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COTTON GROWTH UNDER POTASSIUM DEFICIENCY STRESS IS INFLUENCED BY PHOTOSYNTHETIC APPARATUS AND ROOT SYSTEM

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Abstract

Due to rapid depletion of soil potassium (K) and increasing cost of K fertilizers in Pakistan, the K-use efficient crop genotypes become very important for agricultural sustainability. However, limited research has been done on this important issue particularly in cotton, an important fibre crop. We studied the growth and biomass production of three cotton genotypes (CIM-506, NIAB-78 and NIBGE-2) different in K-use efficiency in a K-deficient solution culture. Genotypes differed significantly for biomass production, absolute growth rates (shoot, root, leaf, total), leaf area, mean leaf area and relative growth rate of leaf under K deficiency stress, besides specific leaf area. The relative growth rate (shoot, root, total) did not differ significantly, except for leaf. For all these characters, NIBGE-2 was the best performer followed by NIAB-78 and CIM-506. Shoot dry weight was significantly related with (in decreasing order of significance): mean leaf area, leaf dry weight, leaf area, root dry weight, absolute growth rate of shoot, absolute growth rate of root, absolute growth rate total, absolute growth rate root, relative growth rate leaf, relative growth rate total and relative growth rate shoot. Hence, the enhanced biomass accumulation of cotton genotypes under K deficiency stress is related to their efficient photosynthetic apparatus and root system, appeared to be the most important morphological markers while breeding for K-use efficient cotton genotypes.

Introduction

Cotton yields in Pakistan are highly stagnant with a negligible increase (<10%) despite highest share of cotton (23%) in total fertilizer consumption of Pakistan after wheat, during last 15 years (Anon., 2007). However, fertilizer consumption is only limited to nitrogen (N) and phosphorus (P), while the potassium (K) fertilization is missing altogether (Anon., 2007). Hence, it is highly likely that the stalemate in cotton yields might be attributed to the negligible use of potassium fertilizer (<1.0 kg K_2O ha^{-1}), coupled with the rapid mining of available soil K in Pakistan (Nawaz *et al*., 2006).

Cotton requires K (150 kg ha^{-1}) almost equal to N (156 kg ha^{-1}) to give a yield of ≈ 2500 kg ha^{-1} of ginned cotton (Silvertooth, 2007). Hence, an adequate K supply is crucial during entire cotton growth and development (Makhdum *et al*., 2007), mainly due to its vital role in biomass production (Zhao *et al*., 2001), leaf area expansion, CO_2 assimilation (Reddy *et al*., 2004), photosynthesis, leaf pressure potential, transpiration and water use efficiency (Pervez *et al*., 2004), boll weight and size, lint yield (Akhtar *et al*., 2003) and fiber quality (Pettigrew *et al*., 1996). Hence, K nutrition of cotton appeared to be very indispensable. However, K-fertilization is a very costly business and farmers of the developing countries, such as Pakistan, are always reluctant to involve K in their crop nutrition programs. The situation, thus, demands for workable alternatives for low-K-input crop production. Growing nutrient-use-efficient genotypes is a key strategy for crop

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production under nutrient deficient conditions. Such genotypes perform well under nutrient deficient conditions (Rengel & Damon, 2008) by acquiring adequate amounts of a certain nutrient (uptake efficiency) and/or by utilizing the acquired nutrients more efficiently (utilization efficiency) (Sattelmacher *et al*., 1994). Considerable intra- and inter-specific variation in K uptake and utilization efficiency has been identified among existing genotypes for a variety of crop species, such as wheat (Damon & Rengel, 2007), rice (Yang *et al*., 2003) and maize (Nawaz *et al*., 2006). Zhang *et al*., (2007) reported the differential responses of four conventional and Bt cotton cultivars to K deficiency. Makhdum *et al*., (2007) also highlighted significant differences in four cotton cultivars for biomass production and partitioning between different organs under the influence of K fertilization. Some other researches conducted on K nutrition of cotton emphasized the crucial role that K plays in cotton nutrition (Makhdum *et al*., 2005, Pervez *et al*., 2006; Makhdum *et al*., 2007; Pervez *et al*., 2007; Zhang *et al*., 2007). In this study, we explored the growth and biomass production of three differentially K-use efficient cotton genotypes of Pakistan under K-deficient solution culture to evaluate the role of genotypic variation and various growth parameters for K deficiency tolerance of cotton.

