc resulted in more leaves than plants grown at 100% ETc (Figure 4.4). Cultivar A40 recorded significantly ($P \le 0.05$) longer vines, followed by cultivar A45 and cultivar 199062.1 with the shortest vines (Figure 4.5). The cultivar with the shortest vines (199062.1) branched profusely (Figure 4.6) and recorded significantly high ($P \le 0.05$) number of leaves. Cultivar A40 had more leaves than cultivar A45 but its branches were not significantly different from that of cultivar A45.

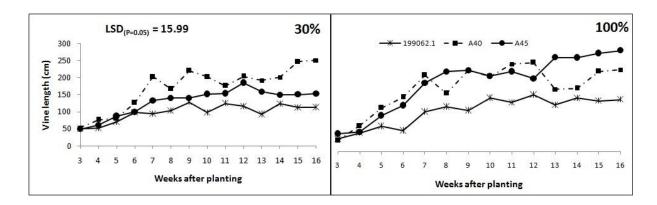


Figure 4.5: Vine length of three sweet potato cultivars grown under different water regimes.

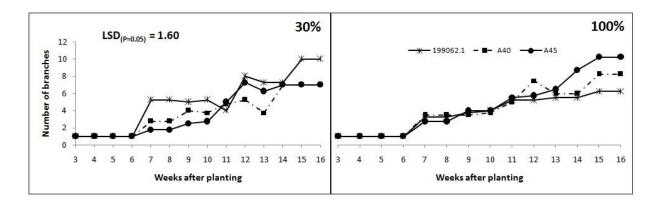


Figure 4.6: Number of branches of three sweet potato cultivars grown under different water regimes.

4.3.3 Yield and yield components

Results of storage roots and storage root girth varied significantly ($P \le 0.05$) among cultivars. Applying water at either level of ETc had no significant effect on storage root yield. The interaction of the two factors was not significant either. Cultivar 199062.1 recorded significantly higher ($P \le 0.05$) storage root yield and thicker roots than the other two cultivars. Storage root yield from cultivars A40 and A45 were statistically similar (Table 4.1). The girth of cultivar A45 was, however, not significantly different from that of cultivar 199062.1.

Biomass yield and harvest index (HI) were significantly ($P \le 0.05$) influenced by cultivar. Cultivar 199062.1 recorded higher biomass and HI than cultivar A40. There was no significant difference in biomass and HI of cultivar 199062.1 and A45. Storage root length showed no variation in response ($P \le 0.05$) to either water regimes or cultivars (Table 4.1). Water regimes did have a significant ($P \le 0.05$) influence on the number of roots per plant. Applying 30% ETc resulted in more storage roots per plant than 100% ETc. Generally, all yield parameters were higher at 30% ETc than at 100% ETc.

Table 4.1: Yield and yield components of three sweet potato cultivars grown under different water regimes.

Water Regime	Cultivar	Biomass (t ha ⁻¹)	Yield (t ha ⁻¹)	Root number plant ⁻¹	Root diameter (cm)	Root length (cm)	HI* (%)
	A40	8.6b	2.9b	3.6b	10.3b	19.6a	35.4b
30% ETc	A45	8.4b	3.3b	3.2b	11.4ab	19.9a	41.5ab
€ H	199062.1	14.3a	7.3a	6.6a	14.7a	18.4a	50.4a
Me	Mean*		4.5a	4.47a	12.3a	19.3a	42.4a
% 0	A40	6.4b	2.2b	2.4a	10.3b	18.8a	32.9a
100% ETc	A45	7.9b	3.6ab	3.2a	12.9ab	19.3a	44.7ab
H	199062.1	8.1b	4.4a	3.6a	13.5a	15.0a	49.9a
Me	ean*	7.4a	3.4a	3.07b	12.3a	17.7a	42.5a
LSD _(P=0.05)	(cultivar)	2.61	1.79	2.29	2.1	3.34	10.04
LSD _(P=0.05)		3.83	2.23	0.54	2.3	3.18	14.10
$LSD_{(P=0.05)}$	(ETc*Var)	4.27	2.68	2.67	2.9	4.50	16.01

^{*}HI = harvest index. Values in the same column sharing the same latter are not significantly different at LSD $_{(P=0.05)}$. Mean* = Significant differences for water level mean.

4.4 Discussion and conclusions

Sweet potato's physiological response at 30% ETc indicated that the plants were experiencing water stress. Plants responded by low SC meaning that low water availability caused stomata to close in order to limit carbon dioxide flow and transpiration losses. This behaviour has been characterised as a drought avoidance mechanism (Farooq et al., 2009). According to Mabhaudhi et al. (2013), SC decreases with increasing water stress. Observations made in this experiment concur with the authors' reports. Sweet potato cultivars grown under 30% ETc recorded low SC values than those grown under 100% ETc. Relative water content (RWC) results followed the same trend as SC results. Even though RWC was not significantly different between water regimes, the 30% ETc recorded lower values than the 100% ETc. Relative water content indicate the balance between absorbed water by plant and consumed through transpiration, therefore, high RWC indicates more resistance to drought stress (Schonfeld et al., 1998). Since there was no significant difference in RWC between the water regimes, it suggests that these sweet potato cultivars are highly tolerant to water stress.

Orange-fleshed cultivars A45 and 199062.1 showed greater stomatal regulation to water stress than cultivar A40, the only white-fleshed cultivar used in this study. This cultivar (A40) was the only cultivar that recorded SC values above the 100 mmol m⁻² s⁻¹ threshold for C3 plants (Flexas et al., 2006). Otherwise the other two cultivars (A45 and 199062.1) recorded values below 100 mmol m⁻² s⁻¹. Stomatal conductance below the threshold led to metabolic impairment in plants (Flexas et al., 2006). Van Heerden and Laurie (2008) also reported low SC (13 mmol m⁻² s⁻¹) from sweet potato cultivars grown under water stress. These authors (van Heerden and Laurie, 2008) further reported that plants avoided metabolic impairment through a mechanism called photorespiration. According to Lawlor and Cornic (2002), photorespiration is the second most important mechanism for removing excess electrons (that may damage photosynthetic apparatus) in C3 plants.

Plant growth was mostly affected by varietal differences than water regimes. Water stress did not have a profound influence on the growth of these cultivars. The results only show leaf number to be sensitive to water availability albeit in an interesting manner. Plants grown under 30% ETc produced more leaves than plants grown under 100% ETc. Branching and vine length was a result of varietal differences. The high number of leaves on plants grown under 30% ETc was another interesting result since reports indicate that plants' morphological characteristics are negatively affected by water stress (Farooq et al., 2009). A

high number of leaves consequently increase the photosynthesis hub. Since the canopy represents the only source of biomass for subsequent partitioning to storage root, this would therefore, improve source sufficiency for yield attainment. Although the number of leaves was high, these cultivars did limit vine extension and branching which resulted into short bushy-growth. Bushy-growth translated to reduced leaf area index (LAI) which is characteristic of drought avoidance in plants (Blum, 2005). It is also possible that sweet potato plants may have developed a ramified root system (Jaleel et al., 2009) which allowed them to support above ground dry mass accumulation. This observation was similar to reports by Saraswati et al. (2004) that water stress reduced stem extension and internodes diameter of sweet potato cultivars; the authors did not mention any increases in leaf number but a decrease in LAI. Leaf size should be of main focus in sweet potato as earlier research reported that it had a negative correlation with apparent photosynthesis (Bhagsari and Brown, 1986). Cultivars with small canopies, short stem length and small leaves were reported by Wilson (1982) to have the capability of giving higher yields than those with long stems and numerous broad leaves. This was later confirmed by Saraswati et al. (2004). Plants grown under 100% ETc had low leaf number but longer vines and more branches. This did not however increase total biomass, instead total biomass was increased in plants grown under 30% ETc.

Storage roots yield recovered from the 30% ETc was higher than in 100% ETc even though the plants under 30% ETc had a lower LAI. Plants grown under 100% ETc may have been experiencing rank growth as reported in cotton (North Carolina State University 2012) where biomass partitioning favoured vegetative than storage root growth. At low water availability (30% ETc), the small canopy tended to partition assimilates more towards storage roots thus higher storage root yield. Generally, all yield parameters were higher at 30% ETc than 100% ETc. Based on these results, the sweet potato cultivars were more drought tolerant. Reports by Mohankumar (2000) also concur with the fact that deficit irrigation strategies such as irrigating sweet potato plants once they had depleted 40% of available soil water tended to give higher yields. Withholding water until 60 days after planting (DAP) was also recommended in order to provide translocation of more photosynthates to the roots and the development of storage roots (Ekanayake et al., 1990). Another contributing factor to the high yield under water stress in this experiment could be the fact that water stress was constant throughout the growth cycle such that plants adapted to low supply of water and maximized its production. This means that if low levels of water are evenly distributed, sweet potato will give higher yields. While 30% ETc was a very low amount of water, the fact that it was applied daily ensured there was some water as opposed to no water at all. Unlike in the field where plants will be subjected to slowly developing water deficit and sometimes subjected to near dryness. Under such conditions drought stress can strike the crop at the most vulnerable growth stage, thus affecting growth and subsequent yield. It would be advisable for farmers to alter their planting dates so that it coincides with even distribution of rainfall and the mid-summer drought coincides with the growth stage that requires a bit of water stress as reported by Mohankumar (2000). The yield results also suggest that these cultivars can be grown in marginal areas, as long as there is even distribution of the low water supply.

When comparing cultivars, it was observed that cultivar 199062.1 was high a yielding cultivar despite water stress. It also displayed the best growth characteristics in terms of branching and number of leaves. Moreover, it contained high CCI and responded to drought stress by reducing SC. These are some of the attributes that are highly appreciated when breeding cultivars for drought tolerance. Cultivar A40 was expected to give higher storage yield since photosynthesis was not substrate constrained due to higher SC results. However, enzyme kinetics does concur with the response of cultivar A40 that if there is excess substrate, enzymatic reactions are limited (Reed et al., 2010). In this case it is suspected that the high levels of CO₂ became the source of inhibition to photosynthesis thus the low yields. Cultivar A45 on the other hand was mostly impartial to these treatments.

