## **EVALUATION OF SELECTED SWEETPOTATO (***Ipomoea batatas***) ACCESSIONS FOR DROUGHT TOLERANCE**

**by**

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# **DECLARATION**

This dissertation is my own work and the results are my own independent investigations and research. I further declare that all sources used or quoted were acknowledged.

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SIGNATURE DATE

## **DEDICATION**

This dissertation is dedicated to the Almighty God for His guidance and provisions, to my loving wife, Folakemi and my two lovely daughters, Olamide and Oreoluwa for their love and encouragement throughout the duration of the project.

Also to my late parents, Chief Patrick Orire and MrsClementinahMofeolaOmotubora, for their immeasurable contribution to my upbringing.

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#### **ABSTRACT**

Sweetpotato (*Ipomoea batatas)* is a major staple food in Africa and the rest of the world where they are discovered to be a good source of carbohydrates, vitamin A, vitamin C and protein. The maximum production potential of the crop is being hampered by severe drought which ravages most parts of Africa.

The main aim of this project therefore is to screen collected accessions of sweetpotatofor drought tolerance in a quick screening method with a view to identify cultivars that can perform well under water stress conditions.

Fifty selected sweetpotato accessions consisting of cultivars and breeding lines collected from the ARC-VOPI gene bank were planted for drought screening in the glass house for 6 weeks during which water was withheld to induce stress. Observations were made on number of dead plants and days to wilting point, the results were analyzed and 12 best performing cultivars were selected for field trials. The field trial was carried out in Lwamondo, Thohoyandou for 6months under rainfed conditions. The experiment was conducted in a complete randomized block design with 6 replicates.Yield data and growth parameters were collected every 8 weeks during the trial period and the data collected was analyzed using ANOVA. The best performing cultivars were Zapallo, Tacna, Ejumula, 2004-9-2 and Ndou. They were therefore recommended for further evaluation in other drought prone areas of the country.

Keywords :sweetpotato, drought tolerance, dry matter, yield.

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## **CHAPTER 1**

#### **1.0 INTRODUCTION**

#### 1.1 **General background**

More than 70 percent of potential yield loses in agriculture worldwide is attributed to adverse environmental factors of which water scarcity represents the most severe constraint (Boyer, 1982). Agriculture is the largest consumer of water in the world and in the drier areas of the world which include South Africa.

The use of water for agriculture can exceed 90 percent of consumption. Global warming is also predicted to affect most severely developing countries where agricultural systems are most vulnerable to climatic conditions and where small increases in temperature are very detrimental to productivity.

The Food and Agricultural Organization of the United Nations (FAO, 2007) estimates that by 2025 approximately 480 million people in Africa could be living in areas with very scarce water, and that as climatic conditions deteriorate, 600,000 km<sup>2</sup> currently classed as moderately constrained will become severely limited.

It is thus essential to improve water use efficiency in agriculture. This will require an integrated approach to water resources management to encourage an efficient and equitable use of the resource, and to ensure sustainability. The identification of crop varieties with increased tolerance to drought is therefore an important strategy to meet global food demands with less water.

Sweetpotato(*Ipomoea batatas)* is a major staple food in Africa, Asia, the Caribbean,

and South America, where they are important sources of carbohydrates, vitamins A and C, fiber, iron, potassium, and protein (Woolfe,1992). Sweetpotato is also used as animal feed.

Increasing recognition of the great potential of the sweetpotato crop as a nutritious food for humans and animals has resulted in intensified research efforts to enhance production and consumption in recent decades (Wolfe, 1992; Yamakawa and Yoshimoto, 2002).

Sweetpotato is especially important in developing countries because it is a highly adaptable crop that generates large amounts of food per unit area and unit time during relatively short rainy periods, giving it an advantage over major staples (Yencho*et al,* 2002, Mwanga*et al,* 2011). Sweetpotato also has flexible planting and harvesting times, tolerates high temperatures and low fertility soils. It is drought tolerant and easy to propagate. Furthermore, compared to other crops, sweetpotato requires fewer inputs and labour making it particularly suitable for households threatened by migration or diseases such as HIV/AIDS (Jayne *et al*., 2004).

Sweetpotato is grown over a broad range of environments and cultural practices and is commonly grown in low-input agriculture systems (Prakash,1994). The plant is sensitive to water deficits particularly during the establishment period including vine development and storage root initiation (Indira and Kabeerathumma, 1988). However, drought is often a major environmental constraint for sweetpotato production in areas where it is grown under rain fed conditions (Anselmo*et al.* 1998). Different cultivars may respond differently to limited quantities of soil water. Selection for good cultivar performance under drought conditions is thus considered to be of major importance.

The adverse effect of drought on the agricultural industry has long been recognized. Drought conditions negatively impact crop survival and yield (Saraswati*et al.,*2004). Apart from the direct effects on yield, the potential advantage of crop management practices such as fertilizer application or pest and disease management can also be affectedby drought. Drought necessitates additional irrigation periods, and this increases the overhead production costs. Lack of sufficient water for sweetpotato especially during its early developmental stage can results in low tuber yields (Ekanayake*et al.,*2004) and unacceptable tuber quality. Prolonged period of drought can also considerably reduce sweetpotato yield, as well as the quality of roots and causes huge economic losses to farmers. It is therefore necessary to improve the water use efficiency of the crop particularly in areas where water scarcities occur and where supplemental irrigation is needed. In the warmer areas where the crop is cultivated, the effect of water stress is also enhanced by high temperature (Ekanayake*et al.,* 2004).

South Africa is regarded as being susceptible to water stress conditions due to prevalence of drought (Bennie and Hensely, 2001). Research into drought tolerant plants is being intensified in order to minimize its overall impact on the agricultural enterprise. Since it is known that drought conditions severely affect crop survival and yield, and also increase the cost of production, development of drought tolerant sweetpotato varieties will increase profitability to farmers by potentially eliminating irrigation and associated production overhead costs.

