



EFFECT OF MID SEASON DROUGHT STRESS ON ROOT TRAITS, ROOT SHOOT RATIO AND PROLINE CONTENT IN GROUNDNUT GENOTYPES WITH CONTRASTING DROUGHT TOLERANCE

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ABSTRACT

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Maximum yield losses under drought situation coincide with pegging, pod formation and pod development stages in groundnut. Among the eleven genotypes analyzed under midseason drought, TCGS 1157 and MLTG 4 were found to be most drought tolerant and Narayani and Kadiri 6 were found to be highly sensitive to moisture stress. Prominent leaf folding was observed in drought tolerant genotypes such as TCGS 1157 and MLTG 4 to reduce the evapotranspiration losses within 15 days after moisture stress imposition. In contrast, drought sensitive genotypes like Kadiri 6 and Narayani did not exhibit leaf folding phenomenon resulted in withering and drooping of leaves at 65 DAS. TCGS 1157 and MLTG 4 displayed better ability to expand their roots than Narayani and Kadiri 6 under moisture stress situation. The extent of increase in root shoot ratio is more in drought tolerant genotypes (79-103%) when compared to susceptible genotypes (31-53%). The increase in proline content was more in Kadiri 6 and Narayani than in TCGS 1157 and MLTG 4 at 20 days and 30 days after stress imposition. Sensitive genotypes were subjected to severe stress at early stages as they did not show any water saving mechanisms like leaf folding which was prominent in drought tolerant genotypes. This leads to increased evapotranspiration rates in Kadiri 6 and Narayani and thereby synthesizes more proline to maintain osmotic potential. Therefore, the increased levels of proline under drought stress can be better considered as a stress indicator in plants.

KEYWORDS: Genetic variability, PCV, GCV, heritability, genetic advance, pod yield, groundnut.

INTRODUCTION

Groundnut (*Arachis hypogaea* L.) is the most important food and oilseed crop cultivated globally in 25.44 m ha with a production of 45.22 mt (FAOSTAT : Anonymous, 2014). About one-third of the groundnut produced globally is consumed as table purpose and two-thirds are crushed for oil. In India, groundnut productivity is 937 kg/ha and is below world average (1037 kg/ha) (Anonymous, 2014). This low productivity is mainly due to production constraints such as the cultivation of the crop on marginal lands under rainfed conditions, occurrence of dry spells due to vagaries of monsoon and low input-use.

Drought triggers a wide variety of plant responses, ranging from cellular metabolism to changes in growth rates and crop yields. Mid-season drought coincides with critical growth stages such as pegging and pod filling stages (50-100 Days After Sowing (DAS)) causes more yield loss than other phenophases. Groundnut genotypes exhibit a variety of drought avoidance strategies such as

leaf folding, modulation of root and shoot growth and increased production of osmoregulants like proline. The root system is critical to plant adaptation under drought conditions. Root traits, particularly root length and biomass are expected to play an important role in avoidance of drought in receding soil moisture conditions by improving water availability to the plant through more efficient extraction of available soil moisture. In groundnut, water stress stimulates the growth of roots into deeper layers and vegetative growth is completely ceased. In addition, prolonged water stress resulted in smaller leaves, reduced internodal length and no effect on number of nodes. Proline accumulation is a common metabolic response of higher plants to water deficits where it contributes substantially to the cytoplasmic osmotic adjustment.

To bridge the prevailing yield gap, genotypes with good root mining ability and drought tolerance need to be developed which can significantly contribute towards mitigation of drought effects and can provide a long-term solution to rainfed farmers.

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MATERIALS AND METHODS

Plant material

Groundnut genotypes namely TCGS-1398, Kadiri-9, TCGS-1157, TCGS-1073, TCGS-1173, MLTG-4, Kadiri-6, Narayani, TPT-1, ICGV-07132, ICGV-07070 were screened for tolerance to mid season drought in pot culture experiments in rainout shelters. Drought stress is imposed by withholding water from 50DAS to 80DAS coincides with peg formation and pod development under field condition for evaluating the influence of water stress on morphological (leaf folding, root length and root shoot ratio) and biochemical parameter like proline content.

