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Study of root traits of chickpea (*Cicer arietinum* L.) under drought stress

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Roots are among the first defence towards drought with other morpho-physiological and biochemical mechanisms employed by plants. To understand precisely the root traits contribution towards yield, parental chickpea genotypes with well known drought response were field evaluated under drought and optimal irrigation in rain-out shelter. A total of ten genotypes planted in 1.2 m PVC lysimeters were subjected to three water stress levels: high moisture stress, medium water stress, and low water stresses. Root traits, such as root length density, total root dry weight, root dry weight and root: shoot ratio, were measured at 40 days after sowing. The roots were washed and scanned using WinRHIZO software. The ANOVA showed that there was significant difference (P < 0.05) in traits measured amongst test genotypes which included shoot biomass, root biomass, total root length (RL) and root length density (RLD). The results also showed that there were significant variations (P < 0.05) in water regimes and traits decreased with increasing moisture stress from low to high moisture regime. Furthermore, there were variations in root anatomy between the two major chickpea types where majority of the best performing genotypes under low moisture regimes were of the Desi type (e.g. ICC 4958, ICCV 00108, ICCV 92944 and ICCV 92318) as compared to Kabulis which had better and higher response under high moisture regime in this study. These traits could be used for indirect selection for drought tolerance especially in early stages of breeding for drought tolerance which would consequently reduce the cost of multi-location field evaluation in the breeding programs.

Key words: Genotypes, Chickpea *Cicer arietinum* L., drought stress, root traits.

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is the world's third most important grain-legume crop after beans and pea (Food and Agriculture Organisation, 2012). It is particularly an important crop for the farmers mainly living in sub-Saharan Africa (SSA), and south East Asia (SEA). This is because it is a key component in the diets of resource-poor people who cannot afford to supplement their diets with animal protein (International Crops Research Institute for semi-arid Tropics, 2009). In addition, chickpea is also rich in minerals, vitamins, and dietary

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fibre. Globally, total production is approximately 14.2 million tons from an area of 14.8 million ha and a productivity of 0.96 t ha^{-1} (FAOSTAT, 2014). South East Asia, led by India is leading producers, while in East Africa, Ethiopia, Tanzania, Malawi, and Kenya are leading chickpea producers. Worldwide chickpea is largely grown as a rain fed crop (> 90%) in the arid and semi-arid environments in Asia and Africa (Kumar and Abbo, 2001), where the annual rainfall is received during the preceding rainy season (April-September) and the crop grows and matures on a progressively depleting soil moisture profile (Kashiwagi et al., 2013) and generally experiences terminal drought stress (DS).

In many regions of East Africa, chickpea is usually sown during short rains and under stored soil moisture, with very little rainfall during the cropping season; this leads to constantly receding intensities of water deficit as the crop cycle advances, leading to a severe water deficit at crop maturity, reducing yields significantly. These types of receding soil water conditions impose a ceiling on the cropping duration demanding selection for matching duration varieties for the best adaptability and productivity (Saxena, 1987; Ludlow and Muchow, 1990). As a result, terminal drought is considered as the most serious constraint in chickpea production (Pooran et al., 2008). The loss experienced in chickpea production globally due to terminal drought is estimated to be approximately 50% of the potential production (900 million US dollars). In Kenya, however, chickpea production and area under cultivation has fluctuated over the years and it has declined steadily from 51,772 ha in 2000 to only less than 8000 ha in 2016. Similarly, yield per hectare declined from 4.5 to 2.6 t/ha over the same period (FAOSTAT, 2014). The declining production and area are due to drought, pests and diseases, and limited market outlets since the crop is mostly utilized by the Indian community in Kenya (Kimurto et al., 2005: Kosgei, 2015). Therefore there is need of developing drought tolerant genotypes for production in these areas with best adaptability and productivity. Genetic improvement for better drought adaptation can be along-lasting and less-expensive solution for drought management than the agronomic options. But, due to the numerous mechanisms that plants employ to maintain growth under low water supply, understanding yield maintenance under DS becomes increasingly difficult (Tuberosa and Salvi, 2006). Consequently, a trait-based breeding approach is being increasingly emphasized over grain-yield-based breeding for realizing better stability as grain yields are heavily influenced by high genotype × environment (G × E) inter-actions and exhibit low heritability (h^2) (Ludlow and Muchow, 1990). Also, a trait-based breeding increases the probability of crosses resulting in additive gene action (Reynolds et al., 2007; Wasson et al., 2012). However, knowledge of the type and intensity of DS and the various traits and mechanisms employed by the plant to sustain productivity under terminal DS is required

in effective breeding for drought tolerance. This requires knowledge on mechanism such as deep root system, increased partitioning coefficient and conservative water use without reducing the shoot biomass production. Several National and International Consultative Groups on International Agricultural Research (CGIARs) such as International Crops Research Institute for Semi-Arid Tropics (ICRISAT) and International Centre for Tropical Agriculture (CIAT) have breeding programs that have deployed several high throughput phenotyping platforms and strategies to enhance drought tolerance through morpho-physiological and biochemical traits such as root biomass, better water use, canopy temperature depression (CTD), lower leaf development. These have been reported to be associated with drought tolerance in chickpea (Vadez et al., 2012; Nayak, 2010; Kashiwagi et al., 2006).

The impact of various root traits on drought tolerance was found to be high under terminal DS environment, especially in environment where plants solely depend on the stored soil moisture (Ludlow and Muchow, 1990; Kashiwagi et al., 2006; Kashiwagi et al., 2005; Turner et al., 2001; Passioura, 2006; Wasson et al., 2012). Several studies showed that root traits such as deep rooting are related to drought tolerance in chickpea and best genotypes respond by increasing roots deeper in the soil profile (Silim and Saxena, 1993; Benjamin and Nielsen, 2006), common beans (Sponchiando et al., 1989), and soybeans (Kaspar et al., 1978) have enhanced productivity despite low precipitation. In chickpea, Kashiwagi et al. (2006) reported that root development contributes to seed yield under terminal drought conditions as it is noted that root density per se would help in the greater extraction of available soil water. Similar study by Zaman-Allah et al. (2011) showed that in chickpea, there was limited correlation between root length density and yield. In related studies, Kirkegaard et al. (2007) demonstrated through field-based direct root and soil water measurements, that a 30 cm rooting depth increase in root system can capture an extra 10 mm of deep soil water at the grain development stage and result in an extra yield of 500 Kg per hectare. In addition, large root system with greater root prolificacy and rooting depth was shown to influence not only transpiration through soil moisture utilization but also shoot biomass production, harvest index (HI) under terminal DS (Kashiwagi et al., 2006,2013; Zaman-Allah et al., 2011; Purushothaman et al., 2017). But on the contrary, a deeper and more profuse roots alone had been considered not that important for higher grain yields (Vadez et al., 2008) or as a needless biomass partitioning (Passioura, 1983) or as an unnecessary energy loss due to its vigorous respiration compared to the shoot system (Krauss and Deacon, 1994). In cowpeas, more profuse (higher root length density, RLD) and deeper root systems are often viewed as desirable traits for drought adaptation, using a root box method and best cultivars were shown to have a

higher root dry matter per unit of leaf area and a downward movement of roots indicating that they would invest more in deeper rooting for water capture (Matsui and Singh, 2003). In chickpea, greater root density deep into the soil profile and the larger proportion of fine roots compared with field pea and soybean resulted in better exploitation of water stored at lower soil depths (Kashiwagi et al., 2006). In related studies, Saxena (2003) has been using ICC 4958 as a check for root studies due to its greater degree of drought tolerance from its large root traits. Several findings have noted that high root mass has been of concern because the more the roots, the more their efficiency in absorption of water. This gives the plant more advantage in times when less moisture is available in the soil. Krishnamurthy et al. (2003) reported that large root biomass in a mini-core collection of ICRISAT chickpea germplasm had high correlation with drought tolerance and could be used as selection criteria in early generation during breeding.

