# MAIZE GROWTH AND YIELD AS AFFECTED BY DIFFERENT SOIL FERTILITY REGIMES IN A LONG TERM TRIAL

**by**

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**in the Department of Plant Production and Soil Science**

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# DECLARATION

I, Jerry Celumusa Dlamini hereby declare that this dissertation for the degree MSc (Agric): Agronomy at the University of Pretoria is my own work and has never been submitted by myself at any other University. The research work reported is the result of my investigation, except where acknowledged.

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# LIST OF ABBREVIATIONS & ACRONYMS

°C : Degrees Celsius

ANOVA : Analysis of variance

ASI : Anthesis - silking interval

Ca : Calcium

cm : Centimetres

DAE : Days after emergence

ET : Evapotranspiration

FAO : Food and Agriculture Organisation

g : Gram

GI : Germination index

ha : Hectare

$H_2O$ : Water

IITA : International Institute of Tropical Agriculture

ISTA : International Seed Testing Association

K : Potassium

KCl : Potassium Chloride

kg : Kilogram

l : Litre

LA : Leaf area

LAI : Leaf area index

LAN : Limestone Ammonium Nitrate

LSD : Least significant difference

Mg : Magnesium

$mgkg^{-1}$ : Milligram per kilogram

N : Nitrogen

Na : Sodium

$NH_4$-N : Ammonium nitrogen

No. : Number

$NO_3$-N : Nitrate nitrogen

P : Phosphorus

RTW : Root dry weight

SAS : Statistical analysis software

SDW : Seed dry weight

SHW : Shoot dry weight

SME : Seed metabolic efficiency

SMR : Seed metabolic rate

WAE : Weeks after emergence

WAP : Weeks after planting

WUE : Water use efficiency

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## ABSTRACT

Maize is the world's third most important cereal after wheat and rice. It serves as a staple food to more than 1.2 billion of the world population. However, its production is threatened by declining soil fertility; mainly due to low inputs of fertilizers containing major elements to replenish lost soil nutrients and unsustainable soil tillage practices linked to mono-cropping. To examine the influence of N, P and K and residual compost on maize growth and yield, an experiment was carried out at the Hatfield Experimental Farm of the University of Pretoria. Utilizing the long-term maize trial, controls (0 and W), seven inorganic fertilizer treatments (N, P, K, NP, NK, PK & NPK) and seven organic + inorganic fertilizer treatments (WN, WP, WK, WNP, WNK, WPK & WNPK) were used. The influence of these fertilizer and residual compost treatments on maize seed viability (germination), plant growth, reproductive development, pollen performance, grain yield parameters, yield and grain yield water-use efficiency was investigated.

Higher seed viability was associated with balanced soil nutrient status (WNPK & NPK), whilst deficient soil nutrient status (0, N, P & K) resulted in lower seed viability. Plant growth (plant height, total dry mass and LAI) and reproductive development (tassel length, ear length, and days to tasseling and silking) were positively influenced by a balanced soil nutrient status and residual compost. Deficiencies in soil nutrients restricted maize plant growth and delayed reproductive development. This highlighted the importance of a balanced soil nutrient status in attaining a vigorous crop and good reproductive development.

Soil nutrient deficiencies (0, P & K treatments) enhanced the production of pollen (mass per plant), but resulted in low pollen quality (viability and germination). Balanced soil nutrient status (WNPK & NPK) resulted in the production of high quality pollen (viability and germinability), which however had a low mass. In both 2012/2013 and 2013/2014 seasons, maize grain components; cob length, number of kernel rows per cob, number of kernels per row, mass per kernel and mass of 100 kernels were positively influenced by balanced soil nutrient status. Grain yield and water use efficiency were also positively influenced by a balanced soil nutrient status (WNPK & NPK), whilst deficient soil nutrient status had a negative effect.

Keywords: *maize, long-term trial, soil fertility regimes*

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# CHAPTER 1

# GENERAL INTRODUCTION

## 1.1 INTRODUCTION

Maize is one of the most important cereal crops in the world, ranked third after wheat and rice. This crop is widely adapted to variable agro-ecological conditions all over the world (O'Keefe & Schipp, 2009). It is utilized for human consumption, animal feeding and manufacturing of industrial products. In the 2011/2012 season, maize from the USA and China accounted for more than half of the approximately 868 million tons produced globally, with 38.4% and 18.8% respectively. Following closely were Brazil (51 million tons), Mexico (20 million tons), Indonesia, India (17 million tons each) and France (15 million tons) (Table 1.1). South Africa produced the highest yield in Africa of 12 million tons (NCGA, 2012).

**Table 1.1:** Top ten maize producers of the world in the 2011/2012 season (NGCA, 2012).

| Country | Production (tons) |
|---|---|
| United States of America | 333 010 910 |
| China | 163 118 097 |
| Brazil | 51 232 447 |
| Mexico | 20 202 600 |
| Indonesia | 17 629 740 |
| India | 17 300 000 |
| France | 15 299 900 |
| Argentina | 13 121 380 |
| South Africa | 12 050 000 |
| Ukraine | 10 486 000 |
| **Total world production** | **868 060 257** |

Maize serves as a staple food for about 1.2 billion of the world population (IITA, 2009). It is an important economic crop, contributing billions of dollars to the global economy each financial year (FAO, 2012). This crop is popular amongst small-scale farmers, since it does not require expensive and highly specialised farm inputs and implements (du Plessis, 2003).

Poor production systems, harsh weather conditions and scarcity of inputs are amongst the main factors affecting maize production output. Soil nutrient imbalance is one of the most prominent limiting factors in maize production (Mengel & Kirkby, 1987). Yield declines have been noted over the years in some parts of the world, especially on the African continent. This is mainly attributed to low nutrient status, especially of nitrogen (N), phosphorus (P) and potassium (K). This result inter alia from slash and burn farming systems associated with bush fallow, and leaching of soil nutrients. Such systems are presently unsustainable due to high population pressure and other human activities which have resulted in reduced fallow periods (Steiner, 1991).

Inadequate replenishment of primary nutrients, including N and P in the soil, affects maize yields (FAO, 2007). Farming without implementing sustainable soil fertility programmes to replace the nutrients removed by the crops can result in soil nutrient losses of about 22 kg of N, 2.5 kg of P, and 15 kg of K per hectare per year (Sanchez, 1997). These losses are in most cases due to topsoil erosion associated with conventional tillage practices. Insufficient nutrient replenishment can render a previously fertile piece of land un-productive in less than 30 years (Lynam, 1998; Cermak & Smatanova, 2012). Erratic rainfall further exacerbate soil fertility challenges since some nutrients are unavailable to plant roots when water is limited. Soil nutrients such as nitrogen ($NO_3^-$ and $NH_4^-$) are made available to the plant root zone through mass flow processes which mainly require soil moisture (Mengel & Kirkby, 1987). Earlier plant nutrition studies revealed that judicious and proper use of fertilizer can markedly increase maize yield (Hokmalipour, *et al.*, 2010). This highlights the importance of continuous soil fertilization and implementation of sustainable soil fertility management practices in farming.

Soil fertility trials under field conditions are usually a long-term investment, and one can seldom get conclusive results after one or two years of experimentation. Therefore, long term experiments are instrumental in providing a better understanding of soil fertility and its management strategies for sustainable crop yields (Korschens, 2006). Such trials are crucial in understanding the processes of soil fertility changes in the soil over time and its influence on yield (Bationo, *et al.*, 2012). On the other hand, the combined use of organic and inorganic fertilizers was observed to have a capacity to improve maize growth and yield

(Abubenywa *et al.*, 2007; Laekemariam & Gidago, 2013). In this dissertation, a long term soil fertility trial was used to demonstrate the effect of more than 70 years of balanced and unbalanced fertilizer application on maize growth, reproductive development and yield.

## 1.2 PROBLEM STATEMENT

Maize growth and reproductive development normally determine grain yield. In order to sustain high maize yields an understanding of the effect of soil fertility management on crop growth and development in such trials is crucial. This is important to curb yield losses associated with deficient plant growing soil conditions. Prolonged years of different inorganic fertilization under conventional farming practices result in a wide range of soil fertility regimes which dramatically affect maize growth and yield. On the other hand, organic fertilizer incorporated into long term inorganic fertilizer treatments can possibly have a positive influence on maize growth and yield.

## 1.3 HYPOTHESES

- Nutrient deficiencies of N, P or K will negatively affect vegetative development, thus limiting assimilates available to the developing grain kernels. Whilst in comparison, deficient treatments with residual compost will positively affect vegetative development.
- Nutrient deficiency stress of N, P or K will negatively affect seed germination. While the residual compost combined with the deficient treatments will enhance seed germination.
- Nutrient deficiencies of N, P or K will negatively influence pollen mass and quality. While the residual compost combined with the deficient treatments will enhance pollen mass and quality.
- Deficiencies in N, P, or K will result in low water-use efficiency. Whilst in comparison, deficient treatments with residual compost will positively affect grain water-use efficiency.

## 1.4 OBJECTIVES OF THE STUDY

This research aims at determining the influence of inorganic and organic + inorganic fertilizer treatments on:

- seed viability,
- maize growth,
- tasseling and silk appearance,
- pollen mass and quality,
- grain components and yield and
- grain water-use efficiency

within a conventional farming system.

# CHAPTER 2

# LITERATURE REVIEW

## 2.1 VEGETATIVE GROWTH OF MAIZE

After planting, maize seed absorb water from the soil and begin to grow. Emergence occurs when the coleoptile (spike) pushes through the soil surface (Ransom, 2013). Maize plants can emerge within five days in ideal heat and moisture conditions (Bonnet, 1947). At least two weeks may be required from planting to emergence under early season cool conditions. With below average spring temperatures, maize seed may be in the ground for three weeks or more before the seedlings emerge (PANNAR Handbook, 2013). The growing point (stem apex) grows between 2.5 to 3 centimetres below the surface (Hanway, 1971). The seminal root system grows from the seed (Figure 2.1). The seminal roots provide much of the plant nutrients at this stage, but growth slows after emergence as nodal roots begin to grow (Ritchie, *et al.*, 1993; OGTR, 2008). A balanced soil nutrient status amongst other factors promotes optimum seed germination and emergence at this stage (Rouanet, 1987).

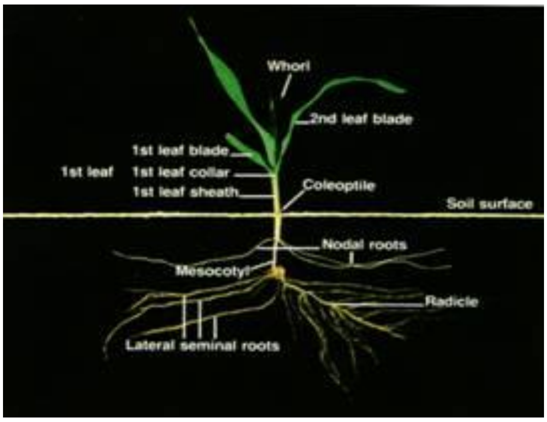

**Figure 2.1:** A young maize plant (http://www.crsbooks.net/student/anatomy.html ).

The young plant develops to the point that the collars start showing on the first leaf (Figure 2.1). This leaf is usually more rounded at the tip than succeeding leaves (OGTR, 2008). Each vegetative stage is determined by counting the visible collars in the sequence; V1, V2, to VN until the tassel emerges (VT) and maximum height is attained (Figure 2.2). When

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counting leaf number at these stages, it is important to consider that leaves may have been lost from the bottom of the plant (du Plessis, 2003; PANNAR Handbook, 2013). At the V1 stage leaves are initiated from a growing point below the soil surface as cell elongation has not yet begun. The initial seminal root system continues to grow and expand with branches and hair roots (Bonnet, 1947). The beginning of the nodal root system may also be visible as bumps at either one or two nodes at the lower end of the coleoptile and above the mesocotyl (PANNAR Handbook, 2013) (Figure 2.1).

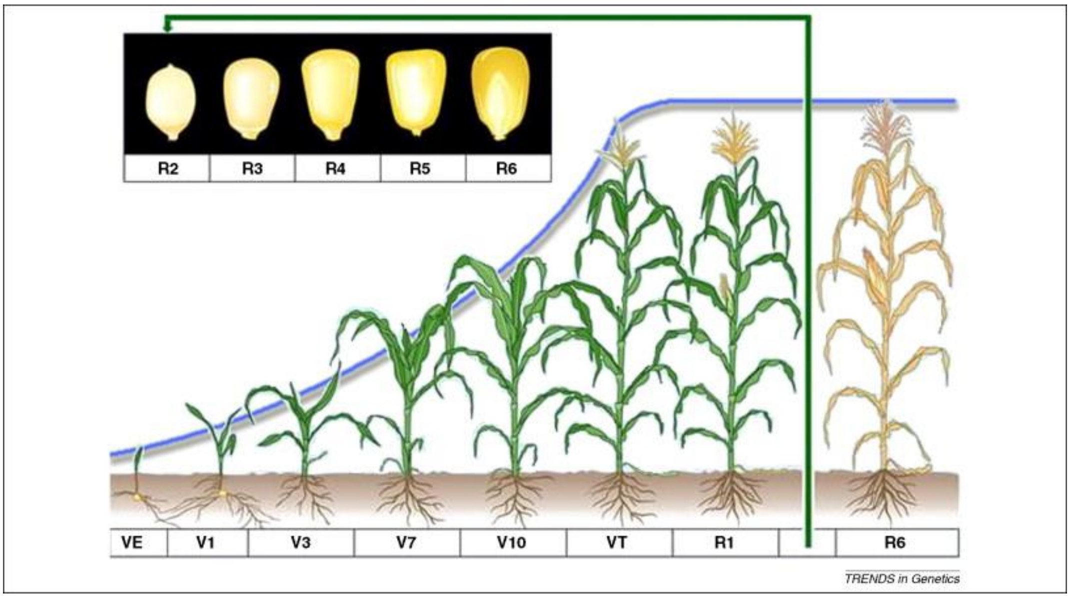


**Figure 2.2:** Growth and development of a maize plant ([http://odells.typepad.com/blog/corn-growth-stages.html](http://odells.typepad.com/blog/corn-growth-stages.html) ).

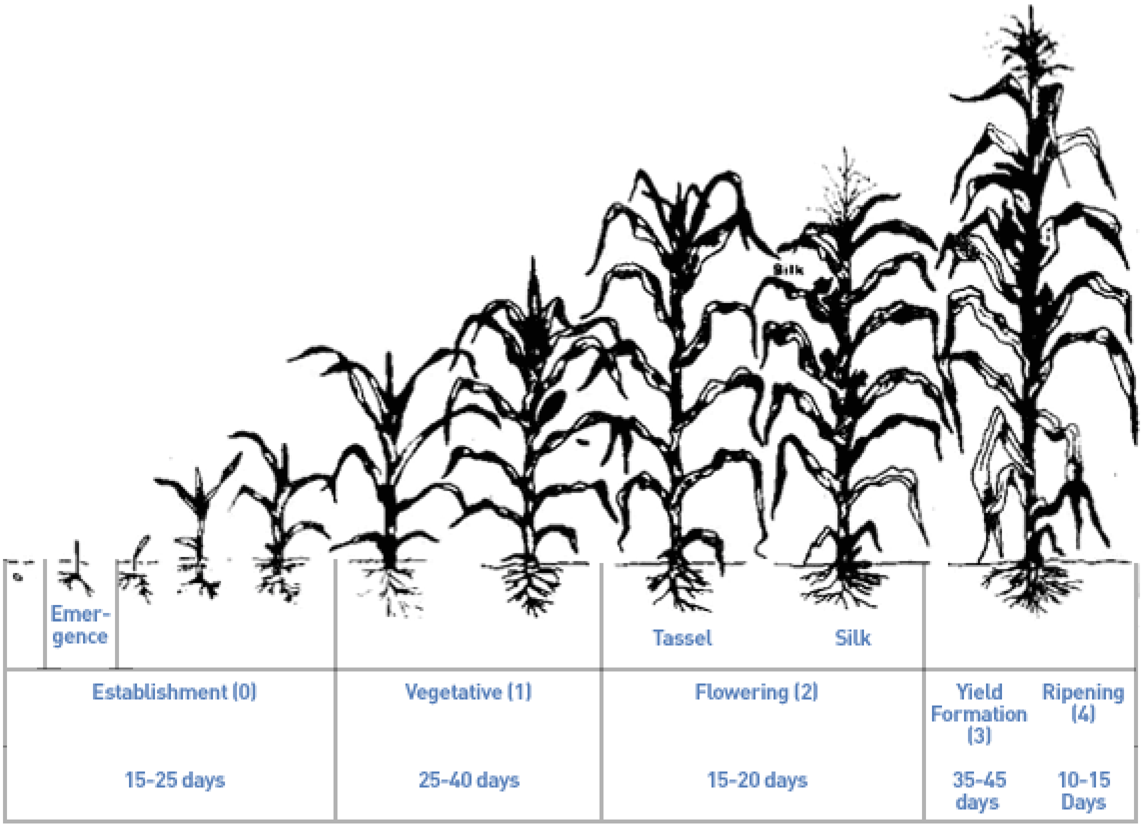
At V3 stage, the stalk (stem) has not elongated much. Root hairs are growing from the nodal roots as seminal roots cease growing. All leaves and ear shoots the plant will ever produce form inside the stalk from V3 to about V5 (Figure 2.2) (Lee, 2012). A tiny tassel forms at the tip of the growing point. Above-ground plant height is typically about 20 cm at this stage. The growing point and tassel rise higher above the soil surface at about the V6 stage. The stalk begins to elongate. The nodal root system grows from the three to four lowest stalk nodes (OGTR, 2008). Some ear shoots or tillers are visible. Tiller (or sucker) development depends on the specific hybrid, plant density, soil fertility and other conditions (O'Keefe & Schipp, 2009; Ramson, 2013).

## 2.2 MAIZE REPRODUCTIVE DEVELOPMENT

A cross-sectional dissection at V9 plant growth stage shows ear shoots (potential ears) (Lee, 2012). These develop from every above-ground node except the last six to eight nodes below the tassel. Lower ear shoots grow fast at first, but only the upper one or two develop to a harvestable ear (Jones & Benton, 1930). The number of kernel rows is also determined by the growing conditions at V9 (Nielsen, 1995; PANNAR Handbook, 2013). The tassel begins to develop rapidly. Stalks lengthen as the internodes grow (Goldsworthy, 1984). At V10, the time between new leaf stages shortens to about every two to three days. The total number of leaves will vary from 12 to over 20; depending on hybrid maturity and genetic make-up (Uchida, 2000; Ramson, 2013).

The potential number of kernels per row is determined between the V12 and V15 stages. Between these stages, the top ear shoot is still smaller than the lower ear shoots, but many of the upper ears are close to the same size. This is the commencement of the most crucial period in determining grain yield (Carvoca, *et al.*, 2003; du Plessis, 2003). Upper ear shoot development overshadows lower ear shoot development (PANNAR Handbook, 2013). Every one to two days, a new leaf stage occurs. Silks begin to grow from the upper ears (Lee, 2012).

At the V17 growth stage, the tips of the upper ear shoots may be visible atop the leaf sheaths. The tip of the tassel may also be visible. Just before tasseling, silks from the basal ear ovules elongate first (Bonnet, 1947). Silks from the ear tip ovules follow. Brace roots (aerial nodal roots) grow from the nodes above the soil surface to help support the plant and take in water and nutrients during the reproductive stages (Bonnet, 1947; Glass, 1989; Nazfiger, 2010).



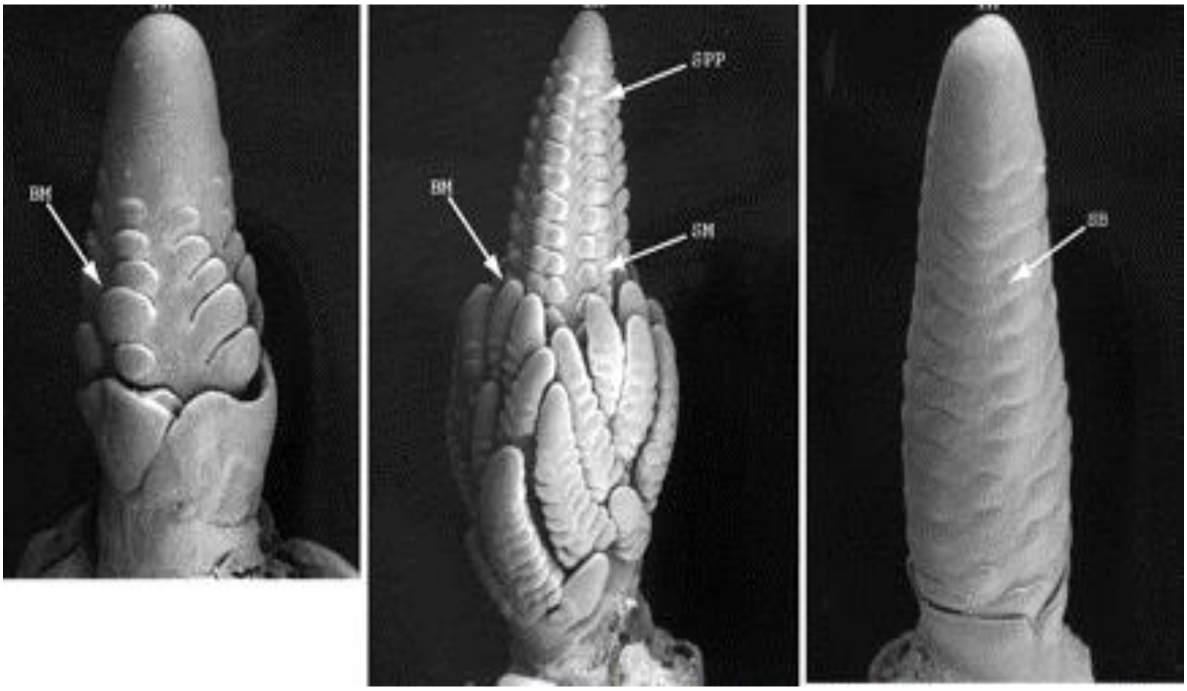
| Establishment (0) | Vegetative (1) | Flowering (2) | Yield Formation (3) | Ripening (4) |
|---|---|---|---|---|
| 15-25 days | 25-40 days | 15-20 days | 35-45 days | 10-15 Days |

**Figure 2.3:** The different development stages of a maize plant (http://www.fao.org/nr/water/cropinfo_maize.html ).

The VT stage is when the last branch of the tassel is completely visible (Ransom & Endres, 2004). VT begins about two to three days before silk emergence; the plant is nearly at its full height (Ramson, 2013) (Figure 2.3). Pollen shed begins, lasting about one week on an individual plant basis and one to two weeks on a field basis (Laekemariam & Gidago, 2012). The interval between VT and R1 can fluctuate considerably depending on the hybrid and the environment. Drought stress lengthens this interval (Russel, 1942; Nazfiger, 2010).

### 2.2.1 Tassel initiation and differentiation

The initiation of plant internode elongation is directly linked to the commencement of tassel differentiation. Differentiation of tassels start with the branch pri-mordia arising in acropetal succession as projections from all the sides of the elongated central axis (Figure 2.4) (Cheng, *et al*., 1983). At the base of the lateral axis, some of the branch initials elongate and become lateral axes of the tassel (Bonnet, 1940). The spikelet divides into two unequal parts during its development; commonly known as spikelet initials. The spikelet initials originate from the initials arising from a higher point on the central axis (Ritter, *et al.*, 2002). The larger initial is always developmentally more advanced than the smaller one; which implies that the central axis will always be more advanced in its development compared to the branches (Bonnet, 1947). Dehiscence of anthers symbolises the completion of the tassel development process (Ritter, *et al.*, 2002).



*IM=inflorescence meristem, BM=branch meristem and SPP=spikelet pair meristem*

**Figure 2.4:** A scanning micrograph showing maize inflorescence meristem elongating and branch meristem, spikelet primordia and spikelet meristem initiation and suppressed tassel bracts; (left) wild-type tassel, (middle) later stage wild-type tassel and (right) tassel with suppressed bracts. (Ritter, *et al.*, 2002).

