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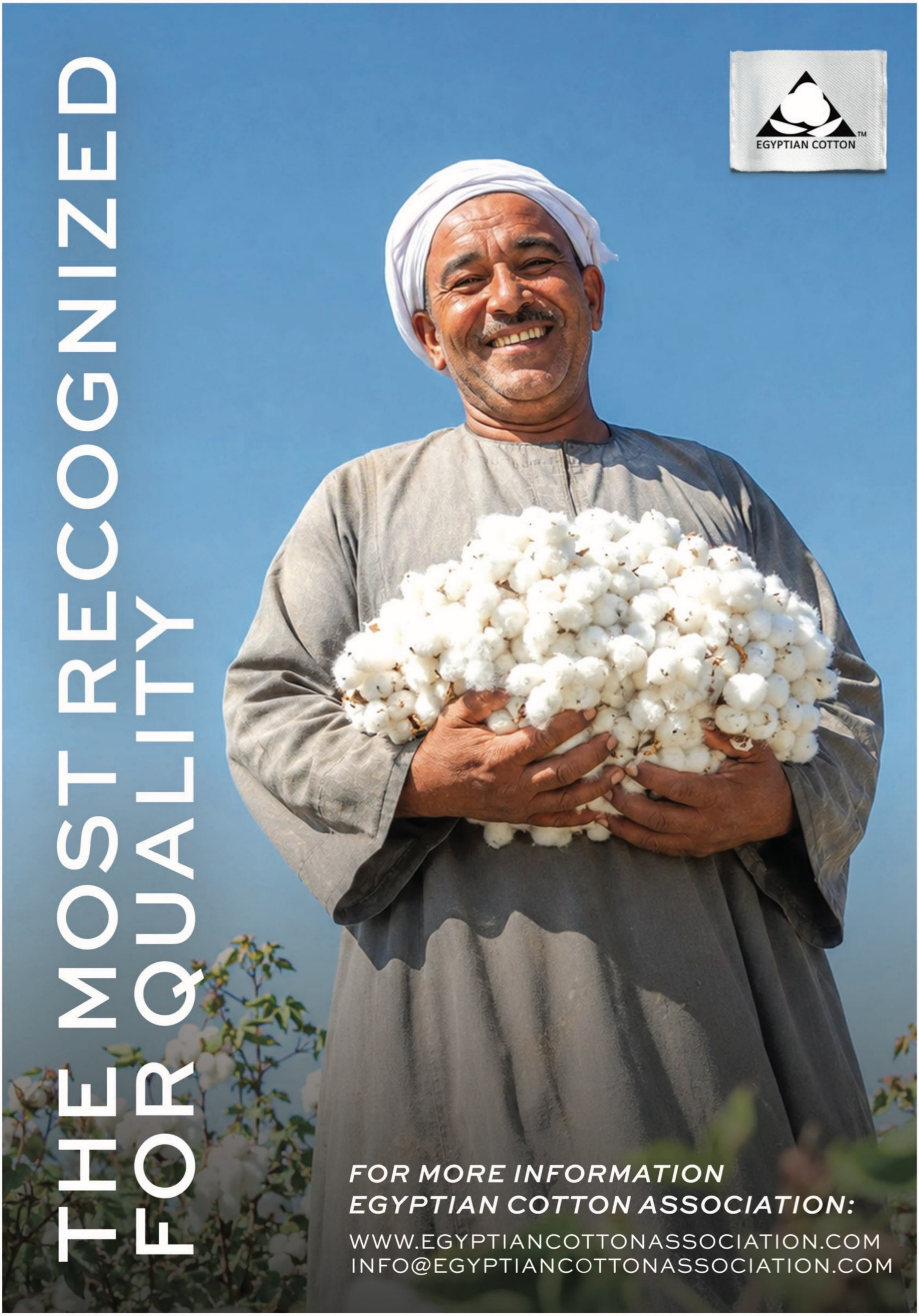
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HOST PLANT RESISTANCE TO WHITEFLY IN COTTON: CURRENT PROGRESS AND FUTURE PROSPECTS

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Abstract

The whitefly, *Bemisia tabaci,* is one of the most destructive insect pests of cultivated cotton (*Gossypium hirsutum* L.) across the tropical and subtropical cotton-growing regions of the world. Beyond direct feeding damage, the insect is the principal vector of Cotton Leaf Curl Virus (CLCuV) and several other begomoviruses that cause severe yield losses and threaten the sustainability of cotton production. Reliance on synthetic insecticides has led to the development of insecticide resistance in whitefly populations, environmental contamination, and the elimination of natural enemies, making host plant resistance (HPR) an increasingly attractive and sustainable alternative for whitefly management. This review synthesizes current knowledge on whitefly biology and the damage it causes, the mechanisms underlying host plant resistance (antixenosis, antibiosis, and tolerance) as originally conceptualized by Painter (1951), the morphological, biochemical, and molecular bases of resistance, and the genetic resources available for breeding resistant cotton cultivars. It further discusses advances in conventional and molecular breeding, including marker-assisted selection, genomic selection, genome-wide association studies, and speed breeding, along with emerging biotechnological tools such as genetic engineering, RNA interference, CRISPR/Cas genome editing, and multi-omics approaches. Finally, the review examines the integration of host plant resistance into integrated pest management (IPM) programs and outlines the major challenges and future prospects for developing durable whitefly-resistant cotton varieties.