Physiological traits of the sweet potato plants reported in this experiment indicated that it is indeed drought tolerant. Stomatal conductance of these cultivars was very low, mostly below the 100 mmol m⁻² s⁻¹ threshold. To avoid metabolic impairment at such low SC values, the cultivars resorted to photorespiration. Growing these cultivars under 30% ETc gives similar results of RWC suggesting that the cultivars are drought tolerant. The cultivar (A40) with high SC but low CCI values indicated substrate inhibition for photosynthesis, which is why it gave lower yields. Water stress limitation to plants growth was to a lesser extent. Leaf number was the only parameter reduced by water stress. This consequently reduced its canopy indicating drought stress avoidance mechanism but gave higher yields than plants with long stems and numerous broad leaves grown under 100% ETc. This indicates that these cultivars are suitable for production under marginal areas as long as there is even distribution of the low water supply. Optimum water supply (at 100% ETc) encourages rank growth on these cultivars, this encourages famers to alter their planting dates such that the stages of growth where plants have to be exposed to water stress coincides with mid-summer drought to break

the rank growth syndrome and increases storage root yield. Cultivar 199062.1 was more tolerant to drought and gave higher yield than the other two cultivars.

CHAPTER 5

EVALUATION OF SELECTED NUTRIENTS AND PHYTOCHEMICALS OF SWEET POTATO (*IPOMEA BATATAS* L.) GROWN IN DIFFERENT AGROECOLOGICAL AREAS

Abstract

Sweet potato's nutritional value outranks most carbohydrate crops. The biochemicals that improve its nutritional quality may be altered by differences in environmental conditions where the crop is grown. A study was done to determine the effect of different agroecological zones (Coast hinterland thornveld; Moist coast, forest, thorn and palm-veld; Moist coast hinterland and ngongoni veld) on starch and β-carotene content and total antioxidants capacity of leaves and storage roots. Chlorophyll and total carotenoids concentration was also determined. Metabolic analysis were conducted on three sweet potato cultivars using FRAP, DPPH, spectrophotometry and enzymatic analysis to determine total antioxidants activity, βcarotene, phytochemicals and starch respectively. Results showed that differences in growth environments altered biochemical content of sweet potato leaves and storage roots. Starch and β-carotene increased under favourable growth environments as displayed by the moist coast hinterland and ngongoni veld. Total antioxidant activity was high in leaves as compared to storage roots. Agro-ecological locations experiencing pronounced drought stress further increased total antioxidant activity in storage roots. Presence of high antioxidant activity even in storage roots makes sweet potato even more important for human nutrition as it can double up as a nutritious food crop with medicinal benefits. Carotenoids and chlorophyll content ware higher in plants grown under favourable environment and decreased in drought stressed locations. This indicated that carotenoids are not always an indicator of drought stress. Differences in climatic conditions do affect sweet potato nutrition and phytochemical content. While favourable conditions improve nutrient content while drought stress increases phytochemical content. Sweet potato leaves and storage roots has a potential to improve food security and human health. The leaves can be consumed as a vegetable while waiting for the storage roots to mature.

Keywords: agro-ecological zones, nutrients, phytochemicals

5.1 Introduction

Sweet potato is an important food security crop and source of essential nutrients, including vitamin A in the orange-fleshed cultivars (Kulembeka et al., 2004, Agili et al., 2012, Laurie et al., 2012). It is highly nutritive, and it outranks most carbohydrate crops in terms of vitamin, minerals, dietary fibre and protein content (Bovell-Benjamin, 2007). It is one of the crops that can still be used to combat malnutrition in developing countries. According to Nafeesa et al. (2012), sweet potato is still grossly underutilised despite its versatility and rich nutritional benefits. Approximately 24.8% of the population in Sub-Saharan Africa are still undernourished (FAO, 2013) and South Africa is no exception. Rural communities are the most affected in South Africa (Wenhold et al., 2012) probably due to disproportionate nutrition uptake. This has led to limited progress toward reaching the first Millennium Development Goal (MDG1) in South Africa (Engesveen et al., 2009).

Agricultural crops that can deliver quick results to address MDG1 are still needed and crops like sweet potato have a quick turnover (Ebregt et al., 2007) and high nutrition in terms of energy and micronutrients (Bovell-Benjamin, 2007). A study by Ahmad et al. (2011) reported that consumers are increasingly conscious about their health and have since started to consider the nutritional benefits, disease prevention and health-promoting properties of foods. This consumer consciousness calls for a critical evaluation of the nutritional composition and health-promoting potential of food crops in order to satisfy their needs. Food security crops like sweet potato, which may still be consumed unprocessed (especially in Africa) (van Jaarsveld et al., 2005) are being promoted since they may contain some health-promoting properties. Health campaigns such as dietary diversification (van Jaarsveld et al., 2005; Laurie and van Heerden, 2012) targeted at promoting traditional and underutilized crops (Modi and Mabhaudhi, 2013) are aimed at improving awareness of rural communities about the importance of healthy diets. Information on other beneficial compounds found in sweet potato roots besides the already known nutrients (starch content, mineral content and β-carotene content) is envisaged to further increase appreciation and consumption. Human consumption of other plant parts such as the leaves can also improve household food security.

Consumption of sweet potato leaves as a vegetable is uncommon in South Africa. A recent study by Laurie and van Heerden (2012) reported good acceptability of utilizing beta-carotene-rich sweet potato leaves as a leafy vegetable among surveyed communities in South

Africa. This highlighted the need to promote the consumption of sweet potato leaves. According to Islam (2006), sweet potato leaves have largely been neglected except for being partially use as livestock feed. The leaves are reported to have high nutrient content and higher concentrations of polyphenols compared with major commercial vegetables such as spinach, broccoli, cabbage and lettuce (Islam, 2006; Mbaeyi-Nwaoha and Emejulu, 2013). Whether these concentrations (polyphenols and nutrients) increase or decrease with response to differences in growth environmental conditions, is still not clear. Thus, there is a need to investigate that.

The absence of empirical information reporting on other chemical responses of sweet potato to differences in environmental conditions presented by different agro-ecological areas in Southern Africa prompted this study. The objective of the study was therefore, to determine the effect of production agro-ecological area on starch and β -carotene content and total antioxidant capacity of storage roots as well as antioxidant activity, total carotenoids and chlorophyll content of young leaves suitable for use as food from three sweet potato cultivars.

5.2 Materials and methods

5.2.1Plant material

Three sweet potato cultivars (A40, A45 and 199062.1) were obtained from the University of KwaZulu-Natal's plant breeding programme. All three cultivars were planted in November at three different agro-ecological locations (Deepdale, Umbumbulu and Richards Bay) during the 2012/13 and 2013/14 planting seasons. Differences in environmental conditions of the three agro-ecological locations are shown in Table 5.1. Planting was done on ridges (~30 cm high) in a randomized complete block design with three replications per location. In all locations, sweet potato was planted on land that had been fallow for at-least one year (Table 5.1). Soil samples were taken prior to planting and analysed for fertility and textural characteristics (Table 5.1). No fertilizer, pesticides or supplementary irrigation was applied for the duration of crop growth at all experimental sites.

Details of weather parameters (maximum and minimum temperatures, reference evapotranspiration and rainfall) were obtained from the Agricultural Research Council –

Institute for Soil, Climate and Water's network of automatic weather stations (Table 5.2). Agronomic conditions and crop management were kept identical in all locations throughout the growth period. Harvesting was conducted at 120 days after planting where marketable storage roots (100-500 g) and young leaf material (up to the fourth youngest leaf) suitable for cooking were selected for chemical evaluation.

Table 5.1: Experimental site description for Deepdale (Agro-ecological/bioresource group 17), Richards Bay (Agro-ecological/bioresource group 1) and Umbumbulu (Agro-ecological/bioresource group 3).

	Deepdale	Richards Bay	Umbumbulu
			Moist coast
Agro-ecological area	Coast hinterland	Moist coast, forest,	hinterland and
(Bio-resource group)	thornveld	thorn and palm-veld	ngongoni veld
Geographical location	28°01'S; 28°99'E	28°19'S; 32°06E	29°98'S; 30°70'E
Altitude (m a.s.l.) ^z	998	30	632
Annual rainfall	7500 – 850 mm	820 – 1423 mm	800 – 1160 mm
Average temperature	18.4°C	22°C	17.9°C
Frost occurrence	moderate	None	Light and occasional
*Soil texture class	clay	Sand	Clay
Clay content	53%	< 5%	>60%
Soil pH	4.4	3.6	4.2
*Soil type	Jonkersberg form (Jb)	Inhoek form (Ik)	Hutton form (Hu)
Soil depth	1.0 m	> 1.0 m	1.0 m
Nitrogen (%)	0.4	0.1	0.3
Phosphorus (mg/L)	5.0	6.0	1.0
Potassium (mg/L)	349.0	18.0	62.0
Organic carbon (%)	4.4	< 0.5	3.4
Previous crop	Fallow (3 years)	Fallow (1 year)	Fallow (1 year)

^{*}Soil Classification, a Taxonomic System for South Africa 1991; ^z metres above sea level.

 Table 5.2: Climatic conditions during the growth season in the different locations.

