

**DEVELOPMENT OF ALUMINIUM TOXICITY
TOLERANCE SELECTION SYSTEM FOR SORGHUM (*Sorghum Bicolor L*)
MOENCH IN ZAMBIA**

BY

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THE DEGREE OF MASTER OF SCIENCE IN PLANT BREEDING AND
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DECLARATION

I, Sally Chikuta hereby declare that this dissertation represents my own work and that it has not been previously submitted for a degree , diploma or other qualification at this or any other University.

Signature:

Date:

APPROVAL

This dissertation of Sally Chikuta has been approved by the University of Zambia as partial fulfillment of the requirements for the award of the degree of Master of Science in Plant Breeding and Seed Systems.

Signature

Date

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DEDICATION

This work is dedicated to my lovely daughters, Sasha and Elly. I love you.

ABSTRACT

Sorghum (*S. bicolor*, L) is a major crop of the hotter and drier regions of the tropics and subtropics grown by resource poor farmers for their subsistence. It can also be cultivated in marginal lands and areas of high rainfall characterized by low pH soils high in aluminium (Al). The overall objective of this study was to characterize selected sorghum varieties for Al tolerance, while the specific objectives were, to determine performance of selected sorghum varieties grown in Al prone environments, to identify root characteristics associated with Al tolerance and to develop a selection criterion for Al tolerance in sorghum. Twenty sorghum genotypes (previously identified) were evaluated at three sites with high soil Al levels and in the laboratory. Genotypes, concentrations and interactions were significantly different ($P \leq 0.001$) for all the parameters studied in the laboratory which included root length, shoot length, number of lateral roots, shoot and root biomass. The correlation between the laboratory attributes and grain yield were highly significant. Direct effects ranged from 0.5 for lateral roots to 0.7 for shoot length. Indirect path effects towards grain yield by root length and root biomass were 0.6 and 0.5 respectively via shoot length. Significant differences ($P \leq 0.05$) were observed in the field study for seven of the nine parameters that were measured and/or derived. Interactions were significant for plant height and grain yield. The entries were significantly different for all the measured and derived parameters except for plant count, pest score and agronomic score. Significant differences were observed for location on days to 50% flowering, plant height, pest score, sundried head weight and grain yield. Associations between the measured and/or derived parameters and grain yield were significant. The direct path effects were low except for head weight which had a significant contribution of 1.4. Direct contributions on yield by other parameters were less than 0.1. Plant height, pest score and agronomic score had significant indirect effects of 0.7, 0.7 and 0.5 respectively via head weight. Results indicate that laboratory attributes can be used to predict high yielding sorghum genotypes suitable for low pH soil with Al toxicity. Selection of head weight and head harvest index would contribute effectively to high yielding sorghum genotypes in low pH soil with high Al. The superior genotypes recommended for Al tolerance are 11, 16, 17 and 20.

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1.0 INTRODUCTION

Sorghum bicolor L. is one of the most important traditional cereal crops of the hotter and drier regions of the tropics and subtropics (Rao et al., 2001). It belongs to the grass family poaceae, tribe *andropogoneae* and sub tribe *sorghastrae*. It is the fifth most important cereal in the world following wheat, maize, rice and barley (Onwueme et al, 1991).

In the developing world the crop is largely grown by resource poor small scale famers for their subsistence. Sorghum can be used for several purposes such as grain for human consumption (68 to 71% carbohydrate) or animal feed in form of hay, silages and pasture. The juice extracted from the sweet sorghum stem is used in production of biofuels, alcohol and molasses while the stalks find use in the manufacture of paper (MACO, 2002; Munyinda et al, 2008).

Zambia requires two different types of sorghum. One type with tropical adaptation for high rainfall areas (1000 to 1400mm) of the north (Chisi et al., 1997). These sorghums should be semi to photo-period sensitive and must possess high tolerance to soil acidity and anthracnose. The second type suitable for the southern regions of Zambia should be sub-tropical in adaptation. These varieties must be photo-period insensitive with good levels of resistance to moisture stress, heat and a number of diseases such as anthracnose, and downy mildew.

Given the current effects of climate change and increased use of marginal lands, sorghum may be the crop of choice for both wet and dry regions with unreliable rainfall (Dogget, 1988). This is because the crop can tolerate hot and dry climatic

conditions but can also be cultivated in areas of high rainfall. However, high rainfall areas are usually associated with low pH soils which result from excessive leaching of soil nutrients. Acidic soils may contain toxic concentrations of aluminium and manganese which are the most important causes of mineral toxicity in soils (Singh, 2005). Aluminium toxicity in plants leads to membrane instability and protein denaturation which are manifested through reduced root growth, root discoloration and lack of lateral roots. Generally aluminium toxicity in plants can be observed at pH levels below 5.5 (Singh, 2005).

Sorghum production in Zambia is restricted by use of land where pH is low and also use of unimproved varieties. Small scale farmers who grow sorghum in Region III, a Region prone to Al toxicity, use unimproved varieties which have low yield potential, are tall and nonresponsive to improved management (MACO, 2002).

Application of agricultural lime to soils having low pH of less than 5 is one way of restoring soil fertility. However, liming is often not economic or practical because of the slow movement of lime especially in the deeper layers of sub soils (Matsumoto *et al.*, 2001). Furthermore, heavy application of lime may have adverse effects on some crops in a rotation or cause deficiencies of certain nutrients (Kochian *et al.*, 2005). Thus, developing cultivars with improved tolerance to acid soil stress offers an alternative solution to address the problem of low yields in sorghum (Jones *et al.*, 2006).

Steps must be taken to find long term solutions to address the problem of low productivity due to Al toxicity. There is need to develop sorghum varieties that are tolerant to Al toxicity and this calls for identification of important plant characteristics

conferring tolerance for use in breeding programmes for Al tolerance in sorghum cultivated in Zambia. In order to increase production levels of Sorghum in acidic soils of Zambia a number of strategies can be adapted.

In this study, the hypothesis was that sufficient genetic variation exists in the sorghum germplasm in Zambia to select for Al tolerance.

The overall objective of this study was to characterize selected sorghum varieties for Al tolerance. The specific objectives were to determine performance of selected sorghum varieties grown in Al prone environments, determine root characteristics associated with Al tolerance and develop a selection criterion for Al tolerance in sorghum.

2.0 LITERATURE REVIEW

Sorghum is relatively undeveloped and has a remarkable array of untapped variability in grain type, plant type, adaptability and productive capacity. It probably has more undeveloped and underutilized genetic potential than any other major food crop (Lost crops of Africa, 1996). More than 30 000 varieties are present in the world's sorghum collections (Leder, 2001).

Certain sorghum varieties have been reported to tolerate high aluminium concentrations found in acidic soils (Mohammadi *et al.*, 2003). However, most sorghum cultivars are not tolerant to high concentrations of Al because sorghum improvement programmes have been conducted at locations with near neutral pH soil.

2.1 Botanical Description of Sorghum

Sorghum belongs to the grass family poaceae, tribe *andropogoneae* and sub tribe *sorghastrae*. The sub tribe has two genera: *Cleistachne* Benth, with four species in southern Africa and India. The other genera are sorghum, which has a wide distribution throughout the warmer regions of the world (Onwueme *et al.*, 1991).

Some common names of sorghum include great millet, guinea corn in West Africa, durra in Sudan, mtama in East Africa, jowal, jola and cholam in India, milo and sorgo in the United States and kaoliang in China (Onwueme *et al.*, 1991).

Sorghum is often an annual crop with a single stem varying in height from 1-5 m. Tillers come out in some cultivars when they are grown as a ratoon crop.

Roots

First a single main root is produced from which a large number of much-branched lateral roots are produced. Many adventitious fibrous roots are formed from the lowest nodes of the stem (Onwueme *et al*, 1991) .

Stem

The stem is usually erect, dry or juicy, insipid or sweet, grooved and nearly oval. The peduncle (top inter node) is not grooved. Young sorghum plants can be distinguished readily from maize plants because of the saw-toothed margins of sorghum leaves. Crown buds give rise to tillers (Onwueme *et al*, 1991).

Leaves

The leaves are alternate in two ranks and the leaf sheaths are 15-55 cm long and they encircle the stem. The midrib is prominent (Onwueme *et al*, 1991).

Inflorescence

The sorghum inflorescence is a loose to dense panicle, usually erect and has many primary branches bearing spikelets. The sessile spikelet of each pair is fertile while the pedicellate spikelet is either sterile or staminate. There are two florets in the fertile spikelet, the lower sterile and the upper fertile. Sorghum is about 95% self-pollinating (Onwueme *et al*, 1991).

Seeds

A well developed panicle may contain as many as 2000 seeds. The seeds are roundish, ovoid to flat, and can be white, pink, red, yellow or brownish. Mature seeds have a black spot near the base. Immature seeds develop the black spot after drying. White or yellow seed grains are generally preferred for food. The other pigmented types are

slightly bitter. The pigmentation is largely in the outer layers of the seed coat and this may be removed by a limited amount of milling (Onwueme *et al*, 1991).

2.2 Origin and distribution of Sorghum

Many annual and perennial species of sorghum are found in the wild form. The greatest variation in the genus is found in the northeast quadrant of Africa, north of latitude 10° and east of longitude 25° E. The crop probably originated here and it's believed that a form (forms) was domesticated in the Ethiopian region some 5,000 or more years ago to produce sorghum bicolor. The cultivated and wild forms were then spread by man throughout Africa (Onwueme *et al*, 1991).

Sorghum was taken from Eastern Africa to India, probably during the first millennium BC and from there to China. Sorghum then spread to the Mediterranean countries. The crop was introduced to the United States from Africa in the mid nineteenth century. It was grown along the Atlantic coast and then carried westward to the drier regions (Onwueme *et al*, 1991).

2.3 Chemical nature of Aluminium (Al)

Aluminum (Al) phytotoxicity is one of the biggest agronomic problems in acid soils. Al is a major constituent of most soils but only when it moves into soluble or exchangeable form can it affect plants. Exchangeable aluminium values may be high in soils with pH below 5.5 but may occur at pH values as high as 6.0 in heavy textured soils (Matsumoto *et al.*, 2001). The critical soil pH, at which aluminium becomes exchangeable in toxic concentration, depends on many factors, including the predominant clay minerals, organic matter level, and concentrations of other cat ions, anions and total salts as well as the species or cultivar of the plant being considered.

Al is primarily in form of insoluble oxides, $\text{Al}(\text{OH})_3$, at neutral pH. However, as the soil becomes more acidic (less than 5.5), phytotoxic forms [$\text{Al}(\text{OH})^{2+}$ and $\text{Al}(\text{OH})^{3+}$] of Al are released from the soil solution (Zhang *et al.*, 2007). This compromises more than 40% of the potentially arable soils in the world (Delhaize *et al.*, 2004). The problem is exacerbated by the use of ammonium fertilizers and acid rain (Beebe *et al.*, 2008). The solubility of the Al compound and severity of their toxic effect on the plant are influenced by many chemicals and physical factors such as pH, oxidation-reduction potential, the composition of clay minerals, organic matter and exchangeable cation concentration. Bal Krishna (2005) reported that soil pH was the major factor that controls Al^{3+} availability and uptake of Al from soil into tea plants. The Al^{3+} ion will be dominant when the soil pH is less than 5.0.

Organic matter in the soil can reduce the toxicity of Al to the plants (Kollmeier *et al.*, 2000). Kollmeier *et al.* (2000) further reported that the critical level of Al in an Alfalfa pot experiment increased as the soil organic matter level increased from 6.6 to 81.6 g kg^{-1} .

Al is present in water, soil and air but most of it is incorporated into aluminosilicate soil minerals and only very small quantities (at submicromolar levels) appear in soluble forms capable of influencing biological systems (Pineros *et al.*, 2002).