Keywords*:* Whitefly (*Bemisia tabaci*), Cotton (*Gossypium hirsutum* L.), Host plant resistance, Antixenosis, Antibiosis, Tolerance, Cotton Leaf Curl Disease (CLCuD), Begomoviruses, Integrated Pest Management (IPM), Marker-Assisted Selection (MAS), Genomic Selection (GS), CRISPR/Cas9, RNA Interference (RNAi), Sustainable agriculture.

1. Introduction

Cotton (*Gossypium hirsutum* L.) is one of the most economically important fibre crops in the world, providing raw material for the textile industry and livelihood for millions of farmers, particularly in South Asia, Africa, and the Americas (Jabran *et al*., 2019). However, cotton production is persistently threatened by a wide range of insect pests, among which the whitefly, *Bemisia tabaci* (Gennadius), stands out as a pest of global significance (Abubakar et al., 2022a). Whitefly is a polyphagous, highly adaptable insect that not only causes direct damage through phloem feeding but also acts as the vector of several plant viruses, most notably the begomoviruses responsible for Cotton Leaf Curl Disease (CLCuD) in South Asia and other viral diseases in different cotton-growing regions (Naveed *et al*., 2023). Cotton is mainly a tropical crop grown in more than 80 countries. The major cotton-producing countries are China, India, the United States, Brazil, and Australia (USDA, 2024/2025), with Pakistan ranking sixth (Databoks, 2025). The global cotton market is projected to grow from USD 43.81 billion in 2025 to USD 62.90 billion by 2034, driven by high demand in China, India, and Vietnam. The global cotton market value is projected to grow from USD 43.81 billion to USD 62.90 billion during the 2026–2034 forecast periods (Renub Research, 2026). This expansion is supported by near-term market conditions, with the USDA (2026) projecting a global supply deficit as consumption reaches 122.9 million bales against 117.6 million bales in production. With this background, increasing production and productivity of cotton becomes crucial in order to meet future demands. However, recent studies reveal that the land used in the cultivation of cotton has substantially shrunk (Harshitha *et al*., 2021), owing to biotic stress leading to yield loss. Cotton plays a significant role in Pakistan's economy and contributes approximately 0.6–0.8% to the national GDP while supporting a large share of textile exports (Pakistan Development Review, 2023; MM News, 2025). Various biotic and abiotic factors have been identified as causing a reduction in cotton yield, leading to a decrease in production of cotton (Rind *et al*., 2020).

The management of whitefly has historically relied on the intensive use of synthetic insecticides. While insecticides provide short-term relief, their repeated and indiscriminate application has resulted in several undesirable consequences, including the evolution of insecticide-resistant whitefly biotypes (Dhillon & Rajna, 2026). Whitefly biotypes,

resurgence of secondary pests, destruction of natural enemy populations, environmental pollution, and increased cost of production (Abubakar *et al*., 2022b; Marabi *et al*., 2025). These concerns have shifted research attention toward more sustainable and environmentally friendly alternatives, among which host plant resistance (HPR) occupies a central position (Pathania *et al*., 2021).

Host plant resistance refers to the inherent ability of a plant genotype to reduce the population, feeding, growth, or reproduction of a pest, or to prevent damage despite pest presence, without any external chemical or biological intervention (Russell, 2013). It is a cost-effective, environmentally sound, and compatible component of integrated pest management (IPM) systems (Zhou *et al*., 2024a). Breeding whitefly-resistant cotton cultivars requires a thorough understanding of whitefly biology, the mechanisms of resistance operating in the plant, the genetic and genomic resources available, and the tools that can accelerate the identification and deployment of resistance genes (Rahman *et al*., 2025).

This review aims to consolidate the current understanding of whitefly biology and its impact on cotton, the mechanisms and genetic basis of host plant resistance (Kumaraswamy & Huang, 2026), The progress made in conventional and molecular breeding, the emerging biotechnological approaches, and the role of host plant resistance within IPM frameworks (Keykhasaber *et al*., 2026). The review concludes with a discussion of the major challenges that continue to hinder the development of durable resistance and the future directions that hold promise for sustainable whitefly management in cotton (Adala *et al*., 2026).



Figure 1. Representative images of whitefly (*Bemisia tabaci*) infestation on cotton: (A) Close-up view of the abaxial surface of a cotton leaf exhibiting whitefly nymphs, chlorotic yellowing, and mechanical damage; (B) Overview of the sampled cotton field plot

2. Whitefly Biology and Its Impact on Cotton

Whitefly (Bemisia tabaci) is one of the most serious insect pests of cotton worldwide. It causes both direct damage through continuous phloem feeding and indirect damage by transmitting several important plant viruses. Understanding its biology, feeding behaviour, and the nature of the damage it causes is essential for developing effective host plant resistance strategies.