According to Saxenaand O'Toole (2002) there are two options for the management of crops in water limiting environments: the agronomic and the genetic management. The former is already alluded to above.Genotypic variability exists for drought

tolerance withsome clones performing better under drought conditions (Ekanayake, 1989). Selection and improvement of adapted genotypes for a particular environment can therefore, be done with the appropriate equipment and using selection criteriaassociated with drought tolerance(Ekanayake,1989). With this genetic management option, drought-tolerant varieties, once developed, would be a low economic input technology that may be readily acceptable to resource-poor, rainfed, small land holding farmers.

Several methods such as measurement of potential relative turgidity, diffusion pressure deficit, chlorophyll stability index, carbon isotope discrimination had been used to evaluate drought and water-use efficiency in crops (Turk and Hall, 1980b; Morgan 1984; Yadava and Patil, 1984; Hall *et.al*., 1990, 1997). These methods were however discovered to be time consuming and therefore not suitable enough for screening large number of cultivars. Studies in the past on drought screening focused predominantly on drought as a whole concept without dealing with the individual component of drought tolerance (Lawan, 1983, Watanabe *et.al*., 1997). These traits can be an indicator to decide a specific screening method. They also develop strategies for screening methods but with less success due to the poor understanding of the concept of drought tolerance and lack of data on the inheritance of stress tolerance in plants. Also, plant defense mechanism varies making it difficult to use one screening method to determine stress. Nevertheless, certain methods based on physiological and phenotypic techniques as described by Bardelo, *et al*. (1995) were employed to determine genotype and environment interactions.

Development of this genetic management technology requires robust, reproducible, simple, and rapid field, pot, and laboratory screening methods for identification of traits of drought tolerance in germplasm, and incorporation of the same in highyielding varieties using conventional and biotechnological tools (Saxena and O'Toole, 2002). Consequently, this study aim to identify drought tolerant sweetpotato varieties from available germplasm by screening selected lines through water stress.

## **Objectives of the study**

- 1. Screening a number of breeding lines and land races for drought tolerance in a quick screening method.
- 2. Evaluation of selected breeding lines and land races for drought tolerance under field condition
- 3. Identification of cultivars that can perform well under water stress conditions without a significant loss of yield and quality.

#### **CHAPTER 2**

#### **2.0 LITERATURE REVIEW**

#### **2.1 Origin and distribution of Sweetpotato**

Sweetpotato (*Ipomoea batatas*) originated from Central America where it was found growing in the wild spreading across the Pacific from Central America and transported to warmer regions of Asia and Africa by Spanish and Portuguese traders (Alleman*et al*., 2004). Sweetpotato is grown in more than 100 countries in tropical, subtropical and temperate climates(Alleman*et al.,* 2004). It ranks as the world's seventh most important crop with an estimated annual production of approximately 121.52 million tons on 9.2 million ha with an estimated average yield of 13.2t/ha (FAO, 2005).

The genus*Ipomoea* consist of 600-700 species (Hauman, 1999). Sweetpotato belongs to the family called *Convolvulaceae* and is a creeping plant that consists of perennial vines and adventitious roots (Kebede*et. al.* 2008).It is usually propagated vegetatively by using both roots and stem cuttings and grown primarily for the edible root which takes about 5-6 months to mature.

Sweetpotato thrives well in sandy-loam and clay loam soils, which must be well drained because of the plant sensitivity to long lasting excessive moisture in the soil (Van den Berg and Laurie, 2004).It is very sensitive to alkaline and saline conditions which influence growth. Soil pH between 5.6 and 6.6 is very good for production (Laurie andNiederwieser, 2004).

China is the largest producer of sweetpotato with 80% of annual world supply (FAO, 2008). It is the third most important root and tuber crop in Sub-Saharan Africa (Ewell

and Mutuura, 1994). Africa produces 11.6 million tons annually with Nigeria being the largest producer followed by Uganda and Tanzania.

Sweetpotato is very rich in Vitamins A, B and C as well as minerals like phosphorous, iron and calcium (Woolfe, 1992). The roots can be boiled, baked or fried.

## **2.2 Sweetpotato production in South Africa**

Sweetpotato was introduced in South Africa in 1652 (Bester andLouw,1992).The average yield of sweetpotato when grown commercially is approximately 49t/ha while on a subsistence farming the average yield is 5-10t/ha (Laurie, 2004). Limpopo, Mpumalanga, Western Cape and Kwazulu Natal provinces are the specific production areas in South Africa. The common varieties are Blesbok, Bosbok, Ribbok and Koedoe.Sweetpotato annual production in South Africa was 62,888t (FAO, 2009).

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# **Table 1: World production of sweetpotato(tons)**

Source (FAOSTAT, Fevrier, 2012)

#### **2.3Drought stress and drought tolerance**

The term 'drought' was defined by (Cregg, 2004) as a meteorological occurrence characterized by below normal rainfall. The phenomena of drought stress is on the other hand defined as a period of insufficient rain which causes injury to crop and leads to a phenomenal reduction in economic yield. It is usually associated with nonavailability or exhaustion of water in the root zone.Drought can be permanent, periodic, or random, occurring early, late, or in the middle of the crop season (Ekanayake, 1990). Drought can also be cumulative or specific and short.

Drought tolerance is defined as the relative yield of a genotype compared to other genotypes subjected to the same drought stress (Hall, 1993). Droughtremains a challenge for researchers due to complexity of factors affecting crop response to drought (CeccarelliandGrando, 1991).