Different forms of Al occur in soil solution; $\text{Al}(\text{OH})^{2+}$ and $\text{Al}(\text{OH})_2$ at pH 4 to 5, Al^{3+} at pH 5.5 to 7.0 and $\text{Al}(\text{OH})_4^-$ at pH 7.0 to 8.0 (Wenzl *et al.*, 2002).

Intensification of the process of Al compounds solubilisation is connected with the degree of soil acidification caused by washing out of alkaline metal ions (Na^+ , K^+ ,

Ca^{2+} , Mg^{2+}) from the soil and a decrease in the pH soil solutions. Al ions translocate very slowly to the upper parts of plants (Ma, 2000).

2.4 Diagnosis of aluminium toxicity in plants

The diagnosis of aluminium toxicity from visual signs in plants is unreliable (Matsumoto *et al.*, 2001), and critical plant concentrations of aluminium are ill defined. The aluminium concentration in leaves of Lucerne is of little value in determining toxicity (Pineros *et al.*, 2002). A value above 150 mg Al/kg DM in sub clover leaves may indicate toxicity (Ma, 2000). Soil exchangeable aluminium concentration is used as a guide to the likelihood of aluminium toxicity. Aluminium levels greater than 15 mg/kg may be a problem and above 50 mg/kg toxic, in which case the economics of liming should be considered to overcome this problem (Wenzl *et al.*, 2002). Testing for Al tolerance in the field has a number of disadvantages some of which include: the presence of other toxic elements and variability in the Al content throughout the field. This constraint can be alleviated by exposing seedlings to known levels of Al in nutrient solution.

2.5 Aluminum (Al^{3+}) Toxicity symptoms in plants

The main symptom of Al toxicity is a rapid inhibition of root growth, which may translate to a reduction in vigor and crop yields (Kochian *et al.*, 2005). Roots injured by high aluminum become stubby and thick, dark colored, brittle, poorly branched and rubberized with a reduced root length and volume (Nguyen *et al.*, 2003). The rapid inhibition of radical growth can be seen in the primary and lateral root apices, which become thick and turn brownish-gray (Rout *et al.*, 2001). These symptoms become evident after a few minutes or hours of the plants being exposed to micro molar

concentrations of Al in hydroponic solutions (Rengel and Zhang, 2003). Radical inhibition coincides with a decline in cell division (Frantzios *et al.*, 2001) and elongation of the root cells, which then induces significant lignification of the cell wall by crossing with pectins (Rout *et al.*, 2001; Jones *et al.*, 2006). This alteration prevents water absorption which is essential for the transport of nutrients through the apoplast, eventually causing a decrease in yield and grain quality (Zheng and Yang, 2005; Raman *et al.*, 2002). The shoot is also inhibited due to limiting supply of water and nutrients.

Furthermore, Al also triggers membrane lipid peroxidation and apoptosis or programmed cell death (PCD) (Pan *et al.*, 2001). It has been reported that prolonged exposure to this element can induce and produce responses of rapid change in other biochemical and physiological processes (Rengel and Zhang, 2003). This is why the symptoms at foliar level resemble phosphorus deficiency, preventing plant growth, turning mature leaves dark green, stems purple and killing leaf apices (Wang *et al.*, 2006). In other cases, Al toxicity reduces calcium (Ca) transport, making young leaves curl, preventing the development and growth of the petiole (Rout *et al.*, 2001). According to some scholars, excesses of Al also induce symptoms of Fe deficiency, which was observed in *Sorghum bicolor* (Rao, 2001), *Triticum aestivum* and *Oryza sativa* (Rout *et al.*, 2001). Several studies indicate that Al affects the normal operation of cell membranes, causing enzymatic disorders and affecting the nuclear DNA (Pan *et al.*, 2001). It acts on the phosphate groups, altering their topology and recognition by polymerase DNA, modifying the entire functioning of the explicative machinery due to the increased rigidity of the double helix (Rout *et al.*, 2001; Zhang *et al.*,

2002). In addition, Al is closely linked to other DNA-associated molecules, such as phosphorylated proteins (histones) (Kochian, 2005). Al interferes in the normal operation of the Golgi apparatus and in the peripheral cells of the apex of intact roots, in their quiescent center, mitotic activity and DNA synthesis (Rout *et al.*, 2001). Al may also affect the mechanism that controls the organization of cytoskeletal microtubules as well as the polymerization of tubulin by delaying disassembly during mitosis (Franzios *et al.*, 2003). This would affect the direction of the microtubules, which is closely related to cell expansion (Zheng and Yang, 2005).

2.6 Aluminum (Al³⁺) tolerance mechanisms

Species can vary in their ability to grow in acid soils with severe Al phytotoxicity (Jones and Ryan, 2003). A study done by Rangel *et al* (2007), on bean seedlings showed a reduction in root growth when sown in high Al concentration. In a similar study done by Musa and Munyinda (1986) on wheat, Al treatment was observed to reduce the root length but increased the number of lateral roots as the Al concentration increased in a nutrient solution. However, there was no differential effect with shoot length across Al concentrations.

Reid *et al* (2001) also found that Al injury to barley was characterized by a decrease in root length with a reduction in the number of lateral roots as Al concentration in the nutrient solution was increased. The reduction in root length exposed to different Al concentrations varies among varieties of the same species depending on level of tolerance to Al concentration (Musa and Munyinda, 1986; Rangel *et al.*, 2007; Reid *et al.*, 2001).

A recent study on sweet sorghum by Munyinda, *et al* (2008) showed a reduction in the root length as well as number of roots as Al concentration was increased. Al tolerance mechanisms have been classified into two main types: a) those that exclude Al from the root cells and b) those that allow Al to be tolerated once it has entered the plant cells (Barceló and Poschenrieder, 2002).

Species in tropical areas are very resistant to Al stress and some of these species can accumulate high concentrations of Al in the leaves, greater than 1% of their dry weight (Jones and Ryan, 2003). However, certain plants referred to as Al accumulators may contain over ten times more Al without any injury i.e. tea plants. The Al content in these plants can reach as high as 30mg per gram of dry mass in older leaves (Matsumoto *et al*, 2001). By contrast, cereals like *Secale cereale*, *Zea mays*, *Hordeum vulgare*, *Triticum aestivum*, X *Triticosecale*, *Sorghum bicolor* and *Avena sativa* do not accumulate high concentrations of Al internally but rather use the Al exclusion mechanism through organic acid exudation (Caniato *et al.*, 2007). This may be one of the most widely used mechanisms by most of the species studied. Nevertheless, important differences are manifested in some of the features of these mechanisms in each species, including the nature of the inducibility by Al, the organic acids released, and whether al-induced gene action plays a role in tolerance (Kochian , 2001).

2.6.1 External tolerance mechanism (exclusion)

Some species detoxify Al in the rhizosphere by exuding organic acid from their roots (Li *et al.*, 2002). This exudation is located in the radical apexes of some species as this is a region which is very sensitive to Al toxicity due to constant cell division and

elongation (Mossor-Pietraszewska, 2001). The organic acids commonly secreted are malate, citrate and oxalate. Malate and citrate are present in all cells given that they are involved in the mitochondrial respiratory cycle (Jones and Ryan, 2003). It suffices to make mention here that sorghum uses the C₄ malate cycle (Lost crops of Africa, 1996). Organic acid levels vary between species, cultivars and even between tissues of the same plant under identical growth conditions. In addition, organic acid biosynthesis and accumulation increase drastically in response to environmental stress (López-Bucio *et al.*, 2000). It has been observed that tolerant genotypes exude a greater amount of organic acids than sensitive genotypes, which would support the notion that organic acid exudation is an Al tolerance mechanism (Delhaize *et al.*, 2004). However, it has been reported that Al-sensitive species of wheat show a greater accumulation in the cortical tissue (5 to 10 times more) than the tolerant genotypes exposed for the same period of time (Kashif *et al.*, 2004). Some organic acids such as citrate, malate and oxalate are able to form stable complexes with Al (Guo *et al.*, 2007), where the Al-citrate complex bond is strongest, followed by the Al-oxalate and Al-malate complexes, which are insoluble and not available for plants (Jones and Ryan, 2003). This is because Al is a metal that tends to form strong complexes with the oxygen donor ligand (Barcelo and Poschenrieder, 2002). The Al-citrate complex is important in terms of plasma membrane transport. Because citrate has been used to desorb Al from root cells and its release has been suggested by some scholars to be an important Al resistance mechanism in snap bean and corn, it is possible that it can cross the plasma membrane (Piñeros and Kochian, 2001).

The transport of these organic acids from the radical cells is mediated by the anionic channel activity in the plasma membrane (Ma *et al.*, 2001). These anionic channels might be Al-activated, which was demonstrated using the patch-clamp technique on isolated protoplasts of wheat and maize radical apexes (Ryan and Jones, 2003). Using anionic channel inhibitors such as niflumic acid would support the existence of these channels as elements for organic acid exudation in response to Al (Piñeros and Kochian, 2001).

A study conducted by Ryan *et al* (2001) demonstrated the Al induced release of root organic acids from Al-resistant genotypes. 36 different wheat cultivars were screened for Al resistance and showed that Al-stimulated malate release correlated with Al-resistance

2.7 Screening methods

Several native and crop species exhibit significant genetic based variability in their responses to Al toxicity. This variability is useful to plant breeders for the production of Al-tolerant crops. Selection and breeding of crops for Al tolerance is a useful approach to increased production on acid soils. For selection of genotypes tolerant to Al, a precise screening technique to evaluate sensitivity of plants to Al is needed. This requires a rapid and reliable system to discriminate between Al-tolerant and Al-sensitive genotypes (Springer, 2004).

Generally, the Al-screening technique can be classified into laboratory screening and field screening. Laboratory screening methods include screening of plants with solution-soaked paper and solution culture methods (Naserian *et al.*, 2007), soil-petri

dish method (Stass *et al.*, 2007), and screening in pots in a greenhouse (Tazeem *et al.*, 2009).

For sorghum, screening in the field (Mohammed and Ezeaku, 2006), and in pots or nutrient solution are commonly used for selection of Al-tolerant genotypes. A rapid screening method is needed to select a large number of new genotypes or new inbred lines in plant breeding, such as solution-soaked paper, solution culture and soil-petri dish methods used to evaluate Al-tolerant sorghum. All of these rapid screening techniques use the response to Al of the rate of seedling germination and root development. However, the method using such growth responses would curtail the accuracy of screening (Yashida and Yashida, 2000). Detection systems not dependant on the rate of seedling or root development, would greatly improve the success of screening procedure (Abdel-Hady, 2006).

Screening by using hematoxylin staining of seedling roots (hematoxylin staining method) which requires less time and simpler pH management than the other methods, is very useful for selection or screening a relatively large population in breeding program. Measurement of Al tolerance is based on the staining pattern of the root. The hematoxylin staining method is a very common technique for the evaluation of Al-tolerance in wheat (Kashif *et al.*, 2004) and barley (Shahinnia *et al.*, 2005), but there have been no reports on the use of hematoxylin staining methods in the screening of Al-tolerant sorghum.

Field screening for Al tolerance would be the best approximate for selecting Al-tolerant plants. In practice, however, reliable ranking of tolerance in the field screening is difficult because the Al concentration in soil may not be uniform and

because environmental factors interact with soil Al to mask the expression of Al tolerance (Naserian *et al.*, 2007). Screening by using the growth response to Al added to the soil in pots at in a greenhouse (referred to as growth-response method hereafter) may be superior in this respect.