Biology and Life Cycle:

Bemisia tabaci, a species complex in the order Hemiptera and family Aleyrodidae, includes cryptic species such as Middle East–Asia Minor 1 (MEAM1) and Mediterranean (MED), both of which are highly invasive and detrimental to cotton crops (Sani et al., 2020). It undergoes a life cycle featuring eggs, four nymphal instars, and adults. Under favorable conditions, the complete life cycle from egg to adult is completed in approximately 16–31 days, which promotes multiple generations per season (Sani et al., 2020). Females lay eggs on the upper and lower leaf surfaces, while the first instar nymph (crawler) is the only mobile stage (Sani et al., 2020). Plant defense strategies aimed at deterring these crawlers can significantly hinder pest establishment.

Feeding Behavior and Damage:

Whitefly nymphs and adults feed on phloem tissue, extracting sap rich in carbohydrates and amino acids, leading to symptoms (Sunidhi *et al*., 2025)Such as chlorosis, leaf curling, premature defoliation, and stunted growth (De Silva *et al*., 2025). A notable sign of infestation is the excretion of honeydew, which promotes sooty mold growth, further impairing photosynthesis and reducing plant vigor. This phenomenon, known as sticky cotton, diminishes the market value of harvested fiber due to processing challenges (Wyckhuys *et al., 2024)*. Additionally, whiteflies serve as vectors for more than 400 plant viruses, mostly begomoviruses (Hussain & Jander, 2026), including Cotton Leaf Curl Virus and other begomoviruses, where even low densities can facilitate rapid disease spread. Infected plants exhibit significant symptoms like leaf curling and stunting, potentially resulting in severe yield losses in affected fields (Xiao *et al*., 2026).

Economic Importance

The economic significance of whitefly in cotton is reflected both in direct yield losses from feeding injury and indirect losses from virus transmission and reduced fiber quality (Jan *et al*., 2025). Bemisia tabaci causes enormous agricultural damage worldwide, with farmers incurring

billions of dollars in losses each year (Hussain & Jander, 2026). In regions such as Pakistan and northern India, epidemics of Cotton Leaf Curl Disease have historically caused yield reductions ranging from moderate levels to near-total crop failure in severely affected years, with cascading effects on the textile industry, farmer income, and rural livelihoods (Sarwar *et al*., 2022). The cost of chemical control adds a further economic burden, as growers may apply insecticides multiple times per season, raising the cost of production (George *et al*., 2024) while offering only partial and often short-lived control due to resistance development in the pest population (Shikha *et al*., 2024). These combined direct and indirect losses make whitefly management, and specifically the development of resistant cultivars, a priority research area for cotton-producing countries (Tauseef *et a*l., 2025).

