Influence of Seed Hardening and Foliar Nutrition on the Seed Yield and Biochemical Parameters of Bt Cotton (RCH-2)

Amit Bijjur1* and K. N. Pawar2
1Ph.D. Department of Crop Physiology
2Professor Department of Crop Physiology
University of Agricultural Sciences, Dharwad-580005
*Corresponding Author E-mail: bijjuramit1617@gmail.com
Received: 22.11.2019 | Revised: 29.12.2019 | Accepted: 7.01.2020

ABSTRACT
A field experiment was conducted at Agricultural Research Station, Dharwad during kharif, 2010-2011 to study the influence of seed pre-sowing hardening with various agro-chemicals (CaCl2, CCC, KNO3, ZnSO4 and water soaking) and foliar nutrition (imposed at 70 and 90 DAS) on the yield and biochemical traits in Bt Cotton (RCH-2) under rainfed conditions. Significant highest yield was recorded in seed hardening @ 2%, and foliar spray @ 0.5% with CaCl2 (2276 kg/ha) followed by seed hardening treatment and foliar spray with ZnSO4 @ 0.5% (2179 kg/ha) as compared to control (1780 kg/ha). The other yield attributing characters viz., plant height (112.7cm), days to 50 % flowering (80.2), days to 50 % boll opening (112.3), dry weight of reproductive parts (159.6g/plant) and number of balls per plant (28.0) increased significantly with the same treatment combination. The significant increase of the biochemical traits viz., proline content, chlorophyll content and chlorophyll stability index in seed hardening at 2% and foliar spray at 0.5% with CaCl2 (109.29mg/g fr.wt., 2.68 mg/g fr.wt and 41.8 respectively) induced the drought tolerance thus increasing the seed yield.

Keywords: Seed hardening, CaCl2, Bt cotton, Foliar spray, proline and chlorophyll stability.

INTRODUCTION
Cotton is the most important cash crop of India. It belongs to the family Malvaceae of cultivated species Gossypium hirsutum. It India, during 2013-14 it was cultivated in an area of 117.27 lakh hectares with production of 390 lakh bales and productivity of 565.36 Kgs per hectare. Cotton accounts for about 45 per cent of the world fibre and supplies 10 per cent world edible oil (Rathore, 2005). In Karnataka, Bt cotton is covering 4.66 lakh ha with 10.15 lakh bales production and 370 kg productivity (Anon., 2011). Though the area of cotton cultivation is increasing the productivity remains almost constant. The area under rainfed cotton predominates over irrigated area and more than 50 per cent of rainfed cotton is grown under low and erratic rainfall conditions.
Besides, the present day cotton varieties, having longer duration of more than 180 days are exposed to more production risks like incidence of disease, pests particularly to bollworms and drought. In addition, premature drying, floral abscission, failure to set boll are some of the factors that leads to poor productivity in Bt cotton. Hence enhancing quality of seed for obtaining better germination and establishing good plant stand is a prime requirement in Bt cotton cultivation. In this view, seed treatment serve as an easy, feasible and economic method which results in modifying the physiological and biochemical nature of seed. Pre-sowing hardening is the result of extensive physiological reorganization induced by dehydration process. Seed treatments serve the purpose of pest control, drought resistance, and disease incidence at least in the early stages of plant growth. Seed hardening accelerates rapid germination and growth rate of seedlings and also it is an important morphological adaptation in plants that helps in drought resistance without loosing productivity. In addition to the designing of seed for enhancing production, the supply of micro nutrients through foliage improves the seed yield. Foliar nutrition is supplementation of nutrients in conjunction with growth stages for improved productivity (Copeland & McDonald, 1995). Foliar application of nutrients during flowering and boll development stage results in effective utilization of the nutrients and enhances the yield. Hence an attempt has been made in the present investigation to evaluate the effect of seed hardening techniques and supply the foliar nutrition on the bio-chemical changes and yield of the Bt cotton (RCH-2).