Leaf folding

The diurnal variation in young leaflets with increased duration of moisture stress was recorded visually at mid season drought stress period (50-80 DAS).

Root length and root shoot ratio

The root length and root shoot ratio were recorded in well watered and respective drought stress imposed genotypes at the time of final harvest using standard methods.

Leaf proline content

Proline content of the leaf was estimated as per the procedure of Bates *et al.* (1973) with slight modifications. 500 mg of fresh leaf sample was finely grounded with liquid nitrogen and macerated with 10ml of 3% (w/v) sulphosalicylic acid. Samples were centrifuged at 8,000 rpm for 15 minutes at 4°C and 2ml of supernatant solution was aliquated into a new test tube. To this, 2 ml each of ninhydrin and glacial acetic acid were added and incubated at 75°C in hot water bath for 1 hour and then cooled to room temperature under running tap water. 4 ml of toluene was added to this contents and stirred uniformly for 30 seconds. Proline collected in upper pink layer was estimated by OD value at 520 nm using UV 2450 visible spectrophotometer. The Leaf proline content was expressed in $\mu\text{g g}^{-1}$ of leaf sample according to the following formula.

$$\mu \text{ moles of proline/g tissue} = \frac{\mu\text{g proline} / \text{ml} \times \text{ml toluene}}{115.5} \times \frac{5}{\text{g sample}}$$

where, 115.5 is the molecular weight of proline.

RESULTS AND DISCUSSION

In the present investigation, eleven genotypes *viz.*, ICGV 07132, ICGV 0707, TCGS 1398, TCGS 1157, TCGS 1073, TCGS 1173, MLTG 4, Narayani, TPT 1, Kadiri 9 and Kadiri 6 were screened for moisture stress tolerance and subsequent recovery after reaching the permanent wilting point in pot culture experiment in rainout shelter (Figure 1).

In groundnut, maximum yield losses under drought situation coincide with pegging, pod formation and pod development stages (mid-season drought) commenced from 50- 80 DAS. The groundnut genotypes under study were categorized into three groups based on drought tolerance and recovery per cent upon watering after reaching permanent wilting point. Among the eleven genotypes analyzed, TCGS 1157 and MLTG 4 genotypes showed early recovery after reaching the permanent wilting point and were grouped into moisture stress tolerant group. In contrast Narayani and Kadiri 6 were highly sensitive to moisture stress and were grouped into moisture stress sensitive group and the remaining genotypes were grouped into intermediate types (Table 1). Puangbut *et al.* (2009) grouped six genotypes into four different groups based on yield response to pre-flowering drought and also reported that two genotypes *viz.*, KK 60-3 and Tifton-8 had moisture stress tolerance with significant yield.

Among the genotypes, TCGS 1157 and MLTG 4 were found to be most drought tolerance and with highest percentage of survival even after reaching permanent wilting point. Based on this, the four contrasting groundnut genotypes *viz.*, TCGS-1157, MLTG 4, Narayani and Kadiri 6 were considered for further studies of drought tolerance submitted to midseason moisture stress (50-80 DAS) in field conditions.

Morphological changes

Leaf folding

In response to moisture stress, groundnut exhibits leaf folding where the opposite leaflets of tetra foliate leaf comes together and orient parallel to each other and thereby reduce the evapotranspiration losses (Reddy *et al.*, 2003). The field was irrigated till it reached field capacity at 50 DAS. Prominent leaf folding was observed in drought tolerant genotypes such as TCGS 1157 and MLTG 4 (Figure 2) to reduce the evapotranspiration losses

within 15 days after moisture stress imposition (*i. e.*, 65 DAS). In this process, the upper photosynthetically active region of opposite leaflets comes together and the lower surface of the leaflets was exposed to both solar and ground radiation (Chung *et al.*, 1997). In contrast, drought sensitive genotypes like Kadiri 6 and Narayani did not exhibit leaf folding phenomenon resulted in withering and drooping of leaves at 65 DAS.