Materials and Methods

The experiment was conducted using solution culture technique in a glass house under natural conditions (mean temperature: 34°C, mean relative humidity: 54%). Three cotton genotypes were used in this experiment viz. CIM-506 (CIM 360 × CP 15/2, released in 2002 by Central Cotton Research Institute (CCRI), Multan, Pakistan), NIAB-78 (DPL 16 × AC 134 F1 Irradiated, released in 1983 by National Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan) and NIBGE-2 (LRA-5166 × S-12, released in 2006 by National Institute for Biotechnology and Genetic Engineering, (NIBGE), Faisalabad, Pakistan). The selection of these genotypes was based upon their differential growth behavior and potassium use efficiency in relation to K nutrition in an early experiment (data unpublished) involving 25 cotton genotypes of Pakistan. We found that under K deficiency stress CIM-506 produced less biomass and had medium K use efficiency, NIAB-78 produced medium biomass and had medium K use efficiency, whereas, NIBGE-2 produced high biomass and had high KUE. The seeds of all three genotypes were delinted by thorough mixing with commercial grade H_2SO_4 in plastic containers @ 1 ml H_2SO_4 per 10 g cottonseed. This was followed by thorough washing of delinted seeds with deionised water and drying under shade. The seeds were surface sterilized with 5% Sodium hypochlorite solution for 5 min. to avoid fungus development. Seed germination was achieved in acid-washed river-bed sand, filled in plastic tubs. Separate tubs were used for the germination of each genotype. Distilled water was sprinkled to maintain optimum moisture contents for seed germination and seedling establishment. Seven days old, uniform sized seedlings were carefully transferred into foam plugged holes made on 1.5 L black-paint-coated plastic jars (13 cm × 12 cm, Ismail Industries Ltd., Hub, Pakistan). These jars were placed over thermopore sheets on an iron table and filled with half strength Johnson's solution (Johnson *et al*., 1957). The solution was modified to contain deficient (0.3 mM) K and renewed twice a week, to avoid changes in the concentration and provide stable supply of nutrients. Deionized water was daily added to jars to cover water loss due to evapotranspiration. Air pumps (Resun AC-9903, China) were used to bubble the air into the nutrient solution to provide oxygen and ensure homogenous nutrient distribution. The pH of nutrient solution was monitored every second day by using a digital pH meter (TOA HM-12P, Japan) and maintained around 5.5 by adding sulphuric acid (H_2SO_4) and/or calcium hydroxide ($Ca(OH)_2$). Harvesting was done twice, i.e. first after 28 days of transplanting (H1) and second after an additional 12 days (H2). Harvested plants were properly washed in distilled water and blotted dry using filter paper. Subsequently, the plants were separated into shoots and roots. The roots were air dried for 12 h at room temperature. Leaf area (LFA) was recorded using area meter (Delta-T Devices Ltd., England). The dry weights of various plant parts were recorded after oven drying at 70°C for 48 h. Mean leaf area (MLA) was calculated by using the formula given by Matsuura *et al*., (2005). Accordingly, MLA = $[LFA_2 - LFA_1]$, $\div$ $[\ln LFA_2 - \ln LFA_1]$, where subscript 2 and 1 represent H2 and H1, respectively, ln is the natural log. Specific leaf area (SLA) was calculated as a ratio of leaf area (LFA) to leaf dry weight (LDW). Absolute and relative growth rates (AGR and RGR, respectively) were calculated in terms of dry weight (W) produced per unit time (T, in days) and by assuming growth in exponential terms, respectively as suggested by Hunt (1978). Accordingly, AGR (g day^{-1}) = $[W_2 - W_1] \div [T_2 - T_1]$ and RGR ([g g^{-1}] day^{-1}) = $[\ln W_2 - \ln W_1] \div [T_2 - T_1]$. The twice replicated experiment was designed in a completely randomized fashion with four repeats. Three plants of each genotype were maintained per jar. The jars of each replication were grouped together on an iron table over white thermopore sheets. Genotypes were randomized within each replicate. Jars were rotated on alternate days to eliminate any environmental influence. The results of both experiments were pooled, due to their homogenity. The analysis of variance and mean separation test, using Tukey’s honestly significant difference, were performed using Statistix for Windows ver. 8.1 (Analytical software© 1985-2005, statistix.com).