Improving the resistance of seedlings to water-deficit stress has a two-fold benefit. The first and direct benefit is that it enables crop establishment through withstanding early season drought (Blum 1996; Passioura, 2012) that happens shortly after successful germination. Similarly, Shaxson and Barber (2003) noted that water from precipitation or irrigation can be lost in the form of crop respiration, soil evaporation and percolation into deeper soil layers. The second advantage is that water stress resistance at early stage can also be indicative of resistance at later growth stages (Comas et al., 2013), which makes root evaluation easier. Also plants can re-access the water that has gone into deep percolation only if they have long and vigorous root growth at early stage. However, many researchers warned the need to be cautious in extrapolating early-stage results for later stage resistance unless it is tested and proved in the field (Passioura, 2012; Wasson et al., 2012; Comas et al., 2013). Munns (2011) noted that root system vigour describes the variation in the rate of root growth that results in the capture of greater volumes of soil water and nutrients. Furthermore, a recent study in wheat re-analyzed the implication of root system size and water capture and concluded that, because of the close link between shoot growth and root growth, the development of a large root system might be better suited to environments where the crop depends on in-season rainfall like the Mediterranean environment, whereas under terminal stress conditions in semi-arid tropics of Asia and Africa, a vigorous root system that is linked to a vigorous shoot, would run the risk of a rapid water depletion of the soil profile and eventually a severe stress during reproduction and grain filling (Watt et al., 2005; Liao et al., 2006; Palta et al., 2011). Hence, two recent modelling studies illustrate this idea and a recent review argues that roots need to be looked at with a view to the whole plant and with a view to resource availability in time and space (Lynch, 2007; Sinclair et al., 2010; Vadez et al., 2012; Comas et al., 2013).

In response to this dilemma, many authors have reported that constitutive traits such as deep root system (Manschadi et al., 2006, Lilley and Kirkegaard, 2011), fine roots with small diameters, root length density (Blum, 2010; Comas et al. 2013), leaf rolling, leaf waxy layer and osmotic adjustment (Blum, 2010) are among the frequently studied traits that confer dehydration avoidance mechanism to plants. Blum (2010) furthermore reported that deeper roots allows the crop to access more water, maintain high stomata conductance and hence photosynthesis, and are indicated by cooler canopies. In this study, both root screening under rainout shelter and field screening at arid and semi-arid lands (ASALs) of Baringo County were conducted to confirm and prove the value and contributions of root traits to improving water use and productivity. The objective of the study is to assess the root variation in selected parental chickpea and identify the key root traits that could contribute to enhancing drought tolerance under water stress conditions semi-arid areas of East Africa.

MATERIALS AND METHODS

Site description

Egerton University, Njoro (0° 22'S, 35º 56'E; altitude of 2,238 m above sea level) has a mean day temperatures of 21 °C, and a mean annual rainfall of 900 to 1,020 mm which falls in a bimodal pattern, with long and short rains (Ondieki et al., 2013; Jaetzold and Schimdt, 1983).

Plant material evaluated

Ten parental genotypes were evaluated for root traits which included four released varieties in Kenya: Chania Desi 1 (ICCV 97105), LTD 068 (ICCV 00108), Chania Desi 2 (ICCV 92944) and a local germplasm commonly referred to as Ngara local. Three advanced lines (ICCV 92318, ICCV 97306 and ICC 3325), two susceptible checks (ICC 283 and ICC 1882) with poor rooting characteristics and ICC 4958 was used as the tolerant check due to its prolific and large root properties (Saxena, 2003). Yield data from field evaluations earlier conducted was included in the study. Table 1 describes the status of the tested plant materials.

Experiment description

The experiment was conducted in Egerton University Field 7 Research Station under rain-out shelter May/September 2013/2014 seasons and a second experiment was conducted during November, 2013/January, 2014 season. The experiment was set in Polyvinyl chloride (PVC) cylinders measuring 120 cm long and 20 cm diameter under rain out shelter. The cylinders were placed in 1.2 m deep cement pits with a spacing of 0.05 m between cylinders, giving a planting density of 20 plants m^{-2} and they were arranged in Randomized Completely Block Design (RCBD) in three replicates. The cylinders were filled with an equal mixture (w/w) of mollic-andosols (forest soil) and sand. The sand was used to decrease the soil bulk density and facilitate root growth and subsequent root extraction. Two seeds of each genotype were sown in the cylinder

Table 1. The status of the tested plant materials.

S/N	Genotype	Type	Status
1	Egerton Chania Desi1 (ICCV 97105)	Desi	Commercial check
2	Leldet 068 (ICCV 00108)	Desi	Commercial check
3	Egerton Chania Desi 2 (ICCV 92944)	Desi	Commercial check
4	ICCV 92318	Kabuli	Advanced breeding lines
5	ICC 4958	Desi	Drought tolerant check (High root length)
6	ICCV 97306	Kabuli	Advanced breeding lines
7	ICC 3325	Desi	Breeding line
8	ICC 283	Desi	Susceptible Breeding line
9	ICC 1882	Kabuli	Susceptible line (Low root length)
10	Ngara local	Desi	Tolerant local accession
11	CAVIR	Kabuli	Spanish Tolerant variety

and irrigated with 2,000 ml water uniformly to achieve uniform emergence. At 14 days after sowing (DAS) water stress treatment was imposed and one seedling was thinned out. There were three water regimes which were imposed: high moisture (75% of near field capacity - FC), medium moisture (50% of near field capacity) and low moisture (25% of near field capacity). This was maintained till 40 DAS (end of vegetative growth). Every two alternate days, 1.5, 1.0, and 0.5 litres of water were used to replenish the high, medium and low moisture levels respectively. Initial calibration of the soil water to be used was done before planting to determine the water holding capacity which ranged between 0.28 to 0.48 cm^3 cm^{-3} lower limit-upper limit respectively for the 0 - 120 cm PVC pipe soil layer and the volume of the water added each time (Ooro et al., 2003; Kimurto et al., 2005). Weeding was done by physically uprooting weedy species once they had emerged.

Measurements on root and shoot traits

Roots were extracted from the PVC pipes by gently washing out the soil particles and other debris at the age of 35 days after sowing (DAS) from the lower end of pipe. When approximately three quarters of the soil–sand mixture was washed away, the cylinders were erected gently on a 2 mm sieve so that the entire root system could be removed. The extracted root system was mostly in one piece with very few small segments of detached roots trapped by the 5 mm sieve. The roots were thoroughly cleaned, separated from the organic debris and straightened by repeated dipping and rising in buckets of clear water, then floating the sample material on water in trays. The entire process was repeated for all the tubes and the roots were separated from the above ground biomass by cutting at the cotyledonary point and put in paper bags for oven for drying to a constant weight as earlier described (Purushothaman et al., 2017). Recovered roots were suspended in a transparent tray with 2 - 3 mm film of water for easy dispersion of roots before scanning. The root system was divided into segments of 15 cm which were placed in the scanning trays. Each root sample was measured using the image analysis system (Win-Rhizo, Regent Instruments INC., Quebec, Canada) following the methodology previously described by Serraj et al. (2004). The roots were kept for oven drying at 70°C for 72 h (to constant weight). The following traits were measured: 1) Shoot dry weight (SDW) (g) – Shoots separated from roots were oven dried at 800°C for 72 h and their weights recorded. The SDW was used as an indicator of plant growth vigour; 2) Root dry weight (RDW) (g) – Scanned roots were oven dried at 80 °C for 72 h and their weights recorded. The RDW was used as an indicator for drought tolerance; 3) Root: shoot ratio (R:S) was calculated using

root and shoot dry weights which was calculated as the ratio of roots dry weight to shoot dry weight; 4) Total dry weight (TDW) (g). This was calculated by combining the SDW and RDW; 5) Total rooting length (TRL) (cm) was measured using an image analysis system (WinRhizo, Regent Instruments Inc., Canada); 6) Specific root length (SRL) was determined by diving root length over root dry mass (RDM) in Mg^{-1} dry (RL/RDM), and 7) Root length density (RLD) was calculated as earlier described by Zaman-Allah et al. (2011) as RLD (cmcm^{-3}) = Length of roots (cm)/volume of soil core (cm3). The soil volume was calculated using the following mathematical expression:

Soil volume= π.r2xh,

Where; π = 3.14; r = Soil core inner radius (20 cm PVC pipe); h = Sub-core height (120 cm).