**MATERIALS AND METHOD**

A field experiment was carried out during kharif season of 2010-2011 at Agricultural Research Station, Dharwad farm under rainfed condition. The trail was laid under RBD with three replications. Seeds were sown by hand dibbling with a spacing of 90 cm × 60 cm. A day before sowing the RCH-2 Bt seeds were soaked separately in 2% CaCl₂, CCC (100 ppm), KNO₃ (0.5%), ZnSO₄ (0.5%) and water at the ratio of 2:1 solution to seeds for 8 hours and seeds were dried under shade and then dried under sun for 1 day prior to sowing. The foliar sprays were given on the 70th and the 90th day of sowing.

Proline content in the leaf tissue was estimated following method as suggested by Bates *et al.* (1973). Total Chlorophyll content in the leaves of different genotypes of cotton was determined calorimetrically as per the DMSO (Di methyl sulfoxide) method of Shoaf and Lium (1976). Green plant pigments are thermo sensitive and degradation occurs when they are subjected to higher temperature. Estimation of Chlorophyll stability index is based on pigment changes induced heating. Chlorophyll destruction commences rapidly at critical temperature at 55 to 56°C. Thus, chlorophyll stability is a function of temperature. This property of chlorophyll stability was found to correlate well with drought resistance. Two clean glass tubes are taken and five grams of representative leaf sample is placed in them with 50 ml of distilled water. One tube is subjected to heat on water bath at 56°C ±1°C for exactly 30 minutes. Other tube is kept as control. The chlorophyll in both the sample is extracted by placing the sample in 7 ml of DMSO at 65°C for 30 min. Decant the supernatant and discard the tissue, make up the volume to 10 ml with DMSO and read the absorbance of the extract at 645 and 663 nm using DMSO as blank.

**RESULTS AND DISCUSSION**

In the present investigation significant differences among the treatments were observed for yield and yield attributing characters of Bt cotton cv. RCH-2 (Table 1). The highest plant height was recorded at seed treatment 2% and foliar spray 0.5% with CaCl₂ (112.7) followed by seed treatment and foliar spray with ZnSO₄ at 0.5% (108.5) and seed treatment and foliar spray with KNO₃ at 0.5%(103.4). The least plant height was recorded with foliar spray at 70 and 90 DAS with CCC (80.6) followed by control (86.4 cm). Manjunath and Dhanoji (2011) also reported on the increase in plant height due to the seed hardening with 2% cacl₂ in chickpea.
The number of days to 50 per cent squaring did not differ significantly among the treatments. But however seed treatment with CaCl$_2$ at 0.5% took the least number of days to 50 per cent squaring (59.70) followed by seed treatment 2% and foliar spray 0.5% with CaCl$_2$ (59.80). The number of days to 50 per cent flowering and days to 50 % boll opening differed significantly among the treatments. Seed treatment 2% and foliar spray 0.5% with CaCl$_2$ took least number of days to 50 per cent flowering (80.20) and 50 per cent boll opening (112.30) followed by seed treatment and foliar spray with ZnSO$_4$ at 0.5% (81.90 & 113.30). While the highest number of days to flowering and boll opening was recorded in control and water soaking. The increased percentage of boll setting from earlier produced flowers increases the seed cotton yield (Ahlawat, 1979, Fry, 1985 and Yu and Huang, 1990).

Dry matter production is an important parameter that determines yield in crops and that of reproductive parts particularly is an important yield contributing character. It is documented that seed hardening treatments, use of growth regulators and chemicals will have their influence on the production of dry matter and the way in which it is partitioned between different parts of the plant. In the present investigation, the reproductive parts dry matter was highest (159.6 g/plant) in seed treatment @ 2% and foliar spray @ 0.5% with CaCl$_2$ followed by seed hardening and foliar spray with ZnSO$_4$ at 0.5% (154.28g/plant) . The least reproductive parts dry matter accumulation was observed in control (102.42 g/plant). Yield improvement in any crop could be attributed to the better partitioning of photo assimilates towards reproductive economic sink. These results are in concurrence with Arjunan and Srinivasan (1989), whose results also revealed that seed treatment with CaCl$_2$ at 1% significantly increased total dry matter produced in groundnut. The number of bolls per plant differed significantly among the treatments. Number of bolls per plant was highest in seed treatment 2% and foliar spray 0.5% by CaCl$_2$ (28.00) and seed treatment and foliar spray with ZnSO$_4$ at 0.5% (26.00) followed by 100 ppm seed treatment and foliar spray with CCC at 20 ppm (24.00) and seed treatment and foliar spray with 0.5% KNO$_3$ (23.00). The least number of bolls per plant was recorded in control (16.00) followed by water soaking (18.00). The increase in Bt cotton yield was significantly more in seed hardening with CaCl$_2$ at 2% and foliar spray at 0.5% (2276Kg/ha) followed by seed treatment with ZnSO$_4$ at 0.5% and foliar spray at 0.5% (2179Kg/ha). Calcium serves as an important membrane stabilizer and specific activator for certain enzyme synthesis (Subbaraman and Selvaraj, 1989). It increases chlorophyll content which leads to better photosynthesis (Muhammad Iqbal et al., 2006). Arjunan and Srinivasan, 1989 reported that seed hardening with CaCl$_2$ 1% recorded significant higher number of pods per plant and pod yield in groundnut.