Results

Highly significant F-values (p=0.0000) from analysis of variance revealed differential growth response of cotton genotypes for biomass production, absolute growth rate, leaf area and mean leaf area, under K deficiency stress. Similarly, the F-value from analysis of variance for relative growth rate of leaf was also significant (p=0.0284). However, the specific leaf area and relative growth rates had non-significant F-values. For all the above characters that were registered significantly different, NIBGE-2 was the best performer (Figs. 1-4). The efficient cultivar NIBGE-2 produced 78% more shoot dry weight, 99% more root dry weight and 101% more leaf dry weight compared to NIAB-78, and in comparison to CIM-506, NIBGE-2 produced 235% more shoot dry weight, 301% more root dry weight and 317% more leaf dry weight. The total dry weight of NIBGE-2 was 81% more than NIAB-78 and 246% more than CIM-506. Likewise, as compared to CIM-506, NIAB-78 produced 88% more shoot dry weight, 102% more root dry weight, 108% more leaf dry weight and 91% more total dry weight (Fig. 1). The data regarding the efficiency of the photosynthetic apparatus (leaves) of cotton genotypes under K deficiency stress (Fig. 2) revealed that the leaf area of NIBGE-2 was 114% more than NIAB-78 and 371% more than CIM-506. Moreover, NIAB-78 had 120% more leaf area than CIM-506. The mean leaf area of NIBGE-2 was 114% more than NIAB-78 and 313% more than CIM-506. The mean leaf area of NIAB-78 was 92% more than CIM-506. As mentioned earlier, the specific leaf area of all three genotypes was statistically alike (Fig. 2). The absolute growth rate of shoot of NIBGE-2 was 89% more than NIAB-78 and 416% more than CIM-506, whereas, NIAB-78 had 172% more absolute growth rate of shoot than CIM-506. The absolute growth rate of root of NIBGE-2 was 10% more

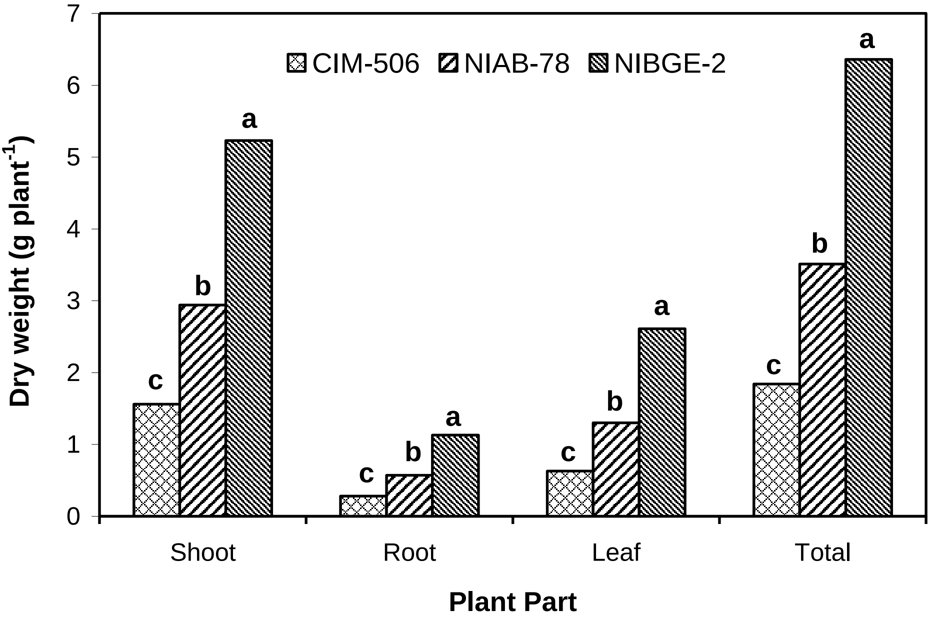

Fig. 1. Biomass accumulation of differentially K-use efficient cotton genotypes under K deficiency stress in hydroponics at seedling stage.

