

SEMINAL ROOT OF MAIZE VARIETIES IN RELATION TO REDUCTION IN THE SUBSTRATE MOISTURE CONTENT

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Abstract

To study the different moisture regimes on root growth of maize varieties, this experiment was conducted at Institute of Biotechnology and Genetic Engineering (IBGE), Agricultural University, Peshawar Pakistan during summer 2008. Four moisture treatments i.e. favorable (100%), moderate (75%), low (50%) and very low (25%) were prepared with sand soil mixtures of 8:1 (w/w). Ten varieties of maize comprising three hybrids and seven synthetic were compared for the seminal root length and elongation rates. Germinated grains were transplanted in glass boxes (23x16x4 cm) filled with substrate having different moisture contents. Grains were separated from the substrate using black cotton cloth to avoid roots disappearance and/or penetration in substrate for elongation rate measurements. Rate of the elongation was calculated by marking root tips on the glass surface at each day on definite time intervals. The results showed that a reduction in root length was common by decreasing substrate moisture from favorable to any level. However, the decrease in root length was moderate to stronger when substrate's moisture contents decreased from below 75%. Different varieties responded differently to the reduction in soil moisture contents showing that potential did exist in genotype for further improvement e.g. identification of marker genes and its transferring to existing commercial varieties to adopt them better in an environment when sowing was accomplished in hot summer months in soils where loss of moisture was relatively high. Reduction in substrate moisture has shown a decrease in root length which might have an almost similar effect on seedling establishment in early development phase and hence can accumulate in yield losses at maturity.

Introduction

Maize (*Zea mays*) is an important summer cereal of the world. It ranks 1st in world for seed production (Long *et al.*, 2006) and 3rd important crop of Pakistan on area basis under cultivation (Anon., 2008). Unlike other industrial world, maize is an important dual purpose crop of the cropping system in Pakistan yielding both seed for human and fodder for the livestock production. Water stress is limiting factor of production of the tropics and sub-tropics. Losses in yield have been reported if young seedlings have been exposed to drought in the early establishment phase of plant development (Saini & Westgate, 2000). Root growth and development of a young seedling is important for its establishment to compete with weeds. Post-emergence dry spells due to hot summer months in June-July in the area is also common. It has been reported that estimates of yield losses in maize due to post-emergence drought stress are as high as those caused by drought stress at anthesis (Edmeades *et al.*, 1989). Selection of proper variety and/or breeding efforts with desired gene may advantageous for the area to breed the variety. Sensitivity of maize seminal roots to soil moisture has already been reported than any other cereal crop (Sharp *et al.*, 2004). Among the several others putative characters, water status of leaf (Amato and Ritchie, 2002) and root (Malik *et al.*, 2002) are interesting traits to know drought tolerance of the plant. Root is major plant organ affected by stress right after sowing the crop (Stuedle, 2000). A rapid development in maize roots was observed during 1st eight week after sowing (Tardieu, 2005). Development of extensive root system may contribute towards drought tolerance of the genotypes in an environment (Wu and Cosgrove, 2000). Differences have been observed in species for root growth and development (Sahnounne *et al.*, 2004) that lead to evolve high yielding new varieties. The response of maize seedling establishment in early stage of growth is subject

to its root system development which could support water and nutrients uptake for the young seedlings (Craine, 2006; Zuo *et al.*, 2006). It has been reported that root number is usually not affected by water deficiency (Sharp *et al.*, 2004; Malamy, 2005) but the elongation rate and root volume might have a significant effect (Sahnounne, 2004; Sharp *et al.*, 2004; Malamy, 2005). Seminal root growth under decreasing moisture supply of maize is, therefore essential to evaluate for average length and elongation rates to get inputs for breeding program for the sustainable production of this important cereals in the region.