According to Ekanayake(1990) a genotype is drought resistant when it produces an economic crop within the limits of its production potential under conditions of limited water availability. Drought resistance and its components are almost constantly being redefined (Blum, 2005). A genotype can be drought resistant due to the mechanisms of drought escape, drought tolerance, drought avoidance, and drought recovery (Levitt, 1972). These mechanisms are not mutually exclusive and provide the crop with the ability to resist drought at any given period during its growth cycle. Escape mechanisms allow the crop to complete the drought sensitive growth stages during periods of adequate moisture or to complete the cycle prior to an onset of a drought (Ekanayake, 1989).

Avoidance is the ability to endure drought or exclusion of a stress by maintaining high water potentials of the plant through higher levels of water absorption due to a

better distributed and larger root system and reducing the water loss by stomata control. Tolerance is the ability to survive an internal stress due to dehydration tolerance or avoidance mechanisms (Ekanayake, 1990)

Drought reduces plant productivity by inhibiting growth and photosynthesis (Taiz*et al.*1998). A positive correlation between photosynthesis rate and crop yield is commonly found (Pooterand Remkes, 1990), but factors changing assimilate partitioning and utilization can reduce this association (Guo*et al.* 2002).

Drought stressimposed at any growth stage during the growing season may reduceyield of tubers. Emergence, tuber initiation, and tuber developmental stages are the most vulnerable stages. A yield reduction is due to a reduced number of tubers set and a poor tuber size distribution. Drought also affects tuber quality by producing growth cracks, elongated or spindly tubers due to alternate maturing and re-growth of the canopy and cyclic cell expansion of tubers. Also transient drought conditions produce more malformed tubers than those exposed to a continuous drought (Ekanayake, 1989).

#### **2.4. Mechanism of drought tolerance in crops**

When a genotype yields better than another under a severe strain of drought, it is relatively more drought resistant (Blum, 2005). The strain of drought is developed when crop demand for water is not met by the supply, and plant water status is reduced. Plants can resist drought by either dehydration avoidance or by dehydration tolerance. Drought resistance in terms of the physiology involved interacts with the magnitude and the timing of the stress.

There are several mechanisms through which plants exhibit resistance or tolerance to drought as described by Blum (2005).

#### **2.4.1 Dehydration avoidance and tolerance**

Dehydration avoidance is defined as the plant capacity to sustain high plant water status or cellular hydration under the effect of drought. Hence, by this mechanism the plant avoids being stressed because plant functions are relatively unexposed to tissue dehydration. Crop plants avoid dehydration by enhanced capture of soil moisture, by limited crop water loss, and by retaining cellular hydration despite the reduction in plant water potential (Blum, 2005).

Dehydration tolerance is defined as the relative capacity to sustain or conserve plant function in a dehydrated state. This is sometimes seen as the second defense line after dehydration avoidance. A legitimate measure of genetic variation in desiccation tolerance is the comparative function at low tissue RWC. Dehydration tolerance as an effective drought-resistance mechanism in crop plants is rare. It exists in the seed embryo, but once germinated the plant loses its tolerance. Extreme desiccation tolerance is known in resurrection plants and some attempts are made in various laboratories to use this tolerance for improving crop plants. Dehydration tolerance, like freezing tolerance, requires that the plant enter a quiescent or a dormant state (Blum, 2005).

## **2.4.2 Enhanced capture of soil moisture**

Deep soil moisture is available and a long root to reach this moisture is simply as effective as a long rope in a deep well. Genetic variation exists in potential root length (maximum root length measured under non-stress and non-restrictive soil

conditions (Blum, 2005). However, when plants are exposed to a drying soil, root morphology and growth can change to the extent that the potential root length, whether it is short or long, becomes irrelevant. In cereals, for example, drying, hard topsoil resists the penetration and establishment of adventitious (crown) roots while existing roots receive all transient assimilates and grow deeper (Blum and Ritchie 1984; Asseng*et al.* 1998).

Shoot/root dry matter ratio increases under drought stress, not because of an increase in root mass but due to a relatively greater decrease in shoot mass. Root mass rarely increases under stress. However, root length and depth may increase in a drying soil even at a reduced total root mass. Hence, total root dry matter or its ratio to shoot dry matter is not helpful information towards selection. It is not absolutely clear whether the capacity for developing longer roots under stress is compatible with a high yield potential phenotype. When all their requirements are effectively supplied, plants do not need a large root. Root mass under very productive drip irrigation systems is relatively small. In such a system a large potential root is a waste of dry matter. However, under conditions of unsecured soil resources, a potentially large root is required to ensure capture of resources under erratic conditions. This form of insurance may pose a load on yield potential if a large root is expressed in large root mass (Blum, 2005).

## **2.4.3 Reduced plant size, leaf area and leaf area index (LAI)**

Reduced plant size, leaf area, and leaf area index (LAI) are a major mechanism for moderating water use and reducing injury under drought stress (Mitchell *et al*. 1998). Often, crop cultivars bred for water-limited environments by selection for yield under stress have a constitutively reduced leaf area. The radioactive energy load on the

canopy (net radiation), of which only a fraction is used for photosynthesis, is dissipated mainly by transpiration. A reduction in transpiration can be achieved by reducing net radiation by way of reflection, namely increasing crop albedo. Various plant-surface structures allow an increase in albedo (Holmes andKeiller 2002).

#### **2.4.4 Water use efficiency (WUE)**

Epicuticular wax or plant glaucousness reduces cuticular conductance and reflects incoming radiation at the ultra violet (UV) and 400–700 nm wavelengths to the extent that leaf temperature and transpiration are reduced without a reduction in stomata conductance. This is expressed in greaterwater use efficiency (WUE) for the glaucous genotype (Premachandra*et al.* 1994). Reduced leaf chlorophyll content expressed in yellowish or pallid green shade of color is indicative of reduced antenna complexes at the Photo system II reaction centre. This reduces photo synthetically active radiation (PAR) absorption and subsequently water use. Such varieties were found adapted to dry and cold conditions (Watanabe *et al.* 1995).