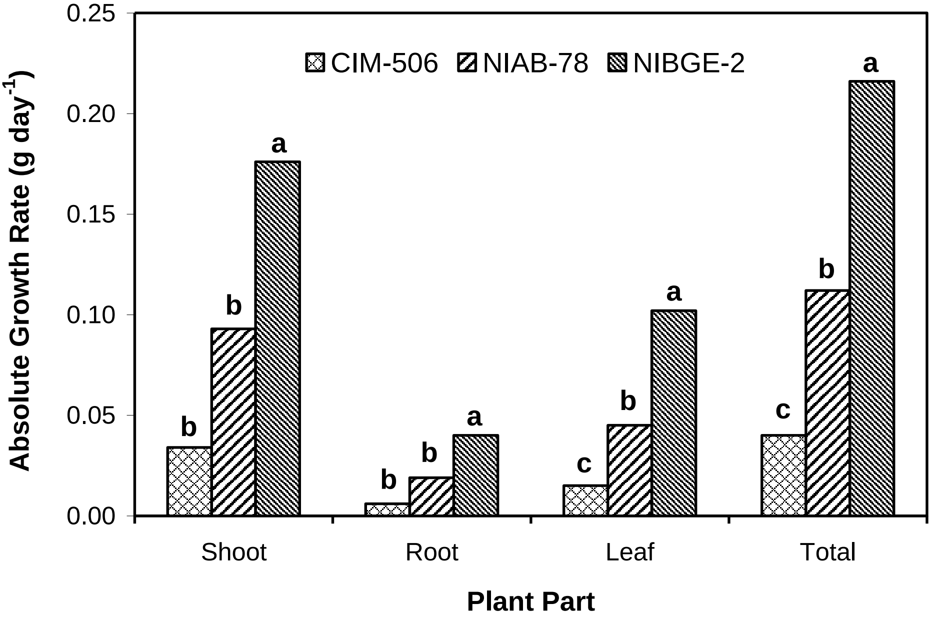

Fig. 2. Absolute growth rate of differentially K-use efficient cotton genotypes under K deficiency stress in hydroponics at seedling stage.

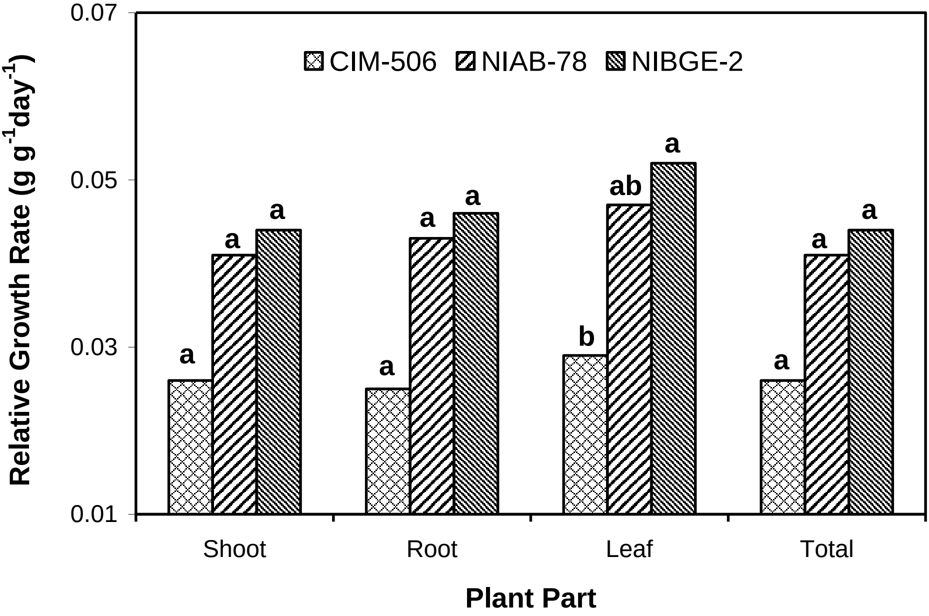

Fig. 3. Relative growth rate of differentially K-use efficient cotton genotypes under K deficiency stress in hydroponics at seedling stage.