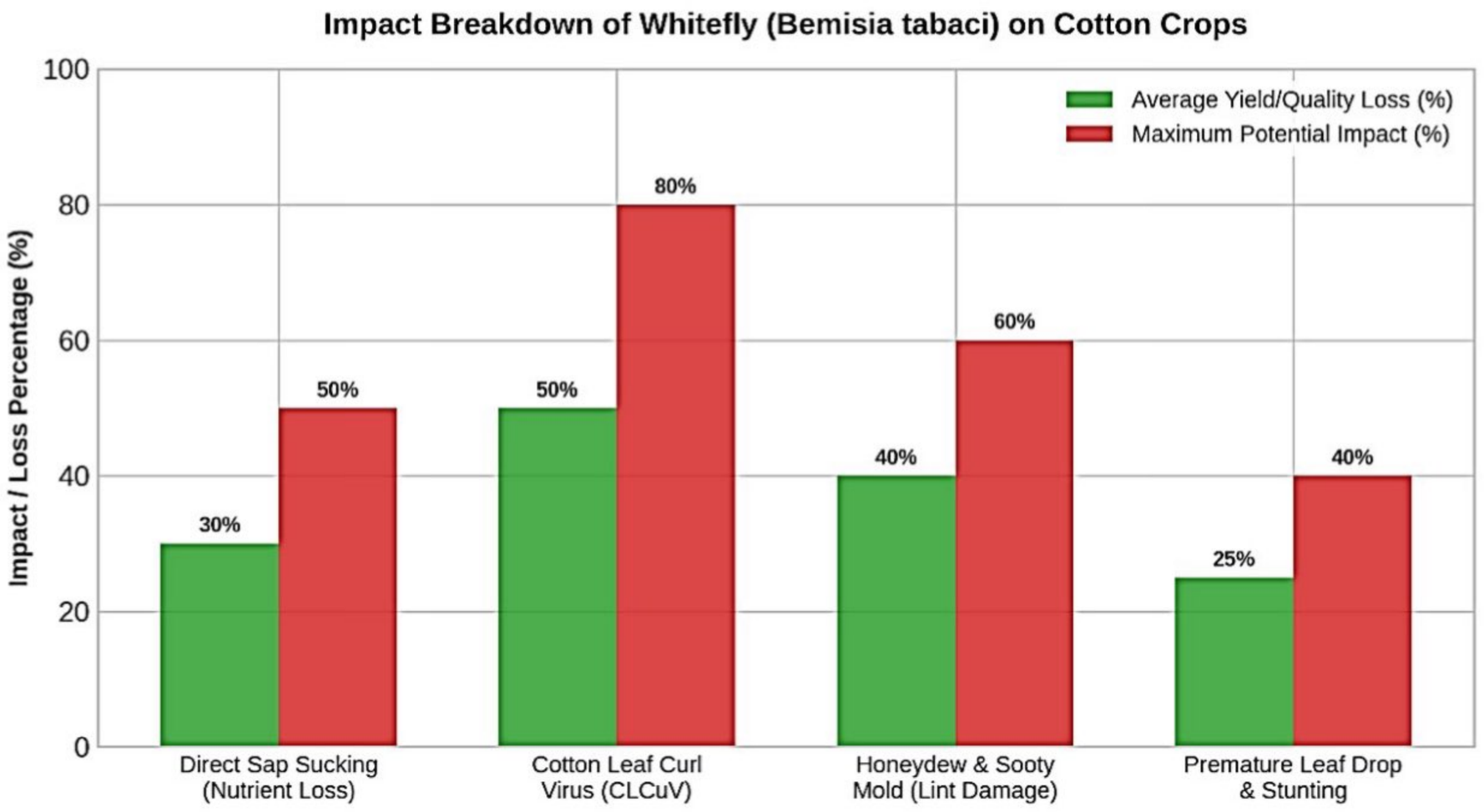


Figure 2: Impact of Breakdown of Whitefly *Bemisia Tabaci* on cotton crop

3. Mechanisms of Host Plant Resistance

The conceptual foundation for understanding host plant resistance to insect pests was established by Painter (1951), who classified resistance mechanisms into three primary categories: antixenosis (non-preference), antibiosis, and tolerance. These mechanisms operate through distinct but complementary pathways that collectively determine the level of protection a cotton genotype provides against whitefly infestation. Antixenosis refers to plant traits that deter

pests from colonization and feeding. At the same time, antibiosis describes the negative physiological effects on pest survival and reproduction, and tolerance enables plants to withstand or recover from pest damage without significant yield loss. Together, these mechanisms form a multi-layered defense system that can be exploited through breeding programs to develop whitefly-resistant cotton cultivars.

Antixenosis, or non-preference, encompasses morphological and physical plant characteristics that discourage whitefly settlement, oviposition, and feeding. In cotton, this resistance is primarily influenced by leaf morphology, including trichome density, leaf shape, and cuticular wax composition (Kaur et al., 2024). Genotypes with lower trichome density on the abaxial leaf surface create smoother surfaces that complicate the insect's ability to settle, anchor, and lay eggs compared to hairy or normal-leaf genotypes. Similarly, okra-leaf and super okra-leaf shapes contribute to whitefly non-preference by creating an open canopy structure with reduced leaf surface area, which deters landing and creates a less favorable microclimate for oviposition (Cheema et al., 2022). The okra-leaf characteristic may provide higher ambient temperature and lower humidity in the cotton canopy, creating a micro-climate unfavorable to whitefly survival. These morphological traits have been frequently utilized in breeding programs to enhance resistance to both whitefly and Cotton Leaf Curl Disease (CLCuD). Antixenosis operates as the first line of defense, reducing initial pest colonization and thereby limiting subsequent population buildup and virus transmission.

Antibiosis, the second mechanism, describes the detrimental physiological effects of resistant plant genotypes on the biology and life cycle of the pest. When whiteflies feed on resistant cotton genotypes, they experience decreased survival rates, prolonged development times, reduced reproductive rates, and smaller body sizes (Baldin et al., 2022; Mrosso et al., 2022). These negative effects are largely attributed to secondary metabolites such as gossypol and phenolic compounds, which disrupt insect digestion and metabolism, causing nutritional deficiencies that hinder nymph development and adult fecundity (Ouaarous et al., 2025). Resistant genotypes typically present higher concentrations of defensive compounds and elevated activities of defense-related enzymes such as peroxidase and chitinase, along with altered phenolic and flavonoid profiles that collectively hinder insect performance (Roychowdhury et al., 2025).

At the molecular level, antibiosis is linked to the differential expression of defense-related genes in the jasmonic acid and salicylic acid pathways, including those encoding proteinase inhibitors that are up-regulated in response to whitefly feeding (Abbas et al., 2024). Unlike antixenosis, which prevents initial infestation, antibiosis reduces the fitness and population growth of whiteflies that do manage to colonize the plant.