Materials and Methods

Experiment was conducted in controlled conditions during summer 2008 in green house at Institute of Biotechnology and Genetic Engineering (IBGE), Agricultural University Peshawar Pakistan. The growth of maize varieties was compared for seminal root length and elongation rate. The experiment was conducted in locally prepared glass boxes. Substrate (8:1 w/w) of sand and soil was thoroughly mixed (Sahnounne *et al.*, 2004) and oven dried at 105°C. Measured quantity of water was gradually added into the known quantity of oven dried substrate until reaching a point of saturation with no more water drained from the substrate. This quantity of the water absorbed by the substrate was referred as point of saturation and the treatment was termed as saturated capacity (M₁). For the subsequent moisture deficit treatments, the water quantity absorbed by M₁ were reduced by 75, 50 and 25%, hereafter, referred as treatments moderate (M₂), low (M₃) and very low (M₄). Using these four different moisture levels treatments of the substrate, 10 maize varieties (Table 1) were compared for root growth and elongation rates. The study was conducted in a Completely Randomized Design (CRD). Each treatment was replicated six times.

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Table 1. Maize varieties studied for root growth and elongation rate under 4 moisture treatments in soil sand mixture.

Name of variety	Type	Seed source
V1 Azam	OPV	CCRI, Nowshera
V2 Jalal	OPV	CCRI, Nowshera
V3 Pahari	OPV	CCRI, Nowshera
V4 Sarhad white	OPV	CCRI, Nowshera
V5 Local hangu	OPV	Hangu city local
V6 Local karak	OPV	Karak city local
V7 No. 974	Hybrids	Monsanto Pvt. Ltd.
V8 Babar	Hybrids	CCRI, Nowshera
V9 Okmas	OPV	CBFB, Okara
V10 P 3025	Hybrids	Pioneer Pvt. Ltd.

OPV: Open Pollinated Variety

CCRI: Cereal Crop Research Institute, Nowshera.

CBFB: Cattle Breeding Farm Bahaulnagar, Okara

Seminal root length (cm) and elongation rate (cm d⁻¹):

The substrates of the desired moisture treatments were separately prepared in different plastic containers (20 kg capacity) by taking known quantity of soil-sand mixture with added pre-calculated water quantities. CaCl₂ (0.5 g litre⁻¹) was added to extend moisture retention of the substrate for the root growth study duration. The substrate in containers was well shaken for about a week on daily basis and filled in gals-boxes. Meanwhile, grains of maize varieties were washed with 0.25% NaCl solution for 10 minutes for any kind of disinfection, rinsed with distilled water and spread in a glass Petri-dish on filter paper sheets to germinate. Grains were allowed to germinate at 25°C in the growth chamber. The different moisture substrates were filled in glass-boxes (23 x 16 x 4cm). Two germinated seeds with about 5-mm uniform radical length were identified, separated and transplanted in the glass-box. The open top surfaces of the glass boxes were carefully replaced for seed placement and covered with thin paper tape (Toshiba) to prevent unnecessary moisture losses from the glass. Grains from substrate within glass-box were bifurcated using black cotton cloth to avoid root disappearance. This setup has facilitated root length measurement by marking tips of roots on the glass. Measurements were taken for maximum four days after transplanting (DAT) the germinated seeds in box. Boxes were inclined at 50° from ground surface to allow root penetration downward. Periodic measurements on daily basis were made by marking root tip position on the glass using different colors. Thin copper wire was used to measure root lengths of young seedlings from emergence till 4th DAT of a germinated seed in boxes. The root tips of all the seminal roots were matched with their positions in boxes and by measuring length of the wire, total seminal root length was calculated. Root elongation rate was measured from differences between the marked lines on the glass surface against position of the root tips. The values of all marked root tips for 2nd, 3rd and 4th day were calculated accordingly and averaged for a mean daily elongation rate.

All data were subjected to ANOVA using General Linear Model (GLM) procedure of SAS. Comparisons between mean of the treatments and their interaction were separated using Least Significant Difference (p<0.05).

Results and Discussion

Root length (cm): Average length of the seminal roots of the different maize varieties in relation to different moisture treatments is shown in Fig. 1. The seminal root length of maize was decreased (p<0.05) when substrate moisture contents decreased from 100 to 25% (Fig. 1a).