Data analysis

Data analysis was performed by GenStat (14th edition) statistical software. The means were separated by least significant difference at P < 0.05. The following statistical model was used:

$$Y_{ijk} = \mu + G_i + S_l + R_j + B_{(kj)} + GS_{il} + \varepsilon_{ijk}$$

Where: Y_{ijk} = observations; μ = mean of the experiment; G_i = effect of the ith genotype; S_l = effect of lth season; R_j = effect of the jth replicate; $B_{(jk)}$ = effect of the kth in complete block within the jth replicate; GS_{il} = effect of ith genotype in l^{th} season and ε_{ijk} = experimental error. The least significant difference was determined at P < 0.05.

RESULTS

Effects of water treatments on root traits for test genotypes under rain shelter

The results for combined analysis of measured root traits showed that there were significant differences (P < 0.05) in the genotype and water treatments (25% low, 50%

Table 2. Mean squares for crop morpho-physiological traits linked to drought tolerance traits under various watering regimes at Egerton research station for season I and II 2014 season.

Source of variation	d.f.	SDW		RDW		TDW	
		SI	SII	SI	SII	SI	SII
G	9	1.90**	2.06***	0.22***	0.08	2.22**	2.69***
WT	2	3.1	0.97	0.28**	0.03	4.95**	0.96
G×WT	18	1.2	0.3	0.41***	0.02	2.22***	0.41
Season							
Error	58	0.59	0.37	0.05	0.03	0.73	0.48
Total	87						
CV%		13.7	9.3	2.4	7.2	10.4	8
l.s.d.0.05 G		0.72	0.58	0.2	0.16	0.8062	0.6562
l.s.d.0.05 WT		0.4	0.32	0.11	0.09	0.4416	0.3594
l.s.d.0.05 G×WT		1.25	1.01	0.35	0.28	1.3964	1.1366

Source of variation	d.f.	R:S		TRL		RLD	
		SI	SII	SI	SII	SI	SII
G	9	22.69*	15.72*	239898*	608663**	0.02*	0.07*
WT	2	9.69*	5.48*	233838*	62334*	0.02*	0.01*
G.WT	18	42.07	7.9	540100	173196	0.05	0.02
Season							
Error	58	12.7	14.86	324615	319953	0.03	0.033
Total	87						
CV%		20	17.4	11.2	12.8	11.2	12.9
l.s.d.0.05 G		7.472	4.705	537.6	533.8	0.02	0.02
l.s.d.0.05WT		4.092	2.577	294.5	292.3	0.01	0.01
l.s.d.0.05 G×WT		12.941	8.149	931.2	924.5	0.03	0.03

Level of significance: ***- 0.001, **- 0.05 and *-0.01, d.f.- degrees of freedom, SI- the first season, SII- the second season, SDW- Shoot dry weight, RDW- Root dry weight, TDW-total dry weight, R:S- Root: to shoot ratio, TRL- Total root length, RLD- Root length density, WT- water treatment, G- Genotype.

medium, and 75% high) (Table 2). Genotype and the interactions between genotype and water treatments and genotype and season affected all the root traits of tested chickpea germplasm. Most of these traits varied significantly amongst test genotypes. The significance of the main effects of genotype (G), water treatment (WT), and genotype × water treatment interaction (GWT) were measured at P < 0.05. The presence of G×S and G×WT for the traits indicated that the output of the traits varied across the seasons and moisture treatment (Table 2).

Effects of water regimes on shoot dry weight (SDW) among test chickpea genotypes

The overall means for each moisture treatment (low to high) across seasons (I & II) showed that drought stress (DS) reduced the shoot dry weight (Table 3). The interaction between water regimes and chickpea genotypes was significant (P < 0.05) on the effect of

shoot biomass accumulation over growing period (Table 3). Overall, moisture stress reduced SDW by 66% under low moisture as compared to high moisture treatment in season I (2013) and by 71% in season II (2013/14) due to the early stage rainfall (long rainfall season) (data not provided) that could have raised RH and delayed stress built up in the rain-out shelter. Overall genotypes varied significantly in SDW both in 2013 and 2013/2014 (Table 3). The overall mean SDW for both seasons combined varied from 0.86 - 0.87 g (ICC 1882 and ICC 283, respectively) to 1.84 - 2.24 g (ICC 4958, ICCV 97306, and ICCV 92318, respectively). In season I it ranged from 0.90 g (ICC 283) to 2.24 g (ICCV 92318) as compared to 0.84 to 2.18 g in season II. There was variation from 0.34 g per plant (ICCV 92318) under low water regime to 2.88 g per plant (ICCV 92318) under high watering regime. Overall the mean SDW in the second season was 1.49 g which was lower than 1.57 g recorded in the season I.

On average, genotype ICCV 92318 attained the highest SDW in season I and season II (2.29 and 2.18 g,

Table 3. Combined means of shoot dry weight (g) under varying watering regimes for season I and II (2013/2014).

Genotype	Season I				Season II				Overall mean
	Low moist	Medium moist	High moist	Mean	Low moist	Medium moist	High moist	Mean	
ICCV 92944	1.52	1.97	2.01	1.83	1.24	1.61	2.06	1.64	1.74
ICCV 00108	1.51	1.61	1.85	1.66	1.48	1.54	1.71	1.58	1.62
ICCV 97105	1.28	1.53	2.05	1.62	1.28	1.45	1.97	1.57	1.59
ICC 4958	1.41	1.97	1.95	1.78	1.66	1.89	2.18	1.91	1.84
ICCV 97306	1.43	2.09	2.82	2.11	1.36	1.79	2.74	1.96	2.04
ICCV 92318	1.04	2.73	3.09	2.29	1.14	2.53	2.88	2.18	2.24
Ngara Local	0.47	1.17	1.64	1.09	0.34	1.19	1.4	0.98	1.04
ICC 1882	0.84	0.91	1.04	0.93	0.53	0.98	1.08	0.86	0.90
ICC 283	0.77	0.85	1.09	0.90	0.77	0.85	0.91	0.84	0.87
ICC 3325	1.28	1.41	1.74	1.48	0.98	1.24	1.56	1.40	1.44
Mean	1.16	1.62	1.93	1.57	1.08	1.51	1.85	1.49	1.53
CV%				10.2				9.3	
l.s.d.0.05 G	*	*	**		**	**	**		
l.s.d.0.05 WT	*	*	*		*	**	*		
l.s.d.0.05 G×WT	*	*	*		*	*	*		

Level of significance ***- 0.001, **- 0.05 and *-0.01, SI- the first season, SII- the second season, G- Genotype, WT- water treatment, G×WT- Genotype x water regime interaction; Moist- Moisture level.

respectively). Genotype ICC 283 and ICC 1882 attained the lowest shoot biomass in both seasons (0.87 - 0.98 g). Under medium and high moisture regimes, SDW was greater by 39.6 and 18.4% (season I) and 40.0 and 71% (season II), respectively, than low moisture regime. On average in both seasons combined, drought tolerant check (ICC 4958) had 101.71 and 111.30% more SDW than susceptible genotype checks ICC 1882 and ICC 283, respectively.