However, at the same time, these reflective properties that are beneficial under drought stress were often associated with reduced photosynthesis and yield potential (Premachandra*et al.* 1994; Sanchez *et al.* 2001). Programmed moderated crop water use has become an important agronomic practice in maximizing crop production in dry land environments that are largely based on stored soil moisture use. According to Blum andNaveh (1976), planting geometry was even found to be effective in reducing water use by increasing plant competition at a given plant density.

### **2.4.5 Osmotic adjustment**

An increasing number of reports also provide evidence on the association between high rate of osmotic adjustment (OA) and sustained yield or biomass under waterlimited conditions across different cultivars of crop plants. Since OA helps to maintain higher leaf relative water content (RWC) at low leaf water potential (LWP), it is evident that OA helps to sustain growth while the plant is meeting transpiration demand by reducing its LWP. 'Osmotic adjustment sustained turgid maintenance and hence the yield-forming processes during moderate and severe water stress' (Ali *et al.* 1999). Beyond the effect on cellular hydration, other putative roles of OA was assembled under the vague term of 'osmoprotection' (Rontein*et al.* 2002). Such is the possible role for cell compatible osmolytes in protecting enzymes against heat inactivation (Paleg*et al*, 1981). Associations between OA and cellular membrane stability under drought stress have been suggested (Riga andVartanian 1999; Chandra Babu*et al*. 2004).

The limited studies of dehydration tolerance in crop plants revealed that genotypic variation in plant recovery from dehydration, as a measure of tolerance, was positively correlated with plant water status retained during desiccation rather than with a capacity to retain function at a dehydrated state. It is also extremely interesting to note the conclusion made by Chaves *et al*. (2002), that 'Differences among species can be traced to different capacities for water acquisition, rather than to differences in metabolism at a given water status'. If all the available literature on crop drought resistance is taken together it can be suggested that both natural selection and selection by man have given preference to dehydration avoidance over

dehydration tolerance as the major strategy for coping with drought stress, with the exception of resurrection plants.

The adaptation of plants to water stress conditions is also being determined by stomata activity, water uptake, morphology of leaves, among other physiological parameters. Plants respond to drought by producing abscisic acid (ABA) which stimulates the closure of stomata to reduce water loss and will automatically affect availability of carbon dioxide for photosynthesis which can cause oxidative injury.In addition to a plant's ability to avoid and/or endure water stress, photosynthetic recovery following rehydration is pivotal to dictate a plant's resistance to drought and to prevent dramatic declines in crop yield (Chaves *et al*., 2009). It was shown that recovery from a severe stress was a two stage process: the first stage occurs during the first hours or days upon re-watering, corresponding to the improvement of leaf water status and stomata reopening (Pinheiro*et al*., 2005; Antonio*et al.,* 2008; Hayano-Kanashiro*et al*., 2009); and the second stage lasts several days and requires de novo synthesis of photosynthetic proteins (Kirschbaum, 1988). Previous stress intensity and/or duration are crucial factors affecting both the velocity and the extent of recovery of photosynthesis (Miyashita *et al*., 2005; Flexas*et al*., 2006).

#### **2.5 Drought screening methods and parameters**

According to International Rice Research Institute (IRRI) 2006) several field and laboratory screening methods have been used successfully to screen for drought resistance, including line-source sprinkler irrigation, rainout shelters, and measurement of drought susceptibility index (DSI). It is however, absolutely important to use a simple and easily repeatable method when screening for drought tolerance in target environment.

The most obvious means to select for improved drought tolerance is to withhold water or reduce irrigation and compare the response of various genotypes through several parameters. The major parameters that have been successfully used in estimating the level of drought tolerance in plants include

#### **2.5.1 Chlorophyll fluorescence**

There is generally a decrease in the rate of photosynthesis during drought stress which results in an increase in abscisic acid concentration leading to stomata closure to reduce water loss that may affect yield (Bower *et.al*. 1992). Abscisic acid major role is the stomata adjustment and its accumulation is known to induce gene expression Skriver*et al*. (1991). Severe drought leads to stomata closure which leads to yield reduction (Turner, 1979).

#### **2.5.2 Relative Water content (RWC)**

Relative water content (RWC) is one of the parameters used to estimate the level of drought tolerance in plants.This determines leaf water status in plants and is a component to consider the ability of a plant that maintains tolerance during drought. It is measured in terms of fresh weight and dry weight (Beadle *et. al*. (1987).

#### **2.5.3 Leaf area**

 The leaf area maintains water balance and it is responsible for the light energy that can be absorbed to provide chemical energy and is determined by stem phonology, morphology, leaf size and emergence (Blum, 1996).

## **2.5.4 Canopy Temperature**

Canopy temperature is measured using an infrared thermometer (IRT). A lower canopy temperature is an indication that a plant has capacity to take up soil moisture content and maintain better water status.

## **2.5.5 Dry matter content (DMC)**

The dry matter content and the moisture can be used as an index to determine stress in crops. Both are a good indicator of drought resistance as a result of its high sensitivity and irritability (Ekanayake*et.al.* 2004).

# **2.5.6 Yield**

The yield of cultivars can be compared after undergoing stress conditions. It is an indicator while selecting because there has to be a correlation between a resistant cultivar and the yield (Turk *et al*.,1980).