In resistant genotypes, the leaf folding helped them to reducing water loss and thereby sustained prolonged drought stress efficiently. The leaf folding characterized by moisture stress greatly contributes to adaptation to the adverse environmental condition (Nautiyal *et al.*, 2008)

Root length

Water stress often stimulates the growth of roots into deeper layers of soil. The average root length of groundnut genotypes subjected to 30 days of water stress (50-80 DAS) was more in all the genotypes when compared to control (Table 1). The best way to measure the root length was through root structures and in this experiment the root length was underestimated as most of them were broken. Drought tolerant genotypes (TCGS 1157 and MLTG 4) displayed better ability to expand their roots than drought susceptible genotypes (Narayani and Kadiri 6) under moisture stress situation (Figure 3). It confirmed the ability of groundnut to deep rooting in soils with low water availability, in order to tolerate the water stress condition (Vorasoort *et al.*, 2003).

The results are in agreement with the fact that groundnut has the capacity to modify the root length and thereby exploiting the available water in deeper soil layers as an important mechanism to avoid drought (Songsri *et al.*, 2008 and Kambiranda *et al.*, 2011). The extent and the pattern of root development were closely related to the ability of the plants to absorb water, enhancement of root growth under drought conditions allows the plant to extract more water from deeper zones may contribute to the drought tolerance in groundnut (Madhusudhan and Sudhakar, 2014).

Root shoot ratio

The groundnut genotypes under the study when subjected to midseason drought showed increased root: shoot ratio when compared to control (Table 2). The drought tolerant genotypes *viz.*, TCGS 1157 and MLTG

4 showed increased root shoot ratio of 103 per cent and 79 per cent respectively over control (well watered). In drought susceptible genotypes (Narayani and Kadiri 6) also the root shoot ratio increased slightly (53% and 31 %) when compared to respective control. The extent of increase in root: shoot ratio is more in drought tolerant genotypes when compared to susceptible genotypes (Table 3). Root shoot ratio in groundnut under water stress directly related to changes in source and sink relationships with the root being a stronger sink than the shoots (Figure 4).

The increased root length and decreased shoot growth under water stress contributes to increased root shoot ratio. The results obtained with root shoot ratio in groundnut genotypes were in consistent with the fact that the plants which exhibits an increase in root shoot ratio under water stress were more drought tolerant (Lloret *et al.*, 1999). This might be due to their ability to maintain osmotic pressure, ability to maximise available water and penetrate into deeper soil horizon.

The degree of reduction in shoot length under water stress may be due to decrease in cell division, elongation and enlargement which might have ultimately leads to the reduction in plant height (Madhusudhan and Sudhakar, 2014).

Proline content

With prolonged moisture stress imposition from 50 to 80 DAS, proline content was increased continuously at all sampling intervals in both drought tolerant and susceptible genotypes in comparison with their respective controls (Table 3). In drought tolerant genotypes like MLTG 4 and TCGS 1175, at 10 days after moisture stress imposition the proline content was 136.3 and 168.3 $\mu\text{g g}^{-1}$ respectively. The accumulation of proline was increased significantly in these genotypes at 20 days (179 and 241 $\mu\text{g g}^{-1}$) and 30 days (262 and 247 $\mu\text{g g}^{-1}$) after moisture stress imposition.