It is important to compare the laboratory screening methods with the field screening methods. There was a correlation between the performance of sorghum in the greenhouse study and grain yield in the field (Abdel-Hady, 2006). The plants that showed severe reduction of shoot or root weight in a greenhouse study showed also low grain yield in the field. There was also a similar genotype response to Al-induced stress in nutrient solution and to acid-soil stress in the field (Shahinnia *et al.*, 2005).

2.8 Path analysis

Path coefficient analysis is one of the reliable statistical techniques which allows the quantification of the direct and indirect effects of characteristics on yield (Dewey and Lu, 1959; Medasir *et al.*, 2009). The types and extent of contribution of the characteristics to grain set or yield is determined with correlation and path analysis. Path analysis requires that cause and effect exists between the variables and the experimenter assign direction in the causal system, either a prior or based on experimental evidence (Dewey and Lu, 1959).

Much of the studies on path analysis in agriculture have been used by plant breeders to assist in identifying traits that are useful as selection criteria to improve crop yield (Medasir *et al.*, 2009; Mohammed and Ezeaku, 2006 and Singh *et al.*, 2003).

3.0 MATERIALS AND METHOD

The study involved field and laboratory experiments. Twenty advanced sorghum lines/entries obtained from Golden valley agricultural research trust under the sorghum breeding research programme were used and these included the entries listed below.

Table 1: List of Sorghum entries evaluated in the 2008/2009 growing season

Entry No.	Entry Name
1	SDS 89426
2	PRGC/E#69414
3	ICSV 1089BF
4	MACIA*DORADO
5	ZSV-18
6	ZSV-30
7	ZSV-31
8	SDS 4378-1-1-1
9	SDS 1023-10-2-4-1-3-2
10	SDS 876-3432(OT)8-2-1
11	[SDS3845×SDS4548]F6-10-2
12	[SDS3845×SDS4548]F6-10-3-2
13	[SDS2690-2×M91057]8-2-1-1
14	SDS 2690-2-3-5-1
15	KSV-7
16	KSV-10
17	KSV-4
18	SDS 4380-S7
19	ZSV-12
20	WP-13

3.1 FIELD EXPERIMENT

The field experiment was conducted in the aluminium prone areas of Masaiti, Mpongwe and Mansa in region III. The Randomised Complete Block Design (RCBD) was used with 3 replications at each site. Each entry was sown in four rows (each 4 m long) per plot with an inter row spacing of 75 cm. After thinning an intra row spacing of 50 cm was left between the plants. The total area per plot was 12 m². 200 Kg ha⁻¹ of Compound D fertilizer (10N: 20P: 10K: 10S) was applied at time of planting to each plot. Urea (46% N) was applied a month after planting at the rate of 100 Kg ha⁻¹ to each plot.

The crop was ready for harvesting after six months from the time of planting.

Soil samples were collected at each of the three sites and these were tested for pH and Al levels.

Parameters that were recorded included days to 50% flowering, Plant ht (cm), plant count, sun dried head, moisture content, Head harvest index, grain yield, Agronomic score (1 for best to 5 for worst), Disease score (1 for most resistant to 5 for most susceptible) and pest score (1 for least damaged to 5 for most damaged).

3.2 LABORATORY EXPERIMENT

The laboratory assessment was done at the University of Zambia, School of Agricultural sciences, Crop Science laboratory. The 20 sorghum entries were tested in nutrient solutions with varying Al concentrations of 0 mg L⁻¹, 4 mg L⁻¹, 8 mg L⁻¹, 12 mg L⁻¹, 16 mg L⁻¹ and 20 mg L⁻¹.

Twenty to thirty seeds of each entry were germinated on tightly covered Petri dishes lined with deionised water moistened filter papers for 48 hours. Seedlings of uniform root length were selected from the Petri dishes and elongated in distilled water in a dish for 24 hours. The seedlings were then transferred to 55 ml test tubes containing nutrient solutions of varying Al levels added as $\text{AlK}(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$ at 0, 4, 8, 12, 16 and 20 mg L^{-1} . Four seedlings were chosen per treatment per entry. The nutrient solutions used in this study was described by Kerridge *et al* (1971) and it provided the following in mg L^{-1} : 48.1 Calcium, 14.6 Magnesium, 42.61 Nitrogen, 23.5 Potassium, 0.02 Sodium, 0.03 Chlorine, 0.03 Manganese, 0.06 Copper, 0.03 Molybdenum, 0.16 Zinc, 0.32 Boron, 1.67 Iron (added as $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$) and 2.0 Phosphorus. The pH was initially adjusted to 4.2 and left unadjusted thereafter.

The Completely Randomised Design (CRD) was used with 4 replications for each entry. One seedling was placed in each test tube and left to grow for 10 days. The seedlings were supported over the nutrient solutions by polythene stoppers. The test tubes were then covered with black polythene bags to prevent algae from growing in the solutions and then placed on test tube racks. The solutions were aerated on a daily basis and changed once after five days. After ten days of growth, measurements of maximum shoot and root length were taken using a 30cm rule. The number of roots per plant were also noted. The plants were then dried in an oven for two days and then weighed to get the root and shoot biomass.

3.3 Statistical Analysis

The analysis of variance (ANOVA) was employed to detect statistical differences among different levels of parameters of interest for both the field and laboratory

experiments using GenStat Release 7.22. The Duncan's Multiple Range Test (DMRT) was used to separate the means. The MSTAT-C statistical software (MSU, 1988) was used to obtain the correlation coefficients and standard partial regression coefficients. The standard partial regression coefficients from multiple regressions were used as direct path coefficients and the indirect effects were determined by multiplying the correlation by the respective path coefficients (Ssango *et al.*, 2004). The conventional path analysis based on the method developed by Dewey and Lu (1959) was used.

4.0 RESULTS

Significant differences were observed for most of the parameters which were measured and/or derived in the field and laboratory analyses as indicated in Tables 3 and 8 respectively. Associations among the parameters are shown in Table 7 for the field analysis and Tables 15 and 16 for the laboratory analysis which indicate the direct and indirect path effects as well as the overall correlation between the measured and/or derived parameters with grain yield.

Soil analysis results for the three sites at which the field experiment was conducted are outlined in Table 2.

4.1 SOIL ANALYSIS RESULTS

Table 2: Soil analysis results for Mpongwe, Mansa and Masaiti

Sample no.	Sample Id	Sample Depth cm	pH CaCl ₂	Al mg/kg
1	Mpongwe	20	4.51	6.90
2	Mpongwe	40	4.91	4.10
3	Mpongwe	60	4.62	0.90
4	Mansa	20	5.10	8.20
5	Mansa	40	4.73	5.40
6	Mansa	60	4.69	2.10
7	Masaiti	20	4.16	10.10
8	Masaiti	40	4.31	7.10
9	Masaiti	60	4.12	2.70

4.2 FIELD EXPERIMENT

Table 3: Mean squares for the parameters measured and/or derived from twenty sorghum entries evaluated at three locations during the 2008/2009 growing season in Zambia

Source	DF	Days to 50% flowering	Plant count (No)	Plant Height (cm)	Disease score	Pest score	Agron score	Sundried Head wt (Tons/ha)	Head Harvest index	Yield (Tons/ha)
Replication	2	65.91	134.5	624.6	1.499	1.2847	0.817	0.0418	0.046792	0.4646
Location (L)	2	3742.94***	1150.4ns	71046.5***	5.6514ns	14.4764**	4.117ns	68.9358*	0.011997ns	32.1257**
Entry (E)	19	129.32***	3343ns	10245.8***	1.3784*	0.5702ns	1.779ns	4.1809***	0.021716***	2.7586***
L * E	38	32.55ns	5011.1ns	1388.5***	0.4935ns	0.4691ns	0.485	0.8451ns	0.007824ns	2.7586***
Error	114	24	12816.1	384.7	0.7543	0.6865	1.5	0.6942	0.005681	0.8112
CV%		6.0	25.1	13.6	14.2	16.4	18.6	28.7	11.9	30.8

***= Significant at $P \leq 0.001$, ** = significant at $P \leq 0.01$, * = significant at $P \leq 0.05$, ^{ns} = not significant, SED= Standard error of difference of means

Table 4: Means of parameters measured and/or derived from twenty sorghum entries evaluated at three locations during the 2008/2009 growing season

Entry	Days to 50% flowering (No)	Plant count (No)	Plant ht (m)	Dizz score	Pest score	Agron score	Sun dried Head Wt (tons/ha)	Head Harvest index	Grain Yield Tons/ha)	
1	81abc	47a	1.3defg	1.6a	1.4a	1.7a	4.7a	0.6ab	3.0ab	
2	78bc	43a	1.9ab	1.7a	1.6a	1.9a	2.9bcdef	0.6ab	2.0abcde	
3	84abc	40a	1.5cde	1.9a	1.6a	1.9a	2.3def	0.5b	1.4cde	
4	78bc	46a	0.9hi	1.6a	1.4a	1.7a	2.2def	0.6ab	1.4cde	
5	81abc	44a	1.4def	1.7a	1.5a	1.7a	2.7bcdef	0.7a	2.1abcde	
6	77bc	52a	1.8abc	1.4a	1.5a	1.7a	4.1abc	0.7a	3.2a	
7	84abc	38a	1.6bcdi	1.5a	1.4a	1.8a	2.9bcdef	0.6ab	2.2abcde	
8	90a	39a	1.2efghi	1.6a	1.6a	1.9a	4.3ab	0.6ab	3.1ab	
9	82abc	39a	1.2efghi	1.7a	1.5a	1.7a	2.7bcdef	0.6ab	1.9bcde	
10	79bc	39a	1.3defg	1.8a	1.5a	1.9a	1.8ef	0.6ab	1.2de	
11	86ab	37a	1.5cde	1.7a	1.5a	2.0a	3.6abcd	0.6ab	2.6abc	
12	82abc	43a	1.8abc	1.8a	1.4a	1.8a	2.7bcdef	0.7a	2.2abcde	
13	85abc	37a	0.9i	1.7a	1.4a	1.8a	2.3def	0.6ab	1.6cde	
14	79bc	41a	1.3defgh	1.7a	1.6a	1.9a	1.6f	0.6ab	1.0e	
15	86ab	36a	1.1fghi	1.7a	1.6a	1.7a	2.4def	0.6ab	1.4cde	
16	77bc	47a	1.9ab	1.7a	1.5a	1.8a	3.0bcdef	0.6ab	2.3abcd	
17	76c	50a	2.0a	1.6a	1.4a	1.8a	3.5abcde	0.6ab	2.5abcd	
18	83abc	45a	1.3defg	1.8a	1.6a	2.1a	2.5cdef	0.6ab	1.6cde	
19	78bc	43a	1.0ghi	1.8a	1.4a	1.5a	2.3def	0.5b	1.4cde	
20	79bc	43a	1.7abcd	1.7a	1.5a	1.8a	3.8abcd	0.7a	3.1ab	
Grand mean	81	42	1.4	1.7	1.5	1.8	2.9	0.6	2.1	
Location means	Mansa	84	42	1.4	1.6	1.5	1.8	2.2	0.6	1.5
	Masaiti	88	39	1.1	1.8	1.7	1.9	-	-	-
	Mpongwe	73	42	1.8	1.7	1.4	1.8	3.7	0.6	2.6

4.2.1 DAYS TO FIFTY PERCENT FLOWERING

Locations were significantly different ($P \leq 0.001$) for days to 50% flowering (Table 3). The overall means (Table 4) were 88, 84 and 73 days for Masaiti, Mansa and Mpongwe respectively indicating that plants in Mpongwe flowered earlier.

Results in Table 3 indicate that the entries significantly differed ($P \leq 0.001$) for days to 50% flowering. The earliest was entry 17 (76 days) and this was followed by entry 16 which was not significantly different from entries 6, 19, 14 and 12 which took between 77 to 79 days to reach 50% flowering (Table 4). The latest was entry 8 which took 90 days to flower but was not significantly different from entries 15 and 11.

