J. Cotton Res. Dev. **29** (1) 65-70 (January, 2015)

Water deficit conditions affect carbon accumulation, photosynthetic and yield contributory parameters of G*ossypium hirsutum* L. and *Bt* cotton

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ABSTRACT: *Gossypium hirsutum* L. (LH 2076) and *Bt* cotton (MRC 7017 BG II) were selected to investigate the effect of water deficit stress on photosynthetic parameters as well as some yield contributory parameters. The role of Corona (CR) on photosynthetic parameters, osmotic adjustments of seedlings under stress (-0.4 and -0.6 MPa) and on yield contributory parameters was evaluated. The contents of chlorophyll (a, b and total) decreased under water deficit conditions. Foliar application of CR increased contents of chlorophyll in both varieties. Carotenoid content was recorded more under stress, CR application further increased it. The decrease in Hill reaction activity under stress resulted in decreased starch content of leaves. Soluble sugar accumulation under stress was improved by CR application that helped in effective osmoregulation. More bolls/plant and boll setting per cent were recorded under control in MRC 7017 as compared with LH 2076; reduced irrigation decreased these in both. Foliar application of CR decreased this adverse effect to some extent and application at 25 DAS was being more effective.

Key words : Boll setting, *Bt* cotton, carbohydrate level, *Gossypium hirsutum,* photosynthetic parameter, water deficit stress

Water is the most common environmental factor that limits crop productivity. Since water resources are limited, efficient use of water volume unit (WVU) is necessary in crop production. The severity and extent of drought depends on hydrometerological factors like soil moisture, infiltration and moisture retention capacity of soil. Water depletion affects cotton growth and the cost of water application and the competitive demands further enhance the attractiveness of water efficient cotton. For each species there is a critical level of water potential below which germination will not occur. This particular character determines the plant stand especially under unfavorable environmental condition such as water stress. PGRs play a key role in internal control mechanism of plant growth by interacting with key metabolic processes such as nucleic acid and protein synthesis.

Photosynthesis plays a major role in determining crop productivity in all plant species and is directly affected by water stress. A major effect of drought is reduction in photosynthesis due to impaired photosynthetic machinery, leaf senescence and is associated with reduction in productivity. Plant growth regulators (PGRs) such as gibberellins are known to regulate storage mobilization and then affect stem elongation, flower initiation, pollen and fruit development (Thomas and Hedden, 2006). The plant extract containing proteinaceous compounds and gibberellins as active ingredients (CR) was presently tested to study its effect on photosynthetic parameters and boll production in selected cotton varieties.

MATERIALS AND METHODS

The seeds of G*ossypium hirsutum L.* (LH2076) and *Bt* cotton (MR 7017 BG II) varieties were procured from the Cotton Section of Department of Plant Breeding and Genetics, PAU, Ludhiana. Seeds were surface sterilized and then treated with Emisan (100mg/100 ml distilled water) for 4h for delinted seeds and for 6-8h for linted seeds to avoid any fungal infection during seed germination. All seeds were germinated

under controlled conditions in petri dishes sterilized in an oven at 100ºC for 1h and lined with autoclaved circular blotting papers. On third day, seedling were shifted to new petri dishes containing blotting sheets wetted with polyethylene glycol (PEG 6000) solutions providing osmotic stress of -0.4 and -0.6 MPa. Distilled water was added to control petri dishes and is indicated as zero stress. Solutions of polyethylene glycol (PEG 6000) for different osmotic concentrations were prepared. After ten days of growth under irrigated and different stress conditions, Corona (CR) @ 2µl/ml and 3µl/ml was sprayed with the help of automizer.

The photosynthetic pigments were extracted from the leaves in 5 ml of the dimethylsulfoxide (DMSO) with extraction period of 12 h in the dark. The concentration of the extracted pigments was calculated from the absorbance values at 665,645 and 480 nm. The chlorophyll content was expressed as chl g/mg fresh weight. Intact chloroplasts were obtained from pre weighted leaves of 15 days old seedlings by gently grinding in extraction medium (0.067 M) phosphate buffer (pH 5.5 containing 0.35 M sucrose) to determine Hill reaction activity. Temperature during extraction was maintained at 0-4ºC and chloroplast suspension was used for ferricyanide reduction. The Hill reaction activity was expressed as decrease in absorbance mg/ chlorophyll/ h.