### 2.2.2 Ear initiation and differentiation

In the early stage of the main stem development, some axillary buds become larger in acropetal succession. Their development is more advanced in the topmost part of the main stem. It is from these axillary shoots that ears develop (Hake, 2014). When ear development starts, their size sequences change such that the topmost shoot may hinder the development of the shoots below (Tollenaar, 1977). Elongation of the growing point of the axillary shoot and differentiation of the lateral projections from the central axis of the ear initial marks the beginning of ear initiation. Bonnet (1947) stated that the number of rows of branch initials that differentiate will determine the kernel row number per ear. The development of ears normally starts later than the initiation of tassel differentiation (Hake, 2014) (Figure 2.4).

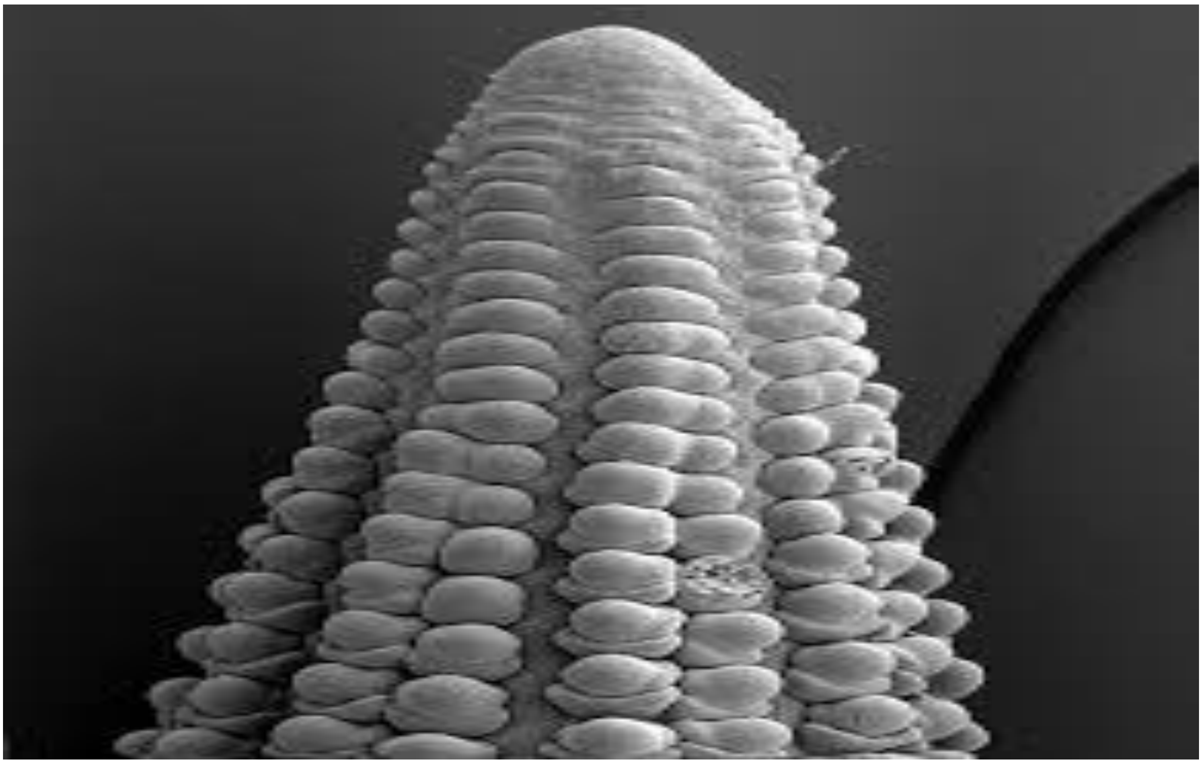


**Figure 2.5:** A scanning micrograph of an ear primordia showing rows of spikelet meristems (Hake, 2014).

## 2.3 MAJOR NUTRIENTS AFFECTING PLANT GROWTH AND YIELD IN MAIZE

Soil nutrient balance is crucial in maize yield improvement. Soil nutrient deficiencies have both direct and indirect effects on plant growth and development. The FSSA (2000) stated that for each ton of maize grain obtained from a field, 15 kg of N, 3 kg of P and 4 kg of K is removed from the soil. By removing a ton of the whole maize plant 27 kg of N, 4.5 kg of P and 20 kg of K is removed from the soil.

### 2.3.1 Nitrogen

Nitrogen is required by maize in higher amounts than any other element, followed closely by K. N is one of the most limiting nutrient elements in crop production, especially in production systems that do not include a legume crop (Aftab, *et al*., 2007). It is a major component of numerous plant biological compounds that play a crucial role in photosynthetic activities and consequently influence crop yield capacity (Cathcart & Swanton, 2003; Zhao, *et al.*, 2005). Adequate N levels result in dark green leaves, whilst, deficiencies cause leaf chlorosis (Padmaja, *et al.* 1999; Tajul, *et al.*, 2013) (Figure 2.6). Deficiencies may also result in slow stunted growth and weak plants; such plants normally mature early which may significantly reduce grain quality and quantity (Benton & Jones, 1930). Maize grain quality and quantity are improved with adequate nitrogen levels in the soil. A legume-cereal rotation reduces the need for supplementing nitrogen in the soil, since the *Rhizobium* bacteria associated with legumes fix substantial amounts of nitrogen (Phillips & Lessman, 1968).

**Figure 2.6:** Nitrogen (N) deficient maize leaves at 6 WAE; yellowing proceeds downwards starting from midrib of older leaves forming a V-shape (https://www.sdstate.edu/ps/extension/soil-fert/corn-deficiency-photos.cfm ).

### 2.3.2 Phosphorus

Phosphorus is an essential element in the production of maize, but it is not required in as high amounts as N. P deficiency is characterized by stunted plants, which may be dark green in colour, with older leaves showing a purple pigmentation (Figure 2.7) (Jones & Benton, 1930). P fertilization is an important factor in increasing grain yield (Phillips & Lessman, 1968). Its availability to plants in the soil may be influenced by soil parent material low in P, soil compaction, low soil pH, soil temperature, soil moisture content, and availability of other mineral elements. Its deficiency during kernel formation in maize may result in poor kernel set, hence affecting grain yield (McVickar & Walker, 1978).

**Figure 2.7:** Phosphorus (P) deficient maize plants; mainly characterised by purple pigmentation on leaves. (http://yara.co.uk/crop-nutrition/crops/maize/crop-nutrition/deficiencies/p/9519-phosphorus-deficiency---maize/ ).

### 2.3.3 Potassium

Potassium is the second important nutrient element required by maize, after N. Scorching of leaf margins is one of the most prominent symptoms of K deficiency (Figure 2.8). It may result in weak and lodged plants and poor kernel set; hence poor quality and quantity of grain (Hanway, 1962; Phillips & Lessman, 1968).

**Figure 2.8:** Potassium (K) deficient maize plants; yellowing of leaves starts from outer margins of older leaves becoming chlorotic and necrotic (http://dopepicz.com/10324054-potassium-deficiency-in-corn-plants.html ).

# CHAPTER 3

# GENERAL MATERIALS AND METHODS

## 3.1 INTRODUCTION

To sustain high maize yields, a detailed understanding of the effect of soil fertility management, on crop growth and development, is crucial. This helps in avoiding nutrient stress which may result in yield losses. A long-term maize fertilizer trial was used for data collection. Established in 1939, this experiment is one of the oldest long-term field trials in the southern African region. Initially, it was established to determine fertilizer requirements for maize on the specific soil type. However, the objectives changed over the years, with more emphasis recently on the sustainability of the different treatments and on a better understanding of how basic production processes are affected by fertilizer treatment combinations. This dissertation focused on the effect of different inorganic (N, P, and K) fertilizer combinations compared to treatments that received three seasons of compost in addition to the inorganic fertilizer treatment combinations. The objectives for this dissertation were to identify the effects of different soil fertility regimes on (i) seed viability; (ii) maize growth; (iii) reproductive development; (iv) pollen mass and quality; (v) grain components and yield; and (vi) grain water-use efficiency.

## 3.2 EXPERIMENTAL SITE

The research was carried out at the Field Trial Section of the University of Pretoria's Hatfield Experimental Farm ($25^0$45'N, $28^0$16'E) situated at an altitude of 1372 m above sea level. The long-term maize fertilizer trial was utilized for data collection during the 2013/2014 cropping season, while historical data for the 2012/2013 cropping season was also incorporated.

## 3.3 EXPERIMENTAL DESIGN AND TREATMENTS

The design of the experiment was a randomised complete block design (RCBD). The bigger trial is a factorial experiment with five factors each at two levels (with and without); water

(W), nitrogen (N), phosphorus (P), potassium (K), and manure (M), combined resulting in 32 treatments with 4 replicates on 128 plots (Nel, *et al*., 1996). The difference between N and WN treatments would thus be that; the WN plots originally received supplementary irrigation while N plots were solely reliant on rainfall, but both received similar amounts of fertilizer. The W treatment was discontinued in 1989, and therefore treatments with and without irrigation (W) can be seen as replicates of each other during the trial period relevant to this dissertation. However, for three seasons; 2002/03, 2003/04 and 2004/05, compost was incorporated into all the W plots. Therefore the W treatments were not used as replicates for the non W treatments. The use of compost thus provided a unique opportunity to investigate the residual effect of compost applied about eight years ago. The objective with the compost treatments was to determine whether application of organic material could rectify soil nutrient imbalances created over decades under the conventional tillage system.

For this dissertation the experimental design remained a factorial trial, but with only 16 selected treatment combinations replicated 4 times, resulting in 64 plots. The treatments were 0, N, P, K, NP, NK, PK, NPK, and W, WN, WN, WK, WNP, WNK, WPK and WNPK; which are further explained in Table 3.1. In the dissertation the treatments will be grouped together as controls (0 and W), inorganic treatments (N, P, K, NP, NK, PK and NPK) and inorganic + organic treatments (WN, WP, WK, WNP, WNK, WPK and WNPK). The gross size of the plots is 8.325 m by 6.30 m (0.005245 ha) and the net size is 7.47 m by 4.93 m (0.003683 ha). As a measure to prevent run-off and transport of soil between plots, soil dikes of about 0.2 m were made around all the plots.

**Table 3.1:** Nutrient treatments applied in the long term maize fertilizer trial.

| No. | Treatment | Code |
|---|---|---|
| 1. | Control (neither inorganic fertilizer nor compost applied) | 0 |
| 2. | Nitrogen only | N |
| 3. | Phosphorus only** | P |
| 4. | Potassium only | K |
| 5. | Nitrogen + phosphorus** | NP |
| 6. | Nitrogen + potassium | NK |
| 7. | Phosphorus + potassium** | PK |
| 8. | Nitrogen + phosphorus + potassium** | NPK |
| 9. | Compost control* (no inorganic fertilizer) | W |
| 10. | Nitrogen + compost* | WN |
| 11. | Phosphorus + compost* | WP |
| 12. | Potassium + compost* | WK |
| 13. | Nitrogen + phosphorus + compost* | WNP |
| 14. | Nitrogen + potassium + compost* | WNK |
| 15. | Phosphorus + potassium + compost* | WPK |
| 16. | Nitrogen + phosphorus + potassium + compost* | WNPK |

***Compost was applied for three (3) seasons between 2003 and 2005 at the equivalent of 5 t ha$^{-1}$ per annum.

** No P fertilizer was applied in this experiment, thus P treatments were actually on residual basis.

## 3.4 SITE DESCRIPTION AND CULTURAL PRACTICES

The soil is classified as a silt clay loam of the Hutton form, belonging to the Suurbekom family (Soil Classification Working Group, 1991). Land preparations were done using a rotovator a week before planting. Fertilizer application was done by hand using the fertilizer types and rates shown in Table 3.2. P application was discontinued provisionally in 1989 since fertilizer applied to P treatments resulted in soil P levels as high as 70 mg kg$^{-1}$.

**Table 3.2:** Fertilizer application rates in the 2013/2014 maize season.

| Fertilizer applied | Fertilizer rates (kg ha$^{-1}$) | Amount applied per plot (kg plot$^{-1}$) |
|---|---|---|
| N(LAN) (%N) | 100 | 1.8 |
| P | 0* | - |
| K(KCl) | 80 | 0.8 |

*No P was applied to the P containing treatments since 1990.

Three seeds per planting hole were planted using a hand-planter (Figure 3.1). Thinning to one seedling per spot was done at two weeks after emergence (WAE) to maintain a 90 cm inter-row and 20 cm intra-row spacing. A hybrid seed containing both the Bt and RR genes namely DKC 7374 BR was planted. The experiment was rain fed; however soil moisture was monitored using a neutron probe meter, whenever precipitation was inadequate it was supplemented with irrigation to prevent water stress. Herbicide was applied using a boom sprayer at three weeks after emergence; this was a mixture of glyphosate (Round-Up Turbo) and acetochlor (Harness-Extra) at 3 l ha$^{-1}$ and 1 l ha$^{-1}$ respectively. No diseases or harmful insects were observed during the season, therefore no pesticides or insecticides were applied. Harvesting was done by hand.

**Figure 3.1:** A hand-planter used during planting of maize in the long term trial field plots (Dlamini JC, 2014).

## 3.5 DATA COLLECTION

### 3.5.1 Meteorological and soil moisture content data

Meteorological data (temperature and rainfall) for the season was obtained from a weather station next to the long term maize trial plots. In addition to rainfall data from the automated weather station, rain gauges were installed over the trial area to monitor rainfall and irrigation. Irrigation was applied via an overhead sprinkler system with water from on-site boreholes. Soil moisture content was monitored on a weekly basis using a neutron probe meter and access tubes. Neutron access tubes were installed at the centre of each of the two plots representing a treatment to determine soil moisture content at 20 cm depth intervals up to a depth of 120 cm.

### 3.5.2 Nutrient analysis

Compost analysis

A very high content of certain nutrients was found in the compost applied for three seasons between 2003 and 2005. N was as high as 11800 mg kg$^{-1}$, P at 14600 mg kg$^{-1}$ and K at 13700 mg kg$^{-1}$ (Table 3.3).

**Table 3.3:** Typical nutrient analysis for the compost applied for three seasons between 2003 and 2005.

| **Nutrient elements** | **N** | **P** | **K** | **Ca** | **Mg** | **Na** |
|---|---|---|---|---|---|---|
| **Content (mg kg$^{-1}$)** | 11800 | 14600 | 13700 | 31200 | 4800 | 2100 |

Soil analysis

Soil samples from each plot were collected before planting in 2013 and the analysis results are shown in Table 3.4. Basic soil analysis was done at the Soil Science laboratory at the University of Pretoria, to quantify the soil nutrient status before planting.

Soil pH

pH was slightly lower in compost + inorganic plots, which averaged at 6.2 compared to inorganic only plots with a mean pH of 5.9. The results confirmed findings by Jobe *et al*., (2007) who observed that most compost material possess a high buffer capacity which lowers soil acidity.

Phosphorus

In order to produce optimum yield grain crops require between 15 and 30 mg kg$^{-1}$ of P in the soil (FSSA, 2000). Residual P treatment plots exhibited higher P content in the soil compared to non-residual P treatments. Some treatments exhibited very low P levels; for instance treatment 0 at 6.0 mg kg$^{-1}$, K at 4.5 mg kg$^{-1}$ and NK at 7.1 mg kg$^{-1}$. By comparison the equivalent residual compost treatments exhibited elevated levels of P values; W with 28.0 mg kg$^{-1}$, WK with 35.7 mg kg$^{-1}$ and WNK with 21.9 mg kg$^{-1}$.

Potassium

Maize requires K levels between 80 and 160 mg kg$^{-1}$ in the soil to obtain optimum yield (FSSA, 2000). K-containing treatments resulted in higher K compared to non K-containing treatments; K (239.1 mgkg$^{-1}$), NK (161.4 mg kg$^{-1}$), PK (185 mg kg$^{-1}$), WK (222.9 mg kg$^{-1}$), WNK (187.9 mg kg$^{-1}$) and WPK (204.2 mg kg$^{-1}$). Residual compost resulted in a higher K-range compared to non-compost containing treatments (Table 3.4).

## 3.6 DATA ANALYSIS

The Statistical Analysis System (SAS) was used to analyse the data collected from the experiment (Statistical Analysis System Institute Inc., 2001) and to create Analysis of Variance (ANOVA) tables. Means were compared using the least significance difference (LSD) test to test probability levels at 5 % (P=0.05) using Tukey's Studentized Range (HSD) Test.

**Table 3.4:** Nutrient status of the topsoil of the different experimental plots as affected by different treatments before planting in the 2013/2014 season.

| **Treatments** | **pH*** | **P**** | **K***** | **Mg***** | **Na***** | **Ca***** | **NH4-N****** | **NO3-N****** | **Total N** |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | **mg kg$^{-1}$** | | | | |
| 0 | 6.9 | 6.0 | 49.9 | 195.0 | 17.1 | 613.4 | 9.7 | 6.1 | 15.8 |
| N | 5.0 | 14.4 | 36.1 | 92.2 | 4.1 | 354.2 | 7.5 | 5.2 | 12.7 |
| P | 6.5 | 38.4 | 33.1 | 203.5 | 6.3 | 64.7 | 12.7 | 6.7 | 19.4 |
| K | 6.9 | 4.5 | 239.1 | 114.2 | 3.9 | 457.2 | 16.8 | 11.6 | 28.4 |
| NP | 5.2 | 41.1 | 27.2 | 114.8 | 2.6 | 437.5 | 13.8 | 9.1 | 22.9 |
| NK | 5.1 | 7.1 | 161.4 | 61.5 | 6.8 | 229.1 | 16.9 | 8.4 | 25.3 |
| PK | 6.5 | 20.7 | 185.2 | 142.7 | 4.2 | 557.6 | 16.6 | 10.1 | 26.7 |
| NPK | 5.2 | 38.4 | 145.3 | 51.0 | 3.3 | 245.4 | 11.2 | 6.6 | 17.8 |
| W | 6.9 | 28.0 | 76.4 | 198.8 | 3.4 | 773.5 | 11.5 | 5.5 | 17.0 |
| WN | 5.6 | 15.4 | 35.9 | 107.7 | 3.7 | 508.8 | 6.6 | 4.4 | 11.0 |
| WP | 6.7 | 59.4 | 63.3 | 181.7 | 4.3 | 862.4 | 12.7 | 8.4 | 21.1 |
| WK | 6.9 | 35.7 | 223.1 | 117.2 | 2.8 | 710.1 | 18.9 | 12.4 | 31.3 |
| WNP | 5.6 | 45.2 | 36.4 | 145.0 | 7.8 | 565.1 | 15.9 | 10.4 | 26.3 |
| WNK | 5.6 | 21.9 | 189.8 | 72.1 | 2.9 | 465.7 | 16.9 | 8.8 | 25.0 |
| WPK | 6.7 | 37.3 | 204.2 | 122.2 | 4.8 | 707.5 | 15.6 | 8.7 | 24.3 |
| WNPK | 6.1 | 41.4 | 137.6 | 85.8 | 5.8 | 418.5 | 13.6 | 6.9 | 20.5 |

* Water ($H_2O$), ** Bray-1, *** Ammonium Acetate Extractable, **** Steam Distillation (Barret *et al.*, 2009)

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# CHAPTER 4

# MAIZE SEED GERMINATION AND SEEDLING EMERGENCE AS AFFECTED BY DIFFERENT FERTILIZER TREATMENTS

## 4.1 INTRODUCTION

Some farmers in rural South Africa use little or no fertilizer in their farming systems and retain seed for use in subsequent seasons (Govender *et al.*, 2008). This practice normally result in poor seed quality, field emergence, growth and yield in various crops (Gosse *et al.* 1986; Vieira *et al.*, 1999). Maximum seed vigour is retained for some time, but begins to deteriorate while seed is still on the plant or in storage (Perry, 1980; Ellis & Pieta-Filho, 1992). Temperature conditions and soil nutrient status may affect seed germination capacity (Ghiyasi *et al.*, 2008). Hence, the main objective of this chapter, is to quantify the effect of different soil fertility regimes on maize seed viability.

## 4.2 MATERIALS AND METHODS

The general procedure for cultivating the maize in the long term fertilizer trial can be found in Chapter 3. In this section only the materials and methods relevant to this chapter will be described. Seed harvested from the field trial in June 2013 were used for thermo-gradient and rotating table seed tests. Hybrid seed (OS) used to establish the crop in 2012 was used as a control for thermo-gradient table trial and fresh hybrid seed was used for the field emergence test.

### 4.2.1 Seed viability tests

*i) Seed germination using a thermo-gradient table*

Two runs of this experiment were carried out in a thermo-gradient table (Type 5008.00) with five covers the (SeedQuest, 2013) located at Phytotron B in the University of Pretoria's Hatfield Experimental Farm. The thermo-gradient table is made up of steel frame of 3 m

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length and 2 m width (Figure 4.1), with both cooling and heating systems installed. The temperature can be set for different zones of the table surface, resulting in a temperature gradient from one side to the other. The minimum temperature was set at 0°C and the maximum at 35°C. This resulted in three temperature zones; 0-11°C (lower limit), 12-23°C (optimum) and 24-35° (upper limit).

**Figure 4.1:** A thermo-gradient table used for the seed tests (Dlamini JC, 2013).

Petri dishes with a double filter paper layer, containing 5 seeds per dish and replicated 10 times, were used (Figure 4.2). In addition to the seed obtained from the field trial, in June 2013, a sample of the hybrid seed was included as a control. The petri-dishes were incubated and seed inspected for coleoptile appearance each morning over the period of 3 to 14 days after incubation. Water was added when dry; especially at the high temperature range to avoid damage of newly appearing shoots. Germination count data obtained was expressed as mean germination index (GI); calculated using equation 4.1 suggested by De Santana and Ranal (2004).

$$GI = \sum_{t=i}^{n} \{[(Exp.days - Di)Gi]\} \;/\; S\ (Seed\ per\ day) \qquad (Eq.\ 41)$$

Where, $n$ =number of germination counts, $D_i$ = number of days until last germination observation, $G_i$ = number of normal germinated seeds and $S$ = number of seeds germinated at the end of the experiment.

**Figure 4.2:** A sample of petri-dishes with maize seeds from the different fertilizer treatments (Dlamini JC, 2013).

*ii) Seedling emergence using a rotating-table*

Seedling emergence was determined at a constant temperature of 30°C in a greenhouse. Soil collected from the different experimental plots was used to fill plastic pots of 0.5 l capacity. Seed harvested from the corresponding plots were planted at a depth of 3 cm; three per pot, each replicated four times. These were then placed on a rotating table in a glasshouse located at the Experimental Farm. Emergence rate was quantified by counting the number of normal seedlings each day from day 3 until no further germination was evident (Agrawal *et al*., 1973; Hampton & TeKrony, 1995; ISTA, 1995). A normal seedling can be defined as one that grows vigorously at the prevailing conditions (Ranal *et al.*, 2009).

At two weeks after emergence (WAE), the seedlings were carefully uprooted after soaking the pots with water overnight, then carefully washed in order to recover most of the roots. Seedling length was measured from the soil surface to the shoot tip, and root length was measured from the soil surface to the tip of the primary root. Shoot and root dry mass (including all seminal roots) was determined by oven-drying seedlings at 65°C until constant mass. Seed metabolic efficiency (SME) was calculated from shoot dry mass (SHW), root dry mass (RTW) and material respired (SMR) using equation 4.2 (Copeland, 1976). Seed metabolic efficiency was determined using equation 4.2 (Rao & Sinha, 1993; Sikder *et al*., 2004):

$$SME = (SHW + RTW)/SMR \qquad (Eq. 4.2)$$

Where, SHW is the shoot dry mass, RTW is the seedling root dry mass and SMR is the seed material respired. Seed material respired (SMR) was determined using equation 4.3 (Copeland, 1976).

$$SMR = SDW - (SHW + RTW + RSW)\ \left(\frac{g}{g}\right) \qquad (Eq. 4.3)$$

Where, SDW is the seed dry mass before germination, SHW is the shoot dry mass, RTW is the seedling root dry mass and RSW is the remaining seed dry mass after germination.