Reduction in the seminal root length of maize was observed moderate (3%) when substrate moisture content decreased from 100 to 75%. A strong reduction of 31% and 38.39% was observed when substrate moisture contents further decreased from 75 to 50% and 50 to 25%, respectively. Different maize varieties were also found different in the seminal root length (Fig. 1b). Variety Azam showed the highest seminal root length (19.07 cm). Variety Jalal (18.83 cm) and Variety Pahari (18.91 cm) did not differ in seminal root lengths from each other. Seminal root lengths of Variety Karak local (18.33 cm), No. 974 (18.32 cm) and P 3025 (18.21 cm) and Sarhad White (17.97 cm) were non-significant from each other. The seminal root length of Hangu local (17.73 cm) and Variety Babar (17.65 cm) was non-significant from each other. Okmas showed the minimum seminal root length (17.22 cm) among the 10 varieties. Interactive effect of the treatments (moisture x varieties) were also found significant (p<0.05) for root length data (Fig. 1c). Root length was observed the highest at favorable moisture (100%) and decreased with reduction in substrate moisture contents for all the varieties. All varieties showed a slight to negligible reduction in length when substrate moisture content decreased from 100% to 75% (moderate). Root length of the varieties decreased at a faster rate when substrate moisture reduced 50% to 25%. This decrease in root length against reduction in the substrate moisture was found positively correlated ($r^2 > 0.90$) for all the varieties and hence the slope of the regression (b) can effectively be used to compare the varieties response against reduction in the substrate moisture contents. Varieties P 3025 and No. 974 showed the highest reduction (b = 0.18), followed by Babar (b = 0.17), Okmas (b = 0.16), Azam (b = 0.15), Jalal (b = 0.14) and Pahari (b = 0.13). Variety Sarhad white did not differ than Hangu local (b = 0.11) for root length decrease against substrate moisture content reduction. Variety Karak local showed the lowest root length reduction (b = 0.10) by decreasing the substrate moisture contents. This slopes of the regressions revealed that varieties with relatively higher 'b' values are susceptible to root length reduction by decreasing moisture contents at the initial establishment phase of a plant growth. The results of this study agree with findings in the literature that seminal root length differed by reduction in the soil moisture contents and/or respond differently for the different genotypes of maize (Malik *et al.*, 2002; Steudle, 2000). Compared to wheat crop, maize has been identified more sensitive to soil moisture reduction (Akmal & Hirasawa, 2004). Literature on root growth also confirms significant interaction of the soil moisture with different genotypes (Sahnoune *et al.*, 2004). Our result has supported the findings of Merah (2001) stating that the substrate moisture reduction from optimum to minimum has a significant effect on root length and elongation rate in maize. From high to moderate reduction the effect was observed but could be quantified towards losses in the seminal roots, as the moisture contents decreased to 50% or higher the effect on root length was pronounced in almost all varieties. On the other hand, local varieties have shown relatively higher resistance to drought because of their small seed size, relatively less plant stature and/or canopy volume. This study also confirmed that variety with relatively smaller seed and plant morphology could stand better under partial drought stress and has potential to identify gene markers for developing a desirable variety for the area having all growth factors in consideration.