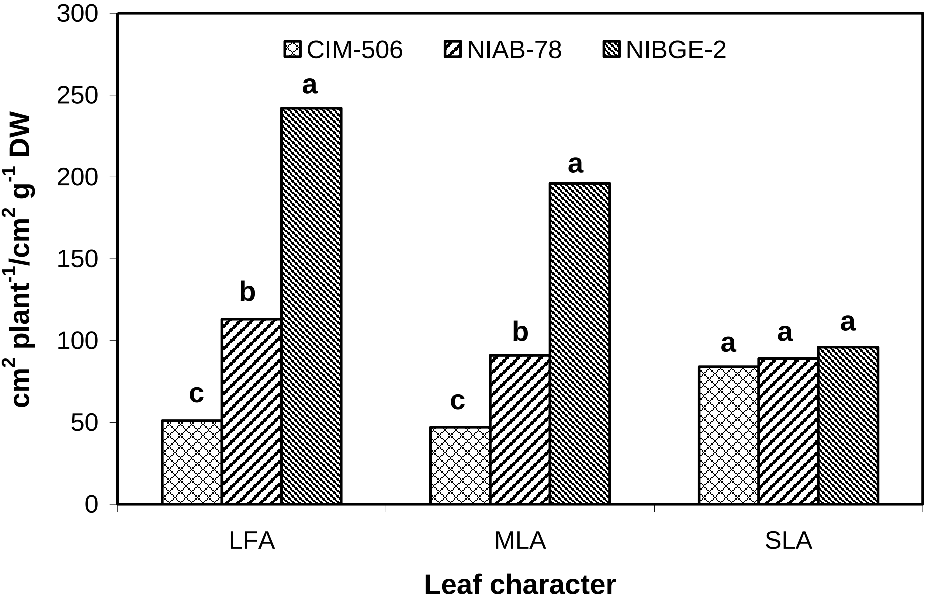

Fig. 4. Leaf characters (LFA: leaf area, MLA: mean leaf area, SLA: specific leaf area) of differentially K-use efficient cotton genotypes under K deficiency stress in hydroponics at seedling stage.

than NIAB-78 and 538% more than CIM-506, whereas, NIAB-78 had 208% more absolute growth rate of root than CIM-506. The absolute growth rate of leaf of NIBGE-2 was 125% more than NIAB-78 and 565% more than CIM-506, whereas, NIAB-78 had 125% more absolute growth rate of leaf than CIM-506. The absolute growth rate on total basis of NIBGE-2 was about 92% more than NIAB-78 and 434% more than CIM-506, whereas, NIAB-78 had 177% more absolute growth rate on total basis than CIM-506 (Fig. 3). The relative growth rate of cotton genotypes was significant only in case of leaf. It was highest for NIBGE-2 while the other two genotypes were non-significant to each other for this parameter. The relative growth rate of shoot of NIBGE-2 was only 8% more than NIAB-78, however, it was 66% more than CIM-506, whereas, NIAB-78 had a 54% more relative growth rate of shoot than CIM-506 (Fig. 4).