		Richards Bay				Umbumbulu				Deepdale						
		Temp	(°C)		(mm)		Temp	Temp (°C) (mm)		Temp(°C)		(mm)				
Season	Months	Max	Min	Rain	ETo	Deficit	Max	Min	Rain	ETo	Deficit	Max	Min	Rain	ETo	Deficit
	November	27.0	17.2	66.9	95.7	28.8	21.3	13.9	165.2	70.6	-94.6	24.2	13.6	117.1	2.9	-114.2
13	December	29.7	9.2	214.7	127.6	-87.1	25.5	16.6	64.7	99.8	54.1	28.5	16.2	76.4	3.8	-72.6
2012/13	January	30.6	19.3	145.4	121.1	-24.3	25.5	16.9	105.6	98.8	-6.8	28.5	16.5	146.9	3.7	-143.2
70	February	30.6	19.2	60.8	119.8	59.0	25.9	16.6	84.5	90.4	5.9	29.8	16.2	89.3	3.8	-85.5
	March	29.3	18.2	75.4	103.1	27.7	24.6	15.6	137.4	89.6	-47.8	28.1	15.4	37.2	3.0	-34.2
				563.2*	567.2*				557.4*	449.2*				466.9*	17.2*	
	November	28.6	18.0	134.0	192.8	58.8	21.3	15.0	117.5	102.0	-15.5	28.0	14.0	84.0	117.5	33.5
14	December	27.2	18.5	114.0	121.7	7.7	23.1	15.5	69.8	97.0	27.2	26.2	15.2	84.4	102.6	18.2
2013/14	January	30.9	20.6	24.8	164.4	139.6	26.2	17.3	119.5	117.8	-1.7	30.2	16.8	108.9	135.5	26.6
70	February	31.7	20.7	30.4	150.2	119.8	26.2	17.3	50.7	107.6	56.9	30.8	16.9	49.1	115.6	66.5
	March	29.4	19.9	157.5	119.6	-37.9	25.7	16.2	85.0	100.5	15.5	29.0	15.7	135.1	99.7	-35.4
				460.7*	748	3.7*			442.5*	524	1.9*			461.5*	57	0.9*

^{*} Rainfall and ETo were totalled not averaged.

5.2.2Analytical procedures

5.2.2.1 Determination of total antioxidant capacity

2.2.1.1 DPPH free radical scavenging assay

Extraction was carried out using perchloric acid. Freeze-dried and ground plant material (leaves and storage roots) of 0.2 g each was mixed with 10 ml of perchloric acid and allowed to stand for 10minutes. The mixture was vortexed for 1 minute before being centrifuged at 12400 x g at 4°C for 10 minutes. The DPPH free radical scavenging activity of each sample was determined according to Wong et al. (2006). Briefly, a 0.1 mM solution of DPPH in methanol was prepared. The initial absorbance of the DPPH in methanol was measured at 515 nm and did not change throughout the period of assay. An aliquot (150 μl) of the extract was added to 2 ml of methanolic DPPH solution. The change in absorbance at 515 nm was measured for 30 min. The antioxidant capacity based on the DPPH free radical scavenging ability of the extract was expressed as μmol Trolox equivalents per 100 gram of plant material on a dry matter basis.

5.2.2.1.2 Ferric reducing ability of plasma (FRAP) assay

Extraction was done as described for the DPPH free radical scavenging assay. The ability to reduce ferric ions was measured using a modified version of the method described by Wong et al. (2006). An aliquot (200 μl) of an extract was added to 2 ml of FRAP reagent (10 parts of 300 mM sodium and 1 part of 20 mM sodium acetate buffer at pH 3.6, 1 part of 10 mM TPTZ solution and 1 part of 20 mM FeCl₃.6H₂O solution). The reaction mixture was incubated in a water bath set at 37°C. The increase in absorbance was measured for 30 minutes at 593 nm. The antioxidant capacity based on the ability to reduce ferric ions of the extract was expressed as μmol Trolox equivalents per 100 gram of plant material on a dry matter basis.

5.2.2.2 Determination of total carotenoid and chlorophyll concentration

Carotenoids and chlorophyll were extracted and determined according to Lichtenthaler (1987) in a dark room. Freeze-dried, ground sweet potato leaf material (0.2 g DM) was placed in centrifuge tubes with 10ml extracting solvent (80% acetone). The mixture was allowed to stand on ice covered with aluminium foilfor10 minutes. Thereafter, the mixture was homogenized with Ultra-Turrax using a 1 minute burst twice and centrifugation at 12500 x g for 5 minutes. The supernatant was decanted into 2ml cuvettes and absorbance read at various wavelengths (663.2 nm, 646.68 nm and 470 nm) as this was a simultaneous extraction of

carotenoids and chlorophyll. Calculations and conversions were done using the equations below (Lichtenthaler, 1987):

$$C_a = (12.25 \text{ x A}_{663.2}) - (2.79 \text{ x A}_{646.8})$$
 Equation 5.1

$$C_b = (21.50 \text{ x } A_{646.8}) - (5.10 \text{ x } A_{663.2})$$
 Equation 5.2

Ca+b =
$$(7.15 \times A_{663.2}) - (8.71 \times A_{646.8})$$
 Equation 5.3

$$C_{x+c} = (1000 \text{ x A}_{470}) - (1.82 \text{ x C}_a) - (85.02 \text{ x C}_b)/19$$
 Equation 5.4

where: $C_a = Chlorophyll a$,

 $C_b = Chlorophyll b$,

 $C_{a+b} = Total Chlorophyll, and$

 C_{x+c} = Total carotenoids.

5.2.2.3 Pigment extraction for β -carotene analysis

The sampling of sweet potato roots for β-carotene was done as described by Rodriguez-Amaya and Kimura (2004). Extraction was then carried out according to the method of the Association of Official Analytical Chemists (AOAC, 1980) with a slight modification. A 5 g sample of macerated sweet potato storage root (with 1 mg of BHT) was added to a conical flask containing 50 ml of 95% ethanol and maintained at a temperature of 70-80°C in a water bath for 20 minutes with periodic shaking. The supernatant was decanted, allowed to cool and its volume was measured and recorded as initial volume. The concentrated ethanol extract was diluted to 85% by adding distilled water and it was further cooled on ice for about 5 minutes. In a separating funnel, 25 ml of petroleum ether was added and the cold ethanol extract was poured over it. The funnel was swirled gently to obtain a homogenous mixture and thereafter allowed to stand until two separate layers were obtained. The bottom layer was run off into a beaker while the top layer was collected into a 250 ml conical flask. The bottom layer was transferred into the funnel and re-extracted with 10 ml of petroleum ether for 5-6 times until the extract became fairly clear. The final extract was measured and transferred into sample bottles before being read at 450 nm.

The concentration of β -carotene was calculated using Beer-Lamberts Law, which states that absorbance (A) is proportional to the concentration (C) of the pigment, as represented by the equation:

A ∞ L (if concentration(C) is constant).

A=ECL; C=A/EL

Equation 5.5

where: C= concentration of carotene,

A= absorbance,

E= extinction coefficient,

L= thickness of cuvettes (path length) = 1cm, and

E of β-carotene = 1.34×10^5 l/mol cm (M=537 g/mol).

5.2.2.4 Starch content determination

Starch was determined using the enzymatic method of Weinmann (1947) with modifications. Freeze-dried, ground material (0.10 g DM) was mixed with 10 ml 80% (v/v) ethanol and homogenized for 60 seconds. Thereafter, the mixture was incubated in a water bath set at 80°C for 60 minutes. Supernatant was suctioned off. These steps were repeated twice then cooled before samples were dried in a Savant Vacuum Concentrator (SpeedVac, Savant, NY, USA).Warm (40 – 50°C) acetate buffer (10 ml) and 200 µl of hexakinase were added to each sample then incubated at 90°C for 30 minutes. Samples were allowed to cool at room temperature before adding 200 µl of G6P-dehydrogenase (G6P-DH) then incubated at 60°C for 20 hrs. Thereafter, samples were vortexed and diluted with distilled water to 200 ml, filtered through Whatman filter paper No. 541. An aliquot (200 µl) of the filtered sample was then taken and diluted further with distilled water to a total of 3 ml. Copper reagent (5 ml) was then added to each sample vortexed and placed in boiling water bath for 20 min. Arsenomolybdate (5 ml) was then added to each sample after cooling, vortexed and left to stand at room temperature for one and a half hours. Samples were diluted (with distilled water) to 200 ml, agitated and read at 750 nm.

5.2.2.5 Measurement of absorbance

Absorbance was measured using a UV–1800 UV–Vis spectrophotometer (Shimadzu, North America) at the respective wavelengths as indicated in the procedures.

5.2.3 Statistical analyses

Data were subjected to analyses of variance (ANOVA) using GenStat® version 14 (VSN International, Hemel Hempstead, UK 2011). Tukey's HSD test was used to separate means at the 5% level of significance.

5.3 Results

5.3.1 Antioxidant activity of the three sweet potato cultivars

Results of the two antioxidant activity assays (FRAP and DPPH) showed significant differences (P < 0.05) among locations, sweet potato cultivars and seasons (Table 5.3). Interaction among all three factors was also significant (P < 0.001) (Tables 5.3 and 5.4). The reducing power (measured using the FRAP essay) and DPPH free radical scavenging activity of storage roots harvested at Richards Bay was higher than for those harvested from Umbumbulu and Deepdale, respectively (Table 5.2). Sweet potato leaf extracts did not follow the same trend. Instead leaves from Deepdale recorded higher (P < 0.001) scavenging activity (Table 5.4) followed by Richards Bay then Umbumbulu. The reducing power of leaf extracts was higher for Umbumbulu sweet potato leaves, whilst Deepdale sweet potato leaves had very low reducing activity.

The leaves of cultivar A40 had high antioxidant activity for both essays. Leaves of cultivars A45 and199062.1 varied within the antioxidant activity assays used. Whilecultivar199062.1 had relatively higher reducing power, cultivar A45 had relatively higher radical scavenging activity. The storage roots followed the same trend when subjected to FRAP assay. Roots of cultivar A40 recorded more reducing powers followed by cultivar 199062.1 then A45. It was also observed that ferric ion reducing activity of the leaves was consistently higher than that of the roots while the opposite was true with the DPPH free radical scavenging. The antioxidant activity of sweet potatoes grown during 2013/14 was higher than those grown in 2012/13.