There were no significant interactions between location and entries for this variable (Table 3).

4.2.2 PLANT COUNT

No significant differences were observed among locations and entries for this variable. Similarly no significant interactions were detected between the locations and entries (Table 3).

4.2.3 PLANT HEIGHT

Locations and entries were significantly different ($P \leq 0.001$) for plant height (Table 3). Plants in Mpongwe were taller (1.8 m) than those in Mansa (1.4 m). The shortest plants were entries were observed in Masaiti (1.1 m).

Entry 17 was the tallest (2.0 m) followed by entry 16 (1.9 m) which was not significantly different from entries 2, 12, 6 and 20 (Table 4). Entry 15 had the shortest plants but it was not different from entries 4, 19, 15, 9 and 8.

Significant ($P \leq 0.001$) interactions between entries and location were observed (Table 3). The entries in terms of plant height responded differently to the prevailing conditions at the locations. These interactions manifested through change in ranking. According to Table 5, entry 12 was the tallest in Mpongwe (2.5 m), second (1.4 m) in Mansa and fourth (1.7 m) in Masaiti. On the other hand entry 2 was also among the tallest in Mpongwe (2.5 m) but third in Masaiti (1.4 m) and fourth in Mansa (1.8 m). Similar changes in ranking are evident among the other entries across the locations.

Table 5: Height Means (m) for the 20 sorghum entries evaluated in Mansa, Mpongwe and Masaiti in 2008/2009 growing season

Entry	Mansa	Mpongwe	Masaiti	Mean
1	1.4 cdefgh	1.6 ef	1.0 de	1.3 defg
2	1.8 abc	2.5 a	1.4 ab	1.9 ab
3	1.4 cdefgh	1.9 cd	1.1 bcde	1.5 cde
4	1.0 h	1.0 g	0.9 de	0.9 hi
5	1.1 efgh	1.8 de	1.4 abc	1.4 def
6	1.6 bcde	2.4 ab	1.5 a	1.8 abc
7	1.6 bcdef	2.1 bc	1.1 bcde	1.6 bcdi
8	1.3 defgh	1.5 ef	0.8 e	1.2 efghi
9	1.1 fgh	1.6 ef	1.0 de	1.2 efghi
10	1.4 cdefg	1.6 ef	1.1 cde	1.3 defg
11	1.6 bcde	1.9 cd	9.5 de	1.5 cde
12	1.7 bcd	2.5 a	1.4 abc	1.8 abc
13	0.9 h	1.0 g	0.8 e	0.9 i
14	1.5 cdefg	1.5 ef	0.9 de	1.3 defgh
15	1.2 efgh	1.1 g	0.9 de	1.1 fghi
16	2.0 ab	2.4 a	1.4 ab	1.9 ab
17	2.2 a	2.5 a	1,2 abcd	2.0 a
18	1.3 cdefgh	1.5 f	1.2 abcd	1.3 defg
19	1.1 gh	1.1 g	0.9 de	1.0 ghi
20	1.5 bcdefg	2.4 ab	1,1 bcde	1.7 abcd
Overall mean	1.4	1.8	1.1	1.4

4.2.4 DISEASE SCORE

Locations were not significantly different ($P \leq 0.05$) for disease prevalence while entries showed significant differences (Table 3).

Despite the significant statistical difference, the entries were very similar in their ability to resist disease as can be seen in the mean separation in table 4.

4.2.5 PEST SCORE

Locations were significantly different (≤ 0.001) (Table 3) for pest infestation with plants at Masaiti being the most affected with a score of 1.7 followed by those at Mansa with 1.5 and then Mpongwe with 1.4 (Table 4).

The entries and locations were not significantly different ($P \leq 0.05$) for pest infestation and there were no significant difference in the interactions between the entries and the location (Table 3).

4.2.6 AGRONOMIC SCORE

The entries and locations were not significantly different ($P \leq 0.05$) for agronomic score neither were the interactions significant (Table 3).

4.2.7 SUN DRIED HEAD WEIGHT

This refers to sun dried weight of heads harvested from a demarcated area of the plot. To facilitate the comparisons these weights were converted into weight per unit area (ton/ha).

The locations and entries were significantly different ($P \leq 0.01$) for sun dried head weight (Table 3). Heads harvested in Mpongwe had higher weights (3.7 tons/ha) than those in Mansa (2.2 tons/ha) as presented in Table 4.

Entry 1 had the highest sun dried head weight (4.7 tons/ha) followed by entry 8 (4.3 tons/ha) and entry 6 (4 tons/ha). Entry 20 was similar to entry 11. Entry 16 had a sun dried head weight of 3 tons/ha and it was not different from means of entries 7, 2, 12, 5 and 9 (Table 4). Entry 14 showed the lowest sun dried head weight of 1.5 tons/ha.

No significant interactions between locations and entries were detected (Table 3).

4.2.8 HEAD HARVEST INDEX

The entries were highly significantly different ($P \leq 0.001$) for head harvest index (Table 3). The means as presented in Table 4 indicated that entries 6, 12, 5 and 20 had higher head harvest indices (0.7) than entries 10, 19 and 3 (0.5). The rest of the entries were intermediates with a harvest index of 0.6.

Locations were not significantly different and there was no interaction between the entries and locations (Table 3).

4.2.9 GRAIN YIELD

The locations and entries were highly significantly different at $P \leq 0.01$ for grain yield (Table 3). The entries in Mpongwe had an average location mean of 2.6 tons/ha and Mansa had a location mean of 1.5 tons/ha.

Entry 6 had the highest yield of 3.2 tons/ha which was not so different from entries 8, 20 and 1. The intermediates were entries 2, 5, 7, 12, 16 and 17. These were similar to each other in terms of grain yield (2.0 to 2.5 tons/ha). Entry 4 had the lowest grain yield (1.0 tons/ha) and was not different from entry 10. This was according to the mean separation indicated in Table 4.

The interactions between entry and location were significantly different as shown in the Table 3.

These interactions were both a change of ranking and differences in the rate of change from one location to the other (Table 6). The rate of change in the yield ranged from -12.5% for entry 18 to 125% for entry 7. Big variations were observed in this regard for the rest of the entries across the locations. Entry 8 was the highest yielding at Mansa but was fourth in ranking at Mpongwe while entry 20 was fourth at Mansa but second at Mpongwe. The highest yielding entry at Mpongwe, entry 6, was the third at Mansa. These shifts in ranking are observed across the locations for all other entries, confirming varietal differential response.

Table 6: Grain Yield means (ton/ha) for the 20 Sorghum Entries Evaluated at Mansa and Mpongwe in 2008/2009 growing season.

Entry	Mansa	Mpongwe	Across location Means
1	1.7 abc	3.7 abc	3.0 ab
2	1.1 bc	2.4 defg	2.0 abcde
3	0.9 c	1.6 ghi	1.4 cde
4	1.1 bc	1.6 ghi	1.4 cde
5	1.3 bc	2.6 defg	2.1 abcde
6	1.9 abc	3.9 a	3.2 a
7	1.3 bc	2.7 def	2.2 abcde
8	2.5 a	3.1 abcd	3.1 ab
9	1.4 bc	2.0 efghi	1.9 bcde
10	9.1 c	1.3 i	1.2 de
11	2.2 ab	2.4 defgh	2.6 abc
12	1.3 bc	2.6 defg	2.2 abcde
13	1.3 bc	1.6 ghi	1.6 cde
14	0.8 c	1.1 i	1.0 e
15	1.0 c	1.7 fghi	1.4 cde
16	1.3 bc	2.9 bcde	2.3 abcd
17	1.6 abc	2.8 cde	2.5 abcd
18	1.6 abc	1.4 hi	1.6 cde
19	0.9 c	1.6 ghi	1.4 cde
20	1.8 abc	3.8 ab	3.1 ab
Overall mean	1.5	2.6	2.1

4.3.11 ASSOCIATIONS AND PATH ANALYSIS OF MEASURED AND DERIVED FIELD PARAMETERS

The overall association between parameters measured and/or derived in the field and yield are indicated in Table 7. As simple correlation does not provide the true contribution of characters towards the yield, these correlations were partitioned into direct and indirect effects as shown in Table 7.

Table 7: Correlation, Direct (Diagonal) and Indirect Effect of measured and derived field parameters on Grain yield

Characters	Head harvest index	Agronomic score	Pest score	Disease score	Sundried Head wt	Plant ht	Plant count	Days to 50% flowering	Correlation with grain yield
Head harvest index	0.049	-0.118	0.002	0.002	-0.619***	0.035	0.005	0.020	0.054ns
Agronomic score	0.031	-0.189	0.005	0.015	0.501*	0.029	0.002	0.009	0.256ns
Pest score	-0.005	0.039	-0.026	-0.059	0.700***	-0.004	0.002	-0.015	0.357ns
Disease Score	0.001	0.028	-0.015	-0.099	0.302	-0.001	0.003	-0.006	0.213ns
Sundried head wt	-0.022	0.067	-0.013	-0.021	1.412***	-0.017	-0.001	-0.011	1.372***
Plant Ht	0.047	-0.016	0.003	0.001	-0.6635***	0.036	-0.001	-0.011	0.143ns
Plant Count	0.02	-0.0318	0.004	-0.025	-0.112	0.013	0.012	0.174	-0.38ns
Days to 50% flowering	0.020	-0.034	0.008	0.011	-0.299	0.382	0.004	0.056	-0.727***