*iii) Seedling emergence in the field*

An *in-situ* seedling emergence count was done after planting hybrid seed, at the long-term trial. This was done to quantify emergence of hybrid seed as influenced by the different fertilizer treatments. Emergence was recorded for only two weeks, in order to quantify the speed of emergence, as at 2 weeks after emergence (WAE) all plots showed 100% emergence. The number of emerged coleoptiles was counted from the first day of emergence until there was no further emergence. Collected data was expressed in emergence rate (equation 4.4) and emergence percentage (equation 4.4) (Ranal *et al.*, 2009).

$$Emergence\ Rate = \left(\frac{number\ of\ normal\ seedlings}{days\ to\ first\ count} + \cdots + \frac{number\ of\ normal\ seedlings}{days\ to\ final\ count}\right) \quad (seed\ per\ day)(Eq.\ 4.4)$$

$$Emergence = \ (No.of\ emerged\ seedlings/No.of\ sown\ seeds) * 100 \quad (\%) \quad (Eq.4.5)$$

## 4.3 RESULTS AND DISUSSION

### 4.3.1 Seed germination index using a thermo-gradient table

*Fertilizer x Temperature interaction effects*

The interaction between fertilizer and temperature treatments significantly influenced maize seed germination index (Figure 4.3). At temperature regime 1-11°C, seed from NPK treated plots resulted in a significantly higher mean germination index (GI) of 1.23 seed per day compared to the control (0) and the other inorganic N, P or K fertilizer treatments. Compared to the other organic + inorganic treatments at the same temperature regime, WNPK had the highest mean GI of 1.41 seed per day. Despite WNPK obtaining the highest mean GI at this temperature regime, it was not significantly different from WP (1.2 seed per day), WNK (1.05 seed per day), WPK (1.03 seed per day), and the original hybrid seed (OS) (1.2 seed per day). Seed obtained from soil deficient in one (NP, NK and NK) or more inorganic nutrients (N, P and K) and the control had a significantly lower mean GI, ranging between 0.45 and 0.78 seed per day. Addition of compost to inorganic fertilizer treatments resulted in a higher mean GI (0.73-1.2 seed per day). Compared to other temperature regimes, the GI values at the 1-11°C regime were on average the lowest. Ranal and De Santana (2006) noted that higher GI values mean higher seedling vigour in relation to the other ($0 < GI \leq. n$)

Germination rate was higher in the 12-23°C regime (Figure 4.3). Compared to the other inorganic fertilizer treatments NPK obtained the highest mean GI of 4.92 seed per day. At this temperature regime, NK (3.81 seed per day) and PK (3.1 seed per day) also performed well. This implies that seed produced under deficient N or P conditions can still perform well if temperature conditions are ideal. Comparing inorganic and organic + inorganic fertilizer treatments, WNPK resulted in the highest mean GI value of 5.41 seed per day. This was, however, not significantly different from NPK (4.92 seed per day) and OS (4.98 seed per day). Comparing GI values from one deficient (NP, NK and PK), two deficient (N, P or K) nutrient element treatments and 0 (1.26 - 3.81 seed per day) with their corresponding organic + inorganic treatments, organic treatments had a higher mean GI (1.53 - 4.66 seed per day). This implies that under deficient soil nutrient conditions, the addition of compost positively influenced maize seed germination vigour. The compost added in the trial filled the void of deficient N, P or K to levels that enhanced seed germination. Compared to other temperature regimes, the 12-23°C temperature regime resulted in the highest mean GI values across all the fertilizer treatments.

Compared to the other inorganic N, P or K fertilizer treatments at temperature regime 24-35°C, NPK had a significantly higher mean GI value of 3.13 seed per day (Figure 4.3). Considering the organic counterparts, WNPK resulted in the highest mean GI of 3.6 seed per day. Organic treatments (1.26-2.82 seed per day) performed better than their corresponding inorganic single, double (N, P or K) and 0 treatments (0.31-1.23 seed per day). With the exception of K and the control, all other fertilizer treatments performed better at temperature regime 24-35°C than the 1-11°C regime. Worth noting is that, seed from NPK and WNPK treatments performed better than the original hybrid seed (OS) under all the three temperature regimes. This could be because the hybrid seed had been stored for about a year at the time of the experiment, which might have deteriorated the vigour.

The results displayed a significant influence of temperature on seed germination regardless of the nutrient status of the soil in which the mother plants were grown. Seed germination may vary under variable soil temperature regimes, since it directly influences the rate of water uptake (Wanjura & Buxtor, 1972; Roberts, 1988; Sikder *et al.*, 2009). Observations by Wilcox & Pfeiffer (2008) stated that extreme temperature conditions, especially low temperatures between 12.3 and 14.5°C may restrict maize seed germination and hence retard plant growth.

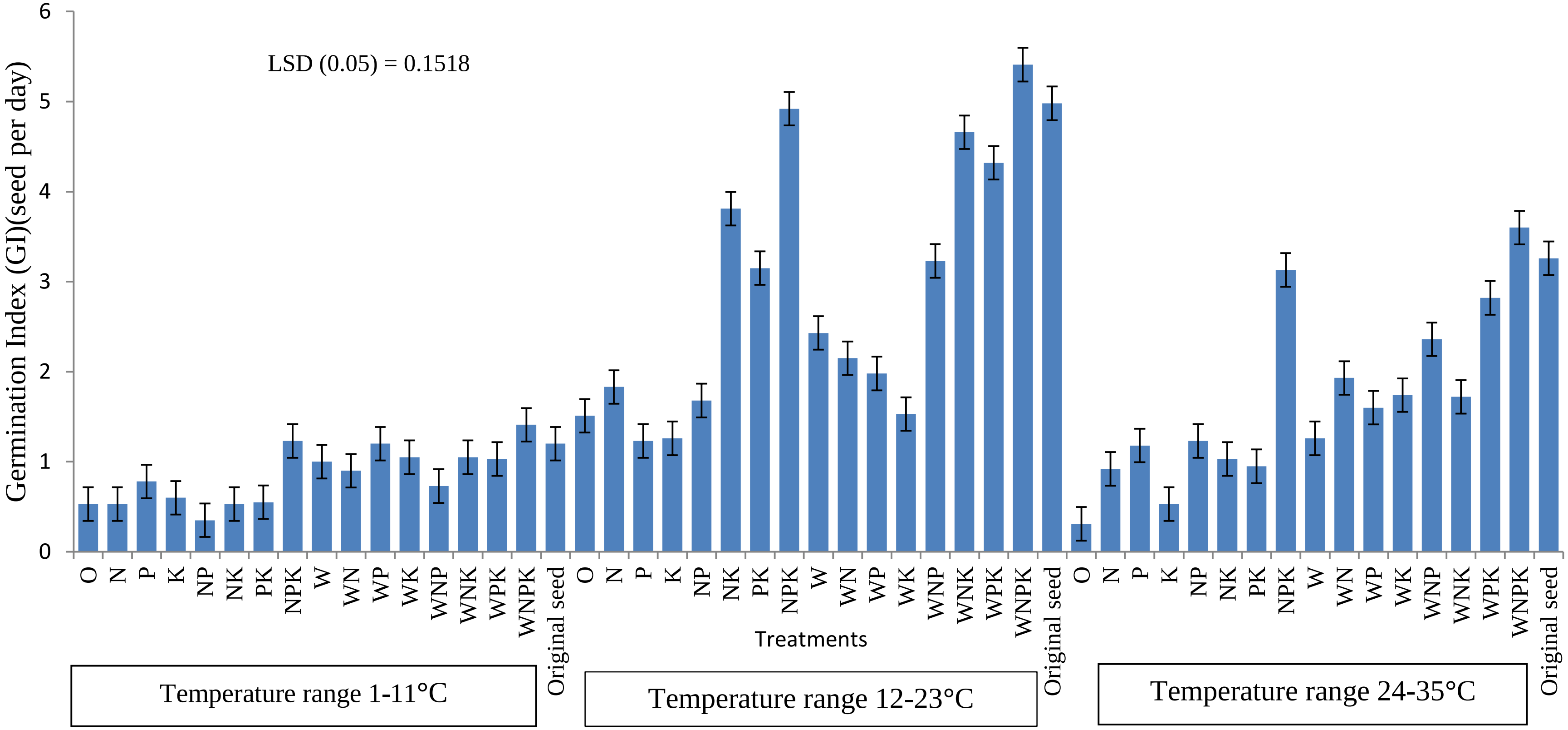


**Figure 4.3:** Germination index (GI) of maize at different temperature regimes as affected by soil nutrient status.

### 4.3.2 Maize seedling emergence

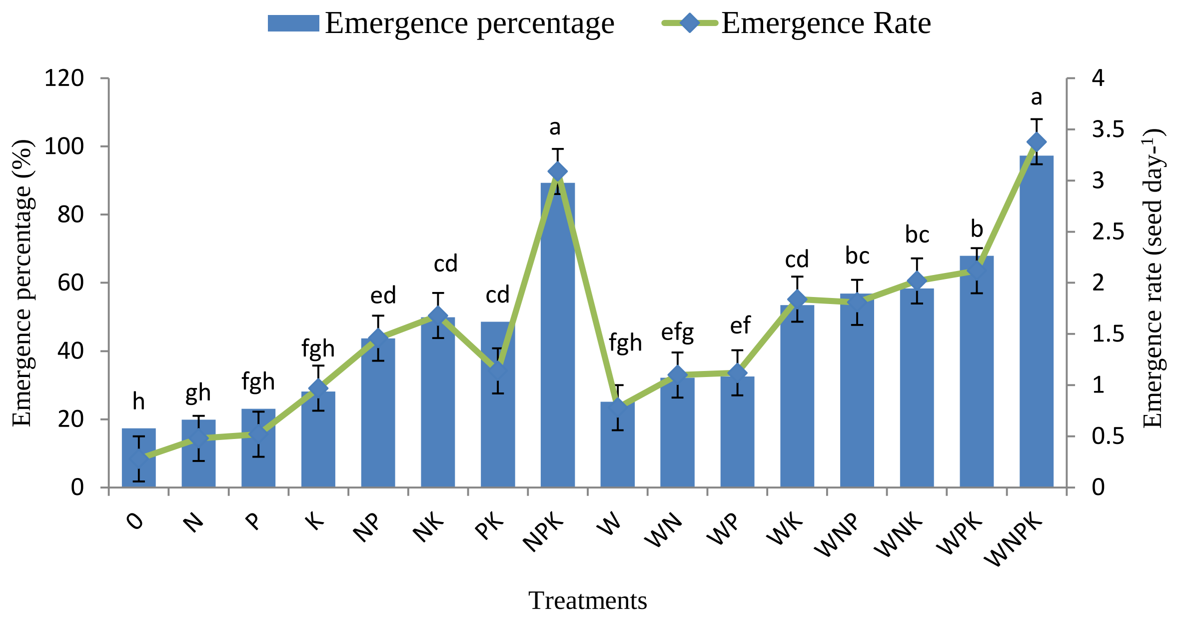
Seedling emergence on the rotating-table

*i) Emergence rate*

Maize seedling emergence was influenced by the different fertilizer treatments. A steady increase in emergence rate with improved soil nutrition was observed (Figure 4.4). Seed from NPK treatment had a higher emergence rate of 3.09 seed day$^{-1}$ compared to the other inorganic fertilizer treatments. Organic (W) treatments exhibited a similar trend. The organic treatments resulted in a higher mean emergence rate range compared to inorganic fertilizer treatments, although mostly not significant. Uneven seedling emergence sometimes demonstrate differences in seed vigour and plant nutrition ranks amongst the most prominent factors influencing seed vigour (Copeland & McDonald, 1995). Cruz-Garcia *et al.,* (1995) reported that a poor crop stand is normally influenced by poor seed quality and may consequently result in low maize yield. The results further confirmed findings by Dornbos Jr, (1995) who noted that soil fertility plays a significant role in seed germination and seedling emergence.

*ii) Emergence percentage*

Emergence percentage was influenced by the different soil fertility regimes (Figure 4.4). An emergence percentage of 89.3% was recorded for NPK treatment, which was higher than the other inorganic fertilizer treatments. There was no statistical difference in emergence percentage between treatment combinations receiving two nutrient elements; NP (43.7%), NK (49.9%) and PK (48.6%). There was also no difference in emergence percentage between the control (0) and plots receiving only one inorganic nutrient element; N (19.9%), P (23.1%), K (28.1%) and 0 (17.3%). However, seed from plots receiving two nutrient elements had higher emergence percentage than those receiving one nutrient element. The organic treatments had a higher emergence percentage than their corresponding inorganic treatments. The results confirm findings by Sun *et al.,* (2007) who observed that seed vigour often affects the potential for its germination, field emergence and seedling establishment.



**Alphabetical letters (abcdefg) are used to show significant differences at p=0.05 for Emergence percentage ,**Vertical bars ($S_e$) = Emergence rate*

**Figure 4.4:** Maize emergence percentage (%) and rate (seed day $^{-1}$) at 2 WAE on a rotating-table as influenced by different fertilizer treatments.

*iii) Seedling growth*

*Shoot length*

Shoots of the NPK treated seedlings were the longest (18.5 cm), whilst the 0 seedlings were the shortest at 10.0 cm (Table 4.1). N-containing treatments tended to produce longer shoots (15.5 – 18.5 cm) than the non-N containing treatments (10.0 to 15.8 cm). This could be because nitrogen is a major component of amino acid; these are responsible for the formation of protoplasm, which is responsible for cell division and thus important in plant growth and development (Marschner, 1989). Hence, if N is available in sufficient quantities in the soil, it often stimulates plant growth and development as a result of increased cell division (Russell, 1942; Uchida, 2000). Soil fertility has a positive influence on plant seedling development (Dornbos Jr., 1995) and well-established seedlings ensure a uniform and vigorous crop stand (Rowden *et al*,. 1981; Harris *et al.*, 1999; Ghasemi-Golezani *et al.*, 2010).

*Root length*

Only minor differences in root length were observed (Table 4.1). Roots from the inorganic N treatment (14.2 cm) were the longest, which were, however, not statistically different.

*iv) Seedling dry mass*

*Shoot dry mass*

Shoot dry mass was not significantly influenced by any of the fertilizer treatments (Table 4.1). Despite the lack in significant differences, inorganic P resulted in the highest shoot dry mass (1.9 g shoot$^{-1}$) amongst the inorganic group. The dry mass of all the other inorganic fertilizer treatments ranged between 0.85 and 1.82 g shoot$^{-1}$. The residual effect of compost did not result in any significant difference in seedling dry mass. Organic treatments resulted in a lower range of shoot masses (0.68 to 1.32 g shoot$^{-1}$) than the inorganic fertilizer treatments (0.85 to 2.0 g shoot $^{-1}$). The results were not in corroboration with findings by Perry (1980) and Ghassemi-Golezani *et al.*, (2010) who observed that differences in seedling shoot dry mass may result from soil nutrient availability, which strongly influences plant seed quality and seedling vigour. The lack of response could be attributed to inability of plants to effectively absorb nitrogen; which is a main growth component, at early plant growth stages (FSSA, 2000).

*Root dry mass*

Root dry mass was influenced by the different soil fertility regimes, although not statistically different (Table 4.1). The treatments, NPK and NP obtained the highest root mass of 0.45 g each. There was a significant difference between the inorganic only and the organic + inorganic treatments. The average root dry mass of inorganic fertilizer treatments were lower than that obtained where organic + inorganic fertilizer was applied.

**Table 4.1:** Shoot and root length and dry mass at two weeks after emergence as influenced by different fertilizer treatments on a rotating table.

| **Treatments** | **Length (cm/seedling)** | | **Seedling mass (grams)** | |
|---|---|---|---|---|
| | **Shoot** | **Root** | **Shoot** | **Root** |
| 0 | $10.0^{b*}$ | $6.9^{c}$ | $0.85^{a}$ | $0.38^{ab}$ |
| N | $16.5^{abc}$ | $14.2^{a}$ | $1.28^{a}$ | $0.25^{ab}$ |
| P | $15.0^{ab}$ | $11.7^{abc}$ | $1.90^{a}$ | $0.31^{ab}$ |
| K | $15.8^{ab}$ | $11.6^{abc}$ | $1.82^{a}$ | $0.30^{ab}$ |
| NP | $17.0^{a}$ | $10.5^{abc}$ | $1.55^{a}$ | $0.45^{a}$ |
| NK | $17.4^{a}$ | $10.0^{abc}$ | $1.48^{a}$ | $0.38^{ab}$ |
| PK | $14.5^{ab}$ | $10.8^{abc}$ | $1.20^{a}$ | $0.28^{ab}$ |
| NPK | $18.5^{a}$ | $10.4^{abc}$ | $1.37^{a}$ | $0.45^{a}$ |
| W | $14.5^{ab}$ | $10.0^{abc}$ | $0.68^{a}$ | $0.13^{b}$ |
| WN | $15.5^{ab}$ | $12.3^{ab}$ | $2.00^{a}$ | $0.33^{ab}$ |
| WP | $13.9^{ab}$ | $10.2^{abc}$ | $1.53^{a}$ | $0.55^{a}$ |
| WK | $14.5^{ab}$ | $11.0^{abc}$ | $1.00^{a}$ | $0.33^{ab}$ |
| WNP | $16.9^{a}$ | $9.9^{abc}$ | $0.90^{a}$ | $0.44^{ab}$ |
| WNK | $17.4^{a}$ | $9.6^{abc}$ | $1.10^{a}$ | $0.45^{a}$ |
| WPK | $13.6^{ab}$ | $8.3^{bc}$ | $1.14^{a}$ | $0.4^{ab}$ |
| WNPK | $17.9^{a}$ | $9.2^{abc}$ | $1.39^{a}$ | $0.47^{a}$ |
| LSD(0.05) | 6.0 | 5.0 | NS | 0.32 |

*In any given column, means followed by the same letter(s) do not differ significantly at 5% level as per Tukey's test. NS = No significant difference.

### *v) Seed metabolic efficiency (SME)*

Seed metabolic efficiency refers to the ability of seed to mobilize and utilize metabolic reserves during the germination process (Bradford & Hsiao, 1982; Rao & Sinha, 1993). Seed with a higher SME are desirable in crop production (Sikder *et al.*, 2009). It positively influences seed germination and seedling establishment in maize production since it determines the amount of food reserves available to the germinating plant (Alofe & Schradder, 1975; Ghasemi-Golezani *et al.*, 2010). It is also a desirable character under water and nutrient stress conditions where emergence may be affected by unfavourable environmental conditions (Wanjura & Buxtor, 1972; Penning de Vries *et al.*, 1979).

The SME of WNPK treatment was significantly lower (0.16 g/g) than that of P or K treatments (Figure 4.5). With the exception of WN and WPK, SME of organic treatments were lower than their inorganic counterparts. The results obtained were not in corroboration with findings by Carvalho & Nakagawa (2000), who stated that a balanced soil nutrient status is crucial where increased amounts of seed reserves are anticipated. He further argued mobilization during germination is not entirely dependent on soil nutrient status, but other factors such as soil moisture also play a significant role; which could have been the case in this experiment as well.

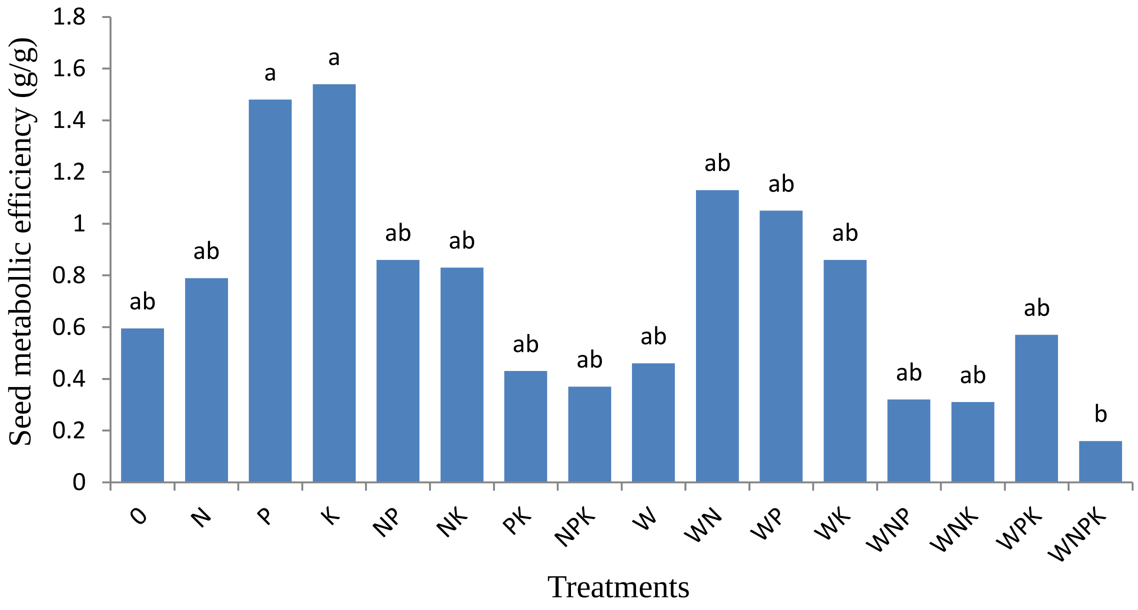


**Figure 4.5:** Seed metabolic efficiency of maize as influenced by different fertilizer treatments.

Seedling emergence in the field

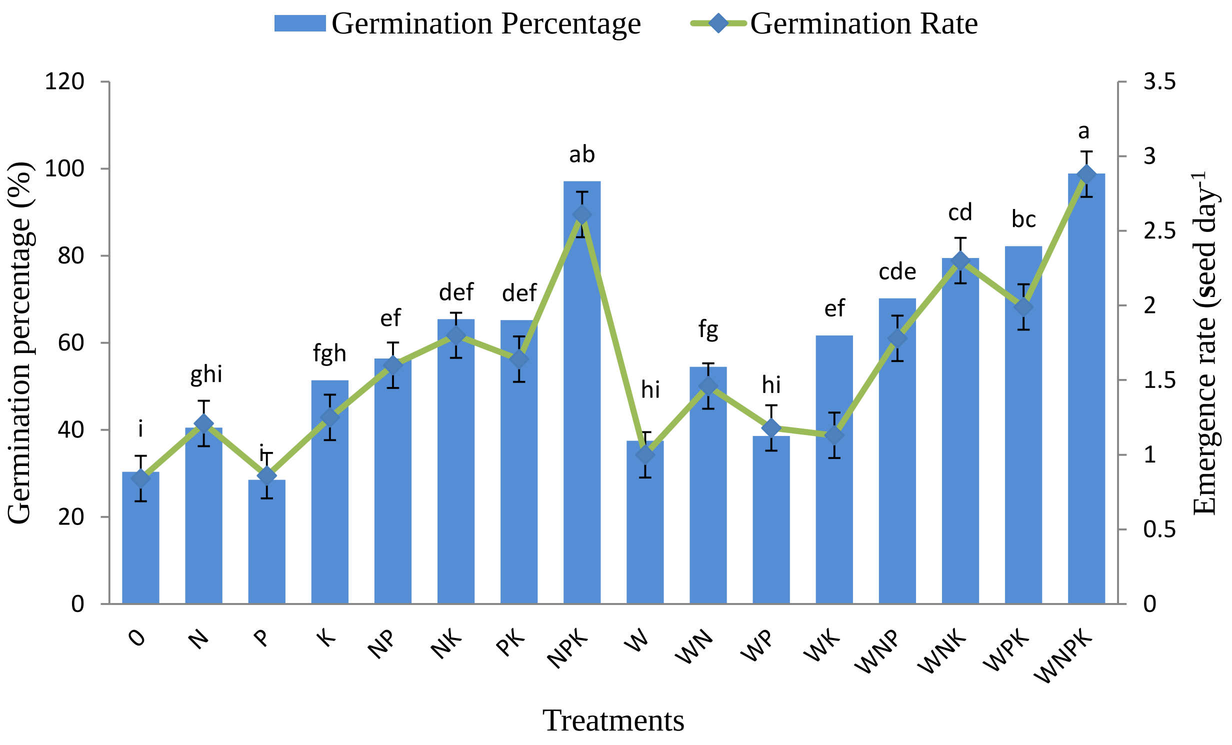
*i) Emergence rate*

The different soil fertility regimes significantly influenced the rate of emergence of maize seedlings from fresh hybrid seed in the field at 2 WAE (Figure 4.6). The NPK treatment resulted in higher mean germination rate (2.61 seed day$^{-1}$) compared to any other inorganic fertilizer treatment. Where two nutrient elements were added together emergence rate tended to be higher (1.6 – 1.8 seed day$^{-}$), compared to single or no fertilizer treatments (0.84 – 1.21

seed day$^{-1}$). This highlighted the importance of a balanced soil nutrient status in the attainment of rapid seedling emergence, which often results in a good crop stand (Hegerty, 1976; Turkmen *et al.*, 2004).