Table 5.3: Total antioxidant activity (μ Mol TE/100 g) of three sweet potato cultivars harvested from different locations determined by DPPH free radical scavenging activity.

			odale	Richard	ds Bay	Umbumbulu		
Season	Cultivars	Leaves	Roots	Leaves	Roots	Leaves	Roots	
3	A40	647.8±0.25	631.9±0.18b	164.3±3.93	587.0±0.05c	268.5±1.04	559.8±0.62c	
2012/13	A45	620.6±0.12	632.5±0.23a	484.4±0.06	619.0±0.26b	309.5±1.21	616.3±0.42a	
200	199062.1	590.2±0.23	629.8±0.38c	443.7±0.23	621.5±0.19a	305.6±1.47	596.7±0.08b	
N	Mean	610.1	631.4	1272.2	609.2	1521.0	590.9	
4	A40	1004.2±10.4	808.0±1.46	1135.1±18.07	1007.4±5.09	993.8±16.8	885.0±0.82	
2013/14	A45	890.4±1.42	864.0±0.41	802.5±1.41	825.9±1.95	870.1±1.45	1003.9±7.09	
20	199062.1	865.7±2.75	950.2±3.69	786.5±1.45	1048.5±3.75	910.0±41.8	988.5±8.16	
N	J ean	1608.1	874.1	1542.2	960.6	1446.2	959.1	
LSD	(cultivar)	1.57	1.76		<u> </u>		1	
LSD	(location)	1.57	1.76					
LSD	(Season)	1.28	1.44					

4.31

3.84

LSD (cultivar x season x location)

^{*}Means in each column with different letters are significantly different (P < 0.05)

Table 5.4: Total antioxidant activity (μMol TE/100g) of three sweet potato cultivars harvested from different locations determined by FRAP essay.

			Deepdale		ds Bay	Umbumbulu		
Season	Cultivars	Leaves	Roots	Leaves	Roots	Leaves	Roots	
8	A40	521.2 ± 0.15 b	178.2±0.03b	1611.1 ± 1.78a	621.7 ±0.89a	$1882.2 \pm 0.84a$	$355.2 \pm 0.08c$	
2012/13	A45	$473.2 \pm 0.14c$	178.0 ± 0.05 b	1274.1± 3.97b	254.3 ± 0.21 b	$1417.1 \pm 1.02b$	$263.4 \pm 0.12b$	
20	199062.1	$836.0 \pm 0.19a$	$256.4 \pm 0.47a$	931.3± 0.28c	$242.4 \pm 0.05c$	$1263.2 \pm 0.26c$	$437.8 \pm 0.21a$	
M	Mean		204.2c	1272.2b	372.8a	1521.0a	352.1b	
4	A40	1004.2± 8.29a	$1399.7 \pm 2.62b$	1135.1± 1.03a	1105.3 ± 3.66 b	$1598.5 \pm 0.07a$	1001.3 ± 0.44 b	
2013/14	A45	890.4± 0.07b	329.8± 0.28c	802.5 ± 0.16 b	$1394.1 \pm 0.83a$	$1180.0 \pm 3.65c$	$841.2 \pm 3.84c$	
20	199062.1	865.7± 0.65c	1506.5± 3.86a	$786.5 \pm 0.71c$	842.9 ± 1.08c	1561.6 ± 0.97 b	$1247.8 \pm 1.72a$	
M	ean	1608.1a	1078.7b	1542.2b	1114.1a	1446.7c	1030.1c	
LSD (cultivar)	1.57	1.23					
LSD (LSD (location)		1.23					
LSD (Season)	1.28	1.00					

3.01

LSD (cultivar x season x location)

3.84

^{*}Means in each column with different letters are significantly different (P < 0.05)

5.3.2 Total carotenoid, β-carotene and chlorophyll content

Total carotenoid, β -carotene and chlorophyll content showed significant (P < 0.001) differences across locations, sweet potato cultivars and seasons. The interaction between all three factors was significant (P < 0.001) (Table 5.5 and 5.6). With respect to differences between locations, storage roots harvested at Umbumbulu contained significantly (P < 0.001) higher β -carotene relative to the storage roots harvested from Deepdale and Richards Bay, respectively. A similar trend was observed for carotenoids and chlorophyll content. These variables (β -carotene, chlorophyll and total carotenoid content) tended to favour the "Moist coast hinterland and ngongoni veld" agro-ecological location (Table 5.1) at mid-elevation (632 m a. s. l.). Very low values were recorded from plants grown at the lowland coastal location (Richards Bay, 30 m a. s. l.). (Tables 5.1, 5.5 and 5.6).

When comparing cultivars, it was observed that cultivar A45 had significantly (P<0.001) higher total carotenoid, β -carotene and chlorophyll content relative to cultivars 199062.1 and A40, respectively. The two orange fleshed sweet potato cultivars (A45 and 199062.1) recorded similar amount of β -carotene content at Richards Bay during the year 2012 (Table 5.6). Significant (P<0.001) increases in β -carotene content on cultivar A45 was only recorded during year 2013 at Richards Bay. Cultivar A40 had the lowest concentrations of β -carotene irrespective of the location. The two phytochemicals (chlorophyll and carotenoids) tended to fluctuate with respect to seasons. Both phytochemicals concentration was high during 2013 than 2012. A positive correlation ($r^2 = 0.76$; p< 0.05) between total carotenoids and chlorophyll contents was also observed, but no relationship between these two phytochemicals in the leaves and β -carotene in the storage roots (Table 5.6).

5.3.3 Starch content

Starch content of the three sweet potato cultivars was significantly (P<0.001) affected by location, seasons and differences in cultivars. The interaction between these factors was also significant (P<0.001). Storage roots planted at Umbumbulu recorded significantly (P<0.001) high starch content than storage roots from Deepdale and Richards Bay (Figure 5.1). Starch content from the two locations (Deepdale and Umbumbulu) was the same. Cultivar A45 had high starch content followed by cultivar A40 then cultivar 199062.1. Growing sweet potato cultivars during 2013 resulted in high starch content than 2012.

Table 5.5: Phytochemicals of three sweet potato cultivars grown in three different locations.

	Sweet potato	Tot	al carotenoids (m	g/g)		Chlorophyll (µg/g)
Season	cultivars	Deepdale	Umbumbulu	R. Bay	Deepdale	Umbumbulu	R. Bay
3	A40	881.8±0.51c	3935.8±0.04c	2574.8±0.24c	3.8±0.01c	33.0±0.02a	18.9±0.01a
2012/1:	A45	3923.0±0.13a	3017.2±4.01a	2269.5±1.13a	44.7±0.05a	24.0±0.01c	15.4±0.00c
20	199062.1	2692.6±0.61b	3147.9±4.70b	2253.9±1.70b	24.3±0.01b	25.1±0.03b	16.8±0.01b
Mean		2499.1b	3367.0a	2366.1c	24.3b	27.4a	17.0c
4	A40	$3922.4 \pm 0.03a$	$3917.8 \pm 0.04a$	2213.6 ± 1.39b	$51.0 \pm 0.03b$	$53.7 \pm 0.01a$	18.3 ±0.02c
2013/14	A45	$3922.4 \pm 0.04a$	$3822.1 \pm 0.03b$	$3924.1 \pm 0.03a$	$51.0 \pm 0.03b$	51.3 ± 0.01 b	47.8 ± 0.03 b
20	199062.1	$3921.9 \pm 0.07a$	$3446.1 \pm 9.62c$	$3923.0 \pm 0.04a$	$51.3 \pm 0.05a$	$44.7 \pm 0.01c$	$49.5 \pm 0.04a$
Me	ean	3922.2a	3762.0b	3353.6с	51.1a	49.9b	38.5c
LSD c	ultivar	1.86			0.02		
LSD lo	LSD location				0.02		
LSD s	LSD season				0.01		
LSD cultivar x le	ocation x season	4.55			0.04		

^{*}Means in each column with different letters are significantly different (P < 0.05)

Table 5.6: β -carotene content and REA for sweet potato cultivars grown in different locations of KZN.

	Sweet potato	β·	carotene (mg/100	g)		RAE ^z (mg/100g)	AE ^z (mg/100g)		
Seasons	lines	Deepdale	Umb	R. Bay	Deepdale	Umb.	R. Bay		
8	A40	2.9±0.05c	0.2 ± 0.05 c	0.98±0.01b	0.2±3.93c	0.0±4.31c	0.1±8.36b		
2012/13	A45	23.2±0.02a	19.8±0.05a	2.3±0.03a	1.9±1.98a	1.6±4.57a	0.2±2.51a		
20	199062.1	10.9±0.0b	5.3±0.10b	2.3±0.08a	0.9±0.56b	$0.4 \pm 8.05 b$	0.2±6.67a		
Me	Mean		8.4b	1.8c	1.0a	0.7b	0.2c		
4	A40	0.6±0.02c	0.4±0.05c	0.5±0.02c	0.0±0.00c	0.0±0.00c	0.0±0.00c		
2013/14	A45	9.2±0.01a	29.6±0.05a	7.3±0.01a	0.8±0.02a	2.5±0.01a	0.6±0.01a		
20	199062.1	6.1±0.03b	7.9±0.10b	4.2±0.04b	0.5±0.02b	0.7 ± 0.01 b	0.4±0.02b		
Me	ean	5.3b	12.6a	4.0c	0.4b	1.1a	0.3c		
LSD c	ultivar	0.02			0.00				
LSD lo	LSD location				0.00				
LSD s	LSD season				0.00				
LSD cultivar x l	ocation x season	0.06			0.00				
	*								

² Retinol activity equivalents. *Means in each column with different letters are significantly different (P < 0.05).

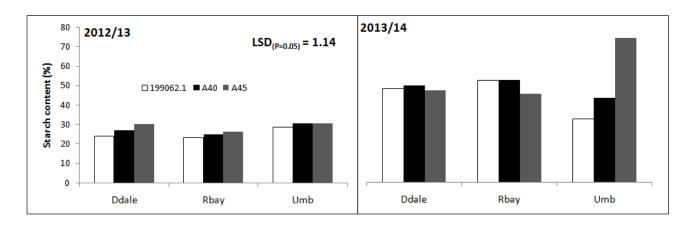


Figure 5.1: Stanch content of different sweet potato cultivars grown at different locations during 2012 and 2013 growing season.

