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Performance of polymer-coated cotton seeds under various moisture stress conditions

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ABSTRACT

Cotton is an important fiber crop in the world and it is mainly cultivated in dryland conditions. Due to climate change, drought is occurs frequently. Even though cotton is cultivated in dryland conditions it is sensitive to drought from germination to harvest. Akshay 65 F1Bt cotton seeds polymer coated with Arcus, Myconate, Genius coat 171, Quick roots, and Genius coat 172 were grown in the field of the Department of Seed Science and Technology, Tamil Nadu Agricultural University (TNAU), Coimbatore, to evaluate the impact of polymers on the physiological parameters under various moisture stress conditions in addition to normal irrigation. Moisture stress was created during the 50% flowering stage, boll initiation stage, and stress at both the 50% flowering and boll initiation stages. Among the moisture stress conditions, moisture stress at 50% blooming and boll initiation stages is critical for the irrigation of the cotton plant. The seeds coated with Quick roots performed better than other polymers and recorded higher physiological parameters such as soluble protein content (7.3 mg/g), leaf area index (2.35), nitrate reductase activity (35.1 mg NO_2/g/h), chlorophyll stability index (73%), and relative water content (72%) in both regular irrigation and various water stress conditions. Polymers are act as a drought-tolerant chemical, and therefore, it can be recommended to increase cotton yield in dryland conditions.

ARTICLE HIGHLIGHTS

- Cotton crop is sensitive to moisture stress at both 50% blooming and boll initiation stages.
- Quick roots polymer coated cotton seeds performed better in normal irrigation and moisture stress conditions.

1. INTRODUCTION

Cotton is the most important cash crop grown in all cotton-growing regions of India and other countries around the world [1]. According to FAO statistics (https://www.fao.org/faostat/en/#data/QCL), China, India, the United States, Brazil, Pakistan, Uzbekistan, Turkey, and Australia are the top eight cotton-producing countries in the world. India accounts for approximately 25% of total global cotton production. It is also known as the “King of fibers” and “White of gold” [2]. Cotton is vital to the livelihoods of an estimated 6 million cotton farmers and 40–50 million people who are employed in related activities such as cotton processing and trade. Cotton fiber and seeds are used in the textile and oil industries as well as for animal feed. Cotton seeds contain a sizable amount of protein (20–40%) and oil (18–24%).

Cotton is sensitive to biotic and abiotic stresses from germination to harvest. Cotton has been further damaged by climate change, putting it in peril of a hostile environment [3]. Crop growth and performance are affected by prolonged heat and moisture stress. Despite using high-quality cotton seeds for planting in the field, they were subjected to several challenges during emergence and establishment, which resulted in low survival and a reduced plant stand. Water stress significantly impacts numerous physiological growth mechanisms during plant life cycles [4]. During the flowering and boll initiation stages, cotton plants demand more water for irrigation [5]. Due to pollen development, the growth of anthers, and sensitivity of fertilization, cotton’s flowering stage is most vulnerable to water stress, which in turn reduces seed output. Besides, during the late flowering stage, pressure caused by a water shortage reduced the blooming rate and boll retention [6]. Withholding irrigation at the boll formation stage

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recorded lesser seed cotton yield [7]. Furthermore, water deficit during the flowering and boll opening stage reduced the seed cotton yield, fiber yield, and ginning percentage [8].

Coating the seeds with polymers is one method to improve a plant's ability to absorb water or its tolerance to drought [9]. Adding polymers to seeds results in desirable seed attributes such as faster water intake and enhanced germination, which are beneficial for better emergence and establishment in undesirable environments. Polymer seed coating is a promising technique for keeping high moisture potential around developing seeds and ensuring soil moisture content does not fall below a critical level. By collecting water from the rhizosphere up to 100–1000 times their weight over time, polymers act as a local reservoir. Then water is released gradually from the reservoir to into the soil until it reaches the plant [10].