*ii) Emergence percentage*

In the field, maize seedling emergence percentage was positively influenced by all the treatments and it exhibited a trend similar to that of emergence rate at 2 WAE (Figure 4.6). Organic + inorganic treatments resulted in a slightly higher germination percentage compared to only inorganic fertilizer treatments. Emergence percentage influenced by inorganic fertilizer treatments ranged between 28.5 and 97.1%, whilst organic + inorganic ranged between 37.5 and 98.9%. The results highlighted the importance of good crop nutrition in the establishment of a vigorous crop stand (Hegarty, 1976; Copeland & McDonald, 1995; Dornbos Jr., 1995).



**Alphabetical letters (abcdefg) are used to show significant differences for Emergence percentage ,**Vertical bars ($S_e$) = Emergence rate*

**Figure 4.6:** Maize seedling emergence (2 WAE) in the field as influenced by different fertilizer treatments.

## 4.4 CONCLUSIONS

Hybrid seed (OS) and seed obtained from NPK and WNPK treated soils resulted in higher germination index (GI) (up to 5.41) regardless of temperature regime. Hence, farmers storing and planting maize seeds carried over from previous seasons should consider seed from plants not deficient in any major nutrient. Balanced soil nutrient status positively influenced emergence under field conditions. These fertilizer treatments have a positive influence on maize seedling length and dry weight (both shoot and root). However, a higher seed metabolic efficiency was reported for plants receiving P and K only.

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# CHAPTER 5

# VEGETATIVE GROWTH AND REPRODUCTIVE DEVELOPMENT OF MAIZE AS AFFECTED BY DIFFERENT FERTILIZER TREATMENTS

## 5.1 INTRODUCTION

More than 70 years of different fertilization inputs resulted in a wide range of soil fertility regimes, which dramatically affect the growth and yield of maize in the long-term maize fertilizer trial. In the quest for a better understanding of low soil fertility effects on the growth and yield, various maize parameters were measured on a regular basis during the growing period of the 2013/2014 season. Classic growth analyses were conducted in order to identify possible explanation for the differences in growth. In order to quantify the impact of soil fertility on reproductive development, the size of the tassels and ears were regularly measured. The dates of emergence and synchronisation of tassels and silks were carefully observed, in order to quantify the influence of the different soil fertility regimes on their time of appearance.

## 5.2 MATERIALS AND METHODS

The general procedures for cultivating maize in the long term fertilizer trial can be found in Chapter 3. In this section only the materials and methods relevant to this chapter are described.

### 5.2.1 Plant growth parameters

From each net plot, a 5 $m^2$ sub-plot was marked out from which all data were collected. Five plants within the 5 $m^2$ sub-plot were randomly selected and permanently marked for non-destructive measurements such as plant height. Plant height was measured weekly from 2 to 8 weeks after emergence (WAE). This parameter was measured using a ruler when the plants were still small and a measuring tape when plants were taller. At 8 WAE, the plants had reached maximum height and the majority of plants had started tasseling. Leaf area was determined through destructive sampling from the demarcated 5 $m^2$ sub-plot every week from the second week until 8 WAE. Using leaves from two plants per plot per week, a Licor Li-3001 leaf area meter was used for leaf area determination. Dry mass was quantified, after measuring leaf area, by oven-drying plant material at a temperature of $65^0$C until constant mass. Leaf area was used to quantify leaf area index (LAI). LAI was then calculated using the equation 5.1 (Gardner *et al*., 1985; Edje 1988);

$$Leaf\ area\ index\ (LAI) = \frac{L_A}{P} \qquad (Eq. 5.1)$$

Where, $L_A$ is the leaf area per plant and P is the area per plant.

Tassel and silk appearance were monitored every morning from the day when the first tassel and silk appeared. Emerged tassels and silks were counted every day until no further tassels and silks appeared. Tassel and ear length were measured at 12 WAE using a ruler. Tassel length was measured from the flag leaf to its tip, and ear length was measured from the stalk to its tip; including the husks.

## 5.3 RESULTS AND DISUSSION

### 5.3.1 Plant height

Maize plant height was significantly influenced by the fertilizer treatments (Figure 5.1). There was an increase in plant height between 2 and 8 weeks after emergence (WAE) in almost all treatments. Comparing the plant height from organic + inorganic fertilizer to that of inorganic only treated plants at 8 WAE, the organic + inorganic treated plants were generally taller. Plant height in organic + inorganic treatments ranged between 154 and 220 cm, whilst in inorganic fertilizer treatments ranged between 101 and 178 cm. WNPK resulted in the tallest plants (220 cm), which however did not differ from WNK and WPK plants (204 cm each). At 8 WAE, NPK (178 cm) and NP (166 cm) resulted in the tallest plants compared to the other inorganic treated plants. Plant height features such as plant stem area and length are appreciable in photosynthetic activities, and are important features to consider when accounting for a plant's total sunlight interception area (Loomis and Williams, 1970; Rajeshwari *et al.*, 2007). The results obtained highlighted the importance of ensuring a balanced nutrient status in obtaining maximum plant growth per unit time.

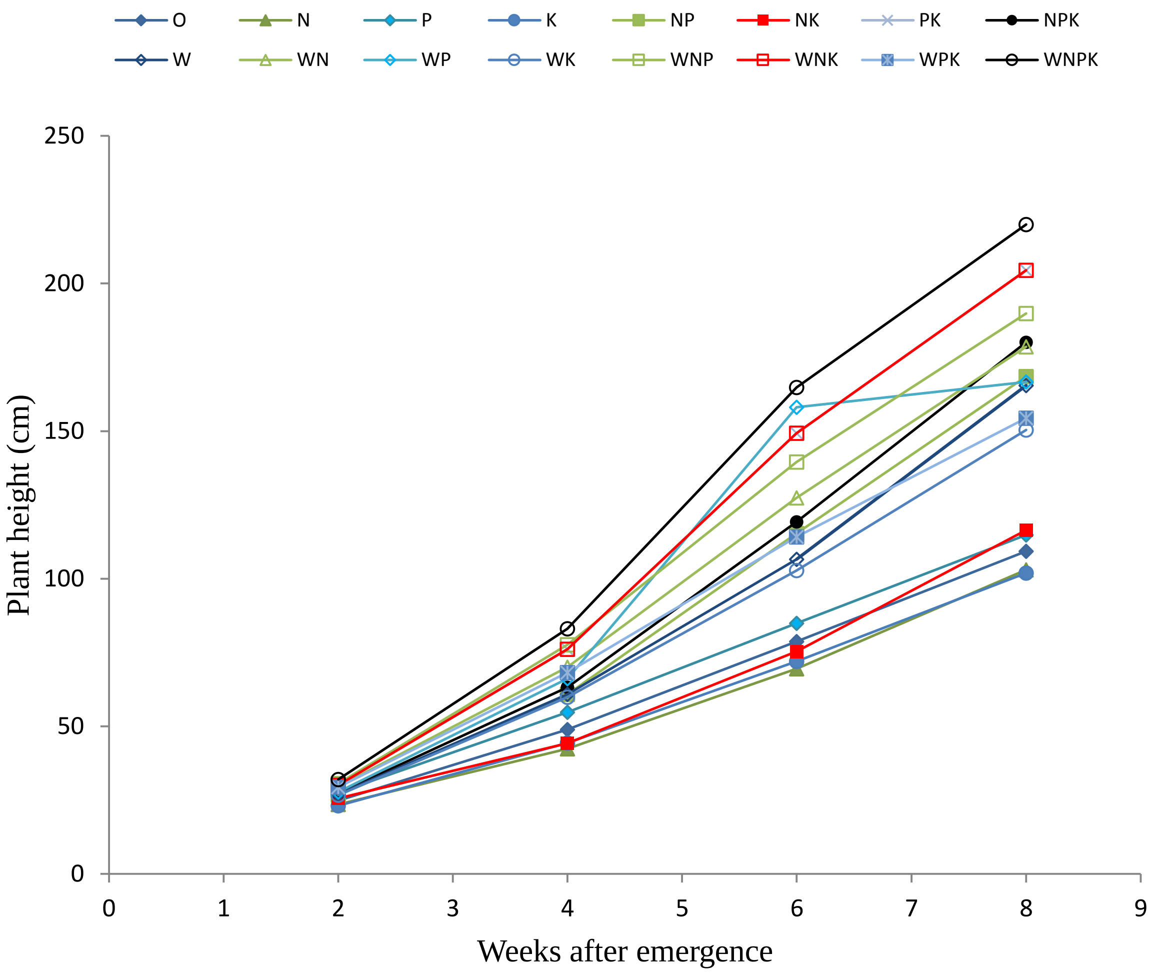

**Figure 5.1:** Maize plant height as affected by different fertilizer treatments.

### 5.3.2 Leaf area index (LAI)

Leaf area index can be described as the leaf area of a crop per unit area on which it stands (Russell, 1942; Edje, 1988). There was an exponential increase in leaf area index from 3 to 8 WAE (Figure 5.3). NPK and NP treatments resulted in significantly the highest leaf area indices compared to the other inorganic fertilizer treatments, with 3.1 and 2.9 respectively. The LAI of 2.4 to 3.8 for the organic + inorganic fertilizer treatments were appreciably higher than those of the inorganic treatments (1.6 to 3.1). The LAI range obtained showed the importance of a balanced soil nutrient status in the development of a healthy and large plant canopy. Previous experiments on maize have shown that, in some varieties LAI between 3 and 4 may be optimal for achieving maximum grain yields (Lindquist *et al.*, 1998). Duncan (1970); Madonni & Otegui, 1996 pointed-out that higher LAI values may be important in the efficient interception of sunlight at low levels of illumination intensities.

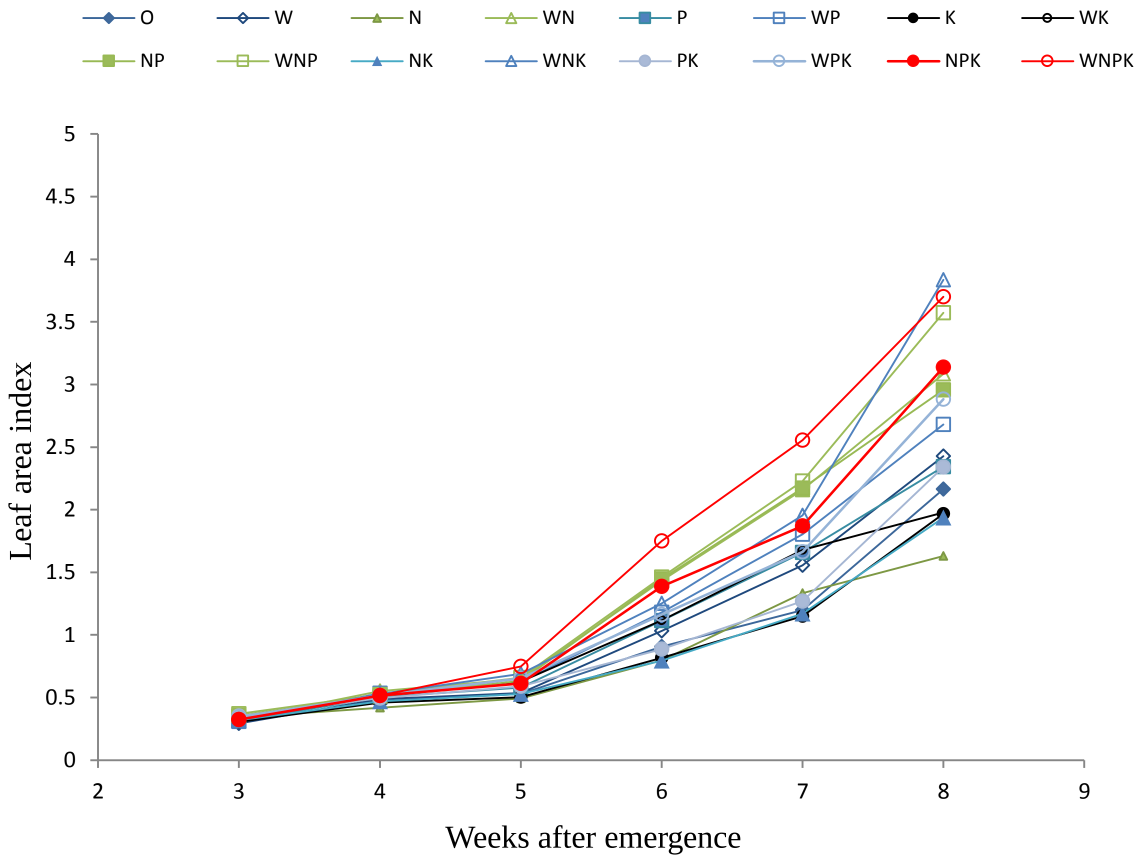

**Figure 5.2:** Leaf area index (LAI) of maize as influenced by different fertilizer treatments.

### 5.3.3 Total dry mass

The differences in dry mass per plant as influenced by inorganic only and organic + inorganic fertilizer treatments were distinct at 8 WAE (Figure 5.3). The dry mass of organic + inorganic fertilizer treated plants ranged between 51 and 108 g, whilst inorganic treatments ranged between 26 g and 71 g. At 8 WAE, the total dry mass (71 g each) obtained from NP and NPK treatments, were the highest compared to the other inorganic treatments.

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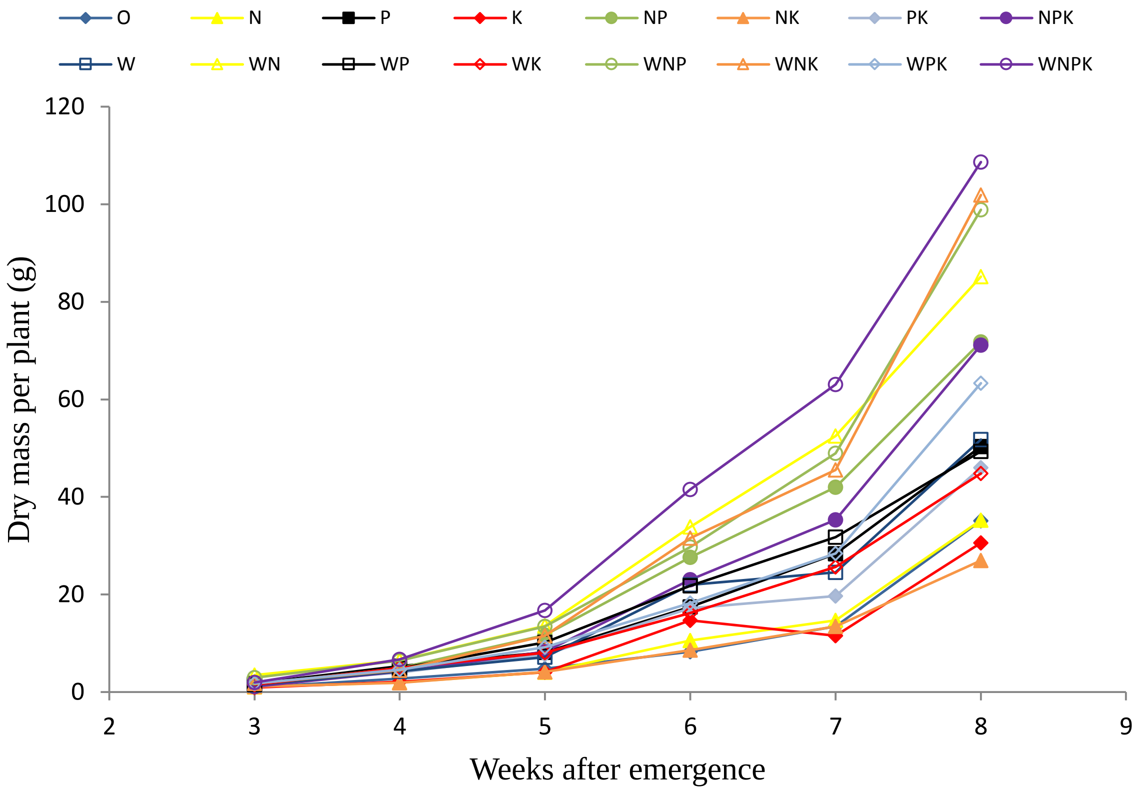

**Figure 5.3:** Total dry mass per maize plant as affected by different fertilizer treatments.

### 5.3.4 Correlation between LAI and total dry mass

The different soil fertility regimes resulted in a strong positive correlation ($r^2$ = 0.9384) between leaf area index and above ground dry mass (Figure 5.4). The size of crop plant's metabolic factory often influences the amount of metabolites which will be available for respiration (Beevers, 1970; Gardner *et al.*, 1985). LAI describes the size of the assimilatory apparatus of a plant stand as one of the primary factors that determine the total dry matter produced by a crop (Kvet *et al.*, 1971). Higher LAI is highly desirable where the total biomass (biological yield) is desirable, especially in forage and fodder crops. The capacity of a plant canopy to efficiently intercept and utilize solar radiation determines the amount of the total dry mass per plant (Gardner *et al.*, 1985). The results confirmed findings by Aase (1977), who observed a high correlation between leaf area index and plant dry mass in barley. Annandale *et al.*, (1987) also found a highly significant correlation ($r^2$ = 0.69) between leaf area index and above ground dry mass of wheat as influenced by varying soil nutrient status. Leaf area expansion per unit area improves solar radiation interception and consequently increasing total dry mass accumulation (Boote *et al.*, 1996; Adelana & Milbourn, 1972). The results are in corroboration with findings by Cock and Yoshida (1973) who observed that the efficient conversion of absorbed radiation into dry matter is significantly reduced by soil nutrient deficiencies, especially in cereal crops.

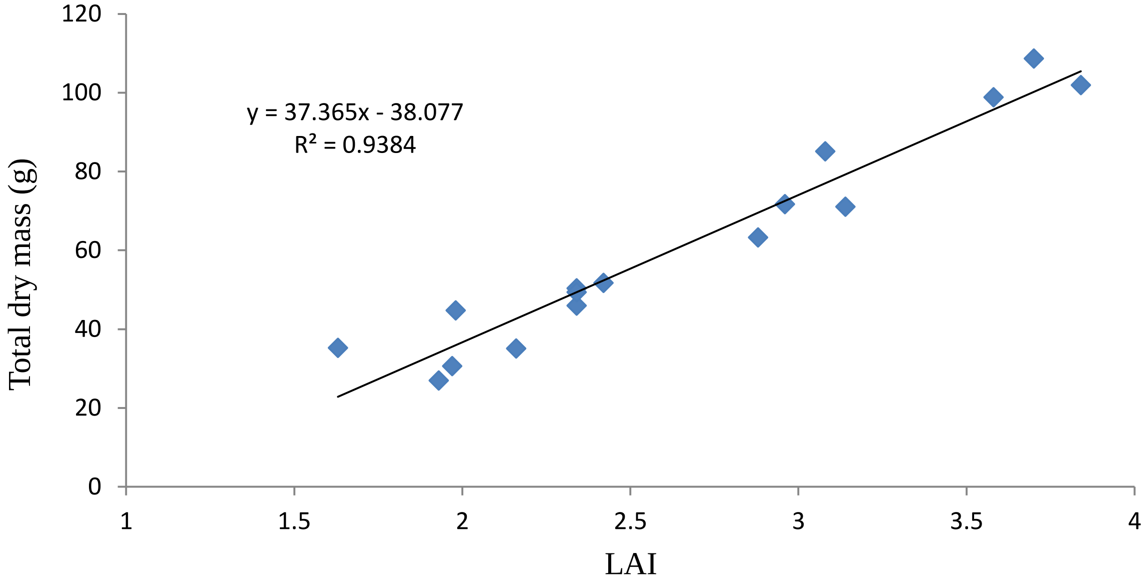

**Figure 5.4:** Relationship between leaf area index (LAI) and total dry mass in maize.

### 5.3.5 Tassel and ear length as influenced by different fertilizer treatments

*Tassel length per plant*

The NPK treatment produced the longest tassels compared to the other inorganic fertilizer treatments, with a final length of 61.7 cm (Figure 5.5). There was no significant difference in tassel length between plots receiving two nutrient elements (NK, NP and PK). Similarly, there was no significant difference in tassel length between plots receiving one nutrient element and the control (N, P, K and 0) ranging between 15.2 and 17.8 cm. Tassels from plants receiving one inorganic nutrient element were shorter than those from plants receiving two elements. The addition of organic material resulted in slightly longer tassels than their inorganic fertilizer treated counterparts; although, the differences were not significantly different from the inorganic treatments.

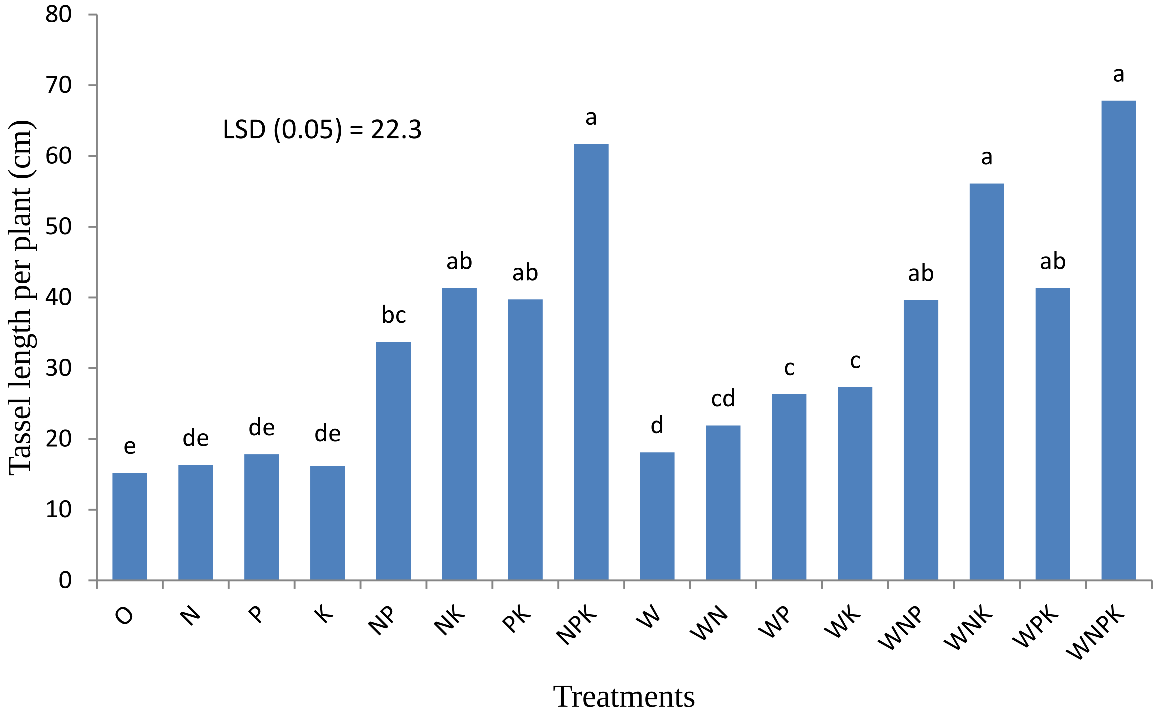

**Figure 5.5:** Tassel length as affected by different fertilizer treatments.

*Ear length*

The NPK treatment produced significantly longer ears compared to the other inorganic fertilizer treatments, with a final length of 51 cm (Figure 5.6). All organic + inorganic fertilizer treated plants produced somewhat longer ears than their corresponding inorganic fertilizer treated counterparts; prominent differences were found between WNP (47.3 cm) and NP (27.8 cm), and WK (31.3 cm) and K (16.3cm). Khan *et al., also* (2008) observed that longer ears are most likely to be found in the integrated organic + inorganic fertilizer programmes due to adequate supply of nutrients from such treatments. Turi *et al.*, (2007) and Ayoola and Makinde (2009) noted that longer ears normally result from combined use of organic and mineral fertilizers and under a balanced soil nutrient status compared to sole inorganic or organic fertilizers. These results were in corroboration with findings by Rajeshwari *et al.*, (2007), who observed a significant increase in maize ear length with an increase in soil nutrient supply, especially nitrogen.