## **CHAPTER 3**

## **3.0MATERIALS AND METHODS**

### **3.1 Study site for pre- screening**

The pre-screening was carried out in the glass house inside Agricultural Research Council, Vegetable and Ornamental Plant Institute, Rodeeplaat, Pretoria. The institute is situated about 25 km from the north of central Pretoria on the Moloto/Kwamhlangaroad (Latitude 25.604 $\mathrm{^{0}S}$ , Longitude 28.345 $\mathrm{^{0}E}$  and altitude 1159m)

## **3.2. Planting materials**

The plant materials used for this study were sweetpotato accessions obtained from the gene bank of Agricultural Research Council - Vegetable and Ornamental Plant Institute (ARC- VOPI, Roodeplaat) Pretoria. (Table 2). The materials consist of old land races, imported cultivars and ARC breeding lines/cultivars. All the cultivars were pre-screened for drought screening.

<b>NO</b>	<b>CULTIVAR</b>	<b>ORIGIN</b>	<b>SKIN COLOR</b>	<b>FLESHCOLOR</b>	<b>STORAGE ROOTSHAPE</b>
1	Wit Blesbok	<b>RSA</b>	Copper	Dark cream	Obovate - Long elliptic
2	Lobed JIII	<b>RSA</b>	Purple	Cream	Long irregular
3	TO-1-1-B	<b>RSA</b>	Purple	Cream	Long irregular - Long oblong
4	Malavuwe III VM-5B	<b>RSA</b>	Purple	White	Long oblong
5	Hlabisa 4	<b>RSA</b>	Pink cream	White	Very long elliptic
6	3 mnde wit	<b>RSA</b>	White	White	Long irregular - Long oblong
$\overline{7}$	6 mnde wit	<b>RSA</b>	White	White	Long irregular - Long oblong
8	Chingowa	Zambia	Cream	Cream	Long oblong - Long irregular
9	Xushu 18	Taiwan	Purple	White	Long elliptic - Oblong
10	Yan Shu 1	Taiwan	Pale purple	White	Heavy oblong
11	Atacama	Peru	Dark purple	White	Obovate - Round elliptic
12	Tacna	Peru	Copper	Pale yellow	Elliptic - Heavy elliptic

**Table 2.Sweetpotato cultivars used for box screening and their characteristics**



### **3.3 Plastic box pre-screening**

The pre-screening of accessions was done at the glass house in ARC-Roodeplaat, in Pretoria, between March and August 2008. Stem cuttings of the 50accessions were planted in plastic boxes of size 155cm  $\times$  77cm  $\times$  23cm. The boxes were filled with a special soil mixture (5: 2: 2 sand: soil : vermiculite). Sweetpotato cuttings of about 30cm long from each accession were cut and planted 2 eyes below the surface and 3 eyes above the surface for uniformity of development. Eight accessions were planted in each box divided into two rows. The plant spacing was 15cm between rows and 10cm between plants and a box contained ten rows (5 plants/row). In each box 8 cultivars and 2 control cultivars planted in each box to serve as positive and negative controls namely Lethlabula (drought tolerant) and Resisto (drought sensitive) based on previous screening by ARC-VOPI. The design was randomized complete block design (RCBD) with 6 replicates consisting of a total of 38 boxes.

The experiment was watered for 10-14 days for establishment after which water was withheld to induce stress. The experiment was concluded exactly 60DAP when 60- 70% of the plants showed severe stress and wilting.



**Fig 1.Sweetpotato plants in boxes after establishment**



**Fig 2.Sweetpotato plants showing signs of water stress.**

# **3.4 Field experiment**

# **3.4.1 Field experiment layout**

The field trial was done at Lwamondo Agricultural Station in Thohoyandou in Limpopo Province, South Africa. It was done in collaboration with the Madzivhandila College of Agriculture between March and September 2009. Thohoyandou was used for the experiment because it is a subtropical area which is ideal for winter planting season when receiving minimal rainfall. It lies at 23.06'S latitude and 30.38' E longitude with an altitude of 618m above sea level. Climate conditions are subtropical with average annual rainfall of 752mm with over 80% occurring between October and December. The average maximum and minimum temperature in the area is between 28.5 and 13.7 $\rm ^{0}C.$ 

The 12 cultivars selected for the trials were Zapallo, Resisto, Ejumula, Tacna, 1999- 3-1, 2004-16-1, 2004-5-2, 2004-9-2, W-119, Phala, 2003-24-2 and Ndou based on the results of the pre-screening trial.

An area of land measuring 20m  $\times$  60m was used for the trial. Field preparation included making ridges of 0.3m high and spaced 1m between the center of the ridges. 120 cuttings of each of the 12 cultivars selected from box screening were planted. Before planting Lime ammonium nitrate (LAN, 28%N) was applied at 150 kg/ha (110g/plot) and Supergrow (1.85% P) was applied at 150 kg/ha (110g/plot) to the field by broadcasting method and incorporated into the soil.

The cuttings were planted the next day in a triple row of 8 plants per row (24 plants/plot) with spacing of 1m between rows and 0.3m between plants and replicated 5 times. The design was randomized complete block design (RCBD). The cultivar Resisto was included as drought sensitive control. Two border rows were planted on each side of each block. The whole plot was watered using overhead irrigation for 7 days to facilitate plant establishment after which water was withheld till the end of the experiment.

### **3.4.2 Field experiment management**

Weeding of was done manually to remove unwanted plants and no further irrigation was applied during the growing period.

#### **3.5 Data collection**

#### **3.5.1Plastic box pre-screening**

Visual observations were made weekly on severely wilted plants using a rating scale of 1-5. Where 1- brown stems, 2- stem wilted 3- bottom leaves dry 4- leaves wilted, 5-vigorous. The number of days to severe wilting of the plants and the number of dead plants were also counted and recorded. The data were collected every week. Data analysis for tolerance parameters were performed with GenStat Release 9.2 and included an analysis of variance (ANOVA) to obtain mean values, and the Student's protected t-LSD test was calculated at the 1% significance level. The multiple t-distribution test procedure of Gupta and Panchapakesan (1979) was performed to group the lines as sensitive, intermediate or tolerant to drought stress.