Polymer fine particles are a "wick" or moisture-attracting substance to increase soil-seed contact [11]. The polymer coating on the seed functions as a temperature switch and acts as a protective covering by regulating the water intake of the seed coating until the soil temperature reaches a predetermined temperature. Seeds coated with polymer might protect against water stress, whereas hydrophilic polymers are mostly employed to speed up the rate of water intake [12].

2. MATERIALS AND METHODS

Polymer materials such as Arcus, Myconate, Genius coat 171, Quick roots, and Genius coat 172 were machine coated onto Akshay 65 F1Bt cotton seeds [Figure 1]. The cotton seeds coated with different polymers were grown under several water stress conditions, including stress during 50% blooming, boll initiation, and 50% blooming and boll initiation phases, coupled with regular irrigation. The physiological parameters comprising the soluble protein content, leaf area index, nitrate reductase activity, chlorophyll stability index (CSI), and relative water content (RWC) were measured.

2.1. Leaf Area Index (LAI)

On 120 DAS, the maximum length and width of the third leaf were measured on 20 plants chosen randomly from each plot. The total number of plant^{-1} leaves was also counted. The leaf area index was computed using the following formula [13].

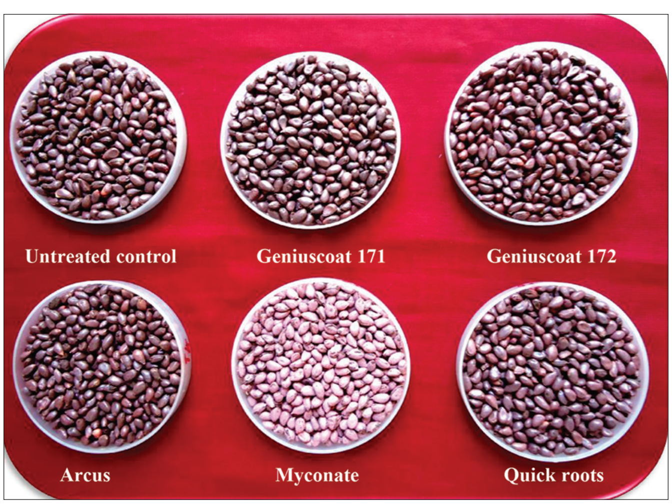

Figure 1: Polymer coated cotton seeds.

$$LAI = \frac{Length\ (cm)\ \times\ Width\ (cm)\ \times\ No.\ of.\ leaves\ \times\ 0.775}{One\ plant\ occupied\ land\ area\ (in\ cm^2)}$$

2.2. RWC

Twenty leaf discs were collected randomly from each treatment and their fresh weight was taken. The weighted leaf disc was floated in distilled water for 4 h to achieve turgidity. After 4 h, the leaf discs were removed, and the excess moisture was removed with blotter paper before measuring the leaf's turgid weight. The leaves were dried in a hot air oven for 48 h at 72°C. Then the dry weight of the leaf was recorded [14].

$$Relative Water Content\ (\%) = \frac{\begin{pmatrix} Fresh\ Weight\ of\ leaf\ disc \\ -\ Dry\ Weight\ of\ leaf\ disc \end{pmatrix}}{\begin{matrix} Turgid\ Weightof\ leaf\ disc \\ -Dry\ Weight\ of\ leaf\ disc \end{matrix}} \times 100$$

2.3. Chlorophyll Stability Index (CSI)

Samples of cotton leaves (250 mg) were collected from each treatment plot and placed in test tubes with 10 mL of distilled water. The test tubes were placed in a 65°C water bath for 30 min. Concurrently, the control tube was kept at the ideal room temperature. Immediately, after the reaction time, a 250 mg leaf sample was taken and macerated with 10 mL acetone (80%). The supernatant was added to a 25 mL volumetric flask filled with 80% acetone. The color intensity was determined in a spectrophotometer at 652 nm [15].

$$Chlorophyll\ stability\ index(\%) = \frac{Total\ leaf\ chlorophyll\ content\ in\ treatment}{Total\ leaf\ chlorophyll\ content\ in\ control} \times 100$$

$$Total\ chlorophyll\ content\ (mg\ /\ g) = \frac{OD \times Volume\ of\ the\ supernatant\ (ml)}{34.5\ \times\ Fresh\ Weight\ of\ leaf\ sample} \times 100$$

2.4. Soluble Protein Content

The soluble protein content of cotton leaf samples was determined [16].