Regardless of moisture, on average, genotypes ICCV 92318, ICC 97306 and ICCV 92944 had 28.6, 2.7 and 2.6% higher SDW than drought tolerant check (ICC 4958) in season I. In season II, ICCV 92318 and ICCV 97306 respectively recorded 21.7 and 10.8% higher SDW than drought tolerant check (ICC 4958) (Figure 1).

On average, in both seasons SDW increased with the increase of the water moisture from 25 to 75% FC (low to high moisture regime). For example, genotype ICCV 92318 had increasing SDW with the increase of moisture. For season I the genotype recorded shoot biomass of 1.04, 2.73 and 3.09 g in the low, medium and high moisture regimes, respectively, in season I as compared to 1.14g, 2.53 g and 2.88 g in the second season respectively. This indicates that ICCV 92318 had better response (176%) to high moisture level. This is in contrast to ICCV 92944 which had lower significance change in SDW with the increase of moisture. For season I the genotype recorded 1.53, 1.97 and 2.01 g in the low, medium and high moisture levels as compared to 1.24, 1.61 and 1.64 g in the low, medium and high moisture levels, thus indicating that ICCV 92944 had low response

(32%); increasing water supply thus can be adopted in regions with a low moisture level.

Effects of varying water regimes on total root biomass (RDW) among test chickpea genotypes

There was significantly large range of variations (P < 0.05) among the tested genotypes for average total root dry weight (RDW) measured during seedling stage in varied water treatments and seasons (Table 4). The interaction between water regimes and chickpea genotypes affected total root dry weight accumulation over growing period. Average RDW varied from 0.27 - 1.63 g in season I to 0.18 - 1.13 g in season II (Table 4). The overall mean RDW was 15% higher in season I (0.55 g) than season II (0.48 g) (Table 4). Moisture stress reduced RDW by 114% under low moisture as compared to high moisture treatment in season I (2013) and by 70% in season II (2013/14) as compared to 54 and 32% under low moisture as compared to medium moisture treatment in season I (2013) and season II (2013/14), respectively.

Under high moisture regimes, RDW was greater by 38.8% (season I) and 32.5% (season II) than medium moisture respectively. In season I, the drought tolerant check (ICC 4958) had 222 and 163% higher RDW than susceptible genotype checks ICC 1882 and Ngara local respectively. In season II, ICC 4958 had 126, 188 and 73% greater RDW than genotypes ICC 1882, ICC 3325 and Ngara local, respectively. Similar trends were observed under low and high moisture regions. Parental

Figure 1. Root traits of test genotypes showing differences in morphology before root scanning under low moisture regimes (25% FC).

Table 4. Mean of root dry weight (RDW) (biomass) (g) for the test genotype under varying watering regimes for season I and II (2013/2014).

Genotype	Season I				Season II				
	Low moist	Medium moist	High moist	Mean	Low moist	Medium moist	High moist	Mean	Overall mean
ICCV 92944	0.31	0.41	0.57	0.43	0.3	0.35	0.44	0.36	0.4
ICCV 00108	0.35	0.41	0.45	0.4	0.27	0.33	0.39	0.33	0.37
ICCV 97105	0.21	0.39	0.46	0.35	0.23	0.3	0.44	0.32	0.34
ICC 4958	0.41	0.54	1.66	0.87	0.45	0.54	0.57	0.52	0.7
ICCV 97306	1.21	1.74	1.93	1.63	0.91	0.94	1.55	1.13	1.38
ICCV 92318	0.26	0.57	0.72	0.52	0.27	0.55	0.67	0.5	0.51
Ngara Local	0.14	0.37	0.48	0.33	0.23	0.27	0.41	0.3	0.32
ICC 1882	0.21	0.28	0.31	0.27	0.16	0.25	0.27	0.23	0.25
ICC 283	0.16	0.32	0.38	0.35	0.14	0.31	0.36	0.27	0.31
ICC 3325	0.29	0.34	0.49	0.37	0.14	0.18	0.21	0.18	0.28
Mean	**0.35**	**0.54**	**0.75**	**0.55**	**0.31**	**0.4**	**0.53**	**0.41**	**0.48**
CV%				5.4				7.2	
l.s.d.0.05 G	*	**	**		**	**	**		
l.s.d.0.05 WT	*	**	*		**	*	*		
l.s.d.0.05 G×WT	*	*	*		*	*	**		

Key: Level of significance ***- 0.001, **- 0.05 and *-0.01, SI- the first season, SII- the second season, G- Genotype, WT- water treatment, G x WT- Genotype x water regime interaction; Moist-Moisture level.

test genotypes varied significantly in RDW both in 2013 and 2013/2014 (Table 4). The overall mean RDW for both seasons combined varied from the 0.25 - 0.28 g (ICC 1882 and ICC 3325 respectively) to 0.70 - 1.38 g (ICC 4958, ICCV 97306, respectively). In season I, RDW ranged from 0.27 g (ICC 1882) to 1.63 g (ICCV 97306) as compared to 0.18 g to 1.13 g in season II (Table 4).

The variation under low and medium water regime was 0.14-0.16 g per plant (Ngara local and ICC 283, respectively) to 1.66-1.93 g per plant (ICC 4958 and

ICCV 97306, respectively) under high watering regime (Table 4). In the second season lower values were recorded: ranging from the 0.41g (ICC ICC 3325) to 0.91 g (ICCV 97306) under lowest moisture regime as compared to 0.21 g per plant (ICC 3325) under low moisture to 1.55 g (ICCV 97306) under high moisture.

On average, genotypes ICCV 97306, ICC 4958, and ICCV 92318 had the highest root biomass (mean 0.86 g) in decreasing order in both seasons (1.38, 0.70 and 0.51 g, respectively), while ICC 1882, ICC 3325, ICC 283, and Ngara local had the lowest root biomass (mean 0.29 g per plant). Commercial varieties ICCV 92944, ICCV 00108, and ICCV 97105 had medium to high root biomass (0.39 g per plant) which was 121% lower than the best performing genotypes and 34% better than worst performing genotypes (Table 4).

Genotype ICCCV 97306 had the highest root dry biomass (mean 1.38 g per plant). This was higher than the drought tolerant check (ICC 4958) by 97 % in both seasons combined and by 87% (season I) and by 117% (season II). Across them moisture treatments the RDW of most test genotypes was increasing with the increase of water level, but was highest for ICC 283, Ngara local and ICCV 92318 which ranged from 97 - 155% RDW increase with increasing moisture from low to highest moisture in both season combined.

Effects of varying water regimes on total dry weight (TDW) (root and shoot biomass) among test chickpea genotypes

The interaction between water regimes and chickpea genotypes affected total dry weight (TDW) at root harvest measured at seedling stage (35DAE) (P<0.01), with significant range of variations among the tested genotypes in varied water treatments and seasons. The overall TDW increased with increasing moisture (25% FC) to 75% FC) with mean root and shoot biomass being 5.5% higher in season I (2.12 g) than season II (2.01 g). Overall moisture stress reduced TDW by 76% under low moisture as compared to high moisture treatment in season I (2013) and by 24% in season II (2013/2014) as compared to 43 and 21% under low moisture as compared to medium moisture treatment in season I (2013) and season II (2013/2014), respectively. Overall, the total shoot and root biomass (TDW) varied from the 1.41 g (ICC 1882) to 3.42 g (ICCV 97306). The mean TDW in the season I was 5.1% higher (2.12 g) than that recorded in the season II (2.01 g).