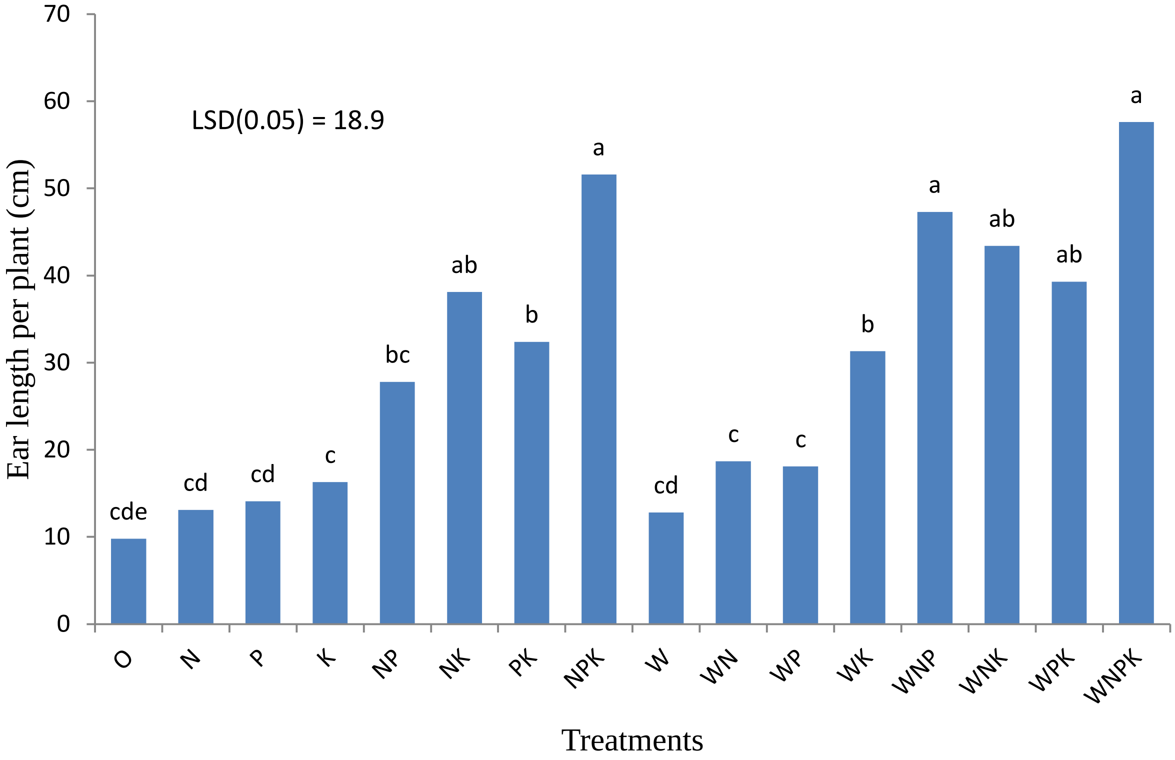

**Figure 5.6:** Maize ear length as influenced by different fertilizer treatments.

*Correlation between plant height and ear length*

There was a weak positive correlation ($r^2 = 0.1714$) between plant height and ear length as influenced by different soil fertility regimes (Figure 5.7). Nanda *et al.*, (1981) observed that traits such as plant height and ear length are more dependent on genetic make-up more than on growing conditions of plants. Hansen and Baggett (1977) reported that maize plant height is determined by the number and length of nodes; whose meristematic elongation is influenced by both growing conditions and genetics. Therefore, if a plant is well-developed and vigorous, longer ears are most likely, the result of a bigger size of the photosynthetic body, which supports ear development (Baynes & Brawn, 1973).

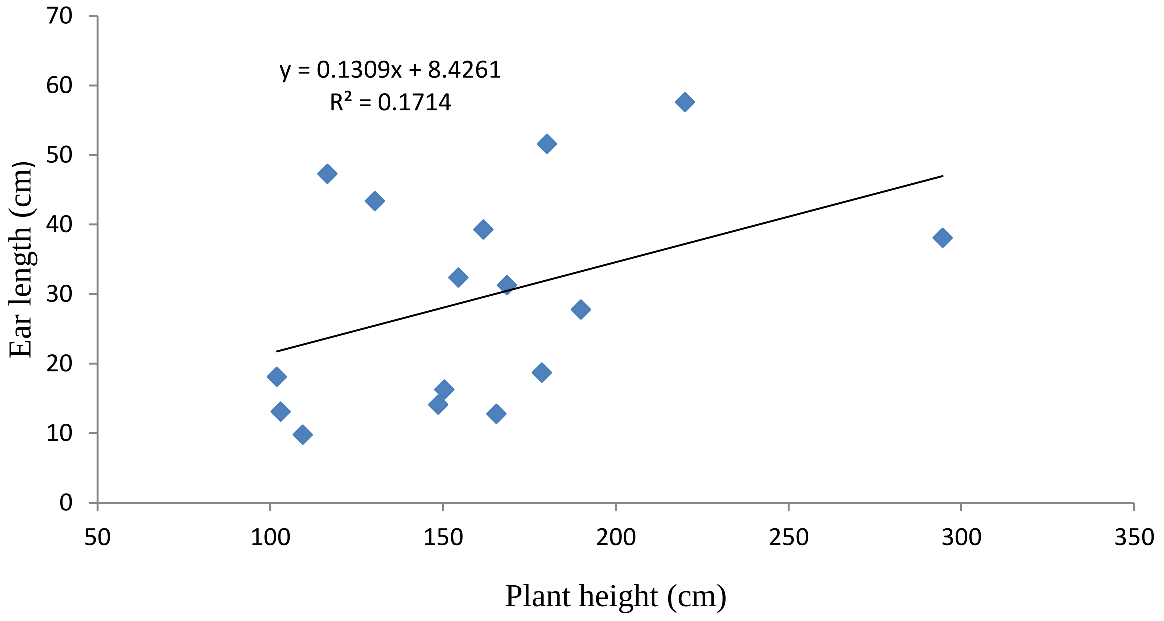

**Figure 5.7:** Relationship between plant height and ear length as influenced by the different fertilizer treatments.

*Correlation between tassel length and ear length*

There was a strong positive correlation ($r^2$ = 0.907) between tassel and ear length as influenced by the different fertilizer treatments (Figure 5.8). Under normal conditions when tassels are big and prominent the ear is likely to be smaller (Stoller USA, 2014). It could not be ascertained whether this is as a result of the plant sending more sugars to the prominent top most parts of the plant; resulting in less sugars available to the ears (Kwabiah *et al*., 2003; Abuyenywa *et al*., 2007).

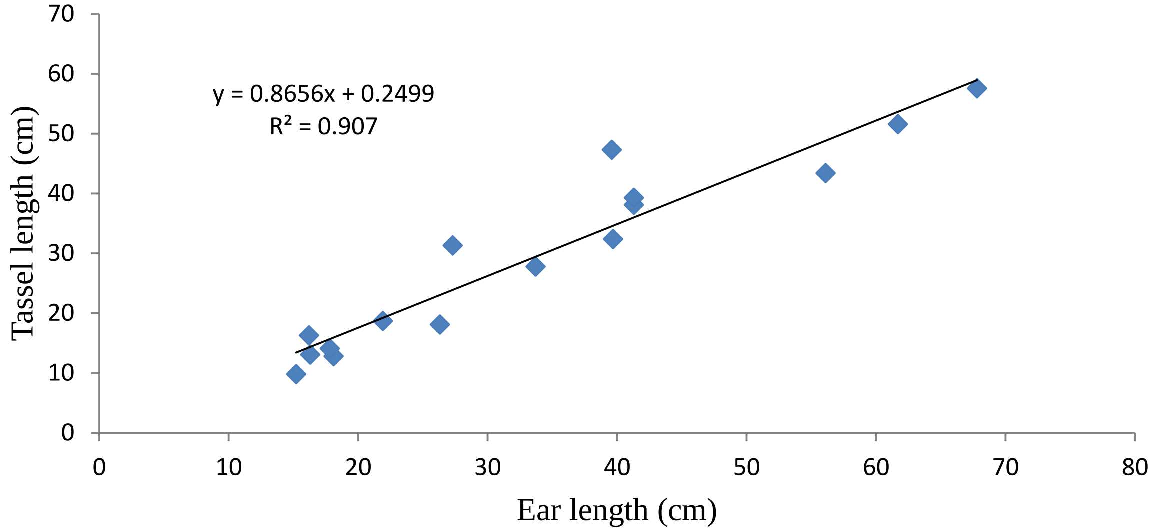

**Figure 5.8:** Relationship between tassel and ear length as influenced by different fertilizer treatments.

### 5.3.6 Tassel and silk appearance as influenced by different fertilizer treatments

*Tassels emergence*

In maize, tasselling normally occurs 2 to 3 days before silk emergence; varying between genotypes (Carvaco *et al*., 2003). The appearance of tassels was affected by the different soil fertility regimes (Figures 5.9 - 5.12). At 9 WAE more plants from plots receiving NPK, WNP and WNPK had tasselled than any other treatment (Figure 5.9). In a study by Ayoola and Makinde (2009) it was observed that under a balanced soil nutrient status maize plants tassel earlier in general, while under unfertilized and control treatments tassel emergence was delayed. At 9 WAE, less than 4 % of plants had tasselled in the P and WNK treatments, whilst up to about 10 % had already tasselled in the NP and WN treatments.

At 10 WAE some plants from all treatments had tasselled, while plants in the WNP and WNPK treatments were already shedding pollen. Worth noting is that, organic + inorganic fertilizer treatments tasselled earlier than their inorganic only counterparts (Figure 5.10).

At 11 WAE, plants from the NPK treatment started shedding pollen as well. Organic + inorganic treatments had between 60.3 % and 88.3 % tasselled plants at this stage, whilst inorganic treatments had between 52.3 % and 97.2% (Figure 5.11)

At 12 WAE NPK, WNK, WPK and WNPK treatments had 100% of their plants tasselled and almost all treatments had some pollen shedding (Figure 5.12). Plants in organic + inorganic treatments had a higher number of tassels emerged compared to the inorganic only counterparts at this stage. Amongst the inorganic group there were distinct differences in the number of emerged tassels; one nutrient element (N, P and K) compared to two nutrient element treatments (NP, NK and PK); with less tassels present in the control (0) and P and K treatments. With two inorganic nutrient elements, plants produced between 10 to 20% more tassels than where one nutrient element was added.

### *Number of plants with silks*

The rate of silk appearance was significantly influenced by different fertilizer treatments (Figures 5.9 - 5.12). As was observed by Nielsen (2010) and Krisna (2012), silking does lag behind tasselling. There were a few plants silking at 9 WAE but more plants had silked at 12 WAE especially as influenced by NPK, WNK, WPK and WNPK (100 % of plants had tasselled and silked). Silk appearance followed a similar trend to that of tasselling, except at 10 WAE, where the control, N, K and NPK treatments had less than 1% (0 to 0.75%) silking.

### *Anthesis - silking interval as affected by different fertilizer treatments*

There was between 57 % and 80 % plants with silks in all treatments when pollen shedding began. There was no significant difference in the number of tasselled and silked plants as affected by the different soil fertility regimes (Figure 5.11 & Figure 5.12). Synchronization of silking and tasseling is mainly dependent on the genetic make-up of the plant more than any other factors such as soil nutrient status and moisture. Early silk appearance is highly desirable in maize; this ensures that silks receive the first pollen which happens to be highly fertile (Spiertz *et al.*, 2007; Krisna, 2012).

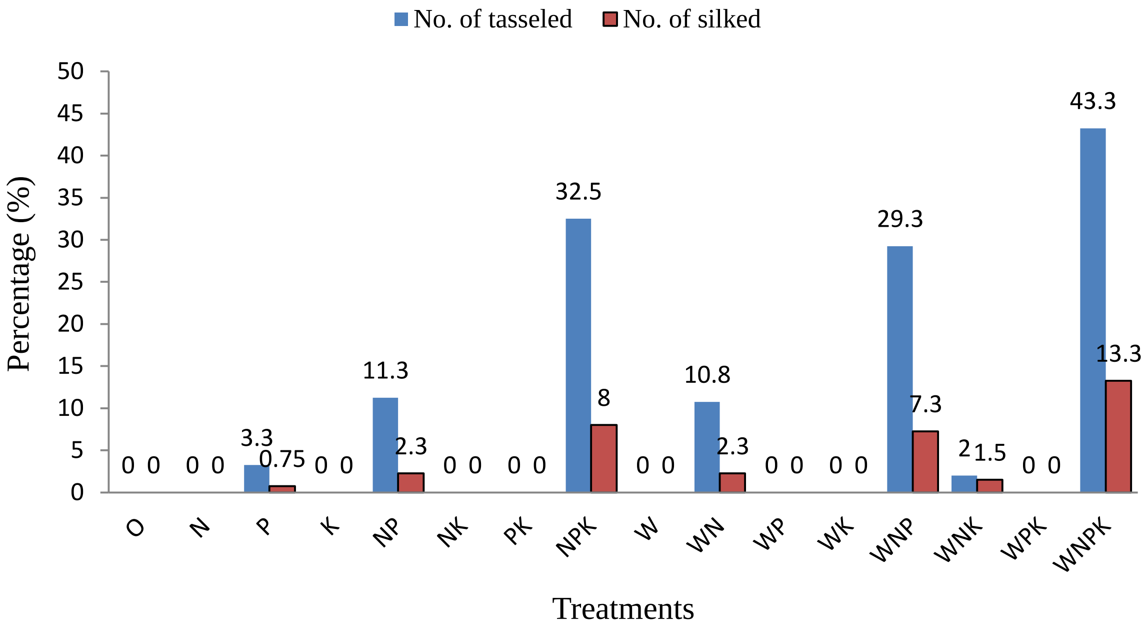

**Figure 5.9:** Tasseling and silking (% of plants) at 9 WAE as influenced by different fertilizer treatments.

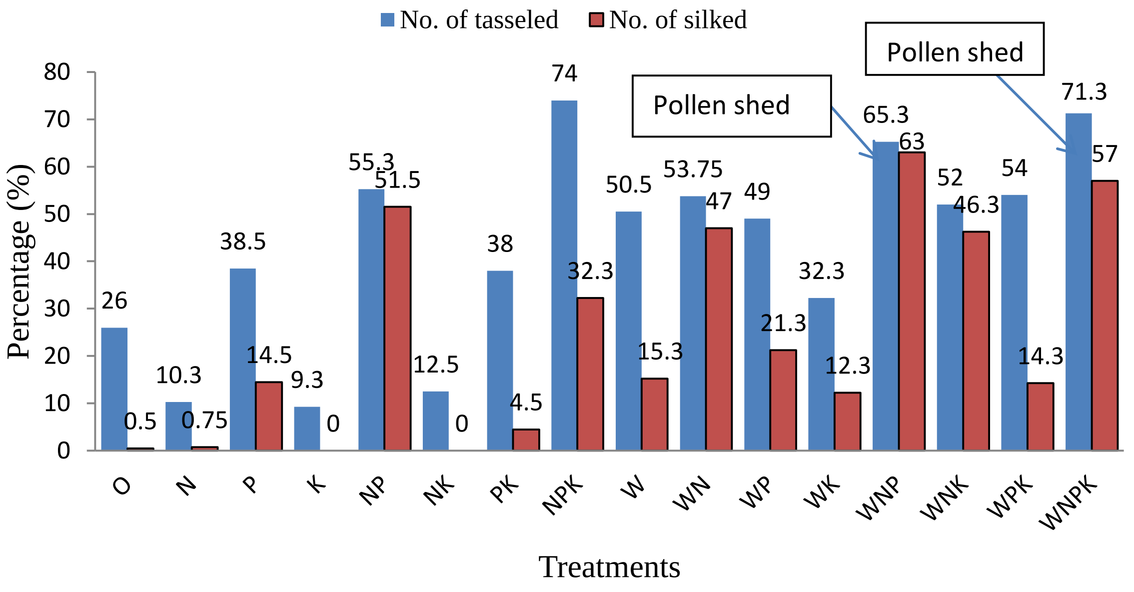

**Figure 5.10:** Tasseling and silking (% of plants) at 10 WAE as influenced by different fertilizer treatments.

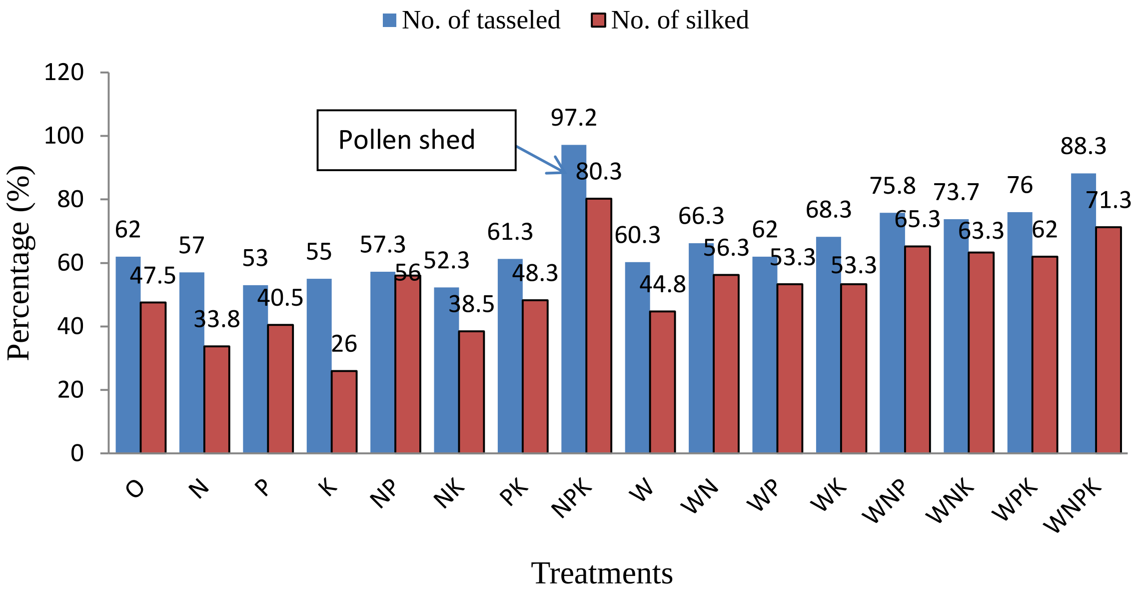

**Figure 5.11:** Tasseling and silking (% of plants) at 11 WAE as influenced by different fertilizer treatments.

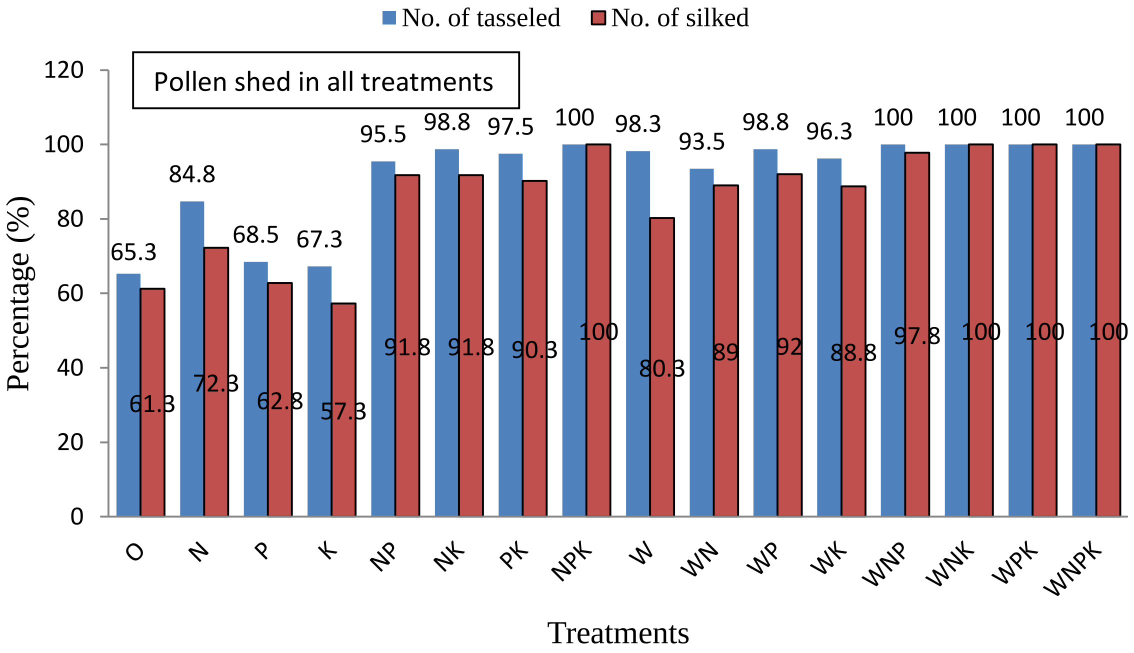

**Figure 5.12:** Tasseling and silking (% of plants) at 12 WAE as influenced by different fertilizer treatments.

## 5.4 CONCLUSIONS

N, P and K fertilizer treatments significantly affected vegetative growth and reproductive development of maize plants. The NPK treatment stimulated vegetative growth and reproductive development compared to treatments receiving only one or two inorganic nutrient elements. Plant height, LAI and total dry mass were always higher in the NPK treatment. The healthy vegetative growth as a result of the NPK fertilizer treatment resulted in an increased size and earlier appearance of maize reproductive structures. The addition of compost for three seasons between 2003 and 2005 resulted in enhanced vegetative and reproductive development compared to the inorganic only counterparts. There was good synchronisation between tassel and silk appearance in all soil fertility regimes. From 9 WAE to 12 WAE nutrient stressed plants lagged behind in flowering; NPK and WNPK had up to 100 % tassels and silks when nutrient stressed plants had less than 80 %.

# CHAPTER 6

# MAIZE POLLEN MASS AND QUALITY AS INFLUENCED BY DIFFERENT FERTILIZER TREATMENTS

## 6.1 INTRODUCTION

A successful grain yield in maize requires the successful transfer of pollen from the male to the female flowers (Delima *et al.*, 2003). A well-formed and fully developed tassel, producing high volumes of pollen, may alleviate inadequacies especially in pollen fertility (viability) (Tranel *et al.*, 2008). Conditions promoting the production of sufficient pollen to fertilize flower ovules are crucial in maize production (Tranel, 2007).