Reagent A – 2 % sodium carbonate in 0.1 N sodium hydroxide

Reagent B – 0.5 % copper sulfate solution in 1 % potassium sodium tartrate

Reagent C – mix 50 mL of A and 1 mL of B before use

Reagent D – Folin–Ciocalteau reagent

A 500 mg sample of fresh cotton leaves was collected from each plot and homogenized with phosphate buffer (10 mL). After centrifugation, the leaf tissues were homogenized for 20 min at 3000 rpm. After taking 0.5 mL of the supernatant, the volume was increased to 1 mL by adding water. Reagent C (5 mL) was added, and it was allowed to settle for 10 min. After that, reagent D (0.5 mL) was added, thoroughly mixed, and incubated for 30 min in the dark to produce a blue color. The intensity of color developed was measured at 660 nm using a spectrophotometer (ELICO SL 159). The standard graph was plotted, and the soluble protein content in cotton leaf samples from different treatments is expressed in mg/g of cotton leaf sample and standard graph (fresh weight).

2.5. Nitrate Reductase Activity (NRA)

Nitrate reductase in the cotton leaf samples was also determined [17]. A leaf sample weighing 500 mg was cut into small pieces. The leaf bits were taken in a test tube with 10 mL of assay medium (8 mL of 0.2 M NaH_2PO_4 + 42 mL of Na_2HPO_4 + 1.01 g of KNO_3 + 1 mL isopropanol and made up to 100 mL). The test tube was placed in a desiccator for 5 minutes to extract the enzyme from the cell, and then the test tube was kept in the dark for an hour to allow the reaction to occur. After an hour, an aliquot (2 mL) was added to a test tube along with zinc acetate (1 mL) and ethanol (2 mL). The sample was filtered using Whatman filter paper. The filtrate was mixed with sulfanilamide (1 mL) and 0.02% NEDH (1 mL) and then the OD value was determined in a spectrophotometer at 540 nm. The OD value was plotted in a standard graph.

KNO_2 was used to make a 50 ppm stock solution, and from which successive solutions of 5, 10, 15, 20, 25, 30, 35, 40, 45, and 50 ppm were made by diluting the stock solution. Run the standard series in the same manner. The sample's OD value was plotted on a standard graph, and the corresponding concentration (X g) was determined.

$$\textit{Nitrate reductase activity} = \frac{}{2} \times \frac{10}{500} \times 1000$$

NRA in the sample was expressed in μg NO_2/g/h.

A split-plot design was used to raise three replications of plants in the field.

2.6. Statistical Analysis

The recorded field experiment values were analyzed [18]. The critical differences have been determined at a 5% probability level. If the F test produced a nonsignificant variation, the results were denoted as NS.