5.4 Discussion and conclusions

Agro-ecological locations and season had a marked influence on the contents of starch, β -carotene, total carotenoid and chlorophyll content of sweet potato cultivars as well as their antioxidant activity. This is an indication that environmental conditions do affect chemical composition of sweet potato (both leaves and storage roots). All aspects of sweet potato nutrition and phytochemicals were higher during 2013 than 2012 growing season. The moist coast hinterland and ngongoni veld (Smith, 2006) where Umbumbulu is located seemed to favour nutritional status of the sweet potato cultivars. This location recorded high starch and β -carotene content suggesting that sweet potato can play an important role both as an energy source and vitamin A supplement and in human nutrition in that area. The starch content was within the range of \leq 30% as reported by Bovell-Benjamin (2007). Fluctuation in its content between locations may be an indication that these nutrients are sensitive to changes in environmental conditions.

Temperature and evapotranspiration (ET_o) (water lost to the atmosphere through surface evaporation and plant transpiration) were the only dominant factors different between these locations. Umbumbulu had an average temperature of 24.5°C during both growing seasons and low ETo with the exception of Deepdale during 2012/13 growing season. The agroecological area of Umbumbulu experiences frequent mist during summer season and this may have contributed to soil water conservation by reducing ETo (FAO, 1989). Average temperatures at Deepdale and Richards Bay were 28.2°C and 29.6°C, respectively which were

3.8°C and 5.1°C, respectively, higher than that of Umbumbulu. An increase in temperature is known to affect partitioning of dry matter in plants (Rotter and van de Geijn, 1999). The low starch content in storage roots from these areas may have been affected by increases in temperature thus starch partitioning and allocation to storage roots was decreased. A similar trend was observed in β-carotene content.

Secondary metabolites, such as phenolics, are generally expected to increase under drought stress conditions and this is believed to be a response to an increase in oxidative damage (Podsedek, 2007; Oh et al., 2009). Richards Bay had high ETo and high temperatures suggesting that there was a water deficit (drought stress). Storage roots harvested from that location recorded high antioxidant activity but the leaves did not follow the same trend as the storage roots. Instead total antioxidant activity was high in the leaves from Deepdale and Umbumbulu. This means that the roots were suffering from prolonged deprivation of water which led to a saturated redox chain and decline of generated adenosine triphosphate (ATP). This is called oxidative stress and it released toxic free radicals which can inactivate enzymes involved in oxidative systems in the plant (Lin et al., 2006). High antioxidant activity means that toxic radicals were being removed to protect plant cells against oxygen toxicity (Perata and Alpi, 1993). Comparison of antioxidant activity of storage roots and leaves revealed that sweet potato leaves had a generally high antioxidant activity. According to Debarry et al. (2005), antioxidant defence is not completely efficient in protecting plants from oxidative burst, free radical damage must be constantly repaired.

Carotenoids are plant pigments that can confer plants with resistance to adverse effects of drought (Jaleel et al., 2009). The concentration of carotenoids was high at Umbumbulu followed by Deepdale then Richards Bay. This was the opposite of total antioxidant activity which increased under drought stress conditions. Results of this nature were also reported by Lin et al. (2006) on sweet potato leaves. These authors discovered that carotenoids were not affected by drought stress, they further reported that increases in total antioxidants may have compensated for the need for carotenoids. Increases in the content of these phytochemicals (carotenoids and total antioxidants) in sweet potato leaves is beneficial to human health since they offer protection against common diseases such as cerebrovascular events, cancer and other age related degenerative diseases (Scalzo et al., 2005; Islam, 2006). All along total antioxidants have been associated with the leaves (especially green leafy vegetables), but

results from this experiment indicate that even the storage roots can have appreciable amounts of antioxidants.

High chlorophyll content in sweet potato leaves and other substances presumably present are likely to have a health-promoting potential since chlorophyll has also been reported to have numerous beneficial biological properties to humans (Ferruzzi et al., 2002; Kizhedath and Suneetha, 2011). High concentration of this phytochemical was also recovered from Umbumbulu planted sweet potato, followed by Deepdale then Richards Bay. This further emphasises that utilization of sweet potato leaves as a vegetable could increase food and nutrition security (micro-nutrient access (vitamin A)) and health. It is beneficial to food security in that it could add to the number of seasonal leafy vegetables used by rural communities who have limited access to produce markets (Vorster et al., 2007). Continuous availability of sweet potato leaves is guaranteed since they (leaves) can be harvested several times during the year even during off-season when storage roots are not available due to unfavourable environmental conditions. The continuous availability and high nutritional benefit will satisfy two of the important pillars of food security.

Storage roots of orange-fleshed cultivars yielded higher β -carotene content than the creamfleshed sweet potatoes as was shown in previous studies (Rautenbach et al., 2010; Laurie and van Heerden, 2012; Laurie et al., 2012). The deeper the orange colour, the greater the β -carotene concentration. Sweet potato cultivar A45 contained more than 15 mg/100g β -carotene, which can provide 100% of the recommended daily allowance (RDA) of vitamin A for children between the ages of 4 – 8 years (IOM 2006).

It is concluded that starch, β -carotene and phytochemicals content and antioxidant activity in sweet potato can be increased or reduced by environmental conditions. Moist coast hinterland and ngongoni veld were more favourable to sweet potato nutritional and phytochemical conditions. Sweet potatoes grown in this agro-ecological area can store starch and phytochemicals (secondary metabolites) better than those grown under harsh environments (Moist coast forest, thorn and palm-veld). Drought stress increased total antioxidant activity in storage roots. Increases in carotenoids may not be due to drought stress. Nutrients and phytochemical concentration is also affected by growth season. The study also demonstrated that sweet potato leaves have a health-promoting potential as they have high content of phytochemicals and antioxidant activity. They can also play a vital role in improving food and

nutrition security as a leafy vegetable while farmers are still waiting for the storage root to mature.

CHAPTER 6

ANTIOXIDANT ACTIVITY AND PHYTOCHEMICAL CONTENT OF SWEET POTATO LEAVES UNDER SIMULATED DROUGHT STRESS

Abstract

Sweet potato leaves are a source of nutrients that can benefit rural communities. However, it is not known whether the nutritional value of sweet potato leaves may be altered by water stress. Therefore, changes in leaf total antioxidant activity and selected nutrients and phytochemical content of three sweet potato cultivars were evaluated in response to simulated water stress conditions [30% and 100% crop water requirement (ETc)]. Leaves were sampled bi-weekly until storage root maturity. Spectrophotometry and FRAP essay were then used for analyses of phytochemicals and total antioxidants, respectively. Leaves of water stressed plants had high phytochemical content and total antioxidant activity as an indication of high oxidative stress. Increases of these phytochemicals in the leaf are an indication of improved health benefits for humans when the leaf is consumed as a vegetable. This also meant that even if sweet potatoes had low yields when grown in marginal areas, their leaves would contribute to food security. The phytochemical content was low in the leaves of fully irrigated plants when compared with water stressed leaves, but they were still higher than that of other common commercial vegetables such as spinach and lettuce. This was an indication that if sweet potatoes were grown in areas where drought stress is not a problem, farmers would benefit from them as healthy leafy vegetables and starch crop from storage roots. Leaves of orange-fleshed cultivars had higher phytochemical content that that of the white-fleshed cultivar.

Key words: water stress, sweet potato leaves, cultivars, phytochemicals

6.1 Introduction

Vegetables are very important in human diets because they supply the body with minerals, vitamins and certain hormone precursors (Antia et al., 2006). Research indicates that nutrients found in fruits and vegetables go beyond preventing deficiencies such as beriberi and rickets in the human body (Oduro et al., 2008); they prevent other diseases not related to nutritional disorders. Other important vitamin precursors such as ascorbic acid and β-carotene have been reported to exist in high concentrations in some vegetables (Gibson, 2005). These are powerful antioxidants needed in large quantities in the body (Labadarios et al., 2005). Sweet potato leaf has been reported to be an excellent source of antioxidants compared to other commercial vegetables such as spinach and broccoli (Islam, 2006). Antioxidants are priced for possessing anti-cancer properties (Podsedek, 2007). The leaf is, however, not common in human diets but largely used in animal feed (Woolfe 1992; Islam 2006). Its potential as a leafy vegetable has been hampered by the fact that it has been used traditionally as a feed for domestic animals and thus considered as a 'poor-man's vegetable' (Antia et al., 2006). Few countries in Asia and Sub-Saharan Africa do use sweet potato leaves as a vegetable though, but it is still an underexploited vegetable. In South Africa, more research has been concentrated on the nutritional improvement and consumption of storage roots (Laurie and Magoro 2008; Laurie et al., 2012). Recently products made out of sweet potato leaves were promoted along with those made from storage roots (Laurie and van Heerden, 2012).

Sweet potato is a drought tolerant crop (Lebot, 2009) and drought stress accounts for 25% total annual yield loss (Placide et al., 2013) compared to > 50% yield loss or complete failure in other crops such as maize under drought stress (Mabhaudhi, 2009). Current predictions on climate change indicate that there will be increased frequency and severity of droughts (Wayne, 2008; Schulze, 2011) in the near future. This gives a greater sense of urgency to promote crops that are resilient to drought and can produce well under such adverse conditions (Modi and Mabhaudhi 2013). Sweet potato is among those crops, and is considered a food security crop because it gives yields even under drought conditions. Moreover, it is generally herbaceous with a lot of green leaves. Research on the nutritional content of the leaves has been conducted (Antia et al., 2006; Islam, 2006; Oduro et al., 2008) and its potential to improve food security has been reported (Agili et al., 2012).

It is therefore vital to know if producing sweet potato under drought stress conditions could alter the nutritional status of the leaves or not. Information generated from that will help

inform communities on the importance of sweet potato leaves in their diets and contribute to food availability throughout the plant growth period, even before the crop (storage roots) matures. This may make promote sweet potato production in marginal areas where water stress is prevalent. The objective of this experiment was therefore, to evaluate the effect of water stress and sequential harvesting on total antioxidant activity, total carotenoids, proline and chlorophyll content of sweet potato leaves at different growth stages.