Tolerance, the third mechanism, refers to the plant's inherent ability to endure or recover from pest infestations without suffering significant yield or quality loss, even when supporting pest populations comparable to those that cause damage on susceptible varieties (Margus et al., 2023). Tolerant cotton genotypes exhibit enhanced compensatory growth, improved photosynthetic efficiency under stress, and physiological adaptations that maintain lint yield despite whitefly pressure (Chandraleka et al., 2025; Ali et al., 2023). Unlike antixenosis and antibiosis, tolerance does not reduce pest populations or prevent virus transmission; rather, it enables the plant to withstand the negative effects of infestation (Jeger, 2023). This mechanism is particularly valuable in integrated pest management (IPM) programs because it provides a buffer against pest damage even when other resistance mechanisms are overcome. At the morphological level, traits such as leaf trichome density, type, shape, color, and cuticular wax composition are crucial determinants of whitefly resistance (Singh et al., 2022), with genotypes featuring sparse trichomes and dissected okra-type leaves showing reduced infestation levels (Daniyal et al., 2025). Biochemically, the accumulation of defensive compounds and increased enzyme activities contribute to tolerance by mitigating the physiological impact of feeding damage (Chamani et al., 2025a). Collectively, antixenosis, antibiosis, and tolerance operate at morphological, biochemical, and molecular levels, providing a comprehensive and durable defense system against whitefly infestation in cotton. The integration of these three mechanisms through strategic breeding programs offers the most sustainable approach for managing whitefly populations and reducing reliance on synthetic insecticides.

Figure 1. Mechanism of resistance (Described by Painter, 1951).

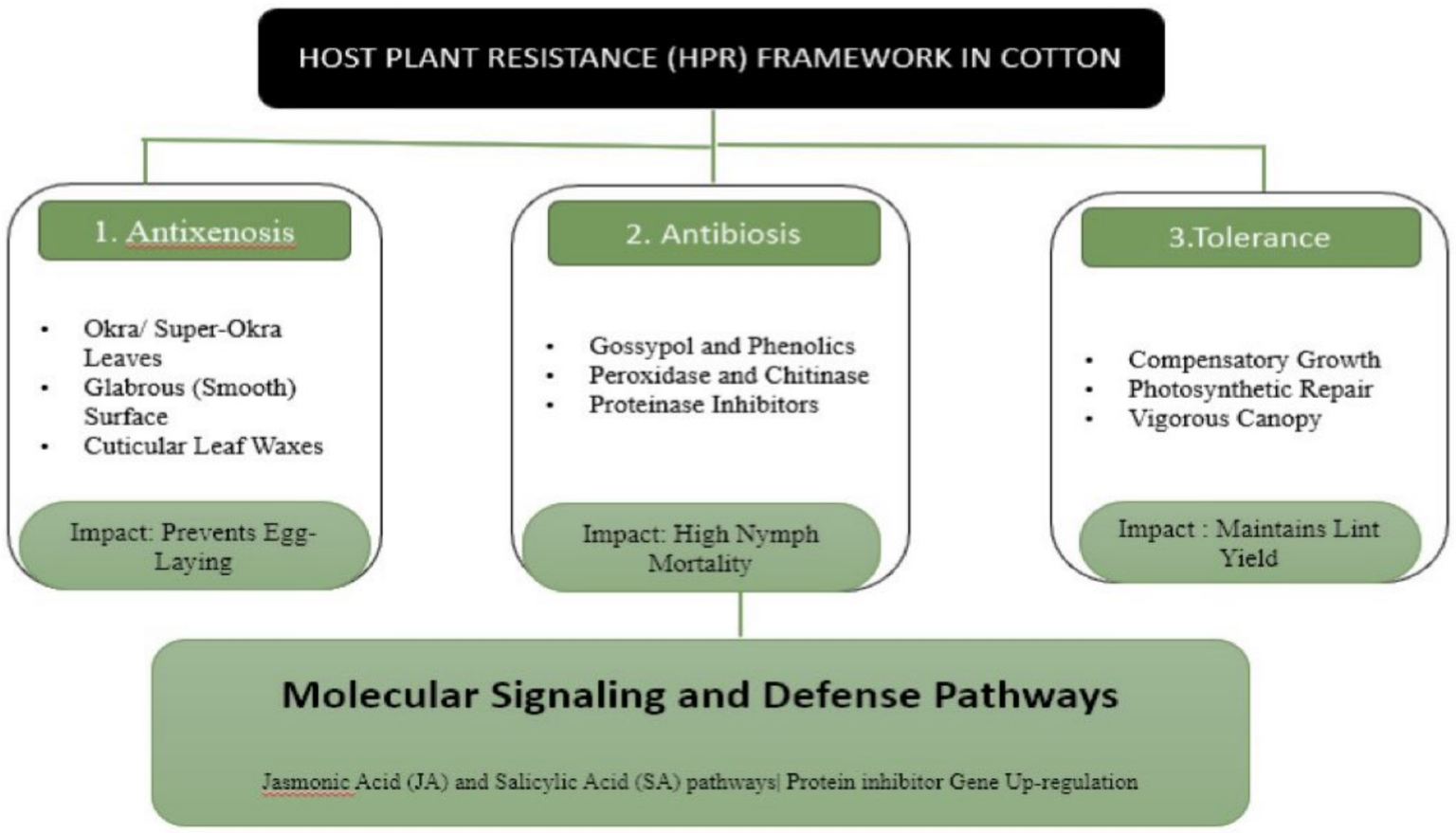


Figure 3: Host Plant Resistance (HPR) FRAMEWORK IN COTTON.