Proline plays an important role as storage compound for carbon and nitrogen, detoxification of NH$_3$, preserving the hydration of proteins in dehydrated tissue, thereby contributing to the survival of cellular functions. In the present study, the proline content was significantly highest (109.29 mg/g fr. wt) in seed treatment @ 2% and foliar spray @ 0.5% with CaCl$_2$. These results are in accordance with Patil (1987), who reported that seed treatment with CaCl$_2$ at 2% significantly increased the proline content in sorghum. Similar results were reported by Amaregowda et al. (1994) in wheat.

The total chlorophyll content determines the photosynthetic capacity and influences the rate of photosynthesis dry matter production and the yield (Krasichkova et al., 1989). In the present study, the chlorophyll content (2.68 mg/g fr. wt) and chlorophyll stability index (41.8) was recorded highest in seed treatment with CaCl$_2$ at 2% and foliar spray at 0.5% and lowest was recorded in control. The results were in accordance with Kadiri and Hussaini (1999) who reported that seed treatment with 100ppm CaCl$_2$ significantly increased chlorophyll stability index in sorghum. Manjunath and Dhanoji (2011) also reported that seed hardening with CaCl$_2$ at 2% increased chlorophyll stability index in chickpea.
Fig. 1: Influence of seed hardening and foliar nutrition on Bt-cotton yield

Table 1: Influence of seed hardening and foliar nutrition on yield (kg/ha) and attributing characters

| Treatments                  | Plant height (cm) | Days to 50% squaring | Days to 50% flowering | Days to 50% boll opening | Dry weight of reproductive parts (g/plant) | No. of bolls/plant | Yield (kg/ha) |
|-----------------------------|-------------------|-----------------------|------------------------|--------------------------|------------------------------------------|------------------|---------------|
| T1                          | 100.3             | 59.70                 | 88.30                  | 118.30                   | 128.41                                   | 22.00            | 1968          |
| T2                          | 92.6              | 62.10                 | 90.40                  | 119.80                   | 126.83                                   | 21.00            | 1903          |
| T3                          | 96.0              | 62.60                 | 90.80                  | 119.90                   | 125.32                                   | 21.00            | 1877          |
| T4                          | 97.2              | 60.90                 | 89.90                  | 119.40                   | 127.22                                   | 22.00            | 1939          |
| T5                          | 112.7             | 59.80                 | 80.20                  | 112.30                   | 159.60                                   | 28.00            | 2276          |
| T6                          | 95.2              | 61.90                 | 82.60                  | 114.20                   | 148.97                                   | 24.00            | 2102          |
| T7                          | 103.4             | 62.80                 | 84.50                  | 115.30                   | 141.23                                   | 23.00            | 2013          |
| T8                          | 108.5             | 60.60                 | 81.90                  | 113.30                   | 154.28                                   | 26.00            | 2179          |
| T9                          | 94.2              | 62.10                 | 86.30                  | 116.40                   | 129.32                                   | 21.00            | 1958          |
| T10                         | 80.6              | 62.30                 | 88.20                  | 117.90                   | 127.32                                   | 20.00            | 1892          |
| T11                         | 92.2              | 62.40                 | 89.60                  | 118.20                   | 124.61                                   | 20.00            | 1846          |
| T12                         | 92.9              | 62.20                 | 87.40                  | 117.60                   | 128.22                                   | 20.00            | 1927          |
| T13                         | 90.1              | 60.80                 | 92.60                  | 115.60                   | 112.63                                   | 20.00            | 1803          |
| T14                         | 86.4              | 62.30                 | 90.80                  | 114.30                   | 102.42                                   | 16.00            | 1780          |
| Mean                        | 95.9              | 61.46                 | 87.39                  | 116.61                   | 131.17                                   | 21.57            | 1962          |
| SEM*                        | 4.0               | 2.21                  | 2.66                   | 1.96                     | 5.50                                     | 0.81             | 33.69         |
| CD @ 5%                     | 11.7              | NS                    | 7.72                   | 5.70                     | 15.98                                    | 2.35             | 98            |