6.2 Materials and methods

6.2.1 Plant material

Three sweet potato cultivars (A40, A45 and 199062.1) were sourced from the University of KwaZulu-Natal's (UKZN) Plant Breeding Department. Two of the sweet potato cultivars (A45 and 199062.1) were orange-fleshed, while the third (A40) was white-fleshed. The cultivars were planted under simulated drought conditions in beds in a growth tunnel at the University of KwaZulu-Natal. The environment in the tunnels is not fully controlled but is designed to mimic sub-tropical conditions (\sim 18/33°C day/night temperatures and 60-80% relative humidity) (Modi 2007). The experimental layout was a split-plot design with two factors; water regimes (30% and 100% crop water requirement (ETc)) as the main plot and cultivars (A40, A45 and 199062.1) as sub-plots. Cultivars were replicated four times. Irrigation scheduling was based on reference evapotranspiration (ETo) and a crop factor (K_c) (Allen *et al.*, 2005). An online automated drip irrigation system was used to apply water in the beds.

6.2.2 Sampling for laboratory analysis

Plant leaf tissue was sampled destructively bi-weekly at midday. Samples were taken from the top three, fully expanded leaves Laboratory analyses involved analysing for beneficial plant metabolites associated with stress acclimation (total antioxidant capacity and proline) and pigment concentration (total carotenoids and chlorophyll content).

6.2.3 Determination of total antioxidant capacity

6.2.3.1 Ferric reducing ability of plasma (FRAP) assay

Extraction was carried out using perchloric acid. Freeze-dried and ground plant material (leaves and storage roots) of 0.2 g each was mixed with10ml of perchloric acid and allowed to stand for 10minutes. The mixture was vortexed for 1 minute before being centrifuged at 12400 x g at 4°C for 10 minutes. The ability to reduce ferric ions was measured using a modified version of the method described by Wong et al. (2006). An aliquot (200 μl) of an extract was added to 2 ml of FRAP reagent (10 parts of 300 mM sodium and 1 part of 20 mM sodium acetate buffer at pH 3.6, 1 part of 10 mM TPTZ solution and 1 part of 20 mM FeCl₃ .6H₂O solution). The reaction mixture was incubated in a water bath set at 37°C. The increase in absorbance was measured for 30 min at 593 nm. The antioxidant capacity based on the ability to reduce ferric ions of the extract was expressed as μmol Trolox equivalents per 100 gram of plant material on a dry matter basis.

6.2.4 Carotenoid and chlorophyll concentration

Carotenoids and chlorophyll were extracted and determined according to Lichtenthaler (1987) in a dark room. Freeze-dried, ground sweet potato leaf material (0.2 g DM) was placed in centrifuge tubes, 10ml of extracting solvent (80% acetone) was added and then allowed to stand on ice covered with aluminium foil for 10 minutes before being homogenized with Ultra-Turrax using 1 minute burst twice and centrifugation at 12500 x g for 5 minutes. Thereafter, the supernatant was decanted into 2ml cuvettes and absorbance read at various wavelengths (663.2 nm, 646.68 nm and 470 nm) as this was a simultaneous extraction of carotenoids and chlorophyll. Calculations and conversions were made using the equations below:

$$C_a = (12.25 \text{ x } A_{663.2}) - (2.79 \text{ x } A_{646.8})$$
 Equation 6.1
$$C_b = (21.50 \text{ x } A_{646.8}) - (5.10 \text{ x } A_{663.2})$$
 Equation 6.2
$$C_{a+b} = (7.15 \text{ x } A_{663.2}) - (8.71 \text{ x } A_{646.8})$$
 Equation 6.3
$$C_{x+c} = (1000 \text{ x } A_{470}) - (1.82 \text{ x } C_a) - (85.02 \text{ x } C_b)/19$$
 Equation 6.4

where: $C_a = Chlorophyll a$,

 $C_b = Chlorophyll b$,

 $C_{a+b} = Total Chlorophyll, and$

 C_{x+c} = Total carotenoids.

6.2.5 Proline content determination

Proline content was determined using the methods of Bates et al. (1973) and Claussen (2005) based on proline's reaction with ninhydrin. Freeze-dried leaf material (0.2g) was homogenized in $10 \text{ m}\ell$ of 3% sulfosalycic acid (w/v). The homogenate was centrifuged at 11000 rpm for 10 min at 4°C. For proline colorimetric determinations, a 1:1:1 solution of proline, ninhydrin acid and glacial acetic acid was incubated at 100°C for 1 hour. The reaction was arrested in an iced bath and the chromophore was extracted with 4 ml toluene and its absorbance was read using a spectrophotometer at 520 nm.

6.2.7 Measurement of absorbance

Absorbance was measured using a UV–1800 UV–Vis spectrophotometer (Shimadzu, North America) at the respective wavelengths as indicated in the procedures.

6.2.7 Statistical analyses

Data were subjected to analysis of variance (ANOVA) using GenStat® version 16 (VSN International, Hemel Hempstead, UK 2011). Tukey's HSD test was used to separate means at the 5% level of significance.

6.3 Results

Total antioxidant activity was significantly ($P \le 0.05$) affected by different water regimes and different cultivars. Interaction between water regimes and cultivars was also significantly ($P \le 0.05$) effective on total antioxidant activity. Water stressed plants recorded higher antioxidant activity than fully irrigated plants. When comparing the three cultivars results showed that total antioxidant activity was higher in cultivar A45 followed by cultivar 1990621 then cultivar A40 (Figure 6.1). Cultivar A45 grown under 30% ETc started with high antioxidant activity which began to decline at six weeks after planting, became constant and increased again at 14 weeks after planting. Antioxidant activity remained constant throughout the

growth period when grown under 100 ETc. Cultivar 199062.1 and A40 were fluctuating throughout the growth period under both water regimes (Figure 6.1).

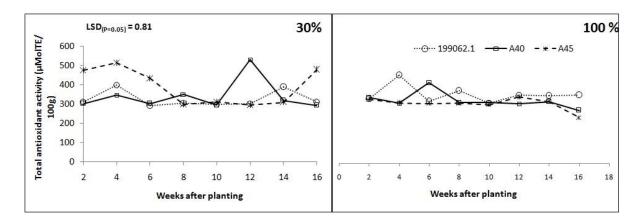


Figure 6.1: Total antioxidant activity of three sweet potato cultivars grown under different water regimes.

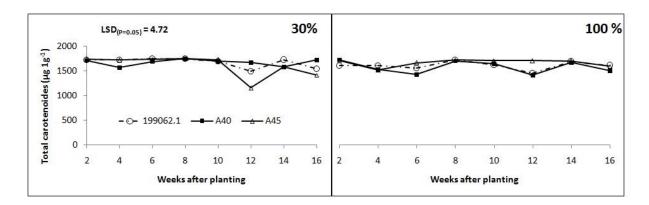


Figure 6.2: Total carotenoid content of three sweet potato cultivars grown under different water regimes.

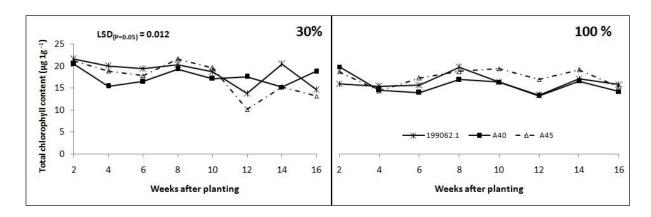


Figure 6.3: Total chlorophyll content of three sweet potato cultivars grown under different water regimes.

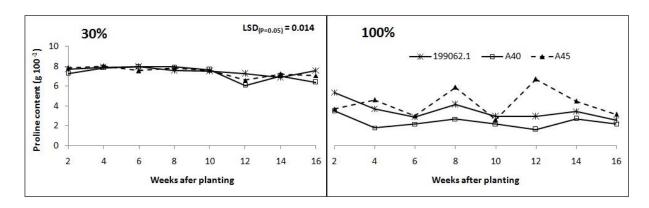


Figure 6.4: Proline content of three sweet potato cultivars grown under different water regimes.

Total carotenoid, chlorophyll and proline contents were significantly ($P \le 0.05$) affected by water regimes and varietal differences. Interaction of the two factors also recorded highly significant ($P \le 0.001$) effect. The water stressed (30% ETc) plants recorded high contents of these phytochemicals than fully irrigated (100% ETc) plants. High water availability did not favour concentration of these phytochemicals. Extracts of cultivar 199062.1 recorded highest contents of total chlorophyll and carotenoids followed by cultivar A45 then A40 (Figure 6.2 and 6.3). Concentrations of carotenoids and chlorophyll were more constant in leaves of plants grown under 100% ETc. It was also noted that the orange-fleshed cultivars (A45 and 199062.1) generally, had higher concentrations of these phytochemicals than the white-fleshed cultivar. Proline content was highly concentrated in cultivar A45, followed by cultivar 199062.1 then A40 (Figure 6.4). Its concentration was constantly high under 30% ETc across all cultivars. Fluctuations in concentration were observed in cultivar A45 under 100% ETc.

6.4 Discussion and conclusions

The results of this investigation showed that water stress does alter the activity and concentration of secondary metabolites in sweet potato leaves. This confirms observations by Ueda et al. (2001) and Alireza et al. (2011) that plants exposed to drought stress respond by showing ingenious adaptations at the physiological level. Plants grown under 30% ETc had high antioxidant activity and carotenoid concentrations indicating that oxidative stress in the leaves caused by water stress. This is a unique defence mechanism for plants to acclimate and increase tolerance under such environment (Xu et al., 2008). Sairam and Saxena (2000) indicated that both enzymatic and non-enzymatic antioxidants in plant cells played an important role in avoiding detrimental effects of free radicals. Antioxidants perform the same function of reducing oxidative damage to the human body (Ismail et al., 2004). High antioxidant activity is more desirable to humans. Their ability to scavenge for free radicals has been attributed to mechanisms such as anti-inflammatory properties, inhibition of enzymes and induction of detoxification enzymes in the human body (Lopez-Martinez et al., 2011). Podsedek (2007) also reported antioxidants as vital in protecting humans against cancer and cardiovascular diseases. Typical commercial leafy vegetables such as broccoli, cabbage and iceberg lettuce have antioxidant activity of 29.4 µMol TE/g, 35 µMol TE/g and 8.8 µMol TE/g respectively (Szeto et al., 2002). The sweet potato leaves under this study recorded an average of 36.5 µMol TE/g and 33.9 µMol TE/g at 30% ETo and 100% ETo, respectively. This was higher than that of spinach which was reported to range between $9.6 - 25.0 \mu Mol$ TE/g (Howard et al., 2002). The antioxidant compares mostly with African leafy vegetables such as Amaranthus hybridus (van der Walt et al., 2009).