***= significant at $P \leq 0.001$, * = significant at $P \leq 0.05$, ns= non significant

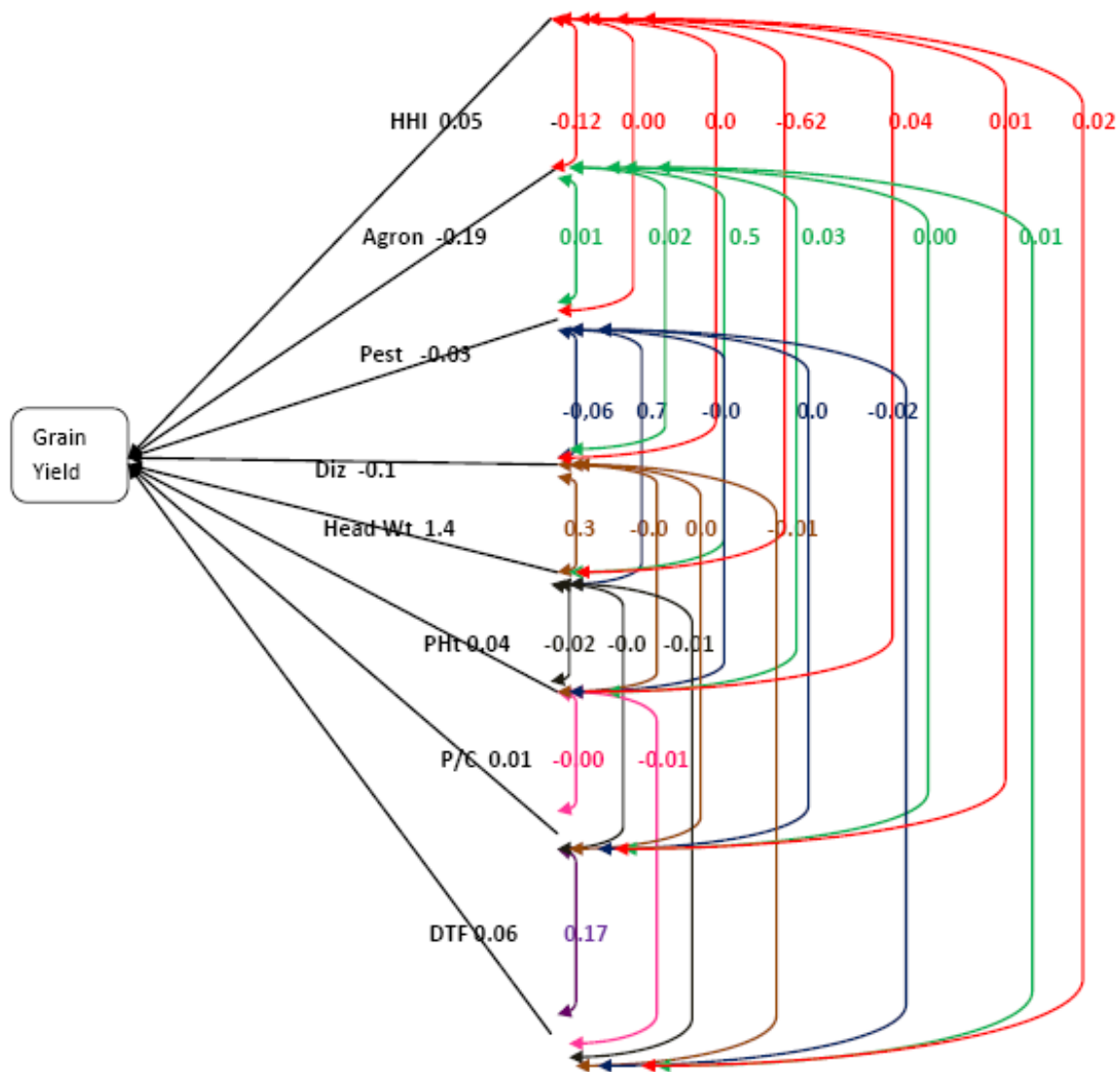


Fig 1: Path diagram showing causal relationships of eight predictor variables with a response variable (grain yield)

The overall correlation between days to 50% flowering and yield was significant at $r = -0.727$ (Table 7). It was partitioned into a direct effect of 0.056 and indirect effects of 0.382 via plant height and -0.299 via disease score, none of which were significant. Indirect effects through other variables were also very low and non significant ($P \leq 0.05$).

The association between plant count and yield was not significant at 5% probability level (Table 7). The overall correlation of $r = -0.38$ was partitioned into a direct effect of 0.03 and very small and not significant indirect effects as shown in Table 7.

There was a low correlation ($r = 0.143$) between plant height and yield (Table 7). This overall correlation was partitioned into a direct effect of 0.036 and an indirect effect of -0.6635 which was significant ($P \leq 0.001$). Other indirect effects were low and not significant.

Disease score had a low association with grain yield at 5% probability level. The overall correlation of $r = 0.213$ (Table 7) was partitioned into a direct effect of -0.099 and an indirect effect of 0.302 through sun head weight. The other indirect effects were low and not significant.

The overall correlation ($r = 0.357$) between grain yield and pest score was not significant (Table 7). The direct effect as it represents the effect of selecting for grain yield using pest score only was also negligible (-0.026). The indirect effects, as they represent the effects of selecting for grain weight, using pest score but through other variables, were negligible except via dried head weight (0.700) (Table 7).

The overall correlation between Agronomic score and grain yield was $r = 0.256$ (Table 7). This correlation was partitioned into a direct effect of -0.189 and a significant indirect effect of 0.5 via sun dried head weight. Other indirect effects were not significant (Table 7).

Head harvest index had an overall correlation $r = 0.054$ with yield (Table 7). The direct effect of the head harvest index towards yield was 0.049. Indirect effects were low and non significant

(Table 7). The direct effect is almost equal to the Correlation thus the relationship between head harvest index and grain yield is true. Selection through head harvest index directly would be effective.

The overall correlation between Sun head weight and yield was very highly significant at $r = 1.372$ (Table 7). This correlation was partitioned into a direct effect of 1.412 and indirect effects through other parameters were low and not significant (Table 7). Since the direct effect is almost equal to the correlation, the implication is that the correlation explains the true relationship and a direct selection through sun dried head weight would be effective.

4.3 LABORATORY EXPERIMENT

Entries were highly significantly different for all the parameters measured in the laboratory as indicated in Table 8. The mean performances of the entries are outline in Table 9.

Table 8: Mean Squares for parameters measured from twenty sorghum entries evaluated in the laboratory at the University of Zambia in October,2010

Source	DF	Root length	Shoot length	# of lateral roots	Root biomass	Shoot biomass
Replication	3	10.742	39.43	617.5	2.466	273.15
Concentration (C)	5	2926.35***	310.14***	84360.8***	187.371***	157.81ns
Entry (E)	19	59.767***	134.34***	567.7***	21.798***	286.26***
E * C	95	16.561***	27.19***	413.6***	6.260***	129.40*
Error	357	5.047	11.78***	1245	1.787	98.38
Cv%		24.6	40.5	41.2	40.5	10.5

***= Significant at $P \leq 0.001$, ** = significant at $P \leq 0.01$, * = significant at $P \leq 0.05$, ^{ns} = not significant, SED=Standard error of differences of means

Table 9: Means of parameters measured from twenty sorghum entries evaluated in the laboratory at the University of Zambia in October, 2010

ENTRY	Root length (cm)	Shoot length (cm)	Number Of lateral roots (No)	Root Biomass (mg)	Shoot Biomass (mg)	
1	7.6bcd	9.4abcd	30a	2.6b	8.4c	
2	9.8abcd	12.2ab	32a	2.5b	10.8bc	
3	7.7bcd	9.4abcd	27a	2.0b	9.6c	
4	8.9abcd	9.5abcd	31a	3.0b	12.3bc	
5	10.9abcd	9.8abcd	26a	3.0b	8.9c	
6	7.4cd	6.4bcd	19a	3.1b	9.1c	
7	7.6bcd	7.3abcd	21a	3.3b	9.3c	
8	7.3d	4.7cd	25a	2.8b	11.3bc	
9	7.3d	6.8abcd	22a	2.5b	8.3c	
10	7.6bcd	3.9d	19a	2.4b	6.0c	
11	11.8a	10.3abc	36a	3.5b	14.4b	
12	11.3ab	9.8abcd	29a	3.9ab	12.2bc	
13	9.6abcd	8.4abcd	28a	3.3b	9.8c	
14	9.4abcd	7.5abcd	26a	3.6b	11.1bc	
15	8.5abcd	7.2abcd	30a	3.5b	7.3b	
16	11.1abc	12.2ab	33a	5.8a	21.3a	
17	11.2abc	12.4a	34a	5.7a	15.5b	
18	9.8abcd	7.0abcd	23a	2.9b	9.0c	
19	7.5bcd	6.6abcd	27a	3.0b	9.2c	
20	10.1abcd	9.3abcd	26a	3.0b	9.7c	
Grand mean	9.1	8.5	27	3.3	10.5	
Concentration means	0 mg L⁻¹	17.5	11.5	73	5.5	12.7
	4 mg L⁻¹	16.2	10.0	63	4.8	11.5
	8 mg L⁻¹	6.2	8.8	17	2.9	9.5
	12 mg L⁻¹	6.0	7.2	6	2.5	9.5
	16 mg L⁻¹	4.4	6.6	2	2.4	10.6
	20 mg L⁻¹	4.4	6.7	1	1.6	9.0

4.3.1 ROOT LENGTH

The concentrations were highly significant at ($P \leq 0.001$) for root length (Table 8). An increase in aluminium concentration resulted in the reduction of the root length with sharp mean reduction

between 0 mg L⁻¹ (17.5 cm) and 4 mg L⁻¹ (16.2 cm). There was a steady decline between 12 mg L⁻¹ (6.0 cm) and 20 mg L⁻¹ (4.4 cm) (Table 9).

The entries responded differently ($P \leq 0.001$) across all Al concentrations (Table 8). According to results presented in Table 9, entry 11 had the longest roots (11.8 cm), followed by entry 12 (11.3 cm), which was not so different from entries 16 and 17. Other entries 2, 4, 5, 13, 14, 15, 18 and 20 had intermediary means ranging from 8.5 to 10.9 cm long. Entry 7 had the shortest roots (7.3 cm) and was similar to entries 9 and 6. Figure 1 shows the response of some of the entries (entries 3 and 5) to varying aluminium concentrations. The pictures show the behavior of entries in terms of root length in concentration 0 (longest roots) through 4, 8, 12 and 16 to 20 mg L⁻¹ (shortest roots).

The interactions between the entries and concentrations were significantly different ($P \leq 0.001$) for root length (Table 8). This indicates that entries were differently affected by the aluminium concentration. According to results in Table 10, there was a reduction, at different rates, in root length in most of the entries from concentration 0 through 4, 8, 12 and 16 to 20 mg L⁻¹. For example the root length for entry 8 reduced as the Al concentration increased from 0 mg L⁻¹ through to 20 mg L⁻¹ concentration by 58%, 34%, 26%, 0.0% and 27% compared to entry 10 whose root length reduced by 41%, 67%, 0.0%, 30% and 4%. Similar variation in the reduction rates of root length were observed among the rest of the entries.

Table 10: Root length means (cm) for the 20 sorghum entries evaluated in the laboratory at University of Zambia in 2009

Entry	0 mg L ⁻¹	4 mg L ⁻¹	8 mg L ⁻¹	12 mg L ⁻¹	16 mg L ⁻¹	20 mg L ⁻¹	Across conc mean
1	14.2cde	12.8cdef	7.3 cdef	3.0 g	4.2 defg	4.3 cdefg	7.6 bcd
2	16.0bcde	20.8ab	7.5 cde	4.7 defg	4.8 bcdef	5.0 bcde	9.8 abcd
3	17.7bcde	12.2 def	4.0 jk	3.8 efg	4.3 def	4.1 cdefgh	7.7 bcd
4	18.5 bcd	14.1 bcdef	5.6 ghij	5.7 cde	4.6 bcdef	4.7 bcdef	8.9 abcd
5	17.1bcde	19.2 abc	10.3 a	5.9 cde	5.9 ab	7.3 a	10.9 abcd
6	13.7 de	13.0 cdef	4.9 hijk	5.9 cde	3.3 fgh	3.6 efgh	7.4 cd
7	13.7 de	15.1 abcde	5.7 ghi	5.4 cdef	2.7 h	3.3 fgh	7.6 bcd
8	19.4 ab	8.2 f	5.4 ghij	4.0 efg	4.0 defgh	2.9 gh	7.3 d
9	14.4 cde	18.5 abcde	2.3 i	2.9 g	2.6 h	3.2 gh	7.3 d
10	20.2 ab	12.0 ef	4.0 jk	4.0 efg	2.8 gh	2.7 h	7.6 bcd
11	20.0 ab	21.9 a	8.8 abc	8.9 ab	5.3 abcd	5.9 b	11.8 a
12	17.8bcde	21.8 a	8.4 bcd	8.1 ab	5.9 abc	5.9 b	11.3 ab
13	14.2 cde	19.1 abcd	6.8 efg	8.4 ab	3.7 efgh	5.3 bcd	9.6 abcd
14	23.6 a	14.5 bcdef	3.5 kl	5.6 cde	3.8 efgh	5.5 bc	9.4 abcd
15	13.2 e	12.7 cdef	7.1defg	8.1 ab	4.8 bcde	5.4 bcd	8.5 abcd
16	18.8 abc	18.5 abcde	9.2 ab	9.6 a	6.7 a	4.0 defgh	11.1 abc
17	20.8 ab	20.0 ab	8.0bcde	8.9 ab	5.4 abcd	4.2 cdefgh	11.2 abc
18	20.7 ab	18.0 abcde	4.2 ijk	6.7 bcd	4.4 def	4.8 bcde	9.8 abcd
19	16.4bcde	12.9 cdef	5.0 hijk	3.3 fg	4.5 bcdef	2.9 gh	7.5 bcd
20	19.4 ab	19.1 abcd	6.4efgh	7.3 abc	4.4 cdef	4.3 cdefg	10.1 abcd
Overall mean	17.5	16.2	6.2	6.0	4.4	4.4	9.1