Treatments

T1 - Seed treatment with CaCl₂ @ 2%
T2 - Seed treatment with CCC @ 100 ppm
T3 - Seed treatment with KNO₃ @ 0.5%
T4 - Seed treatment with ZnSO₄ @ 0.5%
T5 - Seed treatment with CaCl₂ @ 2% and Foliar spray with CaCl₂ @ 0.5% at 70 and 90 DAS
T6 - Seed treatment with CCC @ 100 ppm and Foliar spray with CCC @ 20 ppm at 70 and 90 DAS
T7 - Seed treatment with KNO₃ @ 0.5% and Foliar spray with KNO₃ @ 0.5% at 70 and 90 DAS
T8 - Seed treatment with ZnSO₄ @ 0.5% and Foliar spray with ZnSO₄ @ 0.5% at 70 and 90 DAS
T9 - Foliar spray with CaCl₂ @ 0.5% at 70 and 90 DAS
T10 - Foliar spray with CCC @ 20 ppm at 70 and 90 DAS
T11 - Foliar spray with KNO₃ @ 0.5% at 70 and 90 DAS
T12 - Foliar spray with ZnSO₄ @ 0.5% at 70 and 90 DAS
T13 - Water soaking
T14 - Control

Copyright © July-August, 2020; IJPAB
Table 2: Influence of seed hardening and foliar nutrition on bio-chemical parameters of Bt-cotton

| Treatments | Proline content (mg/g fr. wt.) | Total chlorophyll content (mg/g fr. wt.) | Chlorophyll stability index |
|------------|--------------------------------|----------------------------------------|-----------------------------|
| T1         | 102.31                         | 2.27                                   | 37.9                        |
| T2         | 101.43                         | 2.08                                   | 36.0                        |
| T3         | 100.67                         | 2.01                                   | 35.2                        |
| T4         | 102.09                         | 2.19                                   | 36.4                        |
| T5         | 109.29                         | 2.68                                   | 41.8                        |
| T6         | 95.73                          | 2.31                                   | 40.0                        |
| T7         | 93.29                          | 2.22                                   | 39.3                        |
| T8         | 97.64                          | 2.47                                   | 40.4                        |
| T9         | 104.62                         | 1.96                                   | 37.4                        |
| T10        | 102.26                         | 1.82                                   | 34.1                        |
| T11        | 101.11                         | 1.79                                   | 33.0                        |
| T12        | 104.28                         | 1.90                                   | 36.1                        |
| T13        | 96.92                          | 1.81                                   | 30.1                        |
| T14        | 98.79                          | 1.72                                   | 28.4                        |
| Mean       | 100.75                         | 2.088                                  | 36.2                        |
| SEm+       | 3.76                           | 0.078                                  | 1.3                         |
| CD @ 5%    | 10.94                          | 0.227                                  | 3.9                         |