Carotenoids are pigment molecules contributing to the colour of fruits and vegetables and play an important role in photosynthesis as they are abundant in leaves (van der Walt et al., 2009). They form a key part of the plant's antioxidant defence system (Wahid 2007; Farooq et al., 2009) and can partially help plants withstand adversities of drought (Jaleel et al., 2009). The total carotenoid content of the sweet potato leaves under both water regimes (30% and 100% ETo were at 165 mg 100 g⁻¹ and 162 mg 100 g⁻¹, respectively) ranked higher than that of baby spinach (140 mg 100 g⁻¹) (Bergquist, 2006) and Indian *Amaranthus* sp. (41.5 mg 100 g⁻¹) (Gupta and Prakash, 2009). According to van der Walt et al. (2009), carotenoids have an important biological function, and some of those derived from plant foods are provitamin A

pigments which is converted to vitamin A as per the body requirement. South African children between the ages of 2 and 5 years still suffer from vitamin A deficiency (VAD) (Faber et al., 2002) and a diet with high provitamin A content would improve their condition. A study on children from rural areas of South Africa confirmed that consumption of green leafy vegetable contributed towards vitamin A intake (Faber et al., 2007). Other benefits of carotenoids include protection against eye disease of macular degeneration (Mozaffarieh et al., 2003). Their high concentration in plants grown under 30% ETc indicates beneficial effect of water stress to human health. High chlorophyll content has also been reported to have numerous beneficial biological properties to humans such as anti-inflammatory, control of calcium oxalate and internal deodorization (Ferruzzi et al., 2002; Kizhedath and Suneetha, 2011). Chlorophyll content was expected to increase under fully irrigated plants but this experiment recorded higher concentrations under water stressed plants. It was not easy to compare chlorophyll content for human health benefit in this study because chlorophyll content has all along been extracted for plant physiological studies (Farooq et al., 2009).

Proline is a proteinogenic amino acid usually considered as an osmoprotectant and accumulates in cells to obtain suitable conditions for water uptake under limited water conditions (Ozgur and Mithat 2008). Its accumulation can influence stress tolerance in multiple ways since it functions as a molecular chaperone able to protect protein integrity and enhance the activities of different enzymes (Szabados and Savoure 2009). In humans, proline helps improve recovery from bone fractures since it helps to produce more collagen and cartilage. It is also reported to assist in keeping muscle joints flexible, improve development and maintenance of healthy skin (Guoyao 2013). The authors further indicated that proline is considered a functional amino acid for humans. In the current study, concentration of this amino acid increased in leaves of plants grown under 30% ETc. These results are an indication that sweet potato leaves remain efficient in improving human health even when growing under drought stress conditions. According to Agili et al. (2012) drought stress does not allow sustainable and enduring production of sweet potato storage roots. Since the leaves can still be produced under drought stress conditions this means that they can supplement for losses in storage root yield. Communities located in marginal areas where storage roots formation fails completely can use sweet potato leaves as a vegetable. Where drought is not a problem, communities would have double gains as they will use leaves as a vegetable during the growth period and enjoy the storage roots thereafter. This would ensure food security throughout the growth period of sweet potato crop, unlike with other staple crops where farmers have to wait for the crop to mature since their leaves cannot be utilized as a vegetable; that waiting period opens a gap of food insecurity.

Sequential harvesting did not have much influence on phytochemicals under the two water regimes, it only had an effect on cultivars. Cultivar A45 was the most unstable, its total antioxidant activity and proline content fluctuated a lot (Figure 6.2 and 6.4). This might be an indication that it is a very sensitive cultivar, it responds even to minor changes in environmental conditions. Leaves of orange-fleshed cultivars generally had higher concentration of these phytochemicals than the white fleshed cultivar.

It can be concluded that water stress increases total antioxidant activity, total carotenoid, chlorophyll and proline content in sweet potato leaves. The levels of total antioxidant activity and carotenoid content were higher than that of common green leafy vegetables such as spinach and cabbage. Increases of these phytochemicals in the leaf are advantageous to farmers in marginal areas because they will still be able to benefit from the leaves even if the storage root does not form. Farmers in areas with adequate rainfall will have a double turnover as they will benefit from the leaf as a vegetable and later in the season get to benefit from the storage roots. Leaves of orange-fleshed sweet potato cultivars used in this study contained higher concentrations of phytochemicals than leaves from the white-fleshed cultivar.

CHAPTER 7

GENERAL DISCUSSION

The world's population is expected to increase by one-third in the next 30 years and agricultural production will have to increase by 60% to satisfy the expected demands for food and feed (FAO, 2013). These expectations should be met despite the fact that climate change is already having a negative impact on agriculture and food security due to unpredictability of weather patterns (Schulze, 2011). More productive, resilient and efficient agricultural production that will contribute to the mitigation of climate change is required. Drought tolerant food security crops with wide adaptability and flexible planting and harvesting schedules like sweet potato are considered as future crops under such predictions. This study hypothesised that producing a crop like sweet potato with little losses due to drought stress means it is very capable of ensuring food security during times of drought and in areas that face water scarcity. It was further hypothesised that the nutritional content of the crop cannot be altered by environmental conditions. To test these hypotheses, field and controlled environment experiments were conducted. Field experiments evaluated the effect of different agro-ecological environments and different seasons on physiological responses, growth, yield and nutritional content of sweet potato cultivars growing under low-input cropping system. Controlled environment experiment evaluated the same parameters under water stress conditions.

Adaptation to different environments

Three bioresource groups/agro-ecological areas (Coast hinterland thornveld; Moist coast hinterland and ngongoni veld; Moist coast forest, thorn and palmveld) were used for the experiment and were represented by Deepdale, Umbumbulu and Richards Bay, respectively. Varying environmental conditions at the three locations had a significant effect on plant ecophysiology, growth and yield. Richards Bay environmental conditions were characterized with high evapotranspiration (ETo) which exceeded rainfall received thus created water deficit. Moreover, the predominant soils were sandy with very low permanent wilting point and field capacity. Plants in that location showed signs of being water stressed. Stomatal conductance (SC) and chlorophyll content index (CCI) were relatively low throughout the growing season and this translated into very low biomass in both winter and summer

experiments. The low SC confirms earlier reports that stomatal responses are closely linked to soil water content than to leaf water status (Gowing et al. 1990; Chaves et al. 2002). By closing the stomata, not only does the plant limit water loss through transpiration, it also reduces intracellular carbon dioxide (CO₂) availability (Mabhaudhi et al. 2013; Zhao et al. 2014). This affects photosynthesis since CO₂ is the chief substrate; hence resulting in biomass reduction (Lawlor 2002; Blum 2009). The other two locations, Deepdale and Umbumbulu started with high SC during the juvenile stage but later started to decrease as the crop moved into the vegetative growth stage. This period coincided with increased demand for water by the crop as there were now more leaves transpiring. Decreases in chlorophyll content under drought stress conditions have been associated with oxidative stress and may be the result of pigment photo-oxidation and chlorophyll degradation (Jaleel et al., 2009; Anjum, 2011). The fact that SC and CCI were low in Richards Bay implies that photosynthesis was both substrate and energy limited. Consequently leaves showed rapid signs of chlorosis and later abscission. In the other locations, photosynthesis was not limited by neither chlorophyll degradation nor water deficit and CO₂ flow.

The other two locations (Deepdale and Umbumbulu) had clayey soils and an impeding layer at 50 cm depth. These characteristics promote good water retention capacity and may have limited soil water loss through soil profile. The retained water is believed to have supplemented the plants' metabolic activities after ETo losses. The bioresource groups where the two locations are situated also experience misty conditions which may have reduced the length of exposure to water deficit during the summer season. Since there was no impeding layer in Richards Bay, it is suspected that water was lost through the porous soil profile, ETo and poor water retaining capacity of sandy soils. Plant growth followed a similar pattern as physiological parameters. Low values of CCI and SC resulted in short vines, fewer leaves and fewer branches, another indication of drought stress and plant coping mechanisms as explained by Chaves et al. (2002) and Jaleel et al. (2009). Reduced plant size (height/length, leaf area and leaf area index) was reported by Blum (2005) as the major mechanism for moderating water use and reducing injury under drought stress conditions. The sweet potato cultivars also displayed a tendency to branch in one location and not branch in another but prefer to extend in length or increase leaf number in another location. This was attributed to specific environmental adaptation. It proves the concept of wide adaptation, indicating that adaptation has more of geographical than environmental meaning and it reduces genetic diversity while increasing genetic vulnerability (Ceccarelli, 1994).

Winters in Deepdale and Umbumbulu are characterised with no mist and less rain. The absence of these environmental factors may have hampered growth and vital metabolic activities thus there was poor growth at Umbumbulu and no growth at all at Deepdale. Temperatures below 15°C (Table 3.3) were reported by Laurie and Niederwiser (2004) to cause little growth and below 10°C causes severe retard in growth. This may have been the main reason for the retarded plant growth during the winter season in these locations. Richards Bay exhibited very poor growth in even in winter where it was expected to do better than Umbumbulu, since prevailing temperatures were higher than that of Umbumbulu.

The facts that Deepdale and Umbumbulu recorded high storage root yields suggests that the three sweet potato cultivars can be produced using cost effective agricultural methods, a trait desirable to resource-constrained small-scale farmers. Application of good cultural and agronomic practices such as fallowing and ridging contributed to the increased yields. However, there was an inconsistency at Deepdale over the two growing seasons (2012/13 and 2013/14). Umbumbulu proved to be more consistent compared to Deepdale. Poor planning and not paying attention to weather conditions was the main cause of reduced yields at Deepdale. Planting was done based on historical norm instead of studying weather predictions to help adjust towards planting during warmer dates. This emphasises the importance of using seasonal weather forecasting and adjusting planting dates accordingly. Many farmers lose yield due to such mistakes.