Soluble sugars were estimated and the quantity of sugars was calculated against the standard curve prepared by using pure glucose (10-100µg) and expressed as mg/g dry weight. Starch content was estimated by taking 0.5 ml diluted extact in a test tube and added 4.5 ml of distilled water and 10 ml of cold anthrone sulphuric acid reagent (200 mg of anthrone in 100 ml of cold 95 per cent H_2SO_4, stored at 0ºC) in an ice bath. Heated for 8 min at 100ºC, cooled to room temperature and read absorbance at 630 nm.

Sowing of varieties *Gossypium hirsutum* L. (LH 2076) and *Bt* cotton (MRC 7017 BG II) was done in the Experimental Area of Department of Botany. Cotton crop was raised and treatments were allotted in split plot design. Irrigation was done regularly in control sets but for water deficit conditions, 50 per cent irrigation was reduced in specific treatments. Corona (CR) was applied in different doses at 25 DAS and 125 DAS. The number of mature bolls from all the picks was counted and the cumulative record was maintained for each selected plant. The bolls resulting from the number of flowers were counted and expressed as boll setting per cent.

RESULTS AND DISCUSSION

Two varieties (MRC 7017 and LH 2076) of cotton were exposed to CR application under different moisture deficit levels (0, -0.4 and -0.6 MPa) and contents of chlorophyll a, b and total were estimated in cotyledonary leaves. The pigment content was recorded lesser in cotyledonary leaves under stress which may cause a decrease in photosynthetic activity. Fig. 1 showed that Chl a content decreased under water deficit as compared to controlled conditions. Foliar application of CR had increased the Chl a content in both cotton varieties and showed more values when higher concentration was applied. When CR was applied to -0.4 and -0.6 MPa stress treated plants, higher Chl a content was recorded with CR@ 3µl/ml. The reduction in photosynthetic activity and increase in leaf senescence are symptoms of water deficit stress which adversely affected cotton yield. The genetic differences exist in rate of carbon assimilation for cotton. Okra shaped leaves had higher specific leaf weight and higher leaf chlorophyll concentration compared to normal leaf cultivars.

Fig.1 depicted a decrease in Chl b concentration under water stress conditions. The chlorophyll b content decreased during drought in other varieties also (Pandey *et al.*, 2003). Foliar application of CR@ 2 and 3µl/ml increased Chl b content under the various moisture stress levels (-0.4 and -0.6 MPa). The difference in treatments among selected varieties were recorded more when CR was applied at higher concentration. There was an increase in total chlorophyll content under stress as compared with control

(Fig 2). Foliar application of CR increased total chlorophyll in both varieties. Interaction between varieties and application of CR at various concentrations significantly affected chlorophyll content of leaves. The carotenoid content increased under different moisture stress levels (-0.4 and -0.6 MPa), but increase was more pronounced in MRC 7017 variety (Fig 2). Foliar application of CR increased the carotenoid content at higher concentration. Application of CR@ 3µl/ml was found to promote accumulation of carotenoids in both varieties.

Photosynthetic pigments are important to plants mainly for harvesting light and production of reducing powers. Both the chlorophyll a and b are prone to drought stress (Farooq *et al.*, 2009) that produced changes in the ratio of chlorophyll a and b and carotenoids Farooq *et al.*, 2009). A reduction in total chlorophyll content was also reported in drought stress (Massacci *et al.*, 2008) indicated that water stress caused significant changes in the grana and stroma lamellae, palisade cell walls, number and size of chloroplasts. Earlier, pigment contents were recorded less in cotyledons and leaves of other genotypes under stress. This fact may explain in part the decrease in the photosynthetic process though chlorophyll b content was increased during drought (Pandey *et al.*, 2003).

Carotenoids have additional roles and partially help the plants to withstand adversaries of drought. Carotenoids are a large class of