Treatments
T1 - Seed treatment with CaCl₂ @ 2%
T2 - Seed treatment with CCC @ 100 ppm
T3 - Seed treatment with KNO₃ @ 0.5%
T4 - Seed treatment with ZnSO₄ @ 0.5%
T5 - Seed treatment with CaCl₂ @ 2% and Foliar spray with CaCl₂ @ 0.5% at 70 and 90 DAS
T6 - Seed treatment with CCC @ 100 ppm and Foliar spray with CCC @ 20 ppm at 70 and 90 DAS
T7 - Seed treatment with KNO₃ @ 0.5% and Foliar spray with KNO₃ @ 0.5% at 70 and 90 DAS
T8 - Seed treatment with ZnSO₄ @ 0.5% and Foliar spray with ZnSO₄ @ 0.5% at 70 and 90 DAS
T9 - Foliar spray with CaCl₂ @ 0.5% at 70 and 90 DAS
T10 - Foliar spray with CCC @ 20 ppm at 70 and 90 DAS
T11 - Foliar spray with KNO₃ @ 0.5% at 70 and 90 DAS
T12 - Foliar spray with ZnSO₄ @ 0.5% at 70 and 90 DAS
T13 - Water soaking
T14 - Control

CONCLUSION
It can be concluded from the study that the seed hardening @ 2% and foliar spray @ 0.5% with CaCl₂ significantly enhanced the yield of Bt cotton. The significantly increase in the proline content, chlorophyll content and the chlorophyll stability index were found to enhance the resistance of the plant to adverse environmental conditions especially the drought and promote the productivity. Hence these seed hardening techniques could be used practically to enhance the productivity of cotton under rainfed conditions.

REFERENCES
Ahlawat, I. A. S. (1979). flowering and fruiting behavior in cotton. *Indian J. Agron.*, 25 : 445-448.

Amaregowda, A., Chetti, M. B., & Manjunath, S., 1994, Physiological basis of yield variation due to application of different chemicals in wheat. *Ann. Plant Physiol.*, 8, 24-28.

Anonymous, (2011). All India Coordinated Cotton Improvement Project (AICCIP). *Annu. Rep.*, 2010-11.

Arjunan, A., & Srinivasan, P. S. (1989). Pre-sowing seed hardening for drought tolerance in groundnut (*Arachis hypogaea* L.). *Madras Agric. J.*, 16, 523-526.

Bates, L. S., Waldren, R. P and Terre, I. D., 1973, Rapid determinations of free proline in water stress studies. *Plant and Soil*, 39, 205-208.
Copeland, L. O. (1988). Principles of Seed Science and Technology, Mc Millan Publishing Company, New York.

Fry, R. F. (1985). Earliness factors in three pima cotton genotypes. Crop Sci., 25, 1020-1023.

Kadiri, M., & Hussaini, M. A., (1999), Effect of hardening pre treatments on vegetative growth, enzyme activities and yield of Pennisetum americanum and Sorghum bicolor. Global J. Pure and Appl. Sci., 5, 179-183.

Krasichkova, G. V., Asoeva, L. M., Giller, Yu, E., & Singinov, B. S. (1989). Photosynthetic system of G. barbadense at the early stages of development. Doklady vsesovaznoi ordena Trudovogo krasnogo Znameni Akademii Sel Skokhozya itrvennykh Nauk Imen V. I. Lenina. 12, 9-11.

Majunath, B. L., & Dhanoji, M. M. (2011). Effect of seed hardening with chemicals on drought tolerance traits and yield in chickpea. J. Agric. Sci., 3(3).

Muhammad Iqbal, Mahammad Ashraf, Amer Jamell and Shafiq-ur-Rehman, (2006). Does seed priming induce changes in the levels of some endogenous plant hormones in hexaploid wheat plants under salt stress. J. Integrative Pl. Bio. 48(2), 181-189.

Patil, K. B., Adsule, R. N., & Kachare, D. P. (1999). Influence of foliar spray of methanol on growth, nutrient uptake, yield and quality of groundnut. J. Maharashtra Agric. Univ., 24, 127-130.

Rathore, P. S. (2005). Techniques and Management of Field Crop Production, Agrobios (India) Publishers Jodhpur.

Subbaraman, R and Selvaraj, J. A. (1989). Effect of pre-sowing treatments on seed yield and quality in groundnut JL-24. Seed & farms, 4, 5-9

Yu, S. X., & Huang, Z. M., (1990). Inheritances analysis on earliness components of short season cotton varieties in a G. hirsutum L. Scientia Agric. Sinica, 23, 48-54.