#### **3.5.2 Field experiment**

Data were collected on two plants at the middle rows and the following parameters were recorded and evaluated. The data were collected at 42 days after planting (DAP), 84 DAP, 120 DAP.

#### **3.5.2.1 Plant growth**

Lengths of the shoots were measured using a meter tape and two plants were selected from each plot and measured with the meter tape and the lengths of the shoots were recorded.

#### **3.5.2.2 Dry matter content**

Plants were harvested and the fresh weight of both the roots and the shoots were taken separately using a measuring scale. The fresh roots were washed of soil particles and weighed for fresh weight and then cut into pieces and put in a labeled envelope and thenoven-dried at  $72^0C$  for 24hrs to get the dry weight. The shoots

were placed in a labeled envelope and oven-dried at  $40^0$ Cfor 24hrs to get the dry weight. The weights were recorded in a data sheet. The dry matter was calculated as follows:

Dry matter % = [(Fresh weight – dry weight) / Fresh weight] \*100

## **3.5.2.3 Canopy temperature**

Recordings of the temperature of the canopy were done in the early hours of the morning using an infra-red thermometer (RaytekRaynger ST20). This was taken at 1m from the plot edge and 50cm above the canopy, focusing on the leaves only to reflect exact reading.

### **3.5.2.4 Yield**

The storage roots of the whole plot were harvested at 120DAP. The storage roots were thereafter graded into marketable, unmarketable based on their shapes, sizes, weights and defects. Marketable root mass is greater than 100g with a diameter of 3cm and above when measured and no noticeable pest attack or diseases. Unmarketable roots those less than 100g and the diameter is less than 3cm and those showing defects (cracks, irregular shape) and pest infestation.

## **3.6 Data Analysis**

## **3.6.1. Plastic box screening**

Data were analyzed for Analysis of variance (ANOVA) using Agronomix (2008) computer software. Means were compared by least significant difference (LSD) at 0.001% probability level.

Twelve cultivars were selected for field trials and these are the best performing cultivars during pre – screening. The cultivars selected were Zapallo, Ndou, Ejumula, Tacna, Resisto, 2004-16-1, 2004-5-2, 2004-9-2. 1999-3-1, 2003-24-2, W-119 and Phala.

# **3.6.2 Field experiment**

Data collected were subjected to statistical analysis using Agronomix (2008) computer software. Analysis of variance (ANOVA) was computed and means were compared by the least significance difference at 0.001% probability level.

#### **CHAPTER 4**

## **4.0 RESULTS AND DISCUSSION**

## **4.1 Drought effect on pre-screening**

The results of the pre-screening trial are shown in Table 3. Most cultivars survived between 46 and 60 days before dying. The mean number of days to death was 56.44. The accession with the shortest daysto wilting was Yan Shu1(46.57) and the longest were Atacama and 2004-16-1(60.00).



### **Table 3. Result of plastic box screening experiment**





DTD – days to death, S- Sensitive, I- Intermediate, T- Tolerance, SD- Standard deviation

Most of cultivars started wilting around 57 days when the experiment was almost concluded indicating that the cultivars are generally tolerant to water stress. According to Muhammad *et al* (2011), the physiological responses of plants to a deficit of water include leaf wilting, a reduction in leaf area, leaf abscission, and the stimulation of root growth by directing nutrients to the underground parts of the plants.Observations made on the number of dead plants after the experiment were a better measure to discriminate the entries in terms of drought response. The least number of dead plantswere found in Atacama and 2004-16-1. In contrast entries at the bottom of the table had lost 4 plants during drought stress and were very sensitive to drought stress.The best performing cultivars that were selected for field trials had potential drought tolerance capabilities which allow them to still flourish under prolong water stress. In addition, imported varieties which performed well in similar pre-screening experiments were also selected, as well as recommended cultivars for Lwamondo area Ndou and Resisto and Lethabula serve as controls. It is expected that these cultivars will also show acceptable yield when subjected to periodic moisture stress under field condition. The parameters days to death and nr dead were efficient in distinguishing drought tolerance response of the entries.

Success in breeding for drought tolerance has not been as pronouncedas for many other traits. This is partly due to lack of simple, cheap, and reliablescreening methods to select drought-tolerant plants and progenies from the segregatingpopulations and partly due to the complexity of factors involved in droughttolerance. Singh *et al*. (1999) described a simple wooden box pre-screening method showing good correlation with drought tolerance at vegetative and reproductive stages, to select drought-tolerant plants at the seedling stage. Several experiments on drought screening under greenhouse conditions have also been reported in many crops (Laurie *et al* 2009, Govindaraj*et al* 2010,Pereyra-Irujo*et al* 2007.,Gholami*et al* 2012, Winter *et al* 1988, Ijaz andKhaliq 2007). Wooden box seedling screening is suitable when there are a lot of plants to be screened (Singh *et al* 1997). It is essential in its ability to determine stress at developmental stages. Singh *et al*. (1999) applied this method in screening large number of cowpea cultivars. The parameters used for evaluation were moisture content, flowering, yield and wilting point. The wooden box method was recommended because it is simple and non-destructive and it can easily be used to screen large number of cultivars. Similarly Laurie *et al* (2009) screened large number of sweetpotato cultivars using the plastic box method instead of the wooden box. The accessions were also planted out in the field and screened for drought in rain shelters. Drought tolerance was measured in terms of yield reduction and drought sensitivity index (DSI) of Fisher and Mauer, (1978). The result of the screening in plastic boxes and planting on the field shows there is a relation between number of days to wilting and DSI. The result shows that this method is quick, simple and reliable and can be very effective especially for screening large number of genotypes. Van Heerdenand Laurie, (2008) conducted similar experiment on sweetpotato to determine effect of prolonged

restriction of water on yield, Cultivars were planted in a rain out shelter and irrigated using different water regimes. It was concluded that there was a significant reduction in marketable storage yield with the best yield coming from the cultivars supplied with higher volume of water.