Test genotypes varied significantly in total shoot and root biomass (TDW) in both seasons (2013/2014). The overall mean TDW for both seasons combined varied from 1.20 - 1.25 g (ICC 1882 and ICC 283, respectively) to 2.65 - 3.74 g per plant (in season I) to 1.14-1.18 g for same genotypes to 2.54, 2.74 and 3.42 g for genotypes ICC 4958, ICCV 92318 and ICCV 97306 respectively in

season II (Table 4). In season I, TDW ranged from 0.91 g (ICC 283) to 3.74 g (ICCV 97306) as compared to 0.69 g to 3.36 g in season II.

There was great variation between moisture regimes (25% FC - 75% FC). In first season, under low water regime TDW was lowest ranging from 0.61-0.91 g per plant (Ngara local and ICC 283, respectively) to 3.61, 3.81 and 4.75 g per plant (ICC 4958, ICCV 92318 and ICCV 97306, respectively) under high watering regime (Table 4). As compared to the second season lower values were recorded: ranging from the 0.69 g (ICC 1882) to 2.08, 2.79 and 2.73 g for genotypes ICC 4958, ICCV 92318 and ICCV 97306, respectively (Table 5). Overall the genotype ICCV 97306 had the highest TDW in both seasons combine (3.42 g) while genotype ICC 1882 had the lightest shoot and root weight (1.14 g) followed by ICC 283 (1.18 g) which was 200% and 189% higher, respectively (Table 4). Similar trends were recorded for season I and II

In both seasons, the drought tolerant genotype ICC 4958 had below the average mean (2.54 g) of the two best performing genotypes (ICCV 97306 and ICCV 92318) which had the greatest TDW (3.08 g), 21% lower (Table 4). Drought susceptible genotypes ICC 1882, ICC 3325 and ICC 283 consistently had low TDW under low moisture, medium, and high moisture, respectively. Drought tolerant check (ICC 4958) had 108% greater TDW (mean 2.54 g) than the mean of three susceptible genotype checks (mean 1.22 g) (ICC 1882, Ngara local, and ICC 283) as compared to 152% greater TDW for three best performing genotypes (ICCV 97306 and ICCV 92318) (mean 3.08 g). Regardless of the moisture level, these two genotypes recorded the highest mean TDW in both seasons (3.27 g and 3.08 g respectively). This was higher than the drought tolerant check (ICC 4958) by 21.2% (Table 4). Overall, TDW of most test genotypes was increasing with the increase of moisture applied from 25 - 75% FC, but response varied with highest recorded for genotype ICC 92318 in both seasons combined (Table 4).

Effects of varying water regimes on total root length (TRL) among test chickpea genotypes

The interaction between water regimes and chickpea genotypes significantly (P < 0.05) affected total root growth over the seedling stage growing period (Table 6). The overall means for each moisture treatment (low to high: 25 - 75% FC) across seasons I and II showed that moisture stress reduced the total root length by 61.4% from 1.64 to 1.02 m from high to low moisture in both seasons combined (Table 6). Similarly, moisture stress reduced TRL by 28.8% (1.31 m) from medium (50% FC) to low moisture (25% FC) (Table 6). This varied with seasons: under low moisture TRL decreased by 65.7% as compared to high moisture treatment in season I (2013)

Table 5. Mean of on Total dry weight (TDW) (root and shoot biomass) (g) for the test genotype under varying watering regimes for season I and II (2013/2014).

Genotype	Season I				Season II				
	Low moist	Medium moist	High moist	Mean	Low moist	Medium moist	High moist	Mean	Overall mean
ICCV 92944	1.83	2.38	2.58	2.26	1.54	1.96	2.5	2	2.13
ICCV 00108	1.86	2.02	2.3	2.06	1.81	1.87	1.89	1.91	1.98
ICCV 97105	1.49	1.92	2.51	1.97	1.51	1.75	1.84	1.89	1.93
ICC 4958	1.82	2.51	3.61	2.65	2.4	2.43	2.08	2.43	2.54
ICCV 97306	2.64	3.83	4.75	3.74	1.76	2.73	2.79	3.1	3.42
ICCV 92318	1.3	3.3	3.81	2.8	1.7	3.08	3.36	2.68	2.74
Ngara Local	0.61	1.54	2.12	1.42	1.63	1.46	1.34	1.28	1.35
ICC 1882	1.05	1.19	1.35	1.2	0.69	1.23	1.35	1.09	1.14
ICC 283	0.91	1.17	1.47	1.25	1.48	1.16	1.21	1.11	1.18
ICC 3325	1.57	1.75	2.23	1.85	1.2	1.42	1.15	1.58	1.71
Mean	1.51	2.16	2.67	2.12	1.57	1.91	1.95	1.91	2.01
CV%				11.4					9.62
l.s.d.0.05 G	*	*	*		**	*	*		
l.s.d.0.05 WT	*	*	*		*	*	*		
l.s.d.0.05 G×WT	ns	*	*		ns	*	*		

Key: Level of significance ***- 0.001, **- 0.05 and *-0.01, SI- the first season, SII- the second season, G- Genotype, WT- water treatment, G×WT- Genotype x water regime interaction; Moist- Moisture level.

Table 6. Mean of Total root length (TRL) (cm) for the test genotype under varying watering regimes for season I and II (2013/2014).

Genotype	Season I				Season II				
	Low moist	Medium moist	High moist	Mean	Low moist	Medium moist	High moist	Mean	Overall mean
ICCV 92944	953	1475	1705	1377.7	921	1324	1694	1313	1345.3
ICCV 00108	1162	1380	1778	1440	916	1313	1445	1224.7	1332.3
ICCV 97105	879	1201	1498	1192.7	840	1025	1562	1142.3	1167.5
ICC 4958	1426	1680	1973	1693	1396	1592	1639	1542.3	1617.7
ICCV 97306	1029	1498	2282	1603	1032	1348	2009	1463	1533
ICCV 92318	1020	1329	1856	1401.7	1022	1502	1836	1453.3	1427.5
Ngara Local	1055	1247	1591	1297.7	895	945	1132	990.7	1144.2
ICC 1882	1071	1205	1521	1265.7	950	1029	1090	1023	1144.3
ICC 283	870	1129	1570	1189.7	869	1145	1549	1187.7	1188.7
ICC 3325	1125	1517	1775	1472.3	958	1390	1418	1255.3	1363.8
Mean	1059	1366.1	1754.9	1393.33	979.9	1261.3	1537.4	1259.5	1326.4
CV%				18.4				19.62	
l.s.d.0.05 G	*	**	**		**		*	**	
l.s.d.0.05 WT	*	**	*		*		*	**	
l.s.d.0.05 G×WT	*	*	*		**		*	*	

Key: Level of significance ***- 0.001, **- 0.05 and *-0.01, SI- the first season, SII- the second season, G- Genotype, WT- water treatment, G×WT- Genotype x water regime interaction; Moist- Moisture level.

and by 28.7% in season II (2013/14). This could be due to the early rainfall during the long season rainfall (March-May) (data not provided) that could have raised RH and delayed stress built up in the rain-out shelter as compared to delayed and shorter season rainfall at Egerton during second season (Oct-Feb).