3. RESULTS AND DISCUSSION

Numerous physiological processes are harmed by water stress, as evidenced by measurements of the CSI, nitrate reductase activity, leaf area index, soluble protein, and RWC. A drop in the leaf area index was noted due to a reduction in water availability. The leaf area index is a crucial source of photosynthesis for determining dry matter accumulation and crop yield. When plants are exposed to moisture stress, the leaf area index decreases due to a rapid decrease in leaf elongation and increased leaf mortality. The primary factor influencing the seed cotton yield is the net photosynthetic rate, which is increased by an increase in the leaf area index. A higher leaf area index in the vegetative stage grew steadily during the flowering stage before decreasing at the maturity stage [19]. In the current study, water stress during 50% blooming and boll initiation phases decreased the physiological parameters of the plant. When seeds were coated with Quick roots, a greater leaf area index at the maturity stage was attained under both regular irrigation and moisture stress conditions. Regardless of moisture treatments, seedlings coated with Quick roots at maturity exhibited larger leaf area indices (2.35) than untreated control seeds (2.06). When compared to the stress, skipping watering at both the 50% blooming and boll initiation stages (2.09) recorded a lower leaf area index than not watering at the boll initiation stage (2.24). In compared to the untreated control (1.94), the Quick roots coated seeds (2.25) had the highest leaf area index at maturity under stress during the 50% blooming and boll initiation stages [Figure 2]. The impact of polymer coating, which can enhance growth under drought conditions, is linked to an increased leaf area index. The outcomes are comparable

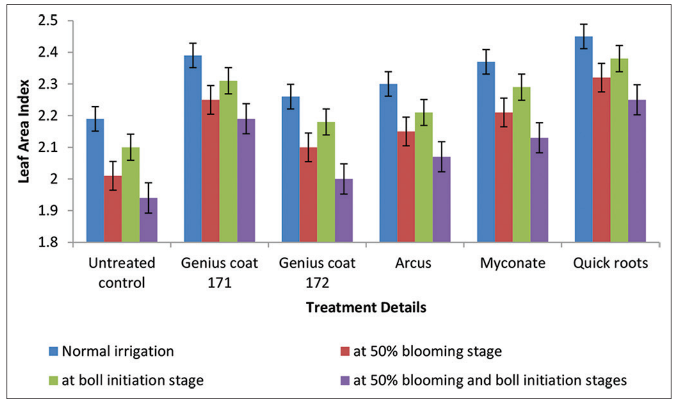

Figure 2: Effect of polymer coating on the leaf area index of cotton plants at maturity under moisture stress conditions.

to those of sweet corn [20] and maize [21]. Water stress during the flowering stage left the cotton plant susceptible and all growth metrics were ceased [7].

The RWC is commonly used indicator to assess plant water status and is thought to reflect the rate of metabolic rate in the tissues. The relationship between RWC and crop yield in water stress conditions provided more support [22]. The CSI is a credible metric for evaluating a plant's ability to withstand drought. The values of the CSI are generally lowered by water stress, with significant variations between irrigation regimes [23]. Similar patterns could be seen in the findings of the RWC and CSI. Both experienced a progressive increase up to flowering and a gradual decline as they matured. Regardless of moisture treatments, the Quick roots coated seeds (72%), compared to the control seed (65%), had a greater RWC at maturity. Compared to stress at both the 50% flowering and the boll formation stages (66%), and skipping irrigation at boll formation stage (70%) recorded higher RWC. RWC at the maturity stage of cotton significantly increased under stress at 50% flowering and boll formation phases, which was the cause of the 11% increase in Quick roots coated seeds compared to the untreated control [Table 1]. RWC considerably enhanced in cotton at the mature stage during the 50% blooming and boll formation phases under stress, which contributed to the 11% increase in Quick roots coated seeds compared to the untreated control.

Polymer-coated seeds recorded a higher CSI than control seeds. At the maturity stage, quick roots coated cotton seeds (72%) had a higher CSI than the control seed (65%). Among the moisture treatments, skipping watering during the boll-formation phase (69%) showed a higher CSI than stress during both the 50% blooming and boll-formation stages (65%). At the 50% blooming and boll formation phases, a significant rise in CSI at the maturity stage of cotton was seen under stress; this observation accounted for a 12% increase in the Quick roots coated seeds compared to the untreated control [Figure 3]. Even under water stress conditions, the Quick roots coated polymer cotton seeds responded effectively, displaying higher values for the CSI and RWC. The ability of the glyoxylate cycle to promote lipid consumption and water absorption depends on the polymer. The findings of the current study are consistent with those for maize [24], periwinkle [25], and wheat and maize [26]. Brinjal seeds coated with polycoat (3 g) and halogen mixture (3 g) performed well in all water-holding capacities from 30% to 80% [27].