4. Sources and Genetic Basis of Whitefly Resistance

The development of whitefly-resistant cotton cultivars depends on the identification and utilization of genetic resources harboring resistance traits. These resources range from cultivated germplasm collections to wild relatives of cotton, each offering unique advantages and challenges for breeding programs. Understanding the genetic architecture of resistance, whether controlled by major genes or polygenic systems, is essential for effective utilization of these resources. Advances in molecular marker technologies, particularly genotyping-by-sequencing (GBS) and quantitative trait locus (QTL) mapping, have significantly enhanced the ability to dissect the complex genetic basis of whitefly resistance and accelerate the development of resistant cultivars.

A comprehensive screening of cultivated cotton germplasm has highlighted various sources of whitefly resistance. Rahman et al. (2025) conducted a comprehensive study using morphological characterization, SSR markers, principal component analysis (PCA), biplot analysis, correlation studies, and cluster analysis to estimate whitefly resistance and yield-related traits among 150 Pakistani genotypes. Field evaluation along with molecular analysis was employed as a comprehensive approach, demonstrating strong genetic control through high heritability estimates in most traits (Rahman et al., 2025). The results described the potential of morpho-molecular screening for selecting superior

genotypes in breeding programs. CA-12 and AGC-155 were recommended as parental lines for developing whitefly-resistant, high-yielding cotton varieties, while Gomal-105 and SLS-87/175 were recommended as whitefly-susceptible parental lines (Rahman et al., 2025). The study also noted that cotton contributes 0.8% to GDP and 60% of textile exports in Pakistan, highlighting the economic importance of developing resistant cultivars (Rahman et al., 2025). Genetic diversity within *Gossypium hirsutum* accessions reveals variability in traits such as trichome density, leaf shape, and levels of antixenosis and antibiosis (Shrestha, 2025). Notably, genotypes exhibiting okra-leaf and super okra-leaf shapes have been recognized as significant contributors to whitefly non-preference. They are frequently used in breeding programs to enhance resistance to both whitefly and CLCuD (Cheema et al., 2022).

The antixenosis mechanism of host plant resistance has been extensively studied in cotton introgression lines. Screenhouse experiments conducted under free-choice conditions assessed nineteen cotton test entries for settling behavior and oviposition preference against ***Bemisia tabaci*** (Shrestha, 2025). The study revealed that introgression line D-12-7-4-P-2 was most preferred for whitefly settling and oviposition, categorized as highly susceptible, whereas synthetic polyploids C1-P-1, C1-P-31, C1-P-20, and C1-P-36 were least preferred for oviposition, falling under the resistant category and exhibiting an antixenosis mechanism of resistance (Shrestha, 2025). Among biophysical parameters, trichome density, leaf area, and vascular bundle length showed significantly positive correlations, whereas leaf lamina thickness was negatively correlated with settling behavior and ovipositional preference (Shrestha, 2025). Biochemically, sugars were predominantly higher in whitefly-susceptible lines, whereas phenols, tannins, and proteins were comparatively higher in resistant genotypes (Shrestha, 2025). Total soluble sugars and reducing sugars registered a positive association, while total soluble proteins, crude proteins, total phenols, and total tannins showed negative correlations with whitefly settling and ovipositional preference (Shrestha, 2025). These findings confirm that the antixenosis mechanism of resistance operates in cotton lines and that biophysical and biochemical parameters play a predominant role in imparting resistance against *B. tabaci* **(Shrestha, 2025).**

Wild and semi-domesticated *Gossypium* species represent valuable

genetic resources containing resistance traits. **Xu et al. (2026)** conducted a genomic reconstruction of upland cotton domestication, revealing that modern upland cotton most probably originated from a single domestication event that unfolded in three successive stages. The first stage (D1), around 2,800 years before present, resulted in the emergence of the earliest domesticated form, **race** *punctatum***, from** *yucatanense* **(Xu et al., 2026). Approximately 2,600 years before present, a second domestication event (D2) led to the establishment of race** *latifolium* **as cultivation areas expanded (Xu et al., 2026). The study confirmed the rich genetic diversity among these landraces and established that race** *latifolium* **is the source of modern cultivated cotton varieties (Xu et al., 2026). Notably, the ~18.16% genome-wide introgression from** *G. barbadense* **represents postdomestication genetic supplementation that does not alter the core single-origin domestication trajectory (Xu et al., 2026).** *Gossypium barbadense* accessions are also valuable genetic resources containing resistance traits (Abdelmoghny et al., 2025). These wild relatives exhibit higher gossypol levels and trichome variability, but their breeding use is limited due to linkage drag, cross-incompatibility, and undesirable agronomic traits that require extensive backcrossing to eliminate (Eghlima et al., 2025; Nurzyńska-Wierdak, 2023).