There was significant variation ($P < 0.05$) in TRL among test genotypes both in 2013 and 2013/2014 (Table 6). The

overall mean TRL for both seasons combined varied from 1144.2-1167.7 cm (Ngara local and ICCV 97105, respectively) to 1427.5 cm (ICCV 92318, ICCV 97306, and ICC 4958, respectively). In season I TRL ranged from 1189.7cm (ICC 283) to 1693.0 cm (ICC 4958) as compared to 990.7 cm to 1617.7 cm in season II (Table 6). In season I, TRL varied from 870.0 cm (ICC 283) under low water regime to 2282.0 cm (ICCV 97306) under high watering regime (Table 6) as compared to 869.0 cm (ICC 283) under low moisture to 2009 cm (ICCV 97306 under high moisture in second season. Overall the mean TRL in the second season was 1295.5 cm which was 11% lower than recorded in the season I (1393.3 cm).

On average, TRL increased with the increase in soil moisture from low to high moisture regime. For example, in season I, the mean TRL recorded was 1059, 1366.1 and 1754.9 cm in the low, medium and high moisture regimes, respectively, as compared to 979.9, 1261.3 , and 1537.4 cm in the low, medium and high moisture levels, respectively, in second season. In both seasons, the drought tolerant genotype, ICC 4958 had above average mean (1617.7 cm) of the two best performing genotypes (ICCV 97306 and ICCV 92318) which had the longest TRL (1533 cm and 1427.5 cm), which was respectively 6 and 13% higher (Table 6). Drought susceptible genotypes ICC 1882, ICC 3325, ICC 283, and Ngara local consistently had low TRL under low moisture, medium and high moisture, respectively. Drought tolerant check (ICC 4958) had 35% greater TRL (mean 1617.7 cm) than the mean of four worst performing susceptible genotype checks (mean 1200.9 cm) (ICC 1882, Ngara local, ICC 3325, and ICC 283). In contrast ICC 4958 had 12% higher TRL than the best performing tolerant genotypes (ICCV 97306, ICCV 92318, and ICCV 92944) with mean of 1435.1 cm.

Regardless of the moisture level, these three genotypes recorded the highest mean TRL in both season combined (1533 cm and 1427.5 and 1345.3 cm, respectively. Consistently, genotypes ICCV 97105 and ICCV 00108 had unexpectedly shorter roots (mean 1249.9 cm) than the drought tolerant check (ICC 4958) and best performing commercial checks by 29.4 and 14.8%, respectively. Overall, TRL of most test genotypes was increasing with the increase of moisture applied from 25 - 75% FC, but response varied with highest (121%) recorded for genotype ICC 97306 in both seasons combined. This shows that this genotype had highest response to increasing water supply.

Effects of varying water regimes and chickpea genotypes on root:shoot (R:S)

The interaction between water regimes and chickpea genotypes significantly (P < 0.05) affected root: shoot root at harvest (Table 7). Overall, water regimes from low to high (25 - 75% FC) had non-significant increase in R:S ratio under low moisture, but increased R:S ratio by 26% from 0.306 to 0.386 from low to high moisture in season I as compared to 2.5% from 0.286 to 0.287 from low to high moisture in season II (Table 7). The mean R:S ratio in the season I was 0.352 which was higher than 0.278 recorded in the season II (Table 7).

In both seasons (2013 and 2013/2014), there was significant variation (P < 0.05) in R:S ratio among test genotypes (Table 7). The overall mean R:S ratio for both seasons combined varied from the 0.190 to 0.212 (mean 0.201) (ICC 3325 and ICCV 97105, respectively) to 0.673 (ICCV 97306 and ICC 4958, respectively). Similarly, in season I, R:S ratio ranged from 0.218 (ICCV 97105) to 0.770 (ICC 4958) as compared to 0.126 to 0.577 (ICC 3325 and ICCV 97306, respectively) in season II (Table 7).

Under low water regime genotypes ICC 3325, ICCV 97105, and ICCV 92944 had lowest R:S ratio while genotypes ICCV 97306, ICC 4858, and Ngara local had highest R:S ratio. Similar trends were observed under medium and high watering regime (Table 6). Overall the mean R:S ratio in the second season was 0.278 which was 35% lower than that recorded in the season I (0.386). On average, genotype ICCV 97105 had the lowest average R:S ratio amongst commercial genotypes in both season (0.212) while ICC 3325 had the lowest average R:S in both seasons (0.180).

Drought tolerant check (ICC 4958) and best performing genotype (ICCV 97306) had 146% R:S ratio (mean 0.63) than worst performing susceptible genotypes (mean 0.256) (ICC 1882, ICC 3325, and ICCV 92318). The increase in R:S ratio of most test genotypes was not consistent with increase in moisture applied from 25 - 75% FC as most traits measured. The highest response was recorded for genotype ICC 97306 in both seasons combined, showing that this genotype had highest response to increasing water supply.

Effects of varying water regimes and chickpea genotypes on root length density (RLD)

The interaction between water regimes and chickpea genotypes did not affect root length density, but there were significant differences between test genotypes across water regimes (Table 8). The overall means for each moisture treatment (low to high: 25 - 75% FC) across seasons I and II showed that RLD reduced with increasing moisture stress. In both seasons combined, RLD was reduced by 34.5% when moisture was decreased from high to low moisture regime. Similarly in season I, RLD was reduced by 35.4% (from 0.218 to 0.161 cm cm^{-3}) under medium to low moisture regime as compared to 33% (0.290 cm cm^{-3} to 0.218 cm cm^{-3}) under high to medium moisture. In the same way, in season II, moisture stress reduced RLD by 27.5 and 30%

Table 7. Mean of root:shoot (R:S) ratio under varying watering regimes for season I and II (2013/2014).

Genotype	Season I				Season II				
	Low moist	Medium moist	High moist	Mean	Low moist	Medium moist	High moist	Mean	Overall mean
ICCV 92944	0.204	0.208	0.284	0.235	0.242	0.217	0.214	0.222	0.228
ICCV 00108	0.232	0.255	0.243	0.243	0.182	0.214	0.228	0.209	0.226
ICCV 97105	0.164	0.255	0.224	0.218	0.18	0.207	0.223	0.206	0.212
ICC 4958	0.291	0.274	0.851	0.49	0.271	0.286	0.261	0.272	0.381
ICCV 97306	0.846	0.833	0.684	0.77	0.669	0.525	0.566	0.577	0.673
ICCV 92318	0.25	0.209	0.233	0.226	0.237	0.217	0.233	0.227	0.227
Ngara Local	0.298	0.316	0.293	0.302	0.676	0.227	0.293	0.311	0.306
ICC 1882	0.25	0.308	0.298	0.287	0.302	0.255	0.25	0.263	0.275
ICC 283	0.182	0.376	0.349	0.387	0.182	0.365	0.396	0.32	0.354
ICC 3325	0.227	0.241	0.282	0.253	0.143	0.145	0.135	0.126	0.19
Mean	0.306	0.331	0.386	0.352	0.286	0.267	0.287	0.278	0.315
CV%			10.28					9.86	
l.s.d.0.05 G	*	**	**	**	**	*			
l.s.d.0.05 WT	ns	**	*	ns	*	*			
l.s.d.0.05 G×WT	*	*	*	*	**	*			

Key: Level of significance ***- 0.001, **- 0.05 and *-0.01, ns-non-significant, SI- the first season, SII- the second season, G- Genotype, WT-water treatment, G×WT- Genotype ×water regime interaction; Moist- Moisture level.

Table 8. Mean of root length density (RLD) (cm cm^{-3}) under varying watering regimes for season I and II (2013/2014).