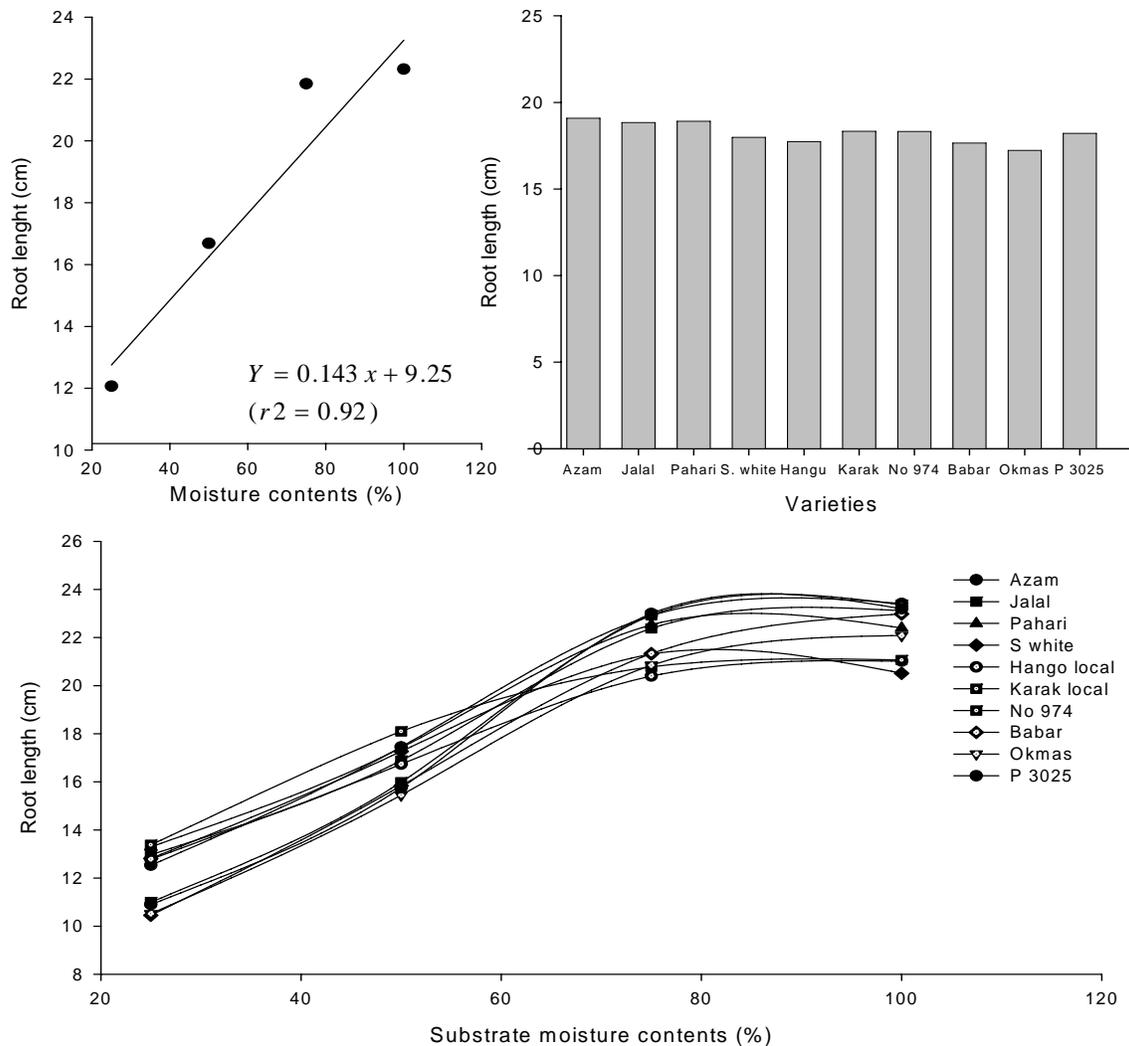


Fig. 1. Seminal root length (cm) of different maize genotypes under four moisture content (%). Different windows of the figure shows mean moisture levels (upper left box), maize varieties (upper right box) and treatment interaction (n = 14).

Root elongation rate (cm day^{-1}): Seminal root elongation rate (RER cm day^{-1}) of different maize varieties was affected by different moisture regimes (Fig. 2). The favorable moisture treatment (100%) showed the highest (6.29 cm day^{-1}) seminal RER, which was significantly decreased to 5.56 cm day^{-1} under moderate treatment (75%), followed by a further significant reduction (4.23 cm day^{-1}) under the low treatment (50%). The minimum (2.21 cm day^{-1}) RER was observed at very low (25%) moisture treatment (Fig. 2a). Maize varieties were also found significantly different in the seminal RER (Fig. 2b). Karak local was the highest (5.03 cm day^{-1}) in seminal RER. Sarhad White (4.66 cm day^{-1}), Azam (4.65 cm day^{-1}), Pahari (4.62 cm), Jalal (4.60 cm day^{-1}), P 3025 (4.55 cm day^{-1}), No. 974, Babar (4.52 cm day^{-1}) and Okmas (4.51 cm day^{-1}) were not different ($p < 0.05$) in RER. The lowest (4.05 cm day^{-1}) RER was observed for Hangu local. The interactive effect of the moisture regimes and varieties were found significant ($p < 0.05$) for seminal RER (Fig. 2c). As compared to 100% moisture treatment, 75% and 50% moisture treatments also responded uniformly for the reduction trend of RER but with relatively inferior