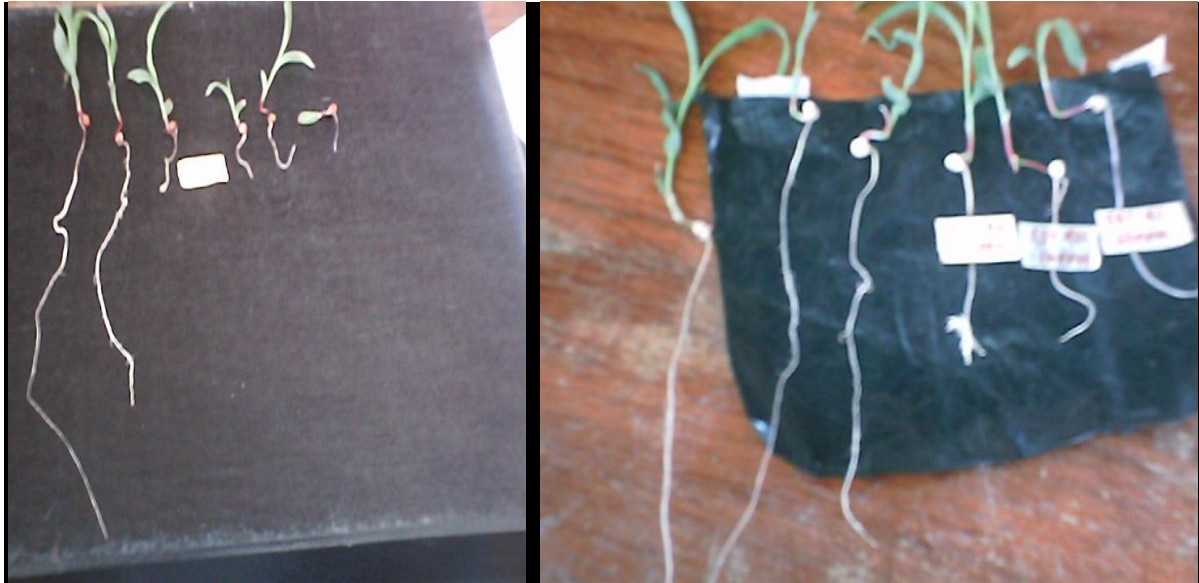


Fig 2: Pictures showing variations in root lengths for entries 3 and 5 respectively from concentration 0 (Longest) through 4, 8, 12 and 16 to 20 mg L⁻¹ (shortest)

4.3.2 SHOOT LENGTH

The concentrations were significantly different ($P \leq 0.001$) for shoot length (Table 8). The mean shoot length decreased steadily as the Aluminium level increased. The average shoot length for concentration 0 was 11.5 cm while it was 10 cm and 8.8 cm for concentrations 4 and 8 mg L⁻¹ respectively. Concentration 16 and 20 mg L⁻¹ had mean shoot lengths of 6.6 cm and 6.7 cm respectively (Table 9).

Entries were also highly significant ($P \leq 0.001$) in relation to shoot length (Table 8). Entry 2 had the longest shoot (12.4 cm) and was not different from entries 16 and 17 .This was followed by entry 11 (10.3 cm). Entry 10 had the shortest shoot (3.9 cm) and the rest were similar to each other with means lying in the range 6.6cm to 9.8cm. This was according to the mean separation in Table 9.

The interaction between entry and concentration was very highly significant ($P \leq 0.001$) for shoot length (Table 8). There was a notable change in magnitude for the entries in the different aluminium concentrations. Entry 4 for instance, had its longest shoots in concentration 0 mg L^{-1} (14.0 cm) followed by 4 mg L^{-1} (11.7 cm) and then 12 mg L^{-1} (7.8 cm). The shoots in concentration 16 mg L^{-1} were 7.4 cm long and 6.1 cm in concentration 12 mg L^{-1} (Table 11).

Table 11: Shoot length means (cm) for 20 sorghum entries evaluated in the laboratory at University of Zambia in 2009

Entry	0	4	8	12	16	20	Across conc.
	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mean
1	10.6 bc	13.2 abc	13.3 b	5.5 defg	7.8 cde	6.1 defgh	9.4 abcd
2	14.5 abc	13.6 abc	9.4 bcde	12.8 ab	13.0 a	10.1 abcd	12.2 ab
3	12.5 abc	12.3 abc	8.6 bcde	5.7 defg	9.3 abcd	7.8 bcdef	9.4 abcd
4	14.0 abc	11.7 abcd	10.4 bc	7.8 abcdefg	7.4 bcdef	6.1 defgh	9.5 abcd
5	14.6 abc	8.8 abcde	4.0 efg	9.8 abcde	11.0 ab	10.6 abc	9.8 abcd
6	3.7 d	9.2 abcde	6.4 cdefg	6.3 cdefg	5.8 defg	7.1 cdefg	6.4 bcd
7	8.9 cd	10.6 abcde	10.0 bcd	7.6 abcdefg	3.6 fgh	3.0 gh	7.3 abcd
8	10.2 bc	4.9 e	6.4 cdefg	3.6 efg	1.0 h	2.2 h	4.7 cd
9	12.2 abc	11.0 abcde	4.7 defg	6.8 bcdefg	2.6 gh	3.9 fgh	6.8 abcd
10	9.40 cd	5.4 de	1.6 g	1.8 g	3.4 fgh	2.0 h	3.9 d
11	11.2 abc	14.4 ab	13.5 b	4.9 defg	4.9 efgh	12.9 a	10.3 abc
12	10.3 bc	14.7 a	9.6 bcd	10.1 abcd	10.0 abc	3.9 fgh	9.8 abcd
13	8.0 cd	11.5 abcd	9.3 bcde	9.2 abcdef	4.9 efgh	7.4 bcdef	8.4 abcd
14	12.6 abc	7.1 cde	4.7 defg	5.3 defg	5.0 efgh	7.6 bcdef	7.5 abcd
15	8.1 cd	7.4 cde	10.4 bc	4.2 defg	4.4 efgh	8.9 abcde	7.2 abcd
16	17.7 a	7.0 cde	19.1 a	7.3 bcdefg	10.4 ab	11.7 ab	12.2 ab
17	16.6 ab	13.0 abc	13.8 b	13.7 a	7.8 bcde	9.9 abcd	12.4 a
18	10.2 bc	9.6 abcde	2.7 fg	7.8 abcdefg	7.1 bcdef	4.6 efgh	7.0 abcd
19	11.1 bc	7.9 bcde	7.5 cdef	3.1 fg	6.3 cdefg	3.9 fgh	6.6 abcd
20	13.0 abc	7.9 bcde	10.4 bc	12.1 abc	7.5 bcdef	5.1 efgh	9.3 abcd
Overall mean	11.5	10.0	8.8	7.2	6.6	6.7	8.5

4.3.3 NUMBER OF LATERAL ROOTS

Concentrations were significantly different ($P \leq 0.001$) for number of lateral roots (Table 8). Generally the number of lateral roots decreased with increase in the aluminium level.

The entries in 0 mg L^{-1} (73 lateral roots) and 4 mg L^{-1} (63 lateral roots) had more lateral roots than those grown in the 8 (17 lateral roots), 12 (6 lateral roots), 16 (2 lateral roots) and 20 mg L^{-1} (1 lateral root) concentrations (Table 9).

The entries showed significant differences ($P \leq 0.001$) in the number of lateral roots across the different concentrations (Table 8). Entry 11 had the most number of lateral roots followed by entries 2, 4, 16 and 17. However; the entries were similar according to the mean separation shown in Table 9.

The interaction between the entries and concentration for number of lateral roots was highly significant (Table 8) implying that the entries were significantly differently affected by the concentration. Entry 2 had 111 lateral roots in concentration 0 mg L^{-1} , 58 in 4 mg L^{-1} , 18 in 8 mg L^{-1} , 3 in 12 mg L^{-1} , 2 in 16 and 20 mg L^{-1} . Entry 14 had 95 lateral roots in concentration 0 mg L^{-1} , 49 in 4 mg L^{-1} , 5 in 8 mg L^{-1} , 6 in 12 mg L^{-1} , 1 in 16 and non in 20 mg L^{-1} (Table 12).

Table 12: Number of Lateral Roots Means for 20 sorghum entries evaluated in the laboratory at the University of Zambia

Entry	0	4	8	12	16	20	Across conc
	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mean
1	69 bcdef	82 ab	25 bc	1 b	2 d	1 c	30 a
2	111 a	58 bcde	18 cd	3 ab	2 d	2 bc	32 a
3	100 ab	52 cde	9d efg	1 b	1 d	1 c	27 a
4	98 abc	78 abc	5 fg	3 ab	2 d	2 bc	31 a
5	67 cdef	54 cde	28 b	4 ab	2 d	1 c	26 a
6	60 ef	48 de	5 fg	2 b	1 d	1 c	19 a
7	62 ef	59 bcde	3 fg	2 b	1 d	1 c	21 a
8	58 ef	73 abcd	16 de	1 b	2 d	1 c	25 a
9	63 def	58 bcde	6 fg	2 b	2 d	2 bc	22 a
10	76 bcdef	36 e	2 g	2 b	1 d	1 c	19 a
11	83 abcde	67 abcd	45 a	3 ab	17 ab	3 ab	36 a
12	62 ef	64 abcd	26 bc	3 ab	19 ab	2 bc	29 a
13	45 f	87 a	26 bc	2 b	6 cd	3 abc	28 a
14	95 abcd	49 de	5 fg	6 a	2 d	1 c	26 a
15	77 bcdef	54 cde	31 b	2 b	12 bc	4 a	30 a
16	70 bcdef	68 abcd	33 b	3 ab	25 a	1 c	33 a
17	74 bcdef	83 ab	28 b	2 b	17 ab	1 c	34 a
18	56 ef	64 abcd	8 efg	3 ab	5 cd	1 c	23 a
19	75 bcdef	65 abcd	12 def	2 b	7 cd	1 c	27 a
20	69 bcdef	64 abcd	12 def	4 ab	5 cd	1 c	26 a
Overall mean	73	63	17	2	6	1	27

4.3.4 SHOOT BIOMASS

Concentrations were not significantly different at 5% probability level for shoot biomass (Table 8).

The entries showed significant differences ($P \leq 0.001$) for shoot biomass (Table 8). However, the mean separation did not indicate any difference in the means as can be indicated in (Table 9).

Significant differences ($P \leq 0.001$) were seen in the interaction between the entries and concentration (Table 8). Entry 9 for instance had a shoot biomass of 15.5 mg in 0 mg L^{-1} , 10.9 mg in 4 mg L^{-1} , 6.9 mg in 8 mg L^{-1} , 5.6 mg in 12 mg L^{-1} , 4.5 mg in 16 mg L^{-1} and 6.1 in 20 mg L^{-1} (Table 13).