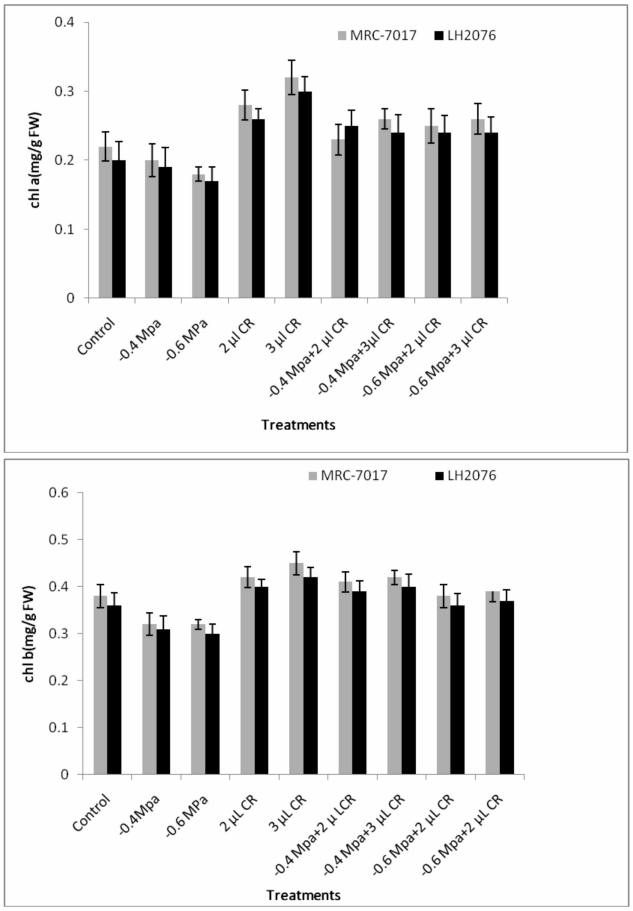

Fig. 1. Effect of CR application on chl a and chl b (mg/g fresh weight) in MRC 7017(*Bt* cotton) and LH 2076 (*Gossypium hirsutum*) at 15 DAS under water deficit conditions.

*Vertical bars indicate standard error

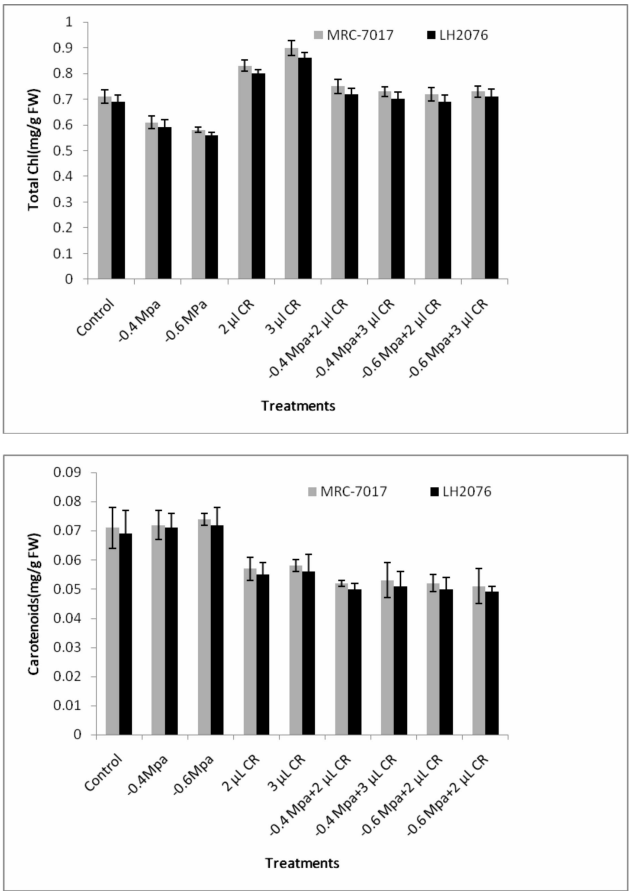

Fig. 2. Effect of CR application on total chlorophyll (mg/g fresh weight) and carotenoids (mg/g fresh weight) contents in MRC 7017 (*Bt* cotton) and LH 2076 (*Gossypium hirsutum*) at 15 DAS under water deficit conditions

*Vertical bars indicate standard error

isoprenoid molecules, which are *de novo* synthesized by all photosynthetic and many non-photosynthetic organisms. They are divided into the hydrocarbon carotenes, such as lycopene and â-carotene or xanthophylls, typified by lutein. Water stress has the ability to reduce the tissue concentration of chlorophylls and carotenoids. Increased contents of carotenoids are important for stress tolerance (Jaleel *et al.*, 2009). Presently, the content was increased in CR treated sets under control as well as stress conditions. Water stress suggestively impaired carotenoid accumulation sooner than the chlorophyll indicating that the metabolic pathway of carotenoid synthesis was sensitive to stress (Ennhali and Earl, 2005). It has been observed that photosynthetic rates are mostly limited by decreased stomatal conductance as well as reduced mesophyll conductance (Flexas *et al.*, 2006) that ultimately resulted in general metabolic impairment due to lower carbon substrate concentration.