values. However, the decreases in RER against reduction in substrate moisture content were in similar fashion for the different varieties but with differences in magnitude. All the varieties showed a downward trend in RER against decrease in the substrate moisture content. The RER of all varieties adversely affected from moderate to severe when substrate moisture contents were reduced from 100 to 75% and thereafter to either 50% to 25%, respectively. The decrease in RER against reduction in substrate moisture was positive correlated ($r^2 > 0.90$) for all the varieties. Hence slope of the regression (b) equation can be effectively used to compare varieties RER against decrease in the substrate moisture. Among group of the varieties, No 974 showed the highest RER ($b = 0.07$), followed by Jalal, Okmas and Hangu local with almost a similar RER value ($b = 0.06$). Varieties Azam, Pahari, Sarhad white, Babar and P 3025 were found similar in RER ($b = 0.05$). The lowest RER ($b = 0.04$) was observed for variety Karak local. Plant responses to reduction in soil moisture are conspicuously due to changes in the growth rate (Steudle, 2000). However, under drought stress conditions growth of root is not suppressed as

reported for shoot growth (Sharp *et al.*, 2004). An increase in root growth with drought has already been reported (Shimazaki *et al.*, 2005). Increase in root growth might be advantageous to plant in drying soil and is important for seedling establishment which is defenseless to drying soil surface (Akmal & Hirasawa, 2004). Research has proved that root depth and density is important to avoid drought stress by the genotype (Sahnoune *et al.*, 2004). Roots in stress conditions have a substantial capacity for osmotic adjustment in the elongating regions (Serraj & Sinclair, 2002; Akmal & Hirasawa, 2004) that helps to maintain turgor in drought conditions. It has been known that seminal root comprises of two regions the mature region and the elongating region. Growth is result of increase in the elongating region that varies in the same genotype to the availability of soil moisture to the seedling at the time of sowing (Akmal & Hirasawa, 2004). The higher the soil moisture contents the greater would be the elongating zone and hence results the higher growth per unit time which consequence relatively the longer seminal roots. We observed an almost similar trend in elongation rates.

When substrate moisture decreased from 100% to 75%, a mild reduction occurred that became rigorous to further reduction by 50% to 25% moisture contents. Interactive effect of the substrate moisture content and varieties revealed that different varieties expressed clear differences in the elongation rate at relatively low moisture contents. Root length is subsequent cause of the elongation rate. As root elongation rate was high at high moisture contents the longest roots were observed (Fig. 3). We observed that high moisture has resulted higher root growth with relatively the longest root length. As the substrate moisture contents decreased from 100% to 75% a reduction in the ratio of root growth to length was observed which considerably decreased by decreasing the substrate moisture to 25% (Sharp & Davies, 1979). The reduction in root elongation rate in relation to reduction in soil moisture is subject to the root diameter, quantity of osmoticum and cell turgor pressure (Akmal & Hirasawa, 2004). Varieties differ in root elongation rate depending on the root diameter, amount and number of osmoticum and the elongation region.