Table 13: Shoot Biomass Means (mg) for 20 sorghum entries evaluated in the laboratory at University of Zambia in 2009

Entry	0	4	8	12	16	20	Across conc
	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mean
1	8.7 bc	13.2 ab	11.4 ab	8.6 b	2.5 c	5.4 de	8.4 c
2	10.9 bc	10.6 b	8.0 ab	15.2 a	9.7 b	10.5 bcde	10.8 bc
3	13.1 abc	11.5 b	8.6 ab	9.9 b	7.2 c	7.1 de	9.6 c
4	14.8 abc	12.1 b	15.6 ab	8.7 b	13.3 b	9.2 bcde	12.3 bc
5	15.2 abc	3.2 b	2.8 ab	11.4 a	9.7 b	11.1 bcd	8.9 c
6	9.2 bc	13.6 ab	8.2 ab	8.4 b	6.3 b	8.8 bcde	9.1 c
7	10.4 bc	10.8 b	15.9 ab	11.9 a	3.1 c	3.9 e	9.3 c
8	14.9 abc	7.5 b	9.0 ab	5.5 b	2.5 c	3.7 e	11.3 bc
9	15.5 abc	10.9 b	6.9 ab	5.6 b	4.5 c	6.1 de	8.3 c
10	14.1 abc	7.0 b	1.0 b	4.3 b	5.2 c	4.6 de	6.0 c
11	9.7 bc	34.9 a	11.9 ab	5.9 b	4.6 c	19.5 a	14.4 b
12	9.9 bc	17.2 ab	10.4 ab	14.2 a	16.0 a	5.4 de	12.2 bc
13	9.8 bc	9.2 b	11.0 ab	10.3a	7.4 c	11.1 bcd	9.8 c
14	20.9 a	8.2 b	7.4 ab	7.6 b	11.1 b	11.5 bcd	11.1 bc
15	6.7 c	7.3 b	5.5 ab	6.4 b	7.8 b	10.2 bcde	7.3 b
16	17.9 ab	7.7 b	20.8 a	10.9 a	15.5 a	15.1 abc	21.3 a
17	16.4 ab	16.1 ab	13.7 ab	17.4 a	14.0 b	15.6 ab	15.5 b
18	9.8 bc	10.9 b	3.5 ab	10.9 a	10.3 b	8.5 cde	9.0 c
19	14.5 abc	9.3 b	8.5 ab	4.7 b	10.5 b	7.5 de	9.2 c
20	11.1 bc	8.1 b	9.2 ab	12.2 a	11.4 b	6.3 de	9.7 c
Overall mean	12.7	11.5	9.5	9.5	10.6	9.0	10.5

4.3.5 ROOT BIOMASS

Concentrations were significantly different for root biomass ($P \leq 0.001$) (Table 9). The root biomass was observed to decrease with increase in Aluminium concentration (Table 9). The average root biomass for concentration 0 mg L^{-1} was 5.5 mg followed 4.8 mg for 4 mg L^{-1} . Concentration 8 and 12 mg L^{-1} had root biomass of 2.9 and 2.5 mg respectively. Entries in 16 and 20 mg L^{-1} concentrations had average shoot biomass of 2.4 and 1.6 mg respectively.

The entries showed significant statistical differences ($P \leq 0.05$) for root biomass (Table 8). Entry 16 was observed to have the highest root biomass of 5.8 mg but it was not different from entry 17 which had a root biomass of 5.7 mg . Entry 12 had a root biomass of 3.9 mg . The rest of the entries were similar with root biomass in the range of 2.0 to 3.6 mg . This was according to mean separation results in Table 9.

The interactions between entries and concentrations were statistically significant at 0.001 probability level (Table 8). The entries were significantly differently affected by the concentration. Results in Table 14 showed that entry 9 had a root biomass of 5.0 mg in 0 mg L^{-1} concentration and 5.1 in 4 mg L^{-1} but 1.8 in 8 mg L^{-1} concentrations. In concentrations 12 , 16 and 20 mg L^{-1} , entry 9 had root biomass of 1.4 , 0.8 and 1.1 mg respectively. Entry 20 had a root biomass of 5.1 mg in 0 mg L^{-1} , 4.7 mg in 4 mg L^{-1} , 3.0 in 8 mg L^{-1} , 2.8 mg in 12 mg L^{-1} , 1.6 mg in 16 mg L^{-1} and 1.1 mg in 20 mg L^{-1} concentration.

Table 14: Root Biomass Means (mg) for 20 sorghum entries evaluated in the laboratory at University of Zambia in 2009

Entry	0	4	8	12	16	20	Across conc
	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mean
1	2.0 f	4.4 bcd	5.0 b	1.9 bcd	1.0 def	1.6 defg	2.6 b
2	2.1 f	4.0 bcd	2.1 efgh	2.4 bcd	3.0 bc	1.7 cdef	2.5 b
3	3.8 def	2.8 d	1.4 efgh	1.4 cd	1.0 def	1.6 defg	2.0 b
4	4.6 cdef	4.3 bcd	3.0 cdef	1.9 bcd	2.8 c	1.5 efgh	3.0 b
5	5.2 cde	4.1 bcd	2.8 cdef	1.6 bcd	3.2 bc	1.0 fghi	3.0 b
6	8.5 ab	5.2 bcd	2.7 cdefg	1.5 bcd	0.18 f	0.9 ghi	3.1 b
7	7.3 abc	5.4 bcd	1.7 efgh	3.5 b	1.1 def	0.8 hi	3.3 b
8	7.0 bc	4.1 bcd	2.4 defgh	1.4 cd	1.7 cdef	0.4 i	2.8 b
9	5.0 cde	5.1 bcd	1.8 efgh	1.4 cd	0.8 ef	1.1 fgh	2.5 b
10	6.5 bcde	3.7 cd	0.5 h	1.1 d	1.1 def	1.6 defg	2.4 b
11	5.2 cde	3.5 cd	4.4 bc	3.3 bc	1.9 cde	3.0 a	3.5 b
12	5.0 cde	4.1 bcd	4.3 bc	3.2 bc	4.6 b	2.4 ab	3.9 ab
13	3.5 ef	5.9 bc	3.0 cde	2.8 bcd	2.1 cde	2.2 bcd	3.3 b
14	9.8 a	3.5 cd	2.2 defgh	1.4 cd	2.6 cd	2.2 bcde	3.6 b
15	5.2 cde	3.3 cd	4.1 bcd	3.0 bcd	3.1 bc	2.2 bcde	3.5 b
16	6.6 bcd	6.9 ab	7.1 a	5.5 a	6.2 a	2.3 abc	5.8 a
17	6.9 bc	8.8 a	5.4 ab	6.1 a	4.4 b	2.4 ab	5.7 a
18	4.3 cdef	6.1 bc	0.8 gh	2.4 bcd	2.4 cde	1.4 fgh	2.9 b
19	6.5 bcde	6.0 bc	1.0 fgh	1.1 d	2.8 c	0.4 i	3.0 b
20	5.1 cde	4.7 bcd	3.0 cdef	2.3 bcd	1.6 cdef	1.1 fghi	3.0 b
Overall mean	5.5	4.8	2.9	2.5	2.4	1.6	3.3

4.3.6 ASSOCIATIONS AND PATH ANALYSIS OF MEASURED PARAMETERS IN THE LABORATORY

The overall correlations among the measured parameters in the laboratory study with root biomass are indicated in Table 15. Highly significant correlations were observed except for number of lateral roots.

Associations between laboratory parameters and grain yield were also observed to be high except between shoot biomass and grain yield (Table 16).

These correlations were partitioned into direct and indirect path effects as shown in Tables 15 and 16.

Table 15: Direct (Diagonal) and Indirect Path Effects of measured parameter in the laboratory analysis and Root biomass

Characters	Shoot biomass	No. of roots	Shoot length	Root length	Correlation With root biomass
Shoot biomass	0.146	0.098	0.066	0.19	0.68***
No. of roots	0.033	0.276	0.018	0.188	0.234ns
Shoot length	0.066	0.054	0.097	0.176	0.554**
Root length	0.044	0.141	0.048	0.363	0.512**

***= Significant at $P \leq 0.001$, ** = significant at $P \leq 0.01$, * = significant at $P \leq 0.05$, ^{ns} = not significant

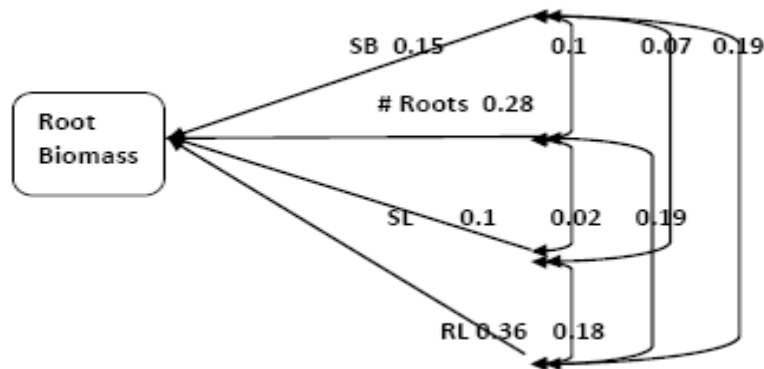


Fig 3: Path diagram showing causal relationships of four predictor variables with a response variable (root biomass)

The overall correlation between root biomass and root length was $r = 0.512$ (Table 15). This correlation was partitioned into a direct effect of 0.363 and indirect affects of 0.141 and 0.048 via the number of lateral root and shoot length pathways respectively. The indirect effect via shoot biomass was 0.044 (Table 15).

The overall association between shoot length and root biomass was $r = 0.554$ (Table 15). The correlation was partitioned into a direct effect of 0.097 while the indirect effects via number of lateral roots , shoot biomass and root length were 0.054, 0.066 and 0.176 respectively (Table 15).

The overall association between number of lateral roots and root biomass was $r = 0.234$ (Table 15). The direct effect of lateral roots was 0.276 while the indirect effect via shoot biomass, shoot length and root length were 0.033, 0.018, and 0.188. Since the direct effect and the overall correlation are almost the same, then a true relationship exists between number of lateral roots and root biomass and direct selection through number of lateral roots would be effective.

The correlation between shoot biomass and root biomass was highly significant $r = 0.68$ (Table 15). The direct effect was 0.145 and the indirect effects were 0.098, 0.066 and 0.19 through number of lateral roots, shoot length and root length.

Table 16: Direct (Diagonal) and indirect effects of parameters measured in the laboratory and field yield

Characters	Shoot biomass	Root biomass	No. of roots	Shoot length	Root length	Correlation with grain yield
Shoot biomass	0.103	0.041	-0.1	0.029	-0.080	0.071
Root biomass	0.021	0.177	-0.406	0.463*	-0.198	0.628***
No. of roots	0.02	0.162	-0.477*	0.356	-0.156	0.624***
Shoot length	0.004	0.132	-0.254	0.687***	-0.238	0.653***
Root length	0.027	0.132	-0.261	0.556**	-0.282	0.713***

***= Significant at $P \leq 0.001$, ** = significant at $P \leq 0.01$, * = significant at $P \leq 0.05$, ^{ns} = not significant

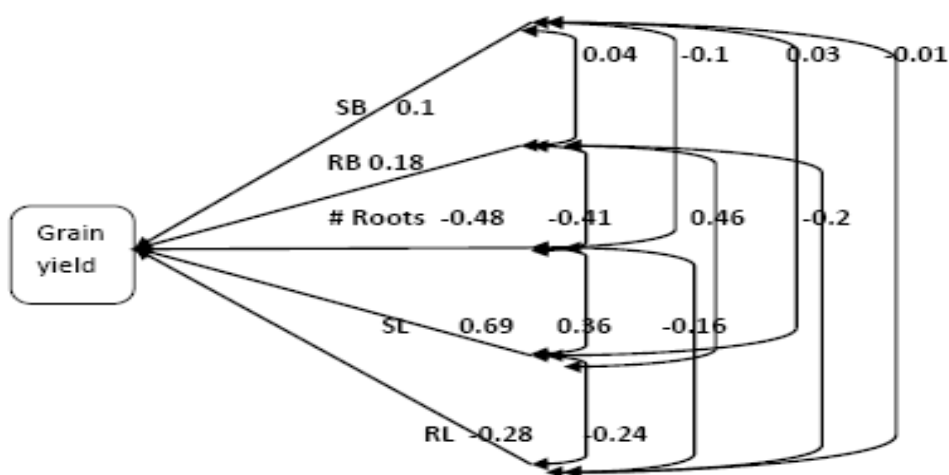


Fig 4: Path diagram showing causal relationships of five predictor variables with a response variable (grain yield)

The overall correlation between shoot biomass and grain yield was $r = 0.071$ (Table 16). The correlation was partitioned into a direct effect of 0.103 and indirect effects of 0.041, -0.1, 0.029 and -0.080 via root biomass, number of lateral roots, shoot length and root length respectively (Table 16 and figure 1). The direct effect of shoot biomass is almost equal to the overall correlation, therefore it can be selected directly.