Fertilizer application did not improve storage roots yield when applied to naturally fertile soils like in Deepdale and Umbumbulu. The innate quality and ability of sweet potato to colonise easily on marginal soils encourages its (fertilizer) application only in continuously cropped and exhausted soils like that of Richards Bay. Adding fertilizers high in K (Bourke 1985; George et al., 2002) is also important to improve storage root yield although most soils in KZN are not deficient of this element (KZNDAE, 2013) as explained in chapter 2.

Sweet potato cultivar A45, an orange-fleshed cultivar, showed more environmental plasticity than the other two sweet potato cultivars (A40 and 199062.1). This indicates that its adoption can contribute towards improved food and nutrition security for resource-constrained and small-scale farmers since it can also supplement for Vitamin A deficiencies. Richards Bay

environment was not suited for the sweet potato cultivars evaluated in this study. All three cultivars experienced limitations to fully expressing their genetic potential. In order to improve sweet potato yields in Richards Bay (where temperatures allows year round production), farmers should be encouraged to practise water harvesting techniques and improve soil water conservation strategies such as increasing the size of ridges, to apply recommended fertilizer amounts, to use more compost at planting in order to improve soil structure and soil water holding capacity.

Drought tolerance

Physiological traits of the sweet potato plants reported in this experiment indicated that it was indeed drought tolerant. When the cultivars were grown under water deficit (30% ETc) they recorded regulated their stomata just as explained by Farooq et al. (2009). Stomatal conductance (SC) of these cultivars was very low, mostly below the 100 mmol m⁻²s⁻¹ threshold. Metabolic impairment at such low SC values, was avoided through a mechanism called photorespiration (van Heerden and Laurie, 2008). Photorespiration is the second most important mechanism for removing excess electrons (that may damage photosynthetic apparatus) in C3 plants (Lawlor and Cornic, 2002). Relative water content (RWC) was also similar to that of plants grown under 100% ETc suggesting that the cultivars are drought tolerant. High RWC indicates more resistance to drought stress (Schonfeld et al., 1998). Chlorophyll content was not affected by water stress indicating that it cannot be used as a measure of physiological adjustment of these sweet potato plants when grown under water stress conditions.

Water stress did not have a profound influence on the growth of these cultivars. The only parameter affected by water stress was the number of leaves which increased under water stress. This was contrary to reports that plants' morphological characteristics are negatively affected by water stress (Farooq et al., 2009; Blum, 2011). A high number of leaves consequently increase the photosynthesis hub which ends up in high yields. Other growth parameters such as branching and vine extension were limited by varietal differences not water stress. Since the canopy represents the only source of biomass for subsequent partitioning to storage root, this would therefore, improve source sufficiency for yield attainment. Yield was increased in the 30% ETc due to the drought avoidance and acclimation characteristics displayed by these cultivars. This indicated that these cultivars are suitable for production under marginal areas as long as there is even distribution of the low water supply.

Plants grown under 100% ETc exhibited rank growth a characteristic common in cotton (North Carolina State University, 2012) where plants tend to partition towards vegetative growth than storage root formation. This indicated that a period of water deficit is needed in order to provide translocation of more photosynthates to the roots and the development of storage roots (Ekanayake et al., 1990; Mohankumar, 2000). This observation encourages famers to alter their planting dates such that the stages of growth where plants have to be exposed to water stress coincides with mid-summer drought to break the rank growth syndrome and increases storage root yield. Cultivar 199062.1 was more tolerant to drought and gave higher yield than the other two cultivars. Cultivar A40 recorded high SC but low CCI and consequently low yields. This was uncommon phenomenon in plants not unless there is substrate inhibition for photosynthesis (Reed et al., 2010). In this case it is suspected that the high levels of CO₂ became the source of inhibition to photosynthesis since it was higher than the enzymes could use, thus the low yields. Cultivar A45 on the other hand was mostly impartial to these treatments.

Nutrients and phytochemicals

Starch, β-carotene and phytochemical concentration as well as antioxidant activity in sweet potato can be increased or reduced by environmental conditions. The moist coast hinterland and ngongoni veld where Umbumbulu is located had more favourable conditions for the production of nutritional and phytochemical substances in sweet potato. Sweet potato grown in this agro-ecological area stored its starch and phytochemicals (secondary metabolites) better than those grown under harsh environments at Richards Bay (Moist coast forest, thorn and palm-veld). The starch content was within the range of $\leq 30\%$ as reported by Bovell-Benjamin (2007) but it was noted that the 2012/13 growing season recorded lower values than 2013/14 season. Fluctuation in its content between locations may be an indication that these nutrients are sensitive to changes in environmental conditions. Temperature was a dominating factor in as far as starch and β-carotene content were concerned. Where temperature was predominantly below 25°C, these nutrients were higher and tended to decrease with increases in temperature. This confirmed reports by Rotter and van de Geijn (1999) which indicated that increases in temperature affects partitioning of dry matter in plants. Contents of β-carotene increased with the depth of orange colour of the storage roots. Cultivar A45 contains high βcarotene than cultivar 199062.1. The β-carotene recovered from cultivar A45 was more than 15 mg/100g, which can provide 100% of the recommended daily allowance (RDA) of vitamin A for children between the ages of 4 - 8 years (IOM, 2006).

Where plants were experiencing drought stress (at Richards Bay), antioxidant activity increased in storage roots. The leaves did not follow the same trend as storage roots, total antioxidant activity was lower than in leaves from other locations. Since drought is associated with high oxidative stress (Lin et al., 2006) even the leaves were expected to record high antioxidant activity, but instead antioxidant activity was high in leaves from Deepdale and Umbumbulu. Carotenoid concentration was high at Umbumbulu followed by Deepdale then Richards Bay. This was opposite of total antioxidant activity which increased under drought stress conditions. Results of this nature were also reported by Lin et al. (2006) on sweet potato leaves. These authors discovered that carotenoids were not affected by drought stress, they further reported that increases in antioxidants abilities may have compensated for the need for carotenoids. Increases in content of nutrients and phytochemicals (carotenoids and total antioxidant) in sweet potato leaves is beneficial to human health since they offer protection against common diseases such as cerebrovascular events, cancer and other age related degenerative diseases (Scalzo et al., 2005; Islam, 2006).

Nutrient and phytochemical concentrations were also affected by growth season. The 2013/14 growth season performed better than 2012/13 growth season. The study also demonstrated that sweet potato leaves have a health-promoting potential as they have high content of phytochemicals and antioxidant activity. They (leaves) could add to the number of seasonal leafy vegetables used by rural communities who have limited access to produce markets (Vorster et al., 2007) thus increase food and nutrition security before the storage root matures.

The potential of sweet potato leaf as a green leafy vegetable was further tested under water stress environment where it was observed that the phytochemical content increased under water stress. Sequential harvesting did not alter its content throughout the 120 days period of growth. Other phytochemicals such as chlorophyll and proline content increased under water stress. As indicated earlier, high antioxidant activity is more desirable to humans and their ability to scavenge for free radicals has been attributed to mechanisms such as anti-inflammatory properties, inhibition of enzymes and induction of detoxification enzymes in the human body (Lopez-Martinez et al., 2011). These cultivars contained levels comparable to most African leafy vegetables such as *Amaranthus hybridus* (van der Walt et al., 2009).

Provitamin A carotenoids can be converted to vitamin A as per the body requirement. Vitamin A deficiency is still a problem in South Africa mainly in children between 2-5 years old (Faber et al., 2002) and a diet with high carotenoids would improve their condition. Consumption of green leafy vegetable contributed towards vitamin A intake (Faber et al., 2007). Proline and chlorophyll concentration also increased in sweet potato leaves grown under water stress. This indicated that consumption of sweet potato leaves grown under marginal conditions will increase chances of healing from bone fractures, internal deodorization (Ferruzzi et al., 2002) and improve development and maintenance of healthy skin (Wu, 2010). The levels of total antioxidant activity and carotenoid content were higher than that of common green leafy vegetables such as spinach and cabbage. Increases of these phytochemicals in the leaf are advantageous to farmers in marginal areas because they will still be able to benefit from the leaves even if the storage root does not form. Farmers in areas with adequate rainfall will have a double turnover as they will benefit from the leaf as a vegetable and later in the season get to benefit from the storage roots. Leaves of orangefleshed sweet potato cultivars used in this study contained high concentrations of phytochemicals than leaves from white-fleshed cultivar.

Phenolics are well known plant secondary metabolites produced under environmental stress, and they are arguably the dominant non-enzymatic plant antioxidants. It appears that one major weakness of the study design is that phenolics were not measured as a stress response variable. This should be done in future studies. Furthermore, sweet potato is well known for its sugar content than starch. Perhaps it would have been a better design to also measure sugar content in storage roots as a response to variable. There were resource limitations to do this and hence it is recommended for future studies.

Conclusions and recommendations

The study was able to demonstrate that sweet potato growth, yield and nutrition are affected by different environmental conditions. Water stress alone does not reduce storage root yield as it was evident from the simulated water stress experiment. It emphasised that sweet potato is drought tolerant and under drought stress conditions it increases its biochemical content as some form of protection against water stress. These biochemicals are beneficial to humans as phytochemicals for healthy lifestyle. Production of these sweet potato cultivars in marginal areas will increase nutritional and health benefits derived from leaves if used as a green leafy vegetable. Even if the storage rot yield will be low, at least farmers would have benefitted from the leaves. These cultivars should not be grown in sandy soil areas without implementing agronomic practices that will improve water retention in the soil. Farmers located in areas with climatic conditions similar to Umbumbulu and Deepdale can adopt these cultivars but pay attention to planting dates and weather forecasting because they tend to be sensitive to changes in environmental conditions, especially temperature. Another study to determine the stage of growth where water stress will trigger high storage root formation and translocation needs to be done in order to avoid rank growth that was observed on these cultivars. The field experiments need to be repeated using more sweet potato cultivars and more locations so that adaptation can be inferred for a wide area. Crop modelling can then be conducted so as to save on time and costs of repeating trials in more locations. Determination of sugar content and other secondary such as phenolics associated with water stress are recommended for future studies on sweet potato.

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