The genetic control of whitefly resistance in cotton is complex, involving many genes with small to moderate effects, while some morphological traits exhibit simpler inheritance patterns (Pokou et al., 2025). Heritability for trichome density and antixenosis traits is moderate to high, indicating that phenotypic selection is feasible. However, the polygenic resistance necessitates careful pedigree selection and multi-location testing to combine multiple mechanisms effectively (Gandhi et al., 2026). Quantitative trait locus (QTL) mapping studies have identified several genomic regions linked to trichome density, leaf morphology, and whitefly populations in cotton by using bi-parental populations from resistant and susceptible genotypes (Rawandoozi et al., 2022).

A genotyping-by-sequencing (GBS) study conducted on 206 cotton plants from a cross between whitefly resistant and whitefly susceptible varieties identified genetic markers linked to whitefly resistance and yield traits (Rahman et al., 2025). This study identified genetic regions associated with whitefly resistance and yield traits, including specific genes on chromosomes A04 and D09 linked to

resistance mechanisms and flower number, respectively (Rahman et al., 2025). The study identified genes involved in whitefly resistance (***At4g27190*** and ***RPPL1***) and genes linked to increased flowering and plant vigor (***GUS1***, ***MBD4L***), paving the way for the development of molecular marker-assisted breeding to create resilient cotton cultivars (Rahman et al., 2025). Key candidate genes implicated in these QTLs relate to trichome development and secondary metabolite biosynthesis (Lapous et al., 2026). The advent of high-quality reference genomes for *Gossypium hirsutum* and *Gossypium barbadense* has enhanced the precision of fine-mapping these regions and facilitated the identification of candidate genes for further validation and marker development (Li et al., 2025).

5. Current Advances in Breeding for Whitefly Resistance

Conventional breeding methods, such as pedigree selection, backcross breeding, and recurrent selection, are essential for developing whitefly-resistant cotton varieties (Dinesh Kumar et al., 2025). These approaches involve phenotypic screening of segregating populations based on traits like trichome density, leaf shape, and whitefly population counts, followed by the selection of superior lines over several generations (Bohorquez-Chaux et al., 2025). The development of resistant cultivars through conventional breeding relies on the identification of stable resistance sources and their systematic incorporation into elite germplasm through repeated crossing and selection cycles (Dinesh Kumar et al., 2025). However, these methods are time-intensive, often taking six to ten years to create stable varieties, and are affected by environmental variability that can hinder precise phenotypic selection (Bohorquez-Chaux et al., 2025). The polygenic nature of whitefly resistance further complicates conventional breeding efforts, as multiple genes with small effects must be accumulated simultaneously to achieve adequate levels of resistance (Dinesh Kumar et al., 2025).

Marker-assisted selection (MAS) utilizes DNA markers associated with resistance QTLs to identify favorable genotypes early in the seedling stage, promoting faster breeding cycles and enhancing selection accuracy for traits with low heritability (Gao & Li, 2025). This method addresses challenges in phenotyping, particularly for complex traits, and enables the integration of multiple resistance mechanisms into a single genetic framework (Gao & Li, 2025). MAS has been successfully employed in cotton breeding programs to select for morphological traits such as

okra-leaf shape and trichome density, which are associated with whitefly non-preference (Dinesh Kumar et al., 2025). The identification of tightly linked markers for resistance QTLs allows breeders to screen large populations efficiently, reducing the time and cost associated with field phenotyping (Gao & Li, 2025). Furthermore, MAS facilitates the pyramiding of multiple resistance genes, enhancing the durability of resistance against genetically diverse whitefly populations (Dinesh Kumar et al., 2025).

Genomic selection (GS) enhances marker-assisted selection by utilizing genome-wide marker data to predict breeding values for complex polygenic traits, such as whitefly resistance (Gao & Li, 2025; Murugan et al., 2026), instead of relying solely on major-effect markers (Crossa et al., 2025). Training prediction models on phenotypic and genotypic data, it enables breeders to select unphenotyped individuals, significantly reducing the breeding cycle and improving genetic gain, crucial for traits influenced by multiple small-effect loci (Dinesh Kumar et al., 2025). The integration of genomic selection with high-throughput phenotyping platforms has further enhanced the efficiency of cotton breeding programs, enabling rapid characterization of large breeding populations (Crossa et al., 2025). Genomic prediction models have been shown to capture small-effect QTLs that are often missed by traditional MAS, making GS particularly valuable for improving polygenic traits like whitefly resistance (Murugan et al., 2026).