Genotype	Season I				Season II				
	Low moist	Medium moist	High moist	Mean	Low moist	Medium moist	High moist	Mean	Overall mean
ICCV 92944	0.16	0.215	0.345	0.24	0.17	0.215	0.255	0.213	0.227
ICCV 00108	0.15	0.25	0.3	0.233	0.2	0.2	0.25	0.217	0.225
ICCV 97105	0.1	0.2	0.25	0.183	0.15	0.15	0.25	0.183	0.183
ICC 4958	0.2	0.25	0.3	0.25	0.1	0.25	0.3	0.217	0.233
ICCV 97306	0.1	0.25	0.35	0.233	0.2	0.2	0.325	0.242	0.238
ICCV 92318	0.15	0.2	0.3	0.217	0.15	0.225	0.275	0.217	0.217
Ngara Local	0.1	0.15	0.2	0.15	0.105	0.17	0.195	0.157	0.153
ICC 1882	0.15	0.18	0.275	0.202	0.1	0.15	0.25	0.167	0.184
ICC 283	0.25	0.23	0.28	0.253	0.2	0.205	0.215	0.207	0.23
ICC 3325	0.2	0.22	0.27	0.236	0.18	0.226	0.25	0.229	0.228
Mean	0.161	0.218	0.29	0.223	0.156	0.199	0.259	0.204	0.214
CV%				13.28					
l.s.d.0.05 G	*	*	*		*	*		*	
l.s.d.0.05 WT	*	*	*		*	*		*	
l.s.d.0.05 G×WT	ns	*	ns		*	ns		*	

Key: Level of significance ***- 0.001, **- 0.05 and *-0.01, S I- the first season, S II-the second season, G- Genotype, WT- water treatment, G×WT- Genotype × water regime interaction; Moist- Moisture level.

under low to medium moisture and from high to medium moisture respectively, indicating that genotypes responded almost uniformly to decreasing moisture; however there was a higher decrease season I than season II (Table 7).

The average RLD of genotypes evaluated varied from 0.153 to 0.184 cm cm^{-3} (Ngara local, ICCV 97105, and ICC 1882) to highest RLD of 0.228 to 0.238 cm cm^{-3} (ICC 3325, ICC 4958 and ICCV 97306) in both seasons combined (Table 8). Except for genotype ICCV 97105,

other commercial checks (ICCV 00108 and ICCV 92944) had 3 and 5.3% lower RLD that drought tolerant check (ICC 4958) and best performing genotype (ICCV 97306), respectively (Table 8).

In both seasons, RLD increased with increase in water regime, but with significant differences between seasons and moisture regimes. On average, genotypes ICCV 97105, Ngara local, and ICC 1882 had the lowest average RLD (mean 0.173 cm cm^{-3}) in both seasons followed by ICCV 92944, ICCV 00108, ICCV 92318, ICC 283, and ICC 3325 (mean 0.225 cm^{-3}), while genotypes ICC 4958 and ICCV 97306 had highest RLD (mean 0.235 cm cm^{-3}). However, in both seasons, ICCV 97306 and ICC 4958 recorded the highest RLD (Table 8).

DISCUSSION

The findings of this study showed that root (and shoot) traits measured had good range of variation among the test chickpea genotypes under the three varied moisture regimes. This is to some extent in agreement to previous studies under both field and lysimetric conditions (Purushothaman et al., 2017; Serraj et al., 2004; Kashiwagi et al., 2005; Lalitha et al., 2015). Genotype and the interactions between genotype and moisture treatments affected most of the root traits (shoot dry weight, root dry weight, total dry weight, root:shoot ratio, total root length and root length density) of tested chickpea germplasm. As expected, increasing moisture stress through reducing moisture supply from high moisture to low moisture (75 - 25% field capacity) reduced most of the measured traits. For example SDW, RDW, TRL, and RLD was reduced by 68, 92, 61 and 34.4% under low moisture as compared to high moisture treatment in both seasons combined, with higher effect in season two than season one. This was probably because of early stage rainfall (long rainfall season) (data not provided) that could have raised RH and delayed stress built up in the rain-out shelter in season one. There was however non-significant effect on R:S ratio under varying moisture indicating that under the water stress levels of this study, the test genotypes could not show significant investments to roots than shoots. Generally, these root traits have clearly differentiated the drought tolerant genotypes from the sensitive ones, and explained why tolerant genotypes have better soil water acquisition under drought stress field conditions. This was earlier demonstrated by these genotypes (ICCV 97306, ICC 4958, and ICCV 92944) which produced higher yields in Chemeron and Marigat in Baringo (Muriuki et al., 2018). For example genotype ICCV 97306 outperformed the tolerant check (ICC 4958) in most root traits measured (RLD, TRL, RDW, and R:S ratio), while susceptible checks (ICC 1882 and ICC 3325) recorded lower root traits values across water regimes. These findings are in agreement with those earlier reported by Lynch (2007)

and Purushothaman et al. (2017) who noted that root architecture is critically important for soil water acquisition and most of the tolerant chickpea genotypes had displayed root growth vigor and deeper soil root proliferation at early to mid-growth period for better adaptation to drought.

They also noted that architectural traits such as basal-root gravitropism (vertical root growth angle), adventitious-root formation (RLD) and lateral branching would offer the advantage in terms of the competition in photosynthate allocation between shoot and root growth and would lead to deep root systems (TRL) without overtly changing root biomass allocation. From this study drought genotypes ICC 4958 and ICCV 97306 had highest RLD (mean 0.235 cm^{-3}), above average mean TRL (1575 cm) and highest root dry mass (RDW) while drought susceptible genotypes (ICC 1882, ICC 3325 and ICC 283, and Ngara local) consistently recorded lower values. Hence one of the options to improve the root systems for drought avoidance is the enhancement of root growth vigour leading to deeper root penetration as shown by the two best performing genotypes. Rooting growth at different depth was however not measured in this study. This suggests that these morpho-physiological root traits (especially TRL) could be used as indirect selection criteria to augment yield-based selection procedures done under field condition. Similarly, Kashiwagi et al. (2005) and Gregory (1988) reported the existence of a large diversity in chickpea rooting depth which ranged from 88 to 126 cm at 35 DAS under long PVC cylinder culture conditions in ICRISAT and from 60 to 150 cm at crop maturity under field conditions respectively.

They observed that their studies also confirmed that previously known drought-tolerant chickpea genotypes such as ICC 4958 possess deep rooting ability. This is in agreement with the findings of this study where drought tolerant check (ICC 4958) had 35% greater TRL (mean 1617.7 cm) than the mean of four worst performing susceptible genotype checks (mean 1200.9 cm) (ICC 1882, Ngara local, and ICC 3325) at 35 DAS under long PVC cylinder culture conditions. These results show that precise targeting of root traits as indicators of yield would consequently lead to faster rates of yield improvement and broadening of genetic base under drought stress in ASALs. This is because, as compared to field evaluation done in several multi-locations, these traits are easier and faster to measure under rain out shelter than grain yield and they can also be observed at/or before flowering (seedling stage) and eliminate susceptible lines from crossing nursery and shorten the time to complete selection cycle. An estimate of yield potential under drought stress simulated conditions can therefore be determined more easily before final harvest.

This is in agreement with Lynch (2007) and Purushothaman et al. (2017) who proposed that breeding for the best combination of root traits mainly profuse RLD

at surface soil depths and RDW at deeper soil layers to be the best selection strategy for an efficient water use and an enhanced terminal drought tolerance in chickpea. In addition, Reynolds and Hunter (2001) noted that in wheat, a deliberate selection with a view to combining synergistic root traits like dry root weight, early seedling vigour, and RLD is likely to achieve results sooner than using grain yield performance alone.