Hill reaction activity is one of the most important vital physiological parameters contributing to plant growth and productivity of crop. In other words, photosynthetic capacity in crop plants is the primary component of dry matter productivity. However rate of photosynthesis varies with change in environmental factors, thereby affecting the

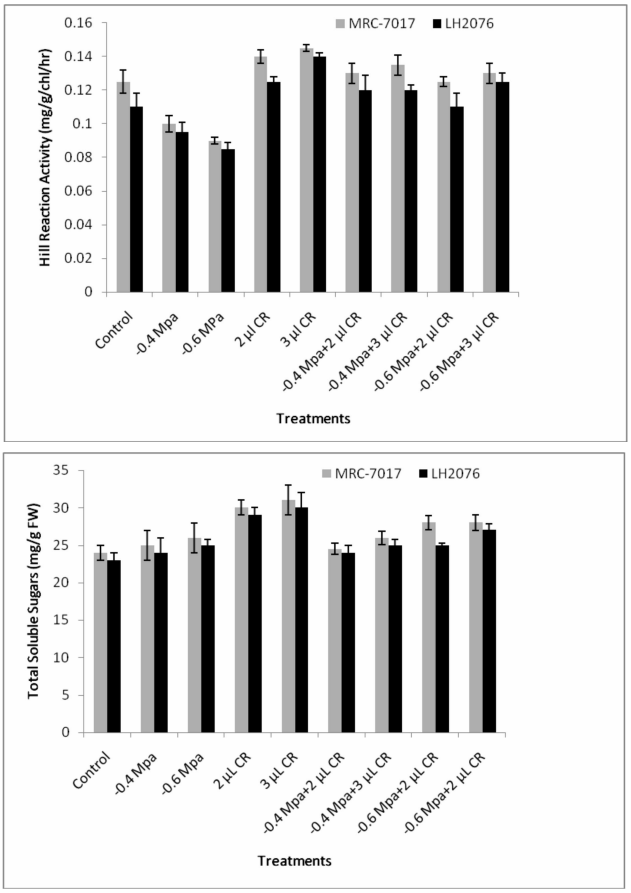

Fig. 3. Effect of CR application on Hill reaction activity (mg/chl/hr) and Total soluble sugars (mg/g fresh weight in MRC 7017 (*Bt* cotton) and LH 2076(*Gossypium hirsutum*) at 15 DAS under water deficit conditions.

*Vertical bars indicate standard error

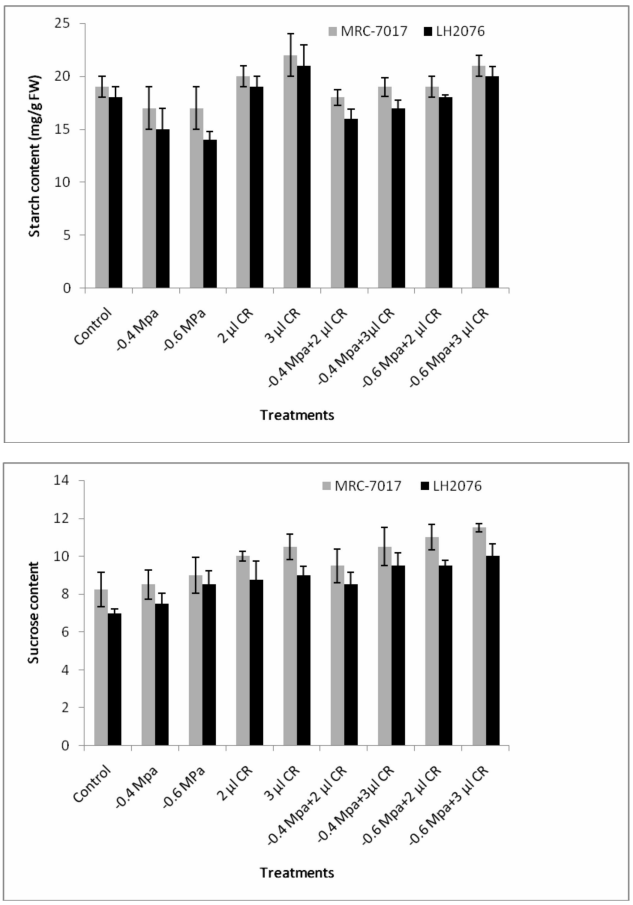

Fig. 4. Effect of CR application on Starch content (mg/g fresh weight) and Surcose content in MRC 7017 (*Bt* cotton) and LH 2076 (*Gossypium hirsutum*) at 15 DAS under water deficit conditions

*Vertical bars indicate standard error

plant growth and yield. A decrease in Hill reaction activity was observed under moisture stress conditions in the selected varieties as compared to controlled condition (Fig 3). Foliar application of CR had increased the Hill reaction activity in both cotton varieties. It has been reported that photosynthetic rates of the leaves decrease as the relative water content and leaf water potential decrease.