Pollen viability has often been defined as the capacity of pollen to live or to germinate (Lincoln *et al.*, 1982). Soil fertility plays a significant role in ensuring high quality pollen (Harper, 1977). The development of angiosperm pollen is completely dependent on the sporophyte for the provision of nutrients (Vasek *et al.*, 1987). Thus, the growing conditions of the sporophyte (parent plant) will influence the quantity and quality of pollen (Freeman & Vitale, 1985; Schlichting, 1986). Pollen quality is determined by two characteristics; pollen fertility and germinability (Johri & Vasil, 1961). Pollen fertility (viability) is the measure of the survival of normal sperm nuclei, while germinability is regarded as the vitality represented by pollen quantitative measurements like the length of the pollen tube (Jensen, 1964). The pollen viability test is instrumental in determining pollen quality and pollen germination quantifies the actual amount of viable pollen (Parfit & Ganeshan, 1989).

## 6.2 MATERIALS AND METHODS

The general procedures for cultivating maize in the long term fertilizer trial can be found in Chapter 3. Pollen production and quality in the long-term trial was investigated during the 2013/2014 season. One tassel per plot (resulting in four replicates per treatment) was collected for quantity and quality analysis.

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### 6.2.1 Pollen mass

Tassels close to pollen shedding, but not shedding yet, were collected in the field between 7 and 8 am daily. These were taken to the laboratory and placed in beakers with water. Plastic bags were placed over the top of the beakers to avoid any pollen falling into the water (Figure 6.1). White sheets of paper were placed underneath the beakers to collect the pollen. After 24 hours, tassels were slightly shaken to dislodge all the pollen still stuck on the anthers. Then the collected material (Figure 6.2) was sieved, weighed and the pollen stored in air-tight vials.

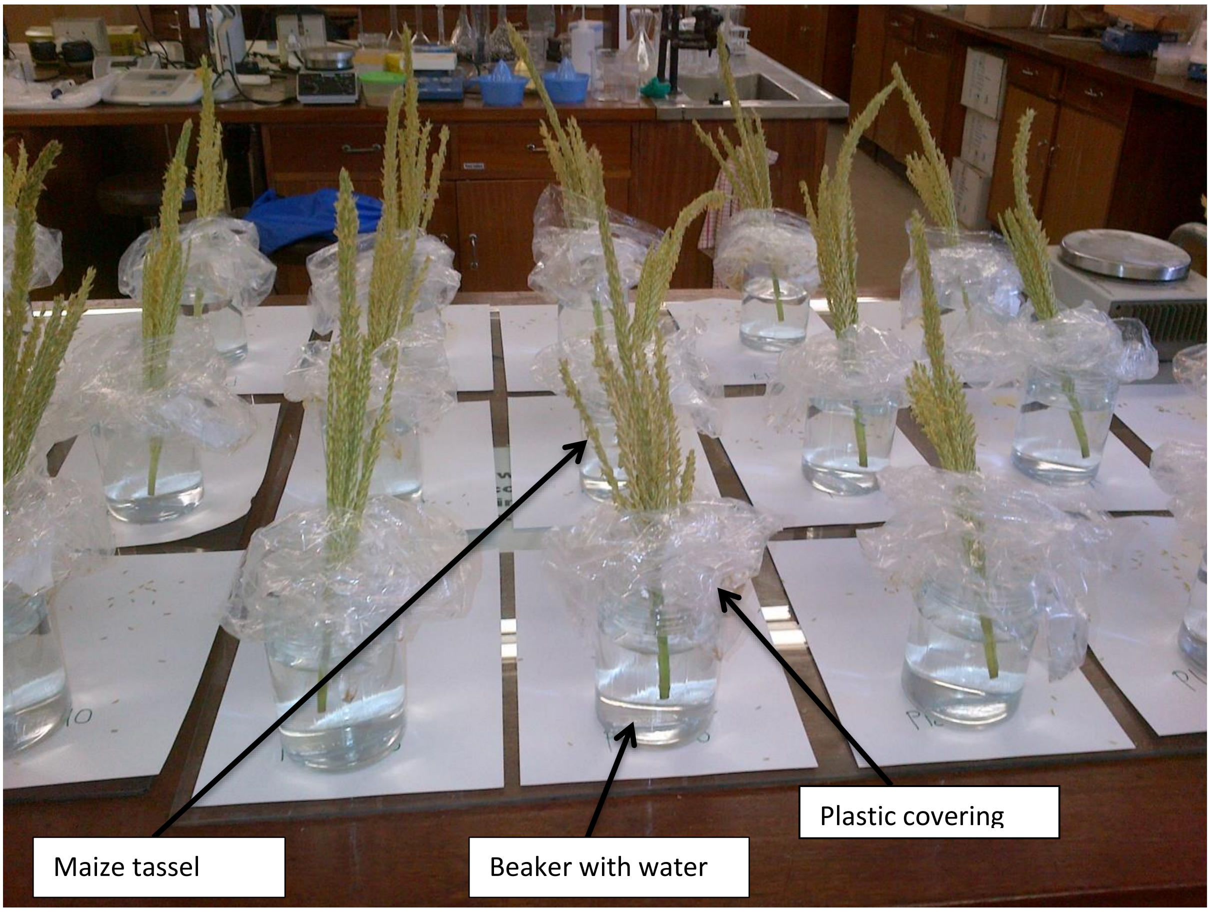

**Figure 6.1:** An illustration of the method used for pollen collection (Dlamini JC, 2014).

**Figure 6.2:** A sample of pollen collected before sieving and weighing (Dlamini JC, 2014).

### 6.2.2 Pollen quality

Pollen quality was determined using two tests; the Alexander's stain method (Oberle & Watson, 1953; Alexander, 1969; Firmage & Dafni, 2001; Pline *et al.*, 2002)), and pollen germination using the hanging-drop method (Werner & Chang, 1981).

*Pollen viability*

A droplet of Alexander's solution (Alexander, 1969) was placed on a glass slide, and then using a sterilized needle, pollen was transferred into the solution. The slides were incubated at 37°C for 30 minutes. Thereafter they were removed from incubation, a cover slip placed over the droplet, and the pollen inspected with a microscope for viability. Pollen grains were considered viable if they turned red, whereas those that remained translucent were regarded as dead (Brewbacker & Kwack, 1963; Werner & Chang, 1981; Shivanna & Rangaswamy, 1992).

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### *Pollen germination*

Pollen germination was determined using the *in-vitro* hanging drop method (Shivanna & Rangaswamy, 1992; Firmage & Dafni, 2001). The germination medium was made up of 10% sucrose solution containing 0.05% boric acid. Each petri dish represented a treatment replicate. A double-folded Whatman no. 42 filter paper was placed at the bottom of each petri dish. A few droplets of the germination medium were used for wetting the filter paper to increase humidity. A droplet of the medium was then placed onto a glass slide; using a sterilized needle pollen was placed into the droplet. Two plastic rods were spaced apart on the wet filter paper. Thereafter, the prepared glass slide was carefully inverted and placed on top of the plastic rods to form a “hanging-drop” (Figure 6.3). Silicone gel was placed on the edges of the petri-dish bottom in order to minimize contaminations and loss of humidity. The petri dishes were left in a well-lit room at room temperature for 24 hours, after which the glass slides were re-inverted and Calberla’s solution (Ogden *et al.*, 1974; Dafni, 1992; Kearns & Inouye, 1993) added to the droplet. A glass cover was then slipped over the droplet and the pollen was studied with a microscope. Pollen was considered to have germinated when the length of the pollen tube was greater or equal to the pollen diameter (Dafni, 1992; Shivanna & Rangaswamy, 1992).

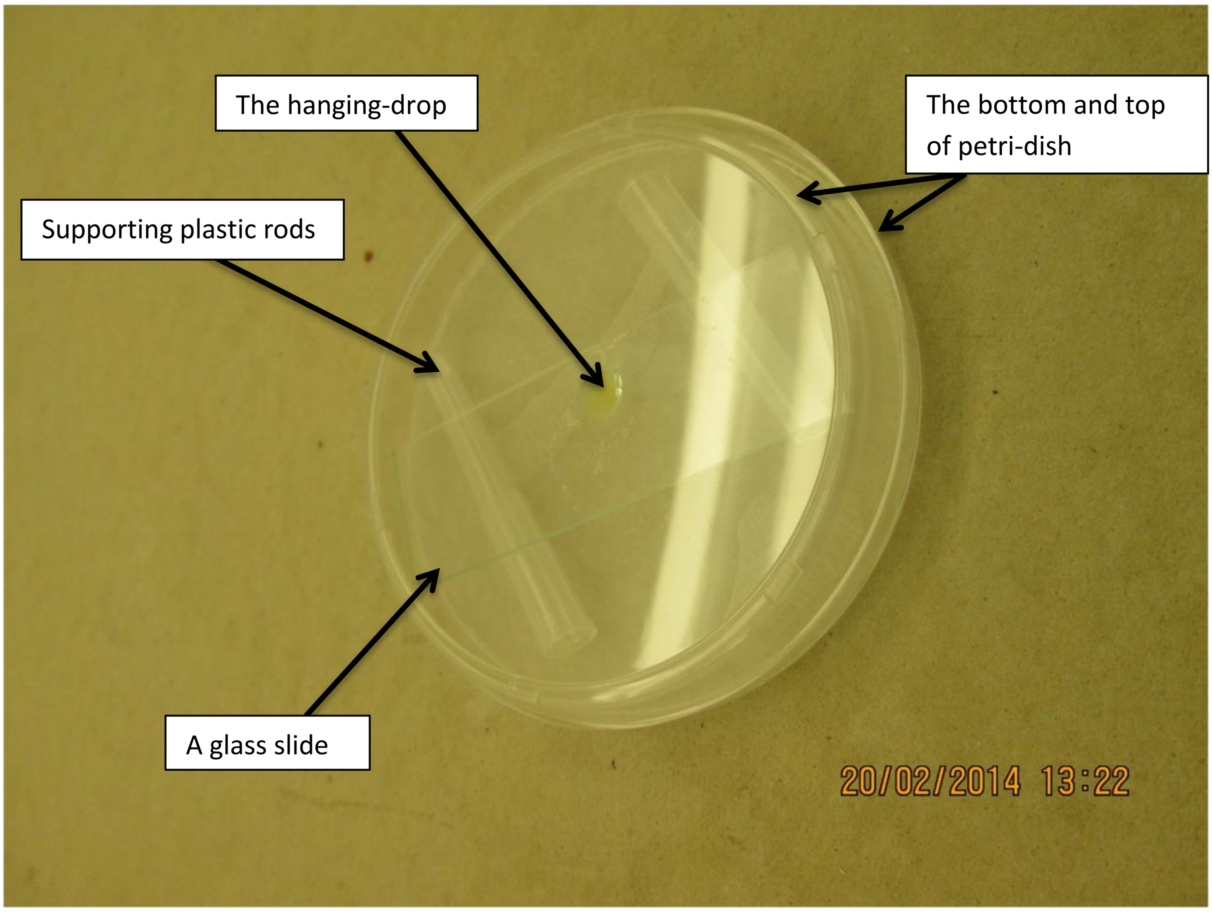


**Figure 6.3:** An illustration of the hanging-drop method (Dlamini JC, 2014).

## 6.3 RESULTS AND DISCUSSION

### 6.3.1 Pollen mass

Tassels from plots treated with N produced the highest pollen mass of 2.8 g (Figure 6.4). This was not significantly different from the other inorganic fertilizer treatments except for NP with a pollen mass of 0.76 g. The residual effect of organic fertilizer (W treatments) resulted in pollen mass ranging between 0.5 and 2.6 g, whilst inorganic fertilizer treatments ranged between 0.7 and 2.8 g. Inorganic treatments, especially where one nutrient element was applied, tended to produce a higher pollen mass than those receiving two or more nutrient elements. The results were in corroboration with findings by Bechoux *et al.*, (2000), Cruden (2000) and Tranel *et al.*, (2008) who observed that under soil nutrient deficiencies, plants tend to produce higher pollen volumes to compensate for inadequacies such as low pollen fertility which are associated with such conditions.

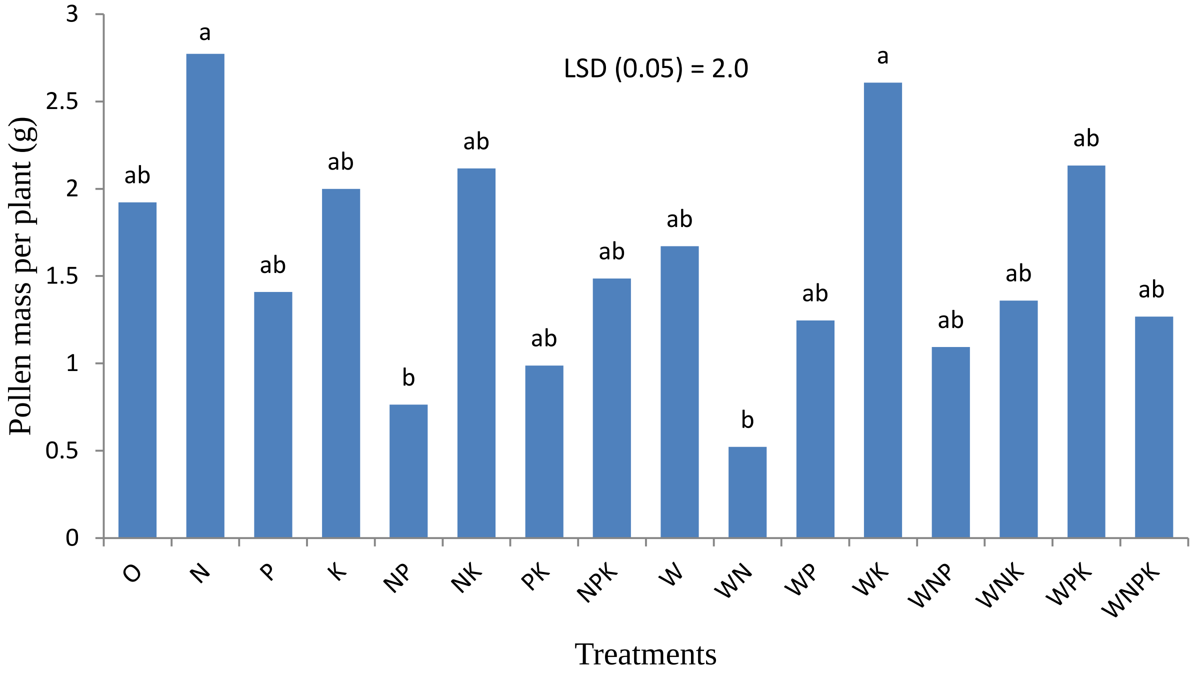


**Figure 6.4:** Maize pollen mass per plant as affected by the different fertilizer treatments.

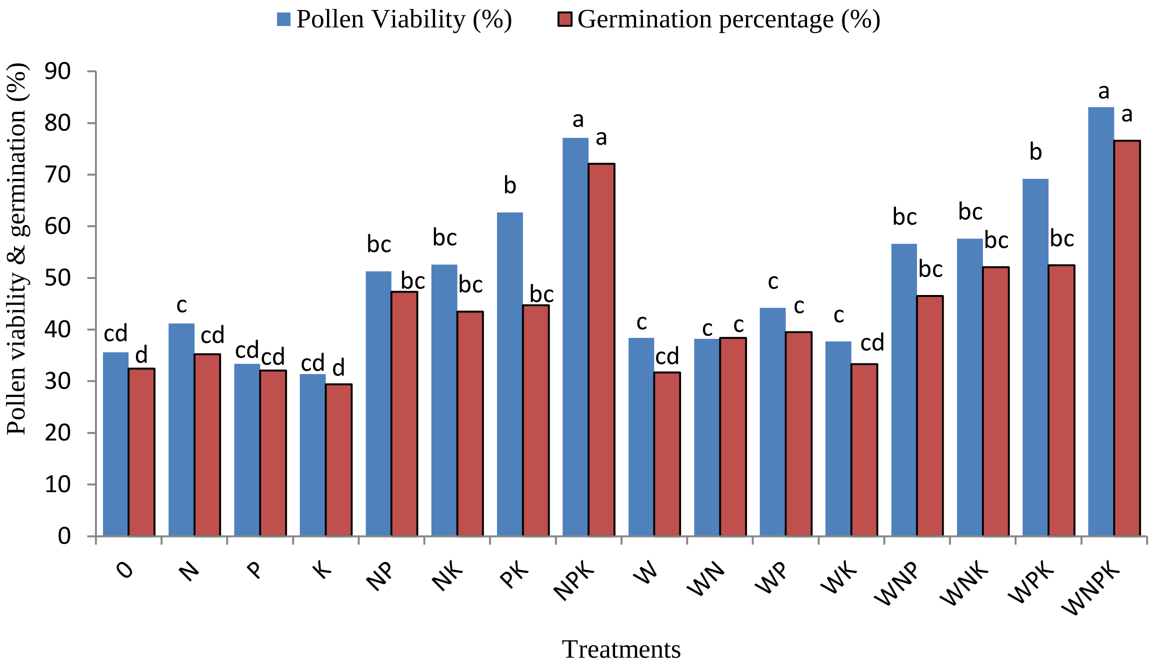
### 6.3.2 Pollen quality

*Pollen viability*

The different soil fertility regimes influenced maize pollen viability. Significantly more viable maize pollen (77.1%) was obtained from the NPK treated plants compared to the other inorganic fertilizer treatments; which ranged between 36 % and 63 %. There was no significant difference between inorganic and the corresponding organic + inorganic fertilizer treatments, although the average viability was somewhat higher for the latter. A deficient soil nutrient status, especially on micronutrients like boron, has been reported to negatively affect flowering, pollen viability, germination, pollen tube growth and seed development (Stosser, 1984; Cakmak & Romheld, 1997). From the results, it is evident that deficient macronutrients can also have a significant negative impact on pollen viability (Figure 6.5). Poor fruit (kernel) set in flowering plants are related to poor pollen characteristics, such as low pollen viability (Pareddy *et al.*, 1989; Eti, 1991). Crop yield can be positively influenced by maintaining conditions favoring high pollen viability (Stanley & Linskens, 1974).

*Pollen germination*

Pollen germination closely reflected pollen viability (Figure 6.5), and was also significantly influenced by the different fertilizer treatments. The NPK treatment resulted in the highest pollen germination percentage of 72.1%. Similarly to pollen viability, there was no significant difference in germination percentage between inorganic and the corresponding organic + inorganic fertilizer treatments. Inorganic treatments resulted in pollen germination percentage ranging between 29.4 and 72.1 %, whilst organic + inorganic fertilizer treatments ranged between 31.7 and 76.6 %. Generally, treatments containing organic + inorganic N resulted in slightly higher germination percentages compared to non N-containing treatments. This could be because additional N from compost had profound effect on pollen performance; (i) Nitrogen influences protein, mRNAs and ribosome concentration of mature pollen (Brewbaker & Kwack, 1963; Tupy, 1982; Willing & Mascarenhas, 1984; Willing *et al*., 1988); (ii) it is a major component of protein macromolecules; (iii) these macromolecules are perceived to be important in pollen germination (Mascarenhas, 1989) and (iv) nitrogen is more often a limiting factor in plant growth and reproductive development (Goh & Haynes, 1986). These then substantiate the possibility that nutrient element deficiencies, especially nitrogen during reproductive development could adversely affect pollen performance (Gunes *et al.*, 2005). The results confirmed findings by Eti (1991), who observed that amongst other factors, pollen germination is highly affected by soil nutrient status.



**Similar letters in bars with similar a colour are not significantly different.*

**Figure 6.5:** Maize pollen viability and germination as influenced by different fertilizer treatments.

*Correlation between pollen viability and pollen germination*

There was a strong positive correlation ($r^2 = 0.9085$) between pollen viability and germination as affected by the different soil fertility regimes (Figure 6.6). Although both pollen viability and germination are highly dependent on various factors such as soil nutrient availability, there is normally a linear relationship between pollen germination capacity and pollen viability in a number of plant species (Grigs *et al*., 1971). Positive relationships between pollen viability and germination are common amongst higher plants growing under different soil nutrient conditions (Norton, 1966; Parfitt & Ganeshan, 1989; Pearson & Harney, 1984).

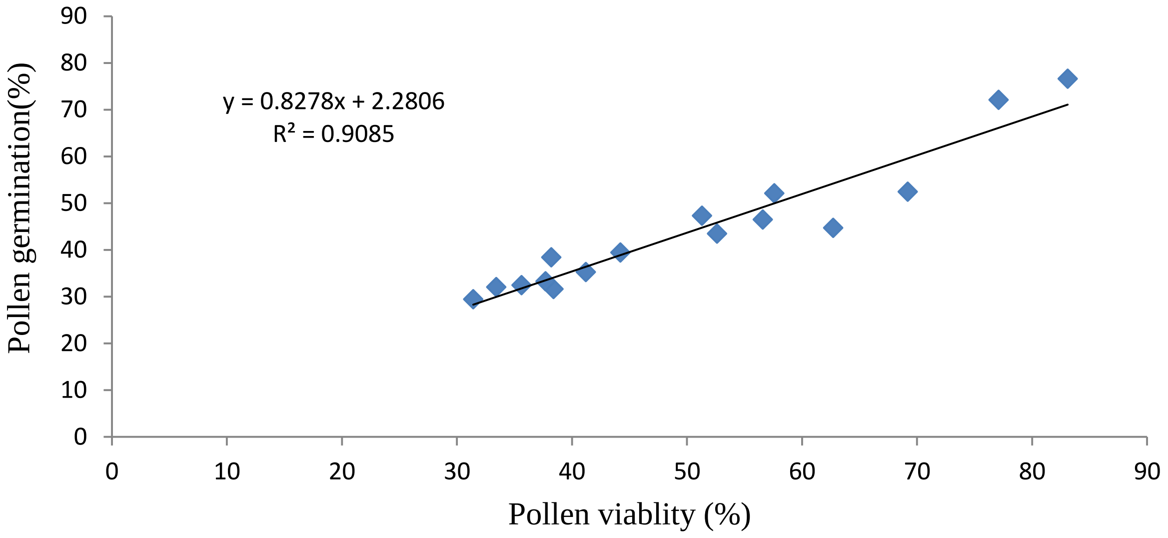


**Figure 6.6:** Relationship between pollen viability and pollen germination as influenced by different fertilizer treatments.

*Other correlations with pollen mass and quality*

A few correlations were tested to quantify the effect of vegetative plant growth on pollen mass and quality as affected by the different soil fertility regimes (Table 6.1). There was a weak positive correlation ($r^2$ = 0.0349) between leaf area index (LAI) and pollen mass. Plants with a low LAI tended to produce a higher pollen mass. Jones and Benton (1930) as well as Uchida (2000) observed that nutrient stress especially on essential elements often result in stunted growth and hence resulting in early maturing plants. Tranel *et al.*, (2008) found that unfavourable environmental conditions induce the production of higher amounts of pollen to compensate for low pollen fertility associated with such conditions. These substantiate the weak positive correlation between LAI and pollen mass. There was also a weak positive correlation ($r^2$ = 0.0034) between LAI and pollen viability.

A negative correlation ($r^2$= 0.0493) was found between pollen mass and pollen viability as affected by the different soil fertility regimes (Table 6.1). The results obtained display that plants growing under inadequate nutrient conditions tended to produce high pollen quantities which were however not as viable. Under nutrient stress conditions, plants produce high pollen quantities to compensate for other pollen inadequacies such as pollen fertility (Tranel *et al.*, 2008). Hence, it could be hypothesized that maize plants that produced less fertile pollen produced it in higher quantities to compensate for the low fertility (low viability) inadequacy of the relevant fertilizer treatments in this experiment.

A very low correlation ($r^2$=0.0084) was found between tassel length and pollen weight (Table 6.1). Tranel *et al.*, (2008) stated that due to surface area, it is expected that longer tassels will produce higher quantities of pollen compared to shorter ones. Goh and Haynes (1986), stated that more than the length of the tassel, pollen quantity is highly dependent on soil nutrient status, especially nitrogen availability. These could then substantiate the low correlation between tassel length and pollen mass which was found in this particular experiment. Tranel *et al.*, (2008) further stated that not only is sufficient pollen quantity required for successful fertilization, but pollen must also be delivered to the female silks while they are still receptive.