Soluble protein (nitrogenous compounds) constitutes 40% of RuBp carboxylase. The RuBp carboxylase enzyme is in charge of fixing CO_2

Table 1: Effect of polymer coating on RWC (%) of cotton plants at maturity under moisture stress conditions.

Polymer treatments (T)	Skipping irrigation				
	Normal irrigation	at 50% flowering stage	at the boll formation stage	at 50% flowering and boll formation stages	Mean
Untreated control	69	65	67	62	65
Genius coat 171	74	70	72	68	71
Genius coat 172	70	66	69	64	67
Arcus	72	67	70	71	68
Myconate	73	69	71	67	70
Quick roots	75	71	73	70	72
Mean	72	68	70	66	
	T	S	T×S	S×T	
SEd	0.537	0.521	1.074	1.111	
CD (P=0.05)	1.086**	1.275**	2.172**	2.234**	

CD: Critical differences, RWC: Relative water content

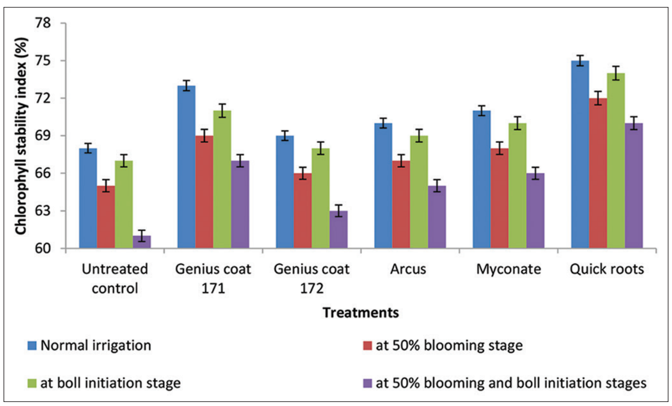

Figure 3: Effect of polymer coating on chlorophyll stability index (%) of cotton plants at maturity under moisture stress conditions.

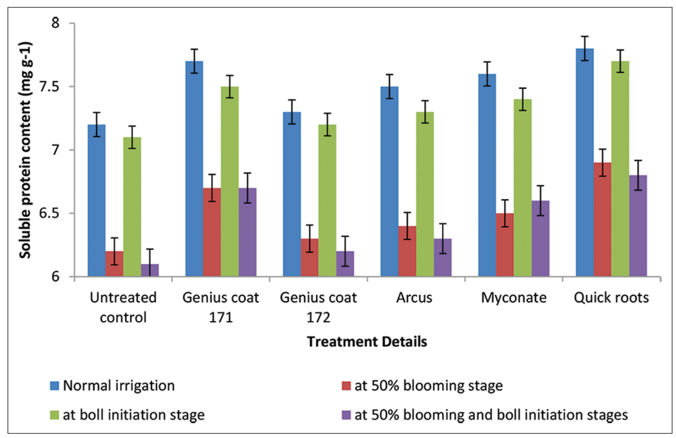

Figure 4: Effect of polymer coating on soluble protein content (mg/g) of cotton plants at maturity under moisture stress conditions.

in higher plant leaves. A rise in soluble protein indicates that plants are better at fixing CO_2 than before. In reaction to water stress, the ability for protein synthesis is significantly reduced. Soluble protein concentration increased throughout the blooming stage but decreased during the maturity stage. It was scarce during the vegetative phase. The soluble protein content in seeds coated with Quick roots at the maturity stage was 13% greater with regular watering compared to stress during at 50% blooming and boll initiation stages [Figure 4]. Cotton seeds coated with quick roots recorded higher amounts of soluble protein (7.3 mg/g) than the untreated control (6.6 mg/g) produced, irrespective of moisture stress treatments. Under water-stress conditions, the deposition of tight-binding inhibitors within the enzyme's catalytic sites may contribute to the lower activity of RuBisCo, which may contribute to the reduced soluble protein concentration [28]. The study concurs with the results of cotton [29] and rice [30].