In vitro screening method proves to be an ideal method to screenlarge set of germplasm with less effort, accurately and the growth pattern differences are due to genotypes with least environmental influences. The method used in this study is similar and it also revealed a numberof drought tolerance cultivars among the 50 cultivars that were pre-screened.

## **4.2 Drought effect on growth and development**

There were significant differences among the cultivars in respect to canopy temperature at this stage. The highest mean canopy temperature was found in W119 with an average of 27.24 while the lowest of 20.62 was recorded in 2003-24-2 (Table 4). These figures are quite low and attest to the drought tolerant capabilities of these cultivars. A lower canopy temperature in drought stressed plant indicate a better capacity for taking up soil moisture and for maintaining better plant water status(Blum, 2009).Blum *et al* (1989) used canopy temperature of drought stressed wheat genotypes to characterize yield stability under various moisture conditions. A positive correlation was found between a drought susceptibility index and canopy temperature in a stressed environment. Stark and Pavek (1987) reported similar report on sweetpotato. Infrared canopy temperature provides an efficient method for rapid non- destructive monitoring of plant response to water stress (Idso*et al*, Jackson et.al.1981). The average stem length of 38cm which is the longest was recorded in Zapallo while the shortest

was found in 2004-5-2 with a length of 28.6cm. Kirk *et. al.,* (2001) indicated in an experiment on eggplant that water stress reduces both stem and internodes diameter. This result is consistent with that of Kirk*et.al*. (2001) as there was a general reduction in both stem length and stem diameter in the studied cultivars under water stress conditions in the field. Ejumula had the highest number of leaves with a mean value of 37.4 while the least mean value was found in both Phala and 2004-16-1**.**

S/N	Cultivar	Canopy	Root dry	Shoot dry	οf Number	<b>Stem</b>
		temperatur	matter $(\%)$	matter (%)	leaves	Length
		$e(^{\circ}C)$				
	Tacna	22.62	19.83	17.94	28.20	31.40
$\overline{2}$	Zapallo	21.50	17.07	15.80	32.80	38.80
3	<b>Ndou</b>	22.02	20.99	19.10	29.00	31.20
4	Phala	24.08	16.10	19.27	16.60	28.80
5	Ejumula	22.66	22.94	19.33	37.40	37.40
6	1999-3-1	21.24	21.93	25.63	21.00	31.60
7	W-119	27.24	23.09	23.06	30.80	32.80
8	2003-24-2	20.62	18.82	21.97	22.60	29.60
9	2004-16-1	25.26	19.20	20.47	16.60	34.40
10	$2004 - 5 - 2$	24.64	15.86	14.17	18.80	28.60
11	2004-9-2	22.76	18.08	15.73	23.20	30.40
12	Resisto	21.02	18.42	24.37	18.20	30.00
	<b>MS</b>	19.61	30.13	63.31	23.78	53.18
	<b>Grand mean</b>	22.97	19.36	19.47	24.60	32.08
	CV%	22.97	16.70	17.14	24.60	32.08
	<b>LSD</b>	5.982	4.1881	4.314	10.16	6.464

**Table 4 Growth parameters collected on 42DAP**

The severity of stress condition became intense at 84DAP (Table 5). There were also little or no differences in the responses of the cultivars to the canopy temperature at that stage and the lowest temperature of 13.5 $^0$ Cwas recorded in 2004-9-2 while the highest was recorded in Ndou  $(17.8^{\circ}C)$ . There was however a significant difference among the cultivars in the stem lengths.The longest length of 40cm was recorded in both Tacna and Ndou while the lowest length was found in 2004-5-2. The highest mean values for number of leaves were recorded in Zapallo (40), the lowest value of 22 was found in 2004-5-2. Moayedi*et al.,* (2007) carried out a drought screening experiment on wheat where a number of cultivars were planted on the field to determine drought tolerance using 4 irrigation regimes. It was discovered that drought stress significantly decreases Relative Water Content(RWC) and has a strong effect on photosynthetic rate and also that the stress leads to increase in leaf and canopy temperature Siddique*et. al* (2010)

Observations made at 120DAP indicated that the cultivars were matured for harvesting as they all show signs of stress with most of the plant already wilting.There was no significant difference in the canopy temperature among the cultivars. 2004-9-2 has the lowest canopy temperature of  $26.2^{\circ}$ C while the highest canopy temperature of  $30-33^0C$  were found in W-119, 2003-24-2, 2004-16-1 and 2004-5-2. There was a noticeable difference in the stem length among the cultivars, the longest stem length of 50cm was recorded in Resisto while the shortest length of 27.0 cm was found in W-119. The highest number of leaves of 60 was found in Zapallo while the lowest number was recorded in 2004-16-1.