**Table 6.1:** Correlations between plant growth parameters and pollen mass and quality.

| Correlation | $R^2$ |
|---|---|
| LAI and pollen weight | 0.0349 |
| Pollen mass and pollen viability | - 0.0493 |
| LAI and pollen viability | 0.0034 |
| Tassel length and pollen mass | 0.0084 |

## 6.4 CONCLUSIONS

Under nutrient stressed soil conditions (0, N, P, K and NK) maize plants tended to produce higher amounts of pollen per tassel. Balanced soil nutrient status (NPK and WNPK) resulted in production of more viable and germinable pollen than in the nutrient deficient treatments. Pollen weight did not influence pollen viability and germinability, while, pollen viability was reflected in its germination capacity.

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# CHAPTER 7

# MAIZE GRAIN PARAMETERS, YIELD AND WATER USE EFFICIENCY AS INFLUENCED BY DIFFERENT FERTILIZER TREATMENTS

## 7.1 INTRODUCTION

In order to obtain and maintain high crop yields under intensive mono-cropping practices, consistent use of fertilizer is crucial (Sharma & Gupta, 1998; Borras *et al*., 2003; Severini *et al*., 2011). Gardner *et al.*, (1985) defined grain yield of maize as the product of variable components including the number of cobs per unit area, the number of grains per cob and the unit grain mass. It is a multifaceted measurable trait that depends on a number of factors that are inherited in a quantitative manner (Hussain *et al.*, 2011). As a quantitative trait, grain yield is significantly influenced by variable environmental factors; which mainly affect kernel row number per cob and kernel number per row (Vasic *et al.*, 2001; Zivanovic *et al.*, 2007; Bovanski *et al.*, 2009). Agronomic practices, such as sustainable soil nutrient management which positively influence any of the grain components, will increase maize yield (Devi *et al.*, 2001; D'Andrea *et al*. 2008).

Water scarcity is one of the key limiting factors of food production under rain fed conditions (Fan *et al.*, 2005). Thus, it is crucial to aim at producing higher food quantities with the available water; improving the ratio of grain yield to total evapotranspiration (ET) per crop per season (Perry *et al*., 2009). In both arid and semi-arid environmental conditions, water stress and nutrient deficiency remain the main limiting factors of primary food production (Lee, 2012). Thus, there is a vital need to shift research focus into identifying means of maximizing crop water-use efficiency under rain fed conditions.

## 7.2 MATERIALS AND METHODS

The general procedures for cultivating maize in the long term fertilizer trial can be found in Chapter 3. In this section, only the materials and methods relevant to this chapter are described.

### 7.2.1 Yield Parameters

Grain yield parameters were quantified at final harvest in both the 2012/2013 and 2013/2014 seasons. These components were collected from the five permanently marked plants situated in the five middle rows of the plot. These included cob length, number of kernel rows per cob, number of kernels per row, mass per kernel and mass of kernels per cob.

### 7.2.2 Grain yield

Grain yield was determined by harvesting from the marked 5m$^2$ sub-plot in both the 2012/2013 and 2013/2014 seasons. The maize was harvested when the plants had turned brown and ears were completely dry. Grain-bearing dry ears were hand-shelled and weighed to obtain the amount of grain yield per plot. Grain yield per treatment was calculated using equation 7.1 suggested by Reddy (2004).

$$Grain\ yield \left(\frac{kg}{ha}\right) = \{Grain\ yield(kg) / \ Area\ harvested\ (5m\ x\ 5m) x\ 10000 \quad (Eq.\ 7.1)$$

Thereafter the results were expressed in tonnes per hectare (ton ha$^{-1}$).

### 7.2.3 Grain yield water use efficiency

Neutron probe access tubes were installed in half of the plots at the beginning of the 2013/2014 season; each treatment was represented by two access tubes. In all the access tubes in the different experimental units, volumetric soil water content was measured every 20 cm up to 120 cm using a neutron water meter Model 503DR CPN Hydroprobe (Campbell Pacific Nuclear, California, USA). Evapotranspiration was measured every week for the duration of the plant growing period. ET was calculated using equation 7.3:

$$\Delta ET(mm) = \Delta S + P + I - R - D \qquad (Eq. 7.2)$$

Where, ΔS (mm) is soil water storage change, P (mm) is precipitation, I (mm) is irrigation rate, R (mm) is surface runoff and D (mm) is deep water percolation. For the duration of the season, precipitation (mm) in form of rainfall was recorded daily using rain gauges which were installed within the plots at the beginning of the 2013/2014 season and also obtained from the automated weather station situated near the field, and the total rainfall received was 329 mm. Soil moisture content was measured weekly using a neutron water meter, in order to determine irrigation water application rate at any point in time. Deep percolation was considered as zero, since only supplementary irrigation was applied during dry spells (no rainfall received for a number of consecutive days). Surface runoff was negligible during the season because the plots have a flat topography and raised dikes (0.2 m) were made on the sides which prevented surface run-off. Water use efficiency was calculated as follows (equation 7.3):

$$WUE\ (kg\ m3) = \frac{Y}{ET} \qquad (Eq. 7.3)$$

Where, Y is grain yield (kg ha$^{-1}$) and ET is the evapotranspiration (mm).

## 7.3 RESULTS AND DISCUSSION

### 7.3.1 Yield Parameters

*Rows per cob*

Nielsen (1995) noted that the number of rows per cob is highly dependent on the genetic make-up of a variety, more than it is influenced by the environmental conditions. In the 2012/2013 season NPK resulted in the highest average number of rows per cob (16.2 rows) amongst the inorganic only fertilizer treatments (Table 7.1). Comparing inorganic and organic + inorganic treatments, WNPK emerged with the highest average number of rows per cob (17.1 rows). This was only significantly different from the control (0) and the N, P or K fertilizer treatments. In the 2013/2014 season, the number of rows per cob was not significantly affected by the different soil fertility regimes (Table 7.2). Despite the lack of significant differences, NPK resulted in the highest average number of rows per cob (15.6

rows) amongst the inorganic fertilizer treatments. Comparing the average number of rows per cob as affected by inorganic and organic + inorganic fertilizer treatments, WNPK obtained the highest (16.9 rows). A lower number of rows were found under soil nutrient deficiencies (notable in treatments 0, N, P and K) which mainly resulted in malformed or relatively smaller cobs.

*Kernels per row*

Andrade *et al.*, (1999) identified the number of kernels per row as one of the main components which directly influence the total grain yield in maize. The different soil fertility regimes significantly influenced the number of kernels per row in both seasons (Tables 7.1 and 7.2). During the 2012/2013 maize season, NPK resulted in the highest average number of kernels per row (42.2 kernels) than any other inorganic fertilizer treatment. This was however, not significantly different from NP (40.3 kernels) (Table 7.1). Comparing inorganic and organic + inorganic fertilizer treatments, NPK maintained the highest average number of kernels per row (42.2 kernels). This was not significantly different from WN (34.7 kernels), WNP (41.3 kernels), WNK (38.9 kernels) and WNPK (39.1 kernels). During the 2013/2014 season, NP had the highest number of kernels per row (41.4 kernels) amongst the inorganic fertilizer treatments. This was however not significantly different from NPK (41.0 kernels), NK (32.8 kernels) and P (34.0 kernels) (Table 7.2). Comparing inorganic and organic + inorganic fertilizer treatments in the same season, WN and WNP emerged with the highest average number of kernels per row (42.1 kernels each).

*Kernels per cob*

In grain crops, kernel number normally results from successive steps that start with the reproductive initiation in the meristems (Bonnet, 1996). The fertilizer treatments significantly influenced the number of kernels per cob in both seasons (Table 7.1 and 7.2). During the 2012/2013 season, NPK resulted in the highest number of kernels per cob (682.5 kernels) amongst the inorganic fertilizer treatments (Table 7.1). Comparing inorganic and organic + inorganic fertilizer treatments, NPK maintained the highest number of kernels per cob (682.4 kernels). This was however, not significantly different from WNP (660.7 kernels), WNK (629.2 kernels) and WNPK (668.7 kernels). In the 2013/2014 season, the NPK treatment produced more kernels per cob (642.3 kernels) compared to any other inorganic fertilizer treatment (Table 7.2). This was however not significantly different from the control (498.4 kernels), P (549.56 kernels), NP (636.38 kernels) and NK (504.19 kernels).

Comparing inorganic and organic + inorganic fertilizer treatments in the 2013/2014 season, WNP resulted in the highest kernel number (674.5 kernels). This was only significantly different from N (415.4 kernels), K (414.0 kernels) and PK (454.4 kernels). Variations in crop grain yield are influenced by the number of kernels per cob (Cox, 1996; DÁndrea *et al.*, 2008), thus, an accurate prediction of the kernel number per cob may be useful in making early estimates of grain yield (Outtar *et al.*, 1987; Otegui & Bonhomme, 1998; Abubenywa *et al.*, 2007). The results were in corroboration with findings by Banziger *et al*., (2002), who observed that a balanced soil nutrient status, especially at flowering (anthesis/silking) stage positively influence kernel number and grain yield in maize.

*Mass per kernel*

Individual kernel mass is one of the most important parameters for the total grain yield in maize (Severini *et al.*, 2011). Kernel mass was not significantly influenced by the different soil fertility regimes in either seasons (Tables 7.1 and 7.2). A higher range of kernel mass (0.285 g to 0.438 g kernel$^{-1}$) was obtained in 2012/2013 compared to the 2013/2014 (0.27 g to 0.39 g kernel$^{-1}$) season. Kernel mass in maize is a result of two kernel growth stages; lag phase (formative stages) and the effective grain-filling phase (Frey, 1981; Madonni *et al.*, 1998). The results were in corroboration with the observations which showed that, a deficient soil nutrient status during plant of growth stages may negatively affect kernel growth in maize (Ouattar *et al*., 1987; Cirilo & Andrade, 1996).

*Mass per cob*

In both seasons, the different fertilizer treatments significantly influenced the cob mass. In the 2012/2013 maize season, NPK resulted in the highest mass per cob (216.9 g cob$^{-1}$) amongst the inorganic fertilizer treatments (Table 7.1). This was not significantly different from cob mass (199.4 g cob$^{-1}$) obtained for the NP treatment. Comparing inorganic and organic + inorganic fertilizer treatments in the same season, WNPK resulted in the highest mass (235 g cob$^{-1}$) (Table 7.1). This was however, not significantly different from NP (199.4 g cob$^{-1}$), NPK (216.9 g cob$^{-1}$), WN (201.9 g cob$^{-1}$), WNP (227.5 g cob$^{-1}$) and WNK (629.2 g cob$^{-1}$) treatments. Similarly to the 2012/2013 season, NPK emerged with the highest cob mass (282.8 g cob$^{-1}$) in the 2013/2014 season (Table7.2). In the 2013/2014 season, NPK maintained the highest cob mass (282.8 g cob$^{-1}$) when comparing inorganic and organic + inorganic fertilizer treatments. This was not significantly different from WN (251.2 g cob$^{-1}$), WNP (232.1 g cob$^{-1}$), WNK (227.5 g cob$^{-1}$) and WNPK (229.2 g cob$^{-1}$) treatments. A slightly higher cob mass range (88 to 282.8 g cob$^{-1}$) was obtained in the 2013/2014 season than the 2012/2013 season (84.6 to 235 g cob$^{-1}$).

*Mass/100 kernels*

The mass of 100 kernels was not significantly affected by the different soil fertility regimes in either seasons (Tables 7.1 and 7.2). Such results were expected, since there were also no significant differences in mass per kernel. The treatments with the highest mass per kernel, especially NPK recorded the highest mass per 100 kernels. The results obtained were in corroboration with findings by Hokmalipour and Darbandi (2011) who observed that NPK fertilizer application has a positive influence on maize seed size and weight. Similarly to mass per kernel, a slightly higher 100 seed mass range (28.75 to 43.75 g 100 seeds$^{-1}$) was obtained in the 2013/2014 season compared to the 2012/2013 season (28.8 g to 38.8 g 100 seeds$^{-1}$).

*Cob length*

Cob length was significantly affected by the different soil fertility regimes. During the 2012/2013 season, NPK and P resulted in the longest cobs (19.2 cm each) than any other inorganic fertilizer treatment (Table 7.1). This was however, only significantly different from K (13.8 cm) and PK (14.2 cm). Comparing inorganic and organic + inorganic fertilizer treatments, NPK and P maintained the longest cobs of 19.2 cm each. They were not significantly different from cob length affected by any of the organic + inorganic treatments. In the 2013/2014 season, P resulted in the longest cobs (19.2 cm) than any other inorganic fertilizer treatment (Table 7.2). This was however only significantly different from K (13.8 cm) and PK (14.1 cm).

**Table 7.1:** Grain yield parameters in the 2012/2013 maize season as influenced by different fertilizer treatments.

| **Treatment** | **Rows/cob** | **Kernels/row** | **Kernels/cob** | **Mass/kernel(g)** | **Mass/cob(g)** | **Mass/100 seeds(g)** | **Cob length (cm)** |
|---|---|---|---|---|---|---|---|
| 0 | 13.5$^{c*}$ | 24.5$^{g}$ | 329.8$^{h}$ | 0.27$^{a}$ | 102.7$^{c}$ | 27.3$^{a}$ | 15.6$^{abc}$ |
| N | 13.6$^{c}$ | 26.6$^{fg}$ | 360.9$^{gh}$ | 0.32$^{a}$ | 114.9$^{c}$ | 31.5$^{a}$ | 14.8$^{abc}$ |
| P | 14.1$^{bc}$ | 28.8$^{efg}$ | 405.9$^{efgh}$ | 0.29$^{a}$ | 102.4$^{c}$ | 28.8$^{a}$ | 19.2$^{a}$ |
| K | 13.7$^{bc}$ | 26.8$^{efg}$ | 365.6$^{fgh}$ | 0.32$^{a}$ | 84.6$^{c}$ | 31.8$^{a}$ | 13.8$^{c}$ |
| NP | 14.9$^{abc}$ | 40.3$^{abc}$ | 599.4$^{abcd}$ | 0.32$^{a}$ | 199.4$^{ab}$ | 31.7$^{a}$ | 17.7$^{abc}$ |
| NK | 15.3$^{abc}$ | 32.8$^{cdef}$ | 504.2$^{cdefg}$ | 0.35$^{a}$ | 130.8$^{c}$ | 34.5$^{a}$ | 16.5$^{abc}$ |
| PK | 14.9$^{abc}$ | 27.8$^{efg}$ | 411.7$^{efgh}$ | 0.30$^{a}$ | 93.1$^{c}$ | 30.0$^{a}$ | 14.2$^{bc}$ |
| NPK | 16.2$^{ab}$ | 42.2$^{a}$ | 682.5$^{a}$ | 0.39$^{a}$ | 216.9$^{a}$ | 38.8$^{a}$ | 19.2$^{a}$ |
| W | 15.3$^{abc}$ | 29.9$^{efg}$ | 456.5$^{defgh}$ | 0.32$^{a}$ | 111.0$^{c}$ | 31.5$^{a}$ | 15.3$^{abc}$ |
| WN | 14.7$^{abc}$ | 34.7$^{abcde}$ | 507.9$^{cdef}$ | 0.35$^{a}$ | 201.9$^{a}$ | 35.3$^{a}$ | 18.9$^{abc}$ |
| WP | 15.1$^{abc}$ | 31.8$^{defg}$ | 480.8$^{defg}$ | 0.29$^{a}$ | 135.4$^{bc}$ | 28.8$^{a}$ | 16.0$^{abc}$ |
| WK | 15.9$^{abc}$ | 33.6$^{bcdef}$ | 534.5$^{bcde}$ | 0.29$^{a}$ | 127.6$^{c}$ | 28.8$^{a}$ | 16.0$^{abc}$ |
| WNP | 16.2$^{ab}$ | 41.3$^{ab}$ | 660.7$^{ab}$ | 0.36$^{a}$ | 232.1$^{a}$ | 36.3$^{a}$ | 17.3$^{abc}$ |
| WNK | 16.2$^{ab}$ | 38.9$^{abcd}$ | 629.2$^{abc}$ | 0.39$^{a}$ | 227.5$^{a}$ | 38.5$^{a}$ | 17.3$^{abc}$ |
| WPK | 15.7$^{abcd}$ | 32.5$^{def}$ | 510.8$^{cdef}$ | 0.31$^{a}$ | 124.4$^{c}$ | 30.5$^{a}$ | 16.5$^{abc}$ |
| WNPK | 17.1$^{a}$ | 39.1$^{abcd}$ | 668.7$^{ab}$ | 0.38$^{a}$ | 235.0$^{a}$ | 37.8$^{a}$ | 18.6$^{ab}$ |
| LSD (0.05) | 2.6 | 7.8 | 145.9 | NS | 65.1 | NS | 4.6 |

*In any given column, means followed by the same letter(s) do not differ significantly at 5% level as per Tukey's test. NS = No significant difference.

**Table 7.2:** Grain yield parameters in the 2013/2014 maize season as influenced by different fertilizer treatments.

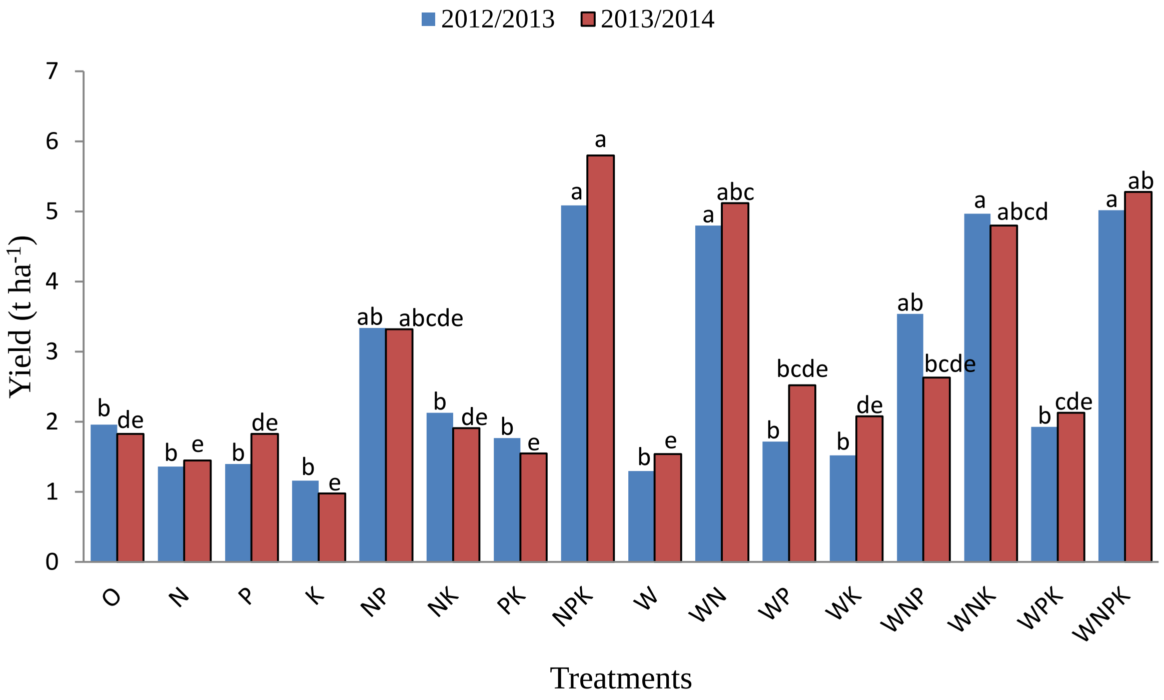
| Treatment | Rows/cob | Kernel/row | Kernel/cob | Mass/kernel(g) | Mass/cob(g) | Mass/100 kernel(g) | Cob length (cm) |
|---|---|---|---|---|---|---|---|
| 0 | $14.3^{a}$ | $30.8^{cd}$ | $498.4^{abc}$ | $0.31^{a}$ | $127.1^{cde}$ | $31.3^{a}$ | $15.7^{ab}$ |
| N | $14.3^{a}$ | $29.3^{d}$ | $415.4^{c}$ | $0.35^{ab}$ | $127.8^{cde}$ | $35.0^{a}$ | $15.3^{ab}$ |
| P | $15.1^{a}$ | $34.0^{abcd}$ | $549.6^{abc}$ | $0.29^{a}$ | $136.5^{cde}$ | $28.8^{a}$ | $19.2^{a}$ |
| K | $15.3^{a}$ | $27.3^{d}$ | $414.0^{c}$ | $0.33^{a}$ | $98.6^{e}$ | $32.5^{a}$ | $13.8^{b}$ |
| NP | $15.4^{a}$ | $41.4^{ab}$ | $636.4^{ab}$ | $0.33^{a}$ | $211.9^{abcd}$ | $32.5^{a}$ | $17.8^{ab}$ |
| NK | $15.3^{a}$ | $32.8^{abcd}$ | $504.2^{abc}$ | $0.39^{a}$ | $145.8^{bcde}$ | $38.8^{a}$ | $15.6^{ab}$ |
| PK | $15.6^{a}$ | $29.2^{d}$ | $454.4^{bc}$ | $0.30^{a}$ | $88.2^{e}$ | $30.0^{a}$ | $14.1^{b}$ |
| NPK | $15.6^{a}$ | $41.0^{ab}$ | $642.3^{ab}$ | $0.44^{a}$ | $282.8^{a}$ | $43.8^{a}$ | $19.2^{a}$ |
| W | $15.6^{a}$ | $32.0^{bcd}$ | $498.8^{abc}$ | $0.31^{a}$ | $118.0^{de}$ | $31.3^{a}$ | $15.3^{ab}$ |
| WN | $15.9^{a}$ | $42.1^{a}$ | $668.6^{a}$ | $0.36^{a}$ | $251.2^{ab}$ | $36.3^{a}$ | $18.2^{ab}$ |
| WP | $15.6^{a}$ | $32.8^{abcd}$ | $510.9^{abc}$ | $0.29^{a}$ | $132.7^{cde}$ | $28.8^{a}$ | $16.0^{ab}$ |
| WK | $16.0^{a}$ | $32.4^{bcd}$ | $518.5^{abc}$ | $0.29^{a}$ | $125.1^{cde}$ | $28.8^{a}$ | $16.0^{ab}$ |
| WNP | $16.0^{a}$ | $42.1^{a}$ | $674.5^{a}$ | $0.36^{a}$ | $232.1^{abc}$ | $36.3^{a}$ | $17.4^{ab}$ |
| WNK | $16.6^{a}$ | $38.9^{abc}$ | $645.3^{a}$ | $0.43^{a}$ | $227.5^{abc}$ | $42.5^{a}$ | $17.8^{ab}$ |
| WPK | $15.4^{a}$ | $33.3^{abcd}$ | $511.1^{abc}$ | $0.31^{a}$ | $110.6^{de}$ | $31.3^{a}$ | $15.9^{ab}$ |
| WNPK | $16.9^{a}$ | $38.9^{abc}$ | $654.8^{a}$ | $0.43^{a}$ | $229.2^{abc}$ | $42.5^{a}$ | $18.4^{ab}$ |
| LSD (0.05) | NS | 9.4 | 188.8 | NS | 25.6 | NS | 4.7 |

*In any given column, means followed by the same letter(s) do not differ significantly at 5% level as per Tukey's test. NS = No significant difference.