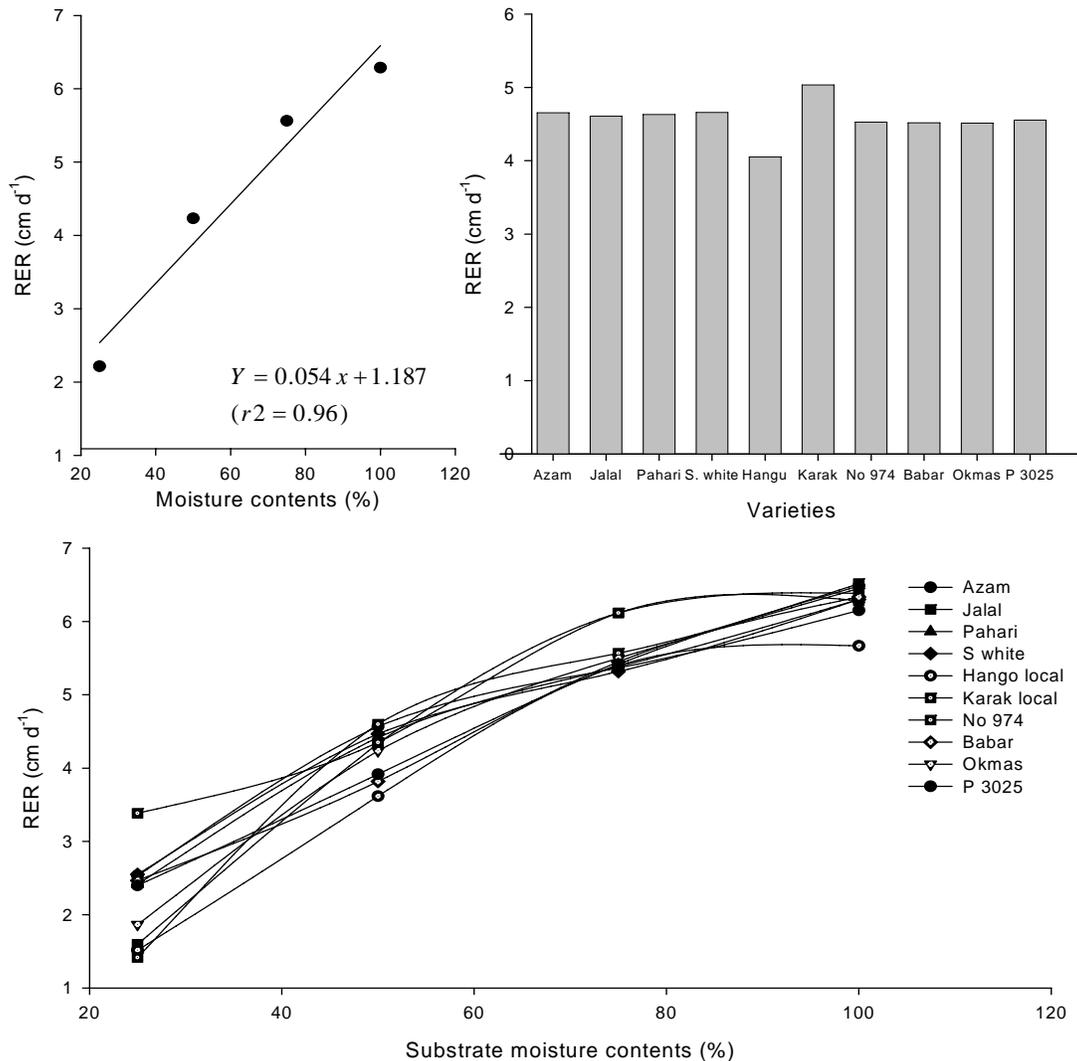


Fig. 2. Seminal root elongation rate (RER cm d⁻¹) of maize genotypes under four moisture contents (%). Different windows of the figure shows mean moisture levels (upper left box), maize varieties (upper right box) and treatment interaction (n = 14).