Furthermore, Kashiwagi et al. (2006) suggested that rooting depth, root biomass, and root length density were identified as most promising traits in chickpea for terminal drought tolerance, as these help in greater extraction of soil moisture. More profuse (higher root length density, and RLD) and deeper root systems are often viewed as desirable traits for drought adaptation. In related legume cowpea crop, Matsui and Singh (2003), reported that tolerant genotypes had higher root dry matter per unit of leaf area and a downward movement of roots while using root pin box method, indicating that they would invest more in deeper rooting for water capture. As in this present study, the possible role of water extraction traits was demonstrated in that study by deeper rooting and higher root length density under decreasing drought stress from low to high moisture regime (25 to 75% field capacity). In related study, Kashiwagi et al. (2006) reported that chickpea genotypes reaching higher yield under terminal stress condition had higher RLD and genetic variability for root penetration rate of 2.5-3.6 cm day^{-1} and RLD of 0.19 -0.30 cm^{-3} among the chickpea mini-core germplasm collection (n = 211) at 35 DAS in similar tall cylinder culture systems (with 120 cm in height and 1.1 g cm^{-3} of bulk density) under rain-fed conditions. For sub-optimal complete extraction of soil moisture, RLD values of <0.5 cm cm^{-3} and <0.4 cm cm^{-3} particularly in Asia have been reported in lysimeters experimentation but even lower values have been reported which range from 0.150-0.252 cm cm^{-3} comparable to the findings of Kashiwagi et al. (2013).

In several related studies, Krishnamurthy et al. (1996), Kashiwagi et al. (2005) and Upadhyaya and Ortiz (2001) reported that genotypes ICC 4958 and ICC 8261 have been identified as the most prolific and deep-rooting chickpea accessions and they have been utilised as breeding materials to introgress these advantageous root traits into well-adapted regional chickpea cultivars for further improving grain yield under drought in semi-arid tropics (Kosgei, 2015; Varshney et al., 2014; Kimurto et al., 2017). In this present study, genotype ICCV 97306 has been identified as most prolific and deep rooting chickpea candidate similar to already identified ICC 4958 and could be used as donor for introgressing these water-uptake enhancing root traits into well-adapted chickpea cultivars for further improving grain yield under drought in semi-arid tropics.

These genotypes could be possessing larger xylems and phloem hence less capillary forces and low hydraulic resistance to water movement from soil to plant tissues through roots, thus increasing more soil water uptake and transport even under dry soils as compared to susceptible checks like ICC 3325 and ICC 1882. In related studies, Purushothaman et al. (2014) and Li et al. (2009) reported that chickpea has been shown to possess the largest number of xylem vessels among 6 major legume crops, hence a largest total xylem passage for water flow of 722 μm^2 in a single chickpea root as compared to 681 μm^2 in cowpea. They however noted that had the narrowest average diameter of 9.5 μm as compared to 14.0 μm in common bean. They noted that root systems with thin xylem vessels can be expected to have more capillary forces and less cavitation, and these are advantageous in terms of soil water uptake and transport even under dry soils. Similarly, Benjamin and Nielsen (2006) noted that as compared to other legumes like beans, chickpea also had relatively large xylem quantity and root biomass and the crop was expected to absorb more plant available soil water (PAW). These indicate that chickpea is more adapted to dense heavier soils in dry lands as compared to common beans.

In this study, it was also noted that most of the genotypes that possessed advantageous root traits and best performing (e.g. RLD of 0.228-0.238 cmcm^{-3}, TRL and RDW) were Desi genotypes (ICC 4958, ICCV 00108, ICCV 92944, and ICCV 92318) except for genotype (ICCV 97105-Desi) and (ICCV 97306-Kabuli) in both seasons combined. These indicate that the root anatomy could be varied among the two major chickpea types of the Desi (brown seed coat in smaller size) and the Kabuli (white seed coat in bolded larger seed size); the Desi could be having restrictive xylem and phloem vessels which could be conservative in water movement into and out of the tissues. This would lead to possible reduction in water loss due to transpiration and increase their performance in limited supply as shown under low and medium moisture regime (50 - 25% FC) compared to Kabulis which had better and higher response under high moisture regime in this study. In earlier studies, Desis had been reported to possess a moderate water uptake when compared to Kabulis, and considered conservative in their water requirement; they adapt well to the receding soil moisture environments than the Kabulis that had access to more water during the major part of their early growth (Berger et al., 2004).

Similarly, Purushothaman et al. (2014) noted that the xylem vessels in Desi were reported to be fewer in number and narrower in diameter compared to the Kabulis which he noted might explain why Desis had a moderate water uptake when compared to Kabulis; they were considered conservative in their water requirement adapting well to the receding soil moisture environments than the Kabulis. Several other studies also show an advantage of having superior root traits for yield under stress conditions (Silim and Saxena, 1993; Price et al., 2002b; Ober et al., 2005; Sarker et al., 2005; Tuberosa et al., 2002; Gowda et al., 2011). One of the important

mechanisms of drought avoidance is the ability of the plant to change its root distribution in the soil and this would vary by cultivar within a species (Benjamin and Nielsen, 2006). Genotype ICCV 92944 recorded best performance across seasons (stability) for this traits and hence indication of better adoption to drought condition.

Furthermore, Kashiwagi et al. (2005) observed cooler leaf canopy temperature estimated by infrared digital thermography at 70 DAS had a significant positive association with seed yield under terminal drought in field-grown chickpea at ICRISAT. This indicates that chickpea genotypes with greater transpiration at this stage would have greater reproductive growth leading to better seed yield under drought environments. They noted that although clear correlations were not consistently detected between leaf canopy temperature and root characteristics at 35 DAS, genotype ICC 4958 (drought tolerant check in this study) recorded a high prolific and deep root system and was one of the most highly transpiring leaf canopies among 16 diverse entries. In other studies, Vadez (2014) noted that in peanut (groundnuts) higher yields were obtained in where more profuse roots in the deeper soil layer were reportedly correlated to higher yield under water stress conditions, indicating that higher root length density (RLD) at depth was responsible for more water extraction. In contrast, drought stress strongly inhibited root growth of chickpea and that root growth ceased after the third week of stress (Tilahun and Sven, 2003). Previous work by Thomas (1995) reported that chickpea plants were found to have lower root length density than barley, but absorbed water more efficiently than barley plants. Amede and Schubert (2003) thus concluded that drought resistance of chickpea was due to the effect of osmotic adjustment, a function of root hydraulic conductivity, which is governed by the diameter and distribution of the meta-xylem vessels of the roots

Conclusion

The findings of this study showed that:

(i) Use of root traits to identify drought tolerance in chickpea during early growth stage significantly contribute to the seed yield in chickpea.
(ii) Increasing rooting depth, root biomass and RLD could increase the uptake of water and yield in chickpea which could be due to relatively large number of xylem vessels and root biomass which enhances better absorption of more plant available soil water (PAW) and superior adaptation to dense heavier soils in dry lands as compared to common beans. There was however variations in root anatomy between the two major chickpea types where majority of the best performing genotypes under low moisture regimes were of the Desi type (e.g. ICC 4958, ICCV 00108, ICCV 92944, and ICCV 92318) as compared to Kabulis which had better

and higher response under high moisture regime in this study.
(iii) Root depth, root biomass and RLD can be used for indirect selection for drought tolerance especially in early stages of breeding for drought tolerance which would consequently reduce the cost of multi-location field evaluation in the breeding programs.
(iv) Genotype ICCV 97306 was identified as most prolific and deep rooting chickpea candidate similar to already identified tolerant check ICC 4958 and could be used as donor for introgressing these water-uptake enhancing root traits into well-adapted chickpea cultivars for further improving grain yield under drought in semi-arid tropics.
(v) Root and shoot growth is closely linked as shown by most genotypes and deeper rooting might lead to faster soil water depletion, which would be a problem for crops depending on stored soil moisture. Hence capturing deep layer water though metabolically expensive is a one-time benefit since any rainfall/irrigation event would wet the profile from the top in progressive drought stress conditions.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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