### 7.3.2 Grain yield

Grain yield in maize is a product of three components; number of ears per unit area, unit grain weight and the number of kernels per ear (Gardner *et al.*, 1985). Increasing or decreasing any of these components will influence the final grain yield (Devi *et al.*, 2001). Yield was significantly influenced by the different soil fertility regimes in both seasons (Figure 7.1). In the 2012/2013 season, NPK resulted in the highest grain yield of 5.09 ton ha$^{-1}$ amongst the other inorganic fertilizer treatments, except NP. Comparing inorganic and organic + inorganic fertilizer treatments in the same season, NPK was not significantly different from the WN (4.8 ton ha$^{-1}$), WNK (4.97 ton ha$^{-1}$) and WNPK (5.02 ton ha$^{-1}$) treatments. Worth noting is that the residual effect of the compost applied in the 2003, 2004 and 2005 seasons almost doubled the yield in some instances; which could be attributed to the additive value of the residual compost to soil nutrients. Compare for instance the WN treatment which yielded 4.8 ton ha$^{-1}$with N (1.36 ton ha$^{-1}$) and the WNK (4.97 ton ha$^{-1}$) with NK (2.13 ton ha$^{-1}$).

In the 2013/2014 season, NPK resulted in the highest grain yield (5.8 ton ha$^{-1}$) of the inorganic fertilizer treatments. NPK maintained the highest grain yield (5.8 ton ha$^{-1}$) when comparing inorganic and organic + inorganic fertilizer treatments in the 2013/2014 season. This was not significantly different from grain yield in the WN (5.12 ton ha$^{-1}$), WNK (4.8 ton ha$^{-1}$) and WNPK (5.3 ton ha$^{-1}$) treatments. Similarly to the 2012/2013 season, addition of compost onto some inorganic fertilizer treatments stimulated grain yield. Compare for instance WN (5.12 ton ha$^{-1}$) with N (1.45 ton ha$^{-1}$) and WNK (4.8 ton ha$^{-1}$) with NK (1.91 ton ha$^{-1}$) and. These were in corroboration with findings by Bationo, *et al*. (2012) who observed that long-term experiments in Africa have for a long time demonstrated the importance of continuous application of mineral fertilizers in maintaining high maize yields. Fakorede and Mock (1979) observed that numerous physiological and biochemical processes interacting throughout the plant growth processes determine grain yield. Grain yield was relatively higher in the 2013/2014 season compared to the 2012/2013 season; which could have been affected by the higher amount of rainfalls which were received in the latter season.



**Similar letters in bars with similar a colour are not significantly different.*

**Figure 7.1:** Grain yield in the 2012/2013 and 2013/2014 maize seasons as influenced by different fertilizer treatments.

### 7.3.4 Grain yield water-use efficiency

An improved understanding of the interaction between a crop, fertilization and precipitation is essential for efficient utilization of the scarce water resource in crop production (Ahmad *et al.* 2002). This is important in ensuring sustainable food production under rain-fed cropping systems currently threatened by climate change (Fan *et al.* 2005). Water use efficiency was significantly influenced by the different soil fertility regimes. Compared to the other inorganic fertilizer treatments, NPK resulted in the highest water use efficiency of 0.91 kg $m^{-3}$. This was significantly different to all other inorganic fertilizer treatments except for NP (0.5 kg $m^3$). These results showed the significance of ensuring balanced soil nutrient status where the efficient use of water is targeted. Comparing inorganic and organic + inorganic fertilizer treatments, NPK obtained the highest water use efficiency of 0.91 kg $m^{-3}$; which could be owed to the positive influence of balanced soil nutrient status in plant's vigorous growth. This was however, not significantly different from the WNPK (0.85 kg $m^{-3}$), WNK (0.69 kg $m^{-3}$), WN (0.65 kg $m^{-3}$) and NP (0.51 kg $m^{-3}$) treatments. Addition of compost resulted in increased water use efficiency in majority of the treatments compared to where inorganic fertilizer was applied. This implies that the incorporation of compost in a long-term plant nutrition field trial is an effective way of increasing water use efficiency of maize. The residual compost added on to long-term inorganic fertilizer treatments in the experiment added soil nutrients, which enhanced vigorous maize growth and water-use efficiency. The compost also added organic matter content into the soil, which improved the soil water-holding capacity.

**Table 7.3:** Grain water-use efficiency in the 2013/2014 maize season as influenced by different fertilizer treatments.

| **Treatment** | **Yield (kg ha$^{-1}$)** | **ET (mm)** | **WUE (kg m$^{-3}$)** |
|---|---|---|---|
| 0 | 1830$^{de}$ | 641.0$^{ab}$ | 0.28$^{bcd}$ |
| N | 1446$^{e}$ | 608.2$^{b}$ | 0.23$^{cd}$ |
| P | 1826$^{de}$ | 656.2$^{ab}$ | 0.26$^{cd}$ |
| K | 978$^{e}$ | 567.6$^{b}$ | 0.16$^{d}$ |
| NP | 3316$^{abcde}$ | 649.0$^{ab}$ | 0.51$^{abcd}$ |
| NK | 1908$^{de}$ | 623.7$^{ab}$ | 0.31$^{bcd}$ |
| PK | 1548$^{e}$ | 641.9$^{ab}$ | 0.23$^{cd}$ |
| NPK | 5802$^{a}$ | 645.9$^{ab}$ | 0.91$^{a}$ |
| W | 1535$^{e}$ | 691.5$^{ab}$ | 0.21$^{d}$ |
| WN | 5118$^{abc}$ | 787.5$^{a}$ | 0.65$^{abc}$ |
| WP | 2516$^{bcde}$ | 643.1$^{ab}$ | 0.38$^{bcd}$ |
| WK | 2080$^{de}$ | 573.9$^{b}$ | 0.37$^{bcd}$ |
| WNP | 2626$^{bcde}$ | 575.1$^{ab}$ | 0.39$^{bcd}$ |
| WNK | 4802$^{abcd}$ | 651.3$^{ab}$ | 0.69$^{ab}$ |
| WPK | 2130$^{cde}$ | 605.7$^{b}$ | 0.36$^{bcd}$ |
| WNPK | 5276$^{ab}$ | 614.3$^{ab}$ | 0.85$^{a}$ |
| LSD (0.05) | 3002.9 | 177.4 | 0.42 |

## 7.4 CONCLUSIONS

The main factors influencing total grain yield in maize; kernel number per row, kernel mass, kernel number per cob, were negatively influenced by deficient soil nutrient stress. These negatively impacted the total yield under deficient soil fertility regimes. Balanced soil nutrient status (WNPK and NPK) ensured the maintenance of higher yields on both seasons by enhancing the healthy development of the maize grain parameters. The addition of compost into inorganic fertilizer treatments significantly stimulated the amount of grain yield. Inorganic fertilizer had a lower grain parameter and yield range compared to treatments with added compost. Balanced soil nutrient status (WNPK and NPK) resulted in higher WUE compared to where one or two inorganic nutrient elements were deficient. Addition of compost in a long-term plant nutrition trial resulted in higher water-use efficiency in maize.

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# CONCLUSIONS

This study was carried out at the Hatfield Experimental Farm situated at the University of Pretoria. Utilizing the long-term maize nutrition trial the effect of different soil fertility regimes on;

- seed viability,
- maize growth,
- pollen shedding-silk appearance,
- pollen quantity and quality,
- grain components and yield, and
- maize water-use efficiency

was determined. The conclusions made on these objectives were as follows:

In **Chapter 4**, seed viability was investigated *in-vivo* and *in-vitro*. These included a seed viability test on a thermo-gradient table, and emergence count in the field and on a rotating table situated in a glass house. The different soil fertility regimes significantly influenced seed emergence and germination in the different experimental observations. Hybrid seed (OS) and seed obtained from NPK and WNPK treated soils resulted in higher germination index (GI) (up to 5.41) regardless of temperature regime. Hence, farmers storing and planting maize seeds carried over from previous seasons should consider seed from plants not deficient in any major nutrient. Balanced soil nutrient status positively influenced emergence under field conditions. These fertilizer treatments have a positive influence on maize seedling length and dry weight (both shoot and root). However, a higher seed metabolic efficiency was reported for plants receiving P and K only.

In **Chapter 5**, a number of growth analyses were employed to investigate the influence of the different fertilizer treatments on maize growth and development. N, P and K fertilizer treatments significantly affected vegetative growth and reproductive development of maize plants. The NPK treatment stimulated vegetative growth and reproductive development compared to treatments receiving only one or two inorganic nutrient elements. Plant height, LAI and total dry mass were always higher in the NPK treatment. The healthy vegetative growth as a result of the NPK fertilizer treatment resulted in an increased size and earlier appearance of maize reproductive structures. The addition of compost for three seasons between 2003 and 2005 resulted in enhanced vegetative and reproductive development compared to the inorganic only counterparts. There was good synchronisation between tassel and silk appearance in all soil fertility regimes. From 9 WAE to 12 WAE nutrient stressed plants lagged behind in flowering; NPK and WNPK had up to 100 % tassels and silks when nutrient stressed plants had less than 80 %.

In **Chapter 6**, maize pollen quality and mass as influenced by the different soil fertility regimes were investigated. Under nutrient stressed soil conditions (0, N, P, K and NK) maize plants tended to produce higher amounts of pollen per tassel. Balanced soil nutrient status (NPK and WNPK) resulted in production of more viable and germinable pollen than in the nutrient deficient treatments. Pollen weight did not influence pollen viability and germinability, while, pollen viability was reflected in its germination capacity.

In **Chapter 7**, the influence of the different soil fertility regimes on grain parameters and yield was quantified. The main factors influencing total grain yield in maize; kernel number per row, kernel mass, kernel number per cob, were negatively influenced by deficient soil nutrient stress. These negatively impacted the total yield under deficient soil fertility regimes. Balanced soil nutrient status (WNPK and NPK) ensured the maintenance of higher yields on both seasons by enhancing the healthy development of the maize grain parameters. The addition of compost into inorganic fertilizer treatments significantly stimulated the grain yield. Inorganic fertilizer had a lower grain parameter and yield range compared to treatments with added compost. Balanced soil nutrient status (WNPK and NPK) resulted in higher WUE compared to where one or two inorganic nutrient elements were deficient. Addition of compost in a long-term plant nutrition trial resulted in higher water-use efficiency in maize.

# SUMMARY

To investigate the effect of the different soil fertility regimes on maize growth and yield, an experiment was conducted at the Hatfield Experimental Farm situated at the University of Pretoria. The long-term maize nutrition trial was utilized in the 2013/2014 season. It was hypothesized that (i) nutrient deficiencies of N, P or K will negatively affect vegetative development; leaf area index, thus influencing the net assimilates available to the developing grain kernels; (ii) nutrient deficiency stress of N, P or K will negatively affect seed germination; (iii) nutrient deficiencies of N, P or K will negatively influence pollen mass and quantity and (iv) deficiencies in N, P, or K will result in low water-use efficiency in maize.

A set of objectives were set to either accept or reject these hypotheses. These objectives entailed observations on maize seed germination, seedling emergence (*in-vitro* and *in-vivo*), plant vegetative growth (plant height per week, dry mass per plant and LAI), and maize plant reproductive development (cob length, tassel length, number of tassels and silks per week). The influence of the different soil fertility regimes on maize pollen quality (viability and germinability), pollen mass per plant, grain parameters and yield was also observed. Fertilizer treatments applied included inorganic (0, N, P, K, NP, NK, PK and NPK) and organic + inorganic (W, WN, WP, WK, WNP, WNK, WPK, WNPK).

Balanced soil nutrient treatments (NPK and WNPK) ensured vigorous maize seed germination compared to deficient soil nutrient status (0, P, K and N). Balanced treatments (NPK and WNPK) stimulated seed emergence (both rate and percentage), resulting in 100 % emergence in less than a week. There was no significant difference in germination index (GI), germination rate and germination percentage as influenced by both NPK and WNPK. Balanced fertilizer treatments also ensured a good maize crop stand within a shorter space of time.

Plant height, dry mass per plant and LAI increased steadily over time. The balanced soil nutrient treatments influenced a steeper rate of growth compared to deficient soil nutrient status. This highlighted the importance of balanced soil nutrients in the development of a vigorous maize plant. NPK resulted in the longest tassels and ears per plant than any other inorganic only nutrient element. The residual compost enhanced soil nutrients, which

stimulated tassel and cob length per plant. WNPK influenced the early appearance of tassels (90 %). Silk appearance was highly influenced by both NPK and WNPK (76 % each). Balanced soil nutrient treatments ensured improved grain parameters, which in turn positively influenced the total amount grain yield per hectare. Balanced soil nutrient treatments (WNPK and NPK) further resulted in higher WUE compared to where one or two nutrient elements were deficient. The addition of compost into inorganic fertilizer treatments influenced higher water use efficiency in maize.

It could be recommended that future research be focused on:

- The determination of seed quality in terms of it being alive or not.
- A study which will consider accounting for the number of pollen grains per tassel and then correlate it with the pollen mass per tassel.

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# APPENDICES

**Appendix 1:** Analysis of variance for maize seed germination index (GI) as affected by different temperature regimes and the soil nutrient status from which the parent plants were grown compared to hybrid seed in a thermogradient table.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 203 | 371.7405980 | | | |
| Replicate | 3 | 0.2654804 | 0.0884935 | 0.63 | 0.5947 |
| Temperature | 2 | 153.4899539 | 76.7449770 | 549.17 | <.0001 |
| Fertilizer | 16 | 122.1925814 | 7.6370363 | 54.65 | <.0001 |
| Temp. * Fert. | 32 | 74.8304627 | 2.3384520 | 16.73 | <.0001 |
| Error | 150 | 20.9621196 | 0.1397475 | | |

**Appendix 2:** Analysis of variance for seed emergence rate (index) in the field as affected by different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 23.20483059 | | | |
| Treatment | 15 | 20.88749256 | 1.39249950 | 29.07 | <.0001 |
| Rep | 3 | 0.16183060 | 0.05394353 | 1.13 | 0.3486 |
| Error | 45 | 2.15550743 | 0.04790017 | | |

**Appendix 3:** Analysis of variance for maize seedlings emergence percentage in the as affected by different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Squar | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 30767.02415 | | | |
| Treatment | 15 | 29042.88732 | 1936.19249 | 53.65 | <.0001 |
| Rep | 3 | 100.10833 | 33.36944 | 0.92 | 0.4366 |
| Error | 45 | 1624.02850 | 36.08952 | | |

**Appendix 4:** Analysis of variance for emergence rate of maize seedlings as affected by the nutrient status of the soil from which the parent plants were grown.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 41.84488436 | | | |
| Treatment | 15 | 40.20908411 | 2.68060561 | 74.83 | <.0001 |
| Rep | 3 | 0.02377242 | 0.00792414 | 0.22 | 0.8812 |
| Error | 45 | 1.61202783 | 0.03582284 | | |

**Appendix 5:** Analysis of variance for emergence percentage of maize seedlings as affected by the nutrient status of the soil from which the parent plants were grown.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 35213.51300 | | | |
| Treatment | 15 | 33856.03105 | 2257.06874 | 94.71 | <.0001 |
| Rep | 3 | 285.05659 | 95.01886 | 3.99 | 0.0133 |
| Error | 45 | 1072.42536 | 23.83167 | | |

**Appendix 6:** Analysis of variance for maize plant dry mass as affected by different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 216013.3783 | | | |
| Treatment | 15 | 166824.6900 | 11121.6460 | 10.92 | <.0001 |
| Rep | 3 | 3351.4375 | 1117.1458 | 1.10 | 0.3603 |
| Error | 45 | 45837.2508 | 1018.6056 | | |

**Appendix 7:** Analysis of variance for maize plant height as affected by different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 104918.2375 | | | |
| Treatment | 15 | 83256.45750 | 5550.43050 | 14.89 | <.0001 |
| Rep | 3 | 4892.65250 | 1630.88417 | 4.38 | 0.0087 |
| Error | 45 | 16769.1275 | 372.6473 | | |

**Appendix 8:** Analysis of variance for maize leaf area index (LAI) as affected by different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 15842.73919 | | | |
| Treatment | 15 | 11016.53694 | 734.43580 | 7.33 | <.0001 |
| Rep | 3 | 320.10530 | 106.70177 | 1.07 | 0.3732 |
| Error | 45 | 4506.09695 | 100.13549 | | |

**Appendix 9:** Analysis of variance for pollen germination as affected by different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 14654.52772 | | | |
| Treatment | 15 | 11549.60270 | 769.97351 | 11.29 | <.0001 |
| Rep | 3 | 37.00309 | 12.33436 | 0.18 | 0.9088 |
| Error | 45 | 3067.92193 | 68.17604 | | |

**Appendix 10:** Analysis of variance for seedling shoots dry mass as affected by different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 26.32966094 | | | |
| Treatment | 15 | 8.93198594 | 0.59546573 | 1.66 | 0.0945 |
| Rep | 3 | 1.29030469 | 0.43010156 | 1.20 | 0.3200 |
| Error | 45 | 16.10737031 | 0.35794156 | | |

**Appendix 11:** Analysis of variance for seedling roots dry mass as affected by different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 1.40457500 | | | |
| Treatment | 15 | 0.64117500 | 0.04274500 | 2.81 | 0.0037 |
| Rep | 3 | 0.07938750 | 0.02646250 | 1.74 | 0.1722 |
| Error | 45 | 0.68401250 | 0.01520028 | | |

**Appendix 12:** Analysis of variance for shoot to root dry mass ratio as affected by different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 1194.174656 | | | |
| Treatment | 15 | 353.6226602 | 23.5748440 | 1.39 | 0.1918 |
| Rep | 3 | 79.4545615 | 26.4848538 | 1.57 | 0.2108 |
| Error | 45 | 761.097434 | 16.913276 | | |

**Appendix 13:** Analysis of variance for seedling' shoot length as affected by different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 525.4288196 | | | |
| Treatment | 15 | 270.1545141 | 18.0103009 | 3.29 | 0.0010 |
| Rep | 3 | 8.7248264 | 2.9082755 | 0.53 | 0.6635 |
| Error | 45 | 246.5494791 | 5.4788773 | | |

**Appendix 14:** Analysis of variance for seedling root length as affected by different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 381.1001215 | | | |
| Treatment | 15 | 162.0891493 | 10.8059433 | 2.82 | 0.0036 |
| Rep | 3 | 46.7423090 | 15.5807697 | 4.07 | 0.0122 |
| Error | 45 | 172.2686632 | 3.8281925 | | |

**Appendix 15:** Analysis of variance for maize pollen quantity as affected by different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 47.69921381 | | | |
| Treatment | 15 | 24.16997627 | 1.61133175 | 3.28 | 0.0010 |
| Rep | 3 | 1.38902318 | 0.46300773 | 0.94 | 0.4288 |
| Error | 45 | 22.14021436 | 0.49200476 | | |

**Appendix 16:** Number of rows per cob in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 1929.63 | | | |
| Treatments | 15 | 1927.3 | 128.483333 | 29.48 | <.0001 |
| Rep | 3 | 2.3875000 | 0.791667 | 0.18 | 0.9083 |
| Error | 45 | 196.125000 | 4.358333 | | |

**Appendix 17:** Number of kernels per cob in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 524532.4844 | | | |
| Treatment | 15 | 440279.7344 | 29351.9823 | 16.57 | <.0001 |
| Rep | 3 | 4523.7969 | 1507.9323 | 0.85 | 0.4734 |
| Error | 45 | 79728.9531 | 1771.7545 | | |

**Appendix 18:** Number of kernels per cob in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 5.30234961 | | | |
| Treatment | 15 | 4.14212686 | 0.27614179 | 10.75 | <.0001 |
| Rep | 3 | 0.00435767 | 0.00145256 | 0.06 | 0.9821 |
| Error | 45 | 1.15586508 | 0.02568589 | | |

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**Appendix 19:** Cob mass in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 140785.2519 | | | |
| Treatment | 15 | 127330.9333 | 8488.7289 | 31.43 | <.0001 |
| Rep | 3 | 1301.7207 | 433.9069 | 1.61 | 0.2011 |
| Error | 45 | 12152.5979 | 270.0577 | | |

**Appendix 20:** Mass per 100 kernels in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 53023.49609 | | | |
| Treatment | 15 | 41421.26859 | 2761.41791 | 10.75 | <.0001 |
| Rep | 3 | 43.57672 | 14.52557 | 0.06 | 0.9821 |
| Error | 45 | 11558.65078 | 256.85891 | | |

**Appendix 21:** Cob length in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 2655.990000 | | | |
| Treatment | 15 | 2239.920000 | 149.328000 | 17.04 | <.0001 |
| Rep | 3 | 21.635000 | 7.211667 | 0.82 | 0.4882 |
| Error | 45 | 394.435000 | 8.765222 | | |

**Appendix 22:** Number of rows per cob in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 93.96484375 | | | |
| Treatment | 15 | 23.15234375 | 1.54348958 | 0.99 | 0.4773 |
| Rep | 3 | 0.94921875 | 0.31640625 | 0.20 | 0.8932 |
| Error | 45 | 69.86328125 | 1.55251736 | | |

**Appendix 23:** Number of kernels per row in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 2302.357500 | | | |
| Treatment | 15 | 1533.662500 | 102.244167 | 7.64 | <.0001 |
| Rep | 3 | 166.413750 | 55.471250 | 4.14 | 0.0112 |
| Error | 45 | 602.281250 | 13.384028 | | |

**Appendix 24:** Number of kernels per cob in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 771975.1034 | | | |
| Treatment | 15 | 491042.7490 | 32736.1833 | 6.04 | <.0001 |
| Rep | 3 | 36952.5229 | 12317.5076 | 2.27 | 0.0931 |
| Error | 45 | 243979.8314 | 5421.7740 | | |

**Appendix 25:** Mass per kernel in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 0.37250000 | | | |
| Treatment | 15 | 0.15875000 | 0.01058333 | 2.94 | 0.0026 |
| Rep | 3 | 0.05187500 | 0.01729167 | 4.81 | 0.0055 |
| Error | 45 | 0.16187500 | 0.00359722 | | |

**Appendix 26:** Mass per cob in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 341398.2850 | | | |
| Treatment | 15 | 232596.3525 | 15506.4235 | 8.67 | <.0001 |
| Rep | 3 | 28274.0097 | 9424.6699 | 5.27 | 0.0034 |
| Error | 45 | 80527.9228 | 1789.5094 | | |

**Appendix 27:** Mass per 100 kernels in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 3725.000000 | | | |
| Treatment | 15 | 1587.500000 | 105.833333 | 2.94 | 0.0026 |
| Rep | 3 | 518.750000 | 172.916667 | 4.81 | 0.0055 |
| Error | 45 | 1618.750000 | 35.972222 | | |

**Appendix 28:** Cob length in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 353.7473438 | | | |
| Treatment | 15 | 174.1923438 | 11.6128229 | 3.45 | 0.0007 |
| Rep | 3 | 27.9576562 | 9.3192187 | 2.77 | 0.0527 |
| Error | 45 | 151.5973437 | 3.3688299 | | |

**Appendix 29:** Grain yield in the 2012/2013 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 181.6811698 | | | |
| Treatment | 15 | 135.6808058 | 9.0453871 | 9.97 | <.0001 |
| Rep | 3 | 5.1694587 | 1.7231529 | 1.90 | 0.1433 |
| Error | 45 | 40.8309053 | 0.9073535 | | |

**Appendix 30:** Grain yield in the 2013/2014 season as influenced by the different fertilizer treatments.

| Source | DF | Sum of Squares | Mean Square | F-Value | Pr > F |
|---|---|---|---|---|---|
| Total | 63 | 183.6811398 | | | |
| Treatment | 15 | 136.6708058 | 9.0453871 | 9.97 | <.0001 |
| Rep | 3 | 5.1694587 | 1.7231529 | 1.90 | 0.1533 |
| Error | 45 | 40.5309053 | 0.9073535 | | |

**Appendix 31:** Grain water use efficiency (WUE) in the 2013/2014 season as influenced by the different fertilizer treatments.

| **Source** | **DF** | **Sum of Squares** | **Mean Square** | **F-Value** | **Pr > F** |
|---|---|---|---|---|---|
| Total | 63 | 18.27233502 | | | |
| Treatment | 15 | 12.78755906 | 0.85250394 | 8.48 | <.0001 |
| Rep | 3 | 0.96059977 | 0.32019992 | 3.18 | 0.0327 |
| Error | 45 | 4.52417619 | 0.10053725 | | |