Discussion

Genotypic variation for K nutrition has been reported in many crops, e.g., maize (Nawaz *et al*., 2006), rice (Yang *et al*., 2003), and wheat (Damon & Rengel, 2007). The differential adaptation of cotton genotypes to adequate and deficient K nutrition has also been reported by many workers (Cassman *et al*., 1989; Dong *et al*., 2004; Makhdum *et al*., 2007; Zhang *et al*., 2007) and our results also confirmed the same. The cultivars behaved quite differently to K deficiency and NIBGE-2 performed better in all growth parameters. Understanding of mechanisms or morphological markers is pre-requisite for long term breeding programs to produce more K efficient crop cultivars. Analysis of plant growth in terms of relative growth rates is an explanatory, holistic and integrative approach to interpreting plant form and function (Hunt *et al*., 2002). Relative growth rate provides a convenient integration of the combined performance of various parts of the plant and is termed as efficiency index of a plant as producer of new material (Hunt, 1982). Ahmad *et al*., (2001) reported that cotton genotypes did not differ for their absolute growth rate and relative growth rate at deficient phosphorus level. Our results, in contrast, report that the genotypes differed significantly for their absolute growth rate, however, the genotypes behaved alike for their relative growth rate, except relative growth rate of leaf. This variation might be due to the differences between the physiology of two nutrients under study resulting in comparatively higher requirement of K as against phosphorus for cotton growth and development (Makhdum *et al*., 2005, Pervez *et al*., 2006; Makhdum *et al*., 2007; Pervez *et al*., 2007; Zhang *et al*., 2007). Leaf growth is considered as the most sensitive physiological process to K deficient conditions (Reddy *et al*., 2000). K deficiency reduces cotton leaf area, leaf weight and leaf area index (Cassman *et al*., 1989). A 14% lower leaf growth rates and 59% lower leaf expansion rates were observed in cotton plants with inadequate leaf K compared to adequate (Reddy *et al*., 2000). Potassium deficient cotton leaf possesses less intercellular air space and fewer chloroplasts in mesophyll cells than the control plants. The chloroplasts are filled with large starch granules, and contain apparently more and greater plastoglobuli and fewer grana. A moderate K deficiency drastically reduces the photosynthesis in cotton (Zhao *et al*., 2001), which is associated with decreased stomatal conductance, increased mesophyll resistance, low chlorophyl content, poor chloroplast ultrastructure, restricted saccharide translocation, and decreased synthesis of RuBP carboxylase (Dong *et al*., 2004). The present study also endorsed the role of better root growth in cotton tolerance to K deficiency stress. The importance of root growth and development in K nutrition of cotton is due to its less dense root system (Gerik *et al*., 1987). Diffusion - the typical mechanism supplying K to plant roots – is severely affected under low K conditions, due to low K diffusion coefficient. Hence, under K deficiency, the root growth rate, root length and root surface area become most important for K acquisition of cotton (Dong *et al.*, 2004). It is highly likely that the efficient cotton genotypes with enhanced root biomass may have better root architecture, more fine roots, more root hairs and increased root exudation, as has been reported by Rengel & Damon (2008). Brouder & Cassman (1990) reported that an efficient root system plays an important role in cotton tolerance to K deficiency stress. The correlation of shoot dry weight (Table 1) was highly significant ($p<0.01$) with various characters in order of mean leaf area (0.95), leaf dry weight (0.94), leaf area (0.93), root dry weight (0.90), absolute growth rate of shoot (0.88), absolute growth rate of leaf (0.85), absolute growth rate on total basis (0.83), absolute growth rate of root (0.71), while significant ($p<0.05$) with relative growth rate of leaf (0.49), relative growth rate of root on total basis (0.49), relative growth rate of shoot (0.45), and non-significant ($p>0.05$) with relative growth rate of root and specific leaf area. Thus, it revealed that enhanced shoot dry weight of cotton genotypes under K deficiency stress was most importantly a function of their leaf characters, i.e., mean leaf area, leaf dry weight, leaf area, absolute growth rate of leaf and relative growth rate of leaf. Next to the photosynthetic apparatus of cotton genotypes, the root characters viz., root dry weight and absolute growth rate of root played important role in determining the shoot dry weight of cotton genotypes under K deficiency stress. In conclusion, cotton tolerance to K deficiency stress is influenced by the genotypic variation, depending upon well developed photosynthetic apparatus (fully expanded leaves) and efficient root system. Hence, these two characters could be considered the most important morphological markers for breeding K-efficient cotton genotypes.

Table 1. Correlation coefficient (r) between shoot dry weight of cotton genotypes and various plant characters under K deficiency stress.

Plant part	r-value	P-value
	Dry weight	
Root	0.90	0.0000
Leaf	0.94	0.0000
	Absolute growth rate	
Shoot	0.88	0.0000
Root	0.71	0.0001
Leaf	0.85	0.0000
Total	0.91	0.0000
	Relative growth rate	
Shoot	0.45	0.0300
Root	0.24	0.2513
Leaf	0.49	0.0140
Total	0.49	0.0146
	Leaf character	
Leaf area	0.93	0.0000
Mean leaf area	0.95	0.0000
Specific leaf area	0.22	0.3078

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