Moderate stress (-0.4 MPa) induced accumulation of more total soluble sugars in the seedlings as compared to control. The level of total soluble sugars further increased when water deficit stress was increased to -0.6 MPa (Fig 3). Higher concentration of CR under water deficit levels showed an increase in total soluble sugar content. An increase in sugar level of stressed seedlings might have helped these in effective osmoregulation. Under control conditions, more content of starch was recorded in MRC 7017. The levels of starch content decreased when water deficit stress was increased from -0.4 to -0.6 MPa (Fig 4). Under different water stress conditions, application of CR increased the starch content as compared to controlled condition. The values were always recorded more in MRC 7017 than in LH 2076. The accumulation of sucrose increased with increase in level of water deficit stress in both cotton varieties. CR application improved sucrose accumulation in both control and stress conditions (Fig 4).

Non structural carbohydrates (NSC) are energy sources for maintaining metabolic activity. The formation of NSC is more likely to occur when carbohydrates accumulated at a rate that exceeds which was used for growth. Carbohydrates produced through photosynthesis, are used by the plants for energy production and are required for assimilation of other compounds such as polysaccharides, cellulose, lignins and proteins. Some of the carbohydrates are translocated to storage organs for future use. GA_3 treatment was reported to enhance the reducing sugars and length of seedlings and imposed water stress affected carbohydrate metabolism and its translocation.

The total bolls that cotton plant bears at maturity is an important yield component having the greater effect on yield. This character is greatly influenced both by physiological and environmental factors. Data in Table 1 depicted that higher bolls/plant were recorded under control condition in MRC 7017 as compared with LH 2076. Reduced irrigation decreased the bolls/plant as compared to control. Foliar application of CR increased the bolls/plant. The effect was more promotary at higher concentration indicating that CR helped in overcoming the adverse effects of water stress when applied at different concentrations in both varieties. Both irrigation rate and application type have been shown to affect boll production and retention (Ritchie *et al.*, 2009).

Boll setting per cent was found to be more

Table 1. Effect of CR application on bolls/plant and boll setting per cent in MRC 7017 and LH 2076 under water deficit conditions

Treatments	Bolls/plant		Boll setting (%)	
	MRC 7017	LH 2076	MRC 7017	LH 2076
T_1 (Control)	24.52	20.09	28.23	22.44
T_2 (50% reduced irrigation at 20 and 80 DAS)	23.15	18.71	26.38	21.11
CD (p=0.05)	V=0.2109,T=0.2109,V×T=NS		V=0.2786,T=0.2786,V×T=NS	
T_1+2 µl CR at 25 DAS	26.61	24.30	32.11	27.49
T_1+3 µl CR at 25 DAS	28.10	25.12	32.69	28.21
T_1+2 µl CR at 125 DAS	24.90	23.10	30.51	24.09
T_1+3 µl CR at 125 DAS	25.11	22.16	30.89	26.59
CD (p=0.05)	V=0.1811,T=0.1280,V×T=0.2561		V=0.2236,T=0.1581,V×T=0.3162	
T_2+2 µl CR at 25 DAS	25.53	23.63	32.01	27.33
T_2+3 µl CR at 125 DAS	24.70	21.62	30.12	23.71
CD (p=0.05)	V=0.2692,T=0.2692,V×T=0.3807		V=0.2727,T=0.2727,V×T=0.3856	

in MRC 7017 in control treatments as compared with LH 2076 (Table 1). Reduced irrigation decreased the boll setting per cent in both the selected cotton varieties. Foliar application of CR at different concentrations decreased adverse effect of stress to some extent. Moisture stress reduced the seed cotton yield and yield components (number of bolls and boll weight). Both irrigation rate and application type have both been shown to affect boll production and retention (Dumka *et al.*, 2004; Whitaker *et al.*, 2008; Ritchie *et al.*, 2009). Present study revealed that CR application at 25 DAS partially ameliorates the adverse effect of water deficit stress on boll retention in selected cotton varieties.

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Recieved for publication : March 21, 2013
Accepted for publication : July 17, 2013