In drought susceptible genotypes like Kadiri 6 and Narayani, at 10 days after stress imposition the accumulation of proline content was 134 and 211 $\mu\text{g g}^{-1}$ respectively. The proline content was increased even more than drought tolerant genotypes at 20 days (288 and 322 $\mu\text{g g}^{-1}$) and 30 days (409 and 432 $\mu\text{g g}^{-1}$) after stress imposition in Kadiri 6 and Narayani respectively (Figure 5). The proline content accumulated in cells may contribute to the maintenance of turgor of cells and the drought sensitive genotypes were prone to loose moisture

Effect of drought stress on root traits, root shoot ratio and proline content in groundnut



Fig. 1. Screening of groundnut genotypes for drought tolerance at 50 DAS in pot culture experiment in rainout shelter



A



B

Fig. 2. Leaf folding displayed in MLTG 4(A) where as prominent drooping in Narayani at 60 DAS (10 days after moisture stress imposition)

Table 1. Root length contrasting groundnut genotypes at the time of harvest in both control and respective moisture stress

S. No.	Genotypes	Root length	
		Control (cm)	Stress (cm)
1	MLTG 4	13.5 ± 1.1	16 ± 1.5
2	TCGS 1157	13.4 ± 1.2	16 ± 2.5
3	NARAYANI	12 ± 0.83	15.2 ± 1.1
4	Kadiri 6	12.6 ± 1.1	14.8 ± 1.3

Table 2. Root shoot ratio of contrasting groundnut genotypes at the time of harvest in both control and respective moisture stress

S. No.	Genotypes	Root shoot ratio	
		Control	Stressed
1	MLTG 4	0.34	0.61
2	TCGS 1157	0.33	0.67
3	NARAYANI	0.36	0.55
4	K 6	0.38	0.50

Table 3. Proline content in contrasting groundnut genotypes at the time of harvest in both control and respective moisture stress

S. No.	Genotypes	50 DAS		60 DAS		70 DAS		80 DAS	
		Control	Control	Stressed	Control	Stressed	Control	Stressed	
1	MLTG 4	10 ± 1.6	8.5 ± 0.6	136.6 ± 1.6	25.5 ± 2.5	179 ± 2.5	37.33 ± 1.5	262.7 ± 1.8	
2	TCGS 1157	7.1 ± 1.07	16.1 ± 1.9	168.3 ± 1.6	26.1 ± 2.46	241.1 ± 12.0	39.9 ± 1.82	247.2 ± 5.3	
3	NARAYANI	1.7 ± 0.19	3.8 ± 0.96	211 ± 2.0	18 ± 3.3	322.8 ± 2.0	69 ± 1.66	432.7 ± 10.8	
4	Kadiri 6	3.43 ± 0.9	5.5 ± 0.9	134.9 ± 3.4	15.73 ± 0.4	288.3 ± 5.3	31.97 ± 0.15	409.6 ± 3.2	

earlier than drought tolerant genotypes and thereby accumulated more proline under prolonged drought situation.

The proline accumulation in drought susceptible genotypes *viz.*, Kadiri 6 and Narayani were increased in two folds than the drought tolerant genotypes *viz.*, TCGS 1157 and MLTG 4 at prolonged moisture stress of 20 to 30 days. Accumulation of free proline content under moisture stress in groundnut genotypes was most probably due to higher rates of proline synthesis than proline oxidation (Ranganayakulu *et al.*, 2015).

Sensitive genotypes were subjected to severe stress at early stages as they did not show any water saving mechanisms like leaf folding which was prominent in drought tolerant genotypes. This leads to increased evapotranspiration rates in Kadiri 6 and Narayani and thereby synthesizes more proline to maintain osmotic potential. Therefore, the increased levels of proline under drought stress can be better considered as a stress indicator in plants (Kaneria and Bishi, 2015).

Similar results were reported by Ranganayakulu *et al.* (2015) in K-134 (drought tolerant) and JL-24 (drought susceptible) groundnut genotypes. In contrast to groundnut, increase in proline content was more in drought tolerant cowpea genotypes than in sensitive genotypes

CONCLUSION

Among the eleven genotypes analyzed under midseason drought, TCGS 1157 and MLTG 4 were found to be most drought tolerant and Narayani and Kadiri 6 were found to be highly sensitive to moisture stress.

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