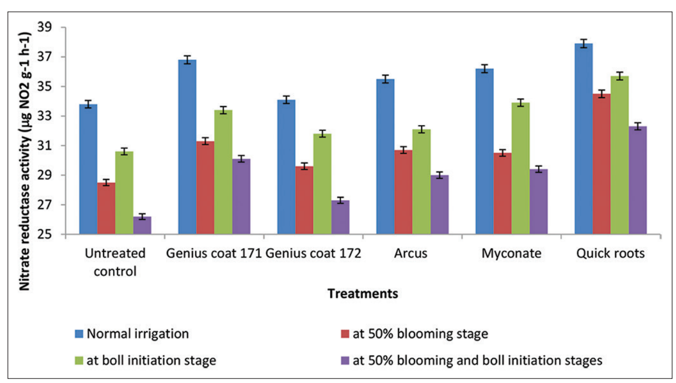

Figure 5: Effect of polymer coating on soluble nitrate reductase activity (mg NO_2/g/h) of cotton plants at maturity under moisture stress conditions.

NRA, an enzyme in plants that regulates nitrogen metabolism, is extremely sensitive to environmental changes. The results of other physiological indicators, such as soluble protein content, RWC, and CSI, confirmed the reduction of NRA. The seeds coated with Quick roots (35 mg NO_2/g/h) had higher RWC at maturity than the untreated control seed (29.7 mg NO_2/g/h), regardless of moisture treatments. Skipping irrigation during at the boll initiation stage (32.9 mg NO_2/g/h) had higher RWC than stress at both the 50% flowering and the boll formation stages (29 mg NO_2/g/h). At 50% blooming and boll formation phases, a significant rise in NRA se at the maturity stage of cotton under stress was seen, accounting for a 19% increase in the Quick roots coated seeds compared to the untreated control [Figure 5]. The findings of the current study are in line with those of research on rice [31] and canola [32]. Water stress causes a significant decline in all physiological characteristics in hybrid rice at critical periods [33]. Red clover seeds that were given a superabsorbent polymer treatment showed stronger field stand in the stress conditions than untreated control seeds [34].

4. CONCLUSION

Irrigation is crucial during the 50% blooming and boll initiation stages for cotton crops to increase yield without changing the physiological metabolism of the crop. The results of the current study demonstrated that polymer coating had a greater impact on plant physiological parameters than control seeds under both normal and moisture stress conditions. Quick roots coated cotton seeds outperformed the other polymers by recording higher physiological parameters under both normal and moisture stress conditions. Polymer coating of cotton seeds with drought-tolerant compounds is a simple and cost-effective technology that farmers can use commercially. Hence, polymers can be recommended for dry land conditions.

5. AUTHORS' CONTRIBUTIONS

Dr. V. Manonmani conceptualized the study. S. Deepika contributed to the data acquisition. Dr. S. Ambika contributed for drafting of the manuscript. Data analysis using the software was by Dr. R. Paramasivam. Dr. K. Mohanraj contributed to preparing the figures for the manuscript. Dr. S. Laksmi contributed for reviewing the manuscript. Dr. S. Kavitha and Dr. Vijaya Geetha contributed for revising the manuscript carefully for final approval. The final manuscript was read and approved by all authors.

6. FUNDING

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7. CONFLICTS OF INTEREST

The authors report no financial or any other conflicts of interest in this work.

8. ETHICAL APPROVALS

This study does not involve experiments on animals or human subjects.

9. DATA AVAILABILITY

All the data is available with the authors and shall be provided upon request.

10. PUBLISHER'S NOTE

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