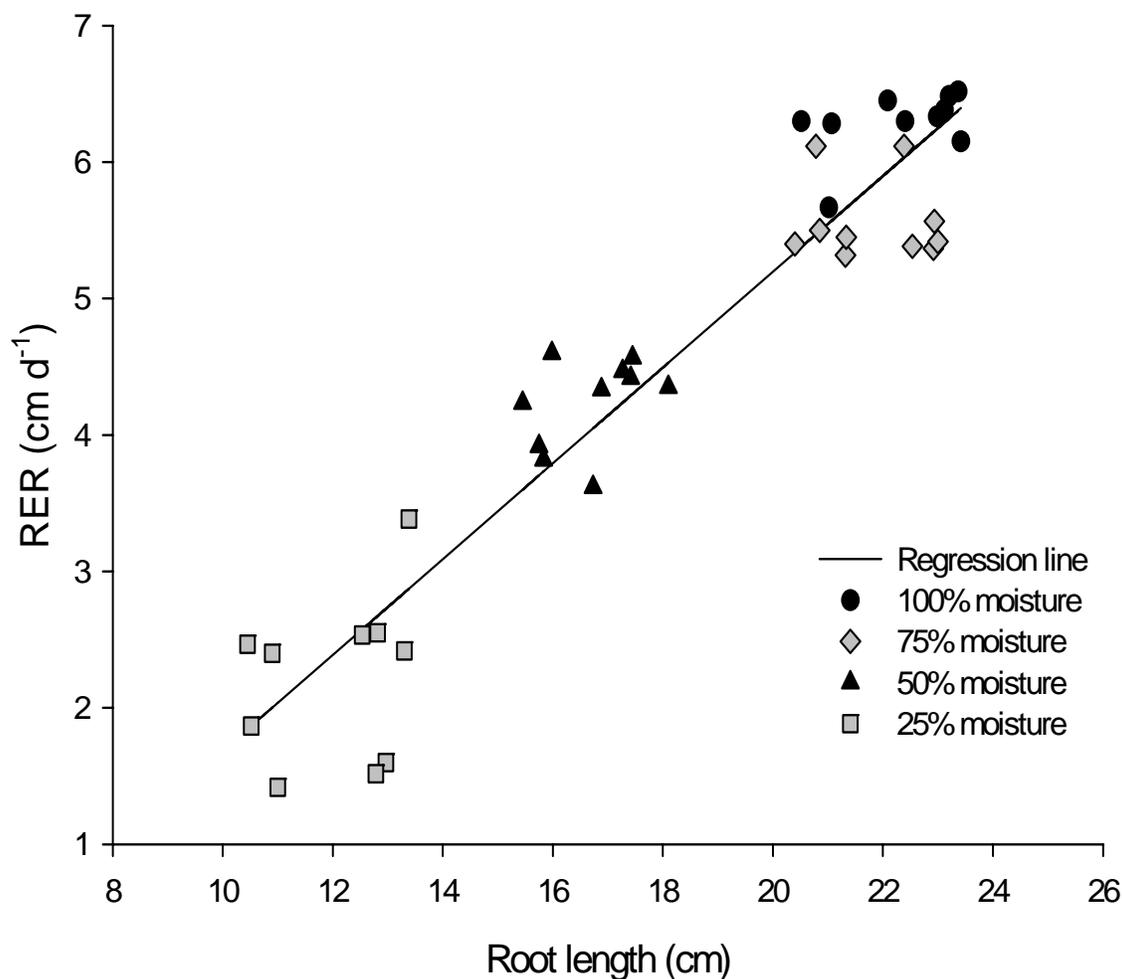


Fig. 3. Relationship of root elongation rate (cm d^{-1}) and seminal root length of maize genotypes under different moisture content. $Y = 0.35x - 1.82$ ($r^2 = 0.90$).

Conclusion

The study suggests that soil moisture is a critical factor affecting germination and seedling. Reduction in the substrate moisture content from field capacity onwards has an adverse effect on root growth and seedling establishment. Losses in the soil moisture from optimum to any extent at sowing time of maize during hot summer month is obvious and might have an adverse effect on the young seedling growth which has a negative effect on plant canopy establishment and finally the seed yield. Different varieties of the genotype responded differently in the different moisture showing that there are certain markers that need to be identified for viable breeding program of the area to develop a variety that could tolerate stress while planting in hot summer months when soil evaporation is expected to be high under the cultural practices and/or technology available for general maize cultivation in the area. Potential does exist in natural biodiversity of the species available for cultivation in an environment but needs to be explored for adoption in yield improvement in area focusing genotypes and environment principals.

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