The overall correlation between root biomass and grain yield was $r = 0.628$ (Table 16). The correlation was partitioned into a direct effect of 0.177 and indirect effects of 0.021, -0.406, 0.463 and -0.198 via shoot biomass, number of lateral roots, shoot length and root length (Table 16).

The overall correlation between number of lateral roots and grain yield was $r = 0.628$ (Table 16). It was partitioned into a direct effect of -0.477 and indirect effects of 0.02, 0.162, 0.356 and -0.156 via shoot biomass, root biomass, shoot length and root length (Table 16).

Shoot length had a significant association with yield at $r = 0.653$ (Table 16). This correlation was partitioned into a direct effect of 0.687 and indirect effects of 0.004, 0.132, -0.254, -0.238 and 0.331. The direct effect of shoot length was almost equal to the correlation suggesting a true relationship with yield. Therefore, a direct selection of Shoot length would be effective.

The overall correlation between root length and grain yield was $r = 0.713$ (Table 16). The correlation was partitioned into a direct effect of -0.282 and indirect effects of 0.027, 0.132, -0.261, 0.556 through shoot biomass, root biomass, number of lateral roots and shoot length (Table 16).

5.0 DISCUSSION

Results from the analysis of variance showed highly significant differences among the genotypes for all the variables measured and/or derived in the Field (Table 3) and Laboratory (Table 8), except for plant count and agronomic score from the field analysis, indicating a presence of genetic variability in the materials tested.

5.1 Field Performance of entries grown in location with low pH

Entries performed differently over the three locations for variables measured. Indeed significant differential responses were also observed for a number of variables.

Plants in the field experiment grew similarly in terms of agronomic score, except that entries were different for plant height (Table 4). The sorghum entries evaluated in the current study were obtained from a breeding programme for aluminium tolerance in agro ecological region III, as such they represented a wide range of genetic diversity for the trait. Differences for agronomic characteristics should be expected as the genotypes used responded differently amongst themselves. In the field trials carried out by the Sorghum Programme of Zambia Agricultural Research Institute (ZARI) differences for this character have always been detected (Annual reports, 2004).

Locations, entries and their interactions were significant for grain yield in the field experiment. The average yield levels observed were 2.6 tons ha⁻¹ at Mpongwe and 1.5 tons ha⁻¹ at Mansa (Table 6), with highest yield of 3.9 tons ha⁻¹ (entry 6) in Mpongwe. Entry 14 recorded the lowest at 0.8 tons ha⁻¹. These yield levels are similar to those reported by other researchers in Zambia, which ranged between 3 and 5 tons ha⁻¹ (Chisi,2003). The differences among the entries for

grain yield could be ascribed to inherent differences among the genotypes tested which were manifested through differences in disease reaction, though the lack of interaction between entry and location pointed to similar reaction by entries to the disease pressure. Diseases in sorghum have been reported to affect grain yield, but that different genotypes will react differently to the disease pressure (Duraes *et al.*, 2001)

Grain yield is related to maturity with late maturing genotypes generally yielding more than the early ones. This is related to the length of assimilate synthesis period, that later maturing genotypes produce more biomass and consequently higher grain yield (Ganesamurthy *et al.*, 2004). In the current study the yields of the entries were not all related to the maturity dates, except for entries 8 and 11 , which were among the late maturing ones and also high yielding (Tables 4 and 6). A similar situation is noted with the early maturing entries that only 4 and 19 conformed to this relationship. Grain yield of the entries thus was not simply determined, rather a number of factors inherent to the genotypes contributed to the yield levels observed. Yield is known to be a complex trait (Singh, 2005) determined by several factors among which the current study may not have measured.

Head parameters derived from the field experiment indicated that all of them sufficiently discriminated the locations and entries. The differences observed, therefore, between locations and among entries may explain the differences for grain yield. Entries 1, 6, 8, 11 and 20 (Table 6) were the highest yielding ones and these were also the ones with the highest in sundried head weight (Table 4). Evidently the yielding ability of the entries can be directly related to these head parameters. Grain yield in sorghum has been reported highly correlated to these parameters (Ojo

et al., 2006) . This result was similar to what Mutegewa et al (1999) found in their study on Sorghum yield components and witch weed in which head weight was positively and highly correlated to grain yield.

The head HI data suggest that while some entries partitioned more assimilates to the head as evidenced by the head weight, they were not partitioned at the same rate between the grain and the supportive structures. Entries 1, 6 and 20 are the only ones that had high head HI and high grain yield. The other entries, 8 and 11, attained the high yields through other mechanism, possibly seed size or others.

5.2 Root reaction to aluminum levels

The root reaction was measured through root length, number of lateral roots, root weight, root biomass and shoot biomass. Root length and root biomass are the best indicators for reaction to aluminum concentrations as they represent the ultimate product of growth and development of roots (Nguyen *et al.*, 2003; Kochian *et al.*, 2005).

There was general decrease in root length with increase in aluminium level across all the sorghum entries. The results obtained showed that Al toxicity inhibited root growth as the concentration increased with the least effect at 4ppm and the most severe effect at 20ppm across all entries. These results were consistent with those obtained by Kochian et al (2005), Munyinda *et al* (2008) and Rangel *et al* (2007) on similar studies of barley, wheat and beans, which showed that an increase in the Al toxicity lead to a reduction in root growth. Further analysis on the twenty entries tested revealed three classes (tolerant, intermediate and susceptible). Entry 11 was the most tolerant followed by entries 12 and 17. Entries 16, 5, 20, 2, 13, 14, 4 and 15 fell in the

intermediate level (Table 9). Entries 1, 3, 7, 10, 18 and 19 were moderately susceptible, entries 6 and 8 were susceptible but entry 9 was the most susceptible.

This inhibition of root growth can be attributed to Al shock and injury which is more severe as the Al toxic concentration increases. The tolerant varieties were relatively less severely inhibited by increase in Al toxicity levels and continued to grow relatively longer roots at higher levels of Al toxic concentration.

There was a reduction in the number of lateral roots as the toxic Al levels were increased. This result coincides with what Kochian *et al* (2005) found out in Barley but its different from the results obtained by Munyinda (1986) for wheat where the wheat varieties showed an increase in the number of lateral roots with increase in Al toxicity levels.

From the results it can be deduced that sorghum stress response to high Al levels is poor unlike other cereals such as wheat, which produce more lateral roots as a survival mechanism. Sorghum does not increase the number of lateral roots (Munyinda *et al.*, 2008) to compensate for the reduction in root length as Al toxicity increases and this explains why there was a reduction in shoot length across all the varieties with increase in Al concentration.

There was a differential effect in the root biomass with increase in Al toxicity levels. This was consistent with the results obtained for the root length. Since the root length reduced with increase in the Al toxicity concentration, the root biomass also reduced with increase in Al level.

There was a reduction in shoot length as well as shoot biomass with increase in Al toxicity level. This coincided with the reduction in root length as the Al toxicity concentration increased

implying that a reduction in root length invariably results in reduced shoot biomass, which was manifested through reduced shoot length. Shoot length was inhibited due to limiting supply of water and nutrients resulting from reduction in root length which are the major source of nutrients from the soil (Zheng and Yang, 2005; Raman *et al.*, 2002).

5.3 Relationships Among field and laboratory measured parameters as indicators of Aluminum Tolerance

The results from path coefficient analysis on field data pointed to the importance of head weight as a determinant of grain yield (Table 7), having high and significant direct and indirect effects. This confirms the results obtained above, suggesting direct relationship between head weight and grain yield. Sundried head weight contributed the most to yield directly. The direct effect was almost equal to the correlation coefficient suggesting that existence of a true relationship between sun dried head weight and yield .Therefore a direct selection of sundried head weight would be effective. El Nagouly *et al* (2000) and Oktem (2008) found positive correlation between grain yield and sundried head and ear weight in their studies on sorghum and sweet corn respectively.

Head harvest index had low correlation with yield. However, the direct effect was equal to the correlation coefficient indicating a true relationship with grain yield. Previous studies have confirmed that head harvest index is an important characteristic correlated to grain yield in grain sorghum. The result found in this investigation was not the same as the one observed by El Nagouly *et al* (2000) on sorghum in which harvest index correlated directly and significantly with yield. Babic *et al* (2008) in his study on maize also found that head harvest index was

positively associated with yield. On account of the equal direct effect and correlation coefficient, head harvest index can be used to select for high yielding plants that are tolerant to Al in low pH soils.

High associations were observed between the root attributes and field yield (Table 16). This implies that laboratory data can be used to determine yield in the field.

Results from the laboratory experiment showed that most of the direct effects were low except for number of lateral roots and shoot length which had high direct effects (Table 16). The direct effect of shoot length on yield was almost equal to the correlation implying that selection of shoot length would be effective for higher field grain yield (Singh and Chaudhary, 1985).

Root biomass had high indirect effects via number of lateral roots and shoot length (Table 16). Root length had a high indirect effect on grain yield through shoot length. This result was different from the one found by Beebe *et al* (2008) on their study on common beans in which greater root biomass under Al stress did not contribute to yield, but greater root length did.

6.0 CONCLUSION

The hypothesis that sufficient genetic variation exists in the sorghum germplasm in Zambia to select for Al tolerance was validated in this study. The genotypes used in the experiment showed sufficient variation in their response to conditions predisposed for Al toxicity. Significant interactions were observed for plant height and grain yield in the field analysis and for all the parameters measured in the laboratory analysis. Entries were significantly different for all parameters except for plant count and agronomic score while locations were significantly different for all parameters with the exception of plant count, agronomic score, disease prevalence and head harvest index.

An efficient selection criterion for Al tolerance in Sorghum that employs deliberate selection of variables that have high and direct effect on yield has been developed. This includes variables through which other traits pass indirectly to contribute significantly to high yield. The results obtained in the field experiment suggested that sundried head weight and head harvest index would contribute effectively to high yielding sorghum genotypes in low pH soils with Al toxicity.

The high positive contribution of the variables in the laboratory analysis to field yield suggests that laboratory attributes can be used to predict high grain yielding genotypes suitable for soil with low pH and aluminium toxicity.

Based on the results obtained in this study, genotypes 1, 2, 3, 6, 8, 11, 12, 16, 17 and 20 showed Al toxicity tolerance and high grain yield. Among these, the top seven in the laboratory were genotypes 2, 3, 11, 12, 16, 17 and 20 while, in the field they were 1, 6, 8, 11, 16, 17, and 20. The

superior genotypes recommended for high rainfall areas are 11, 17, 16 and 20.

7.0 RECOMMENDATIONS

The genotypes that performed well in both the laboratory and field examination should be screened again to validate their performance. Sorghum markers for acidity can be used to hasten the breeding programme and deliver varieties in a shorter period of time

The use of test tubes in the laboratory is very cumbersome. A better system using mini-tanks which are easily aerated should be designed.

More variables relating directly to yield in the field such as 1000 seed weight, panicle length and plant biomass should be studied in order to establish a more effective selection criterion.

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