#### **4.3 Drought effect on dry matter content**

For 42 DAP the highest root dry matter percentage was recorded in W-119 with a value of 23.09%,Ejumula followed with 22.94%. The rest of the cultivars did have a dry matter percentage ranging between 15 and 20 percent. There was a significant difference among the cultivars in the percentageshoot dry matter,the highest was recorded in 1999-3-1 having 25.63% and it was closely followed by W-119 and 2004- 9-2 with 15.75%. The lowest percentage shoot dry matter of 14.17% was recorded in 2004-5-2. The observed root dry matter also follows a similar pattern (Table 4). There was a general reduction in the root dry matter in all the cultivars under water stress conditions. Demagante*et. al.,* (1989) indicated that storage root drymass is correlated positively with vegetative growth.Similarly, Indira and Kabeerathumma (1988), Indirama (1994) and Ekanayakeet.al.(2004) reported a reduction in root dry mass under stress conditions. Indira and Kabeerathumma (1988), Indiramma(1994) and Ekanayake*et al*. (2004) all reported a reduction in root dry mass under water stress condition. The variation in dry matter content can also be dependent on various factors such as soil type, pest, diseases, cultivar and climate (Roseand Vasanthakalam,2011).

Despite the severity of the stress, some cultivars still show good traits reflecting in their dry matter accumulation at 84DAP. The highest root dry matter of 60.90% was recorded in Resisto and this was followed by 2004-5-2 (59.39%) and Ndou (56.85%) (Table5). Most of the rest of the cultivars had mean values ranging between 37 to 49 % with the lowest of 31.08 recorded in Tacna. Equally, Resisto had the highest shoot dry matter and the lowest was found in Tacna.



### **Table 6 Observation made at 120 DAP**

At 120DAPEjumula had the highest root dry matter percentage of 37.49% followed by Phala (37.49%). The lowest of 23.68% was found in 1999-3-1. Other cultivars performed well having values ranging between 23 to 37% (Table 6).

### **4.4. Drought effect on Yield**

Water stress is a common phenomenon and it severely reduces yields of field crops grown under rainfed conditions(Jangpromma*et al.,* 2010a).The marketable yield ranges from 0.96 t/ha to 3.83t/ha (Table 7). The highest marketable yield was recorded in Zapallo (3.83t/ha), Tacna (3.63t/ha), Ndou (3.12t/ha). The lowest marketable yield of 0.58t/ha was found in 2003-24-2.The ANOVA shows that there were significant differences among the cultivars. Also, the three cultivars with the highest marketable yield mostly had the highest total yield. The highest total yield was recorded in Tacna (9.24t/ha), Zappalo (6.16t/ha), 2004-9-2(5.58t/ha). These yield values were comparable to the average yield values of 5-10t/ha normally recorded for sweetpotato grown under subsistence farming under rain-fed conditions. The lowest yield was recorded in 2004-5-2 (1.69t/ha).

S/N	<b>Cultivar</b>	<b>MYLD</b> (t/ha)	T-YLD (t/ha)	<b>Survival</b> <b>Rate (%)</b>	Root dry matter $(\%)$	<b>Shoot</b> dry matter (%)
	Tacna	3.63	9.24	81.66	28.23	28.23
2	Zapallo	3.83	6.16	77.49	33.49	33.49
3	Ndou	3.12	4.50	75.00	31.89	31.89
4	Phala	1.05	1.91	59.99	37.79	37.79
5	Ejumula	2.31	3.81	75.83	35.41	35.41
6	1999-3-1	1.83	3.00	81.66	28.11	28.11
	W-119	1.83	3.23	77.50	34.01	34.01
8	2003-24-2	0.58	1.97	64.16	31.10	31.10
9	2004-16-1	1.31	2.18	68.33	36.15	36.15
10	2004-5-2	0.96	1.69	53.33	29.67	29.67
11	2004-9-2	2.02	5.58	66.66	32.20	32.20
12	Resisto	1.23	2.31	59.16	43.65	43.65
	<b>MS</b>	5.67	25.48	448.54	97.28ns	97.28ns
	GrandMean	1.979	3.801	70.069	33.47	33.47
	CV(%)	31.3	28.48	16.70	32.09	32.09
	LSD	0.7895	1.3799	14.918	13.692	13.692

**Table 7. Yield data collected at harvest at 120DAP**

#### **4.4.1 Drought effect on survival rates**

Cultivars Tacna and 1999-3-1 has the highest survival rate of 81.66% followed by W-119 (77.50%) Zapallo (77.49%), Ejumula (75.83%) and Ndou (75.0%).The lowest was found in 2004-4-2 (53.33%). This result is an indication that these cultivars had a very good mechanism to tolerate water stress.Van Heerdenand Laurie (2008) conducted an experiment on sweetpotato to determine effect prolonged restriction of water on yield. Various cultivars were planted under irrigation using different water regimes with specific nozzles based on the calculated soil water content which was monitored on a daily basis. It was discovered that there was a significant reduction in marketable storage yield with a best yield coming from the cultivars supplied with higher volume of water. Saraswati*et.al.,* (2004) also evaluated sweetpotato cultivars for drought in a pot experiment in a glass house at James Cook University, North Queensland. Yield and yield related parameters such as leaf growth, dry biomass, root dry weight and internodes length were used to successfully determine the drought tolerant cultivars.It was concluded that all the cultivars show reduction in in plant growth and yield as a result of decrease in soil water content. Bourke (1989) reported that drought had the greatest effect on sweetpotato yield during the root bulking phase.

## **CHAPTER 5**

## **SUMMARY AND CONCLUSIONS**

Among the 50 cultivars pre-screened for drought 12 cultivars were found to be drought tolerant based on the number of days to wilting. The cultivars includeTacna, Zapallo, 2004-9-2,Ndou, 2004-16- 1, 2003-24-2, Resisto, W-119, Ejumula, Phala, 2004-5-2, 1999-3-1.

The 12 pre- screened cultivars were further evaluated in field trials and five cultivars (Tacna, Zapallo, 2004-9-2, Ndouand Ejumula) were considered to have the greatest tolerance to water stress.

Based on the above findings, these five best performing cultivars were therefore recommended to be included as parents in the drought tolerant sweetpotato breeding program.

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