Genome-wide association studies (GWAS) offer enhanced marker-trait association identification compared to traditional bi-parental QTL mapping by utilizing historical recombination in diverse germplasm panels (Joshi et al., 2026). Recent evaluations of GWAS panels featuring Gossypium hirsutum accessions have uncovered novel genomic regions and candidate genes related to whitefly resistance, thus supplying breeders with more precise markers for resistance improvement (Zhang, 2025). GWAS enables the identification of resistance-associated loci without the need for developing specialized mapping populations, significantly accelerating the discovery of novel resistance genes (Joshi et al., 2026). The application of GWAS in cotton has identified multiple genomic regions associated with trichome density, leaf morphology, and whitefly population dynamics, providing valuable targets for marker-assisted breeding (Zhang, 2025).

Speed breeding techniques utilize extended photoperiods, controlled temperature, and optimized conditions to

accelerate the generation time of cotton (Kabade et al., 2025). When combined with marker-assisted and genomic selection, these methods can develop whitefly-resistant lines more rapidly. This approach allows for multiple generations within a year, significantly reducing the time needed to create new resistant cultivars (Kabade et al., 2025; Kayess et al., 2025). Speed breeding has been successfully applied in cotton to achieve up to 4–6 generations per year, compared to 1–2 generations under conventional field conditions (Kabade et al., 2025). The integration of speed breeding with genomic selection and CRISPR-based gene editing offers a powerful platform for accelerating the development of whitefly-resistant cotton varieties (Kayess et al., 2025). This approach is particularly valuable for rapidly incorporating resistance genes from wild relatives into elite cultivars and for stacking multiple resistance mechanisms in a single genetic background (Dinesh Kumar et al., 2025).

6. Biotechnological Approaches

Research on genetic engineering for cotton whitefly resistance has focused on introducing genes for insecticidal proteins, plant lectins, and proteinase inhibitors that disrupt the insect's digestive system (Hussain & Jander, 2026). While transgenic Bt cotton effectively manages lepidopteran pests, it is ineffective against hemipteran pests like whitefly, leading to investigations into alternative transgenes and gene-stacking approaches targeting phloem-feeding insects (Math et al., 2026). These alternative approaches include the expression of insecticidal genes under phloem-specific promoters to specifically target sap-sucking pests (Hussain & Jander, 2026). The identification of novel insecticidal proteins from sources such as ferns has also been explored as a strategy for controlling whitefly and other sucking pests (Hussain & Jander, 2026).

RNA interference (RNAi)-based methods present a species-specific approach for managing whiteflies by engineering cotton plants to produce double-stranded RNA that targets critical whitefly genes (Salum et al., 2026). Upon ingestion, this RNA induces gene silencing, disrupting essential physiological functions such as digestion and reproduction, thereby decreasing whitefly survival and reproduction rates (Siddique et al., 2025). RNAi has emerged as a promising tool for controlling CLCuD by targeting the whitefly vector, with studies demonstrating that feeding dsRNA targeting critical genes can cause significant mortality in treated whiteflies (Salum et al., 2026). The use of artificial

microRNA (amiRNA)-mediated resistance targeting multiple whitefly genes has also shown potential for enhancing resistance (Salum et al., 2026). While proof-of-concept studies on host-induced gene silencing (HIGS) have shown promising results, issues like dsRNA stability, delivery efficiency, and off-target effects must be addressed before commercial use (Mishra & Khare, 2026).

CRISPR/Cas-based genome editing serves as a precise tool for modifying genes associated with whitefly resistance in cotton, allowing for alterations without introducing foreign DNA (Sheri et al., 2025). This method can knock out susceptibility genes exploited by pests or fine-tune defense-related gene expression, such as those linked to trichome development or secondary metabolite biosynthesis (Dussutour, 2026). With the availability of well-annotated cotton reference genomes, CRISPR/Cas editing is poised to significantly enhance whitefly resistance in elite cotton varieties (Kangben et al., 2025). CRISPR-Cas9 technology has been successfully applied in upland cotton to generate targeted knockouts of genes such as Mildew Resistance Locus O (GhiMLO3), conferring enhanced resistance to reniform nematode (Kangben et al., 2025). The application of CRISPR/Cas9 for modifying susceptibility genes and defense-related pathways offers a promising approach for developing whitefly-resistant cotton cultivars (Sheri et al., 2025).

The multi-omics approach, combining genomics, transcriptomics, proteomics, and metabolomics, enhances understanding of whitefly resistance in cotton (Luqman et al., 2025). Studies show that differential gene expression in resistant genotypes relates to defense signaling, secondary metabolism, and cell wall reinforcement (Pascual et al., 2025; R. Upadhyay et al., 2025). Furthermore, metabolomic profiling identifies biochemical compounds linked to antibiosis, aiding the transition from broad QTL regions to specific candidate genes for marker development or genetic engineering (Hassanine et al., 2025). The integration of multi-omics data provides a comprehensive understanding of the molecular mechanisms underlying whitefly resistance, facilitating the identification of key regulatory genes and metabolic pathways that can be targeted for genetic improvement (Luqman et al., 2025).

Table 1. Breakdown of Resistance Traits and Breeding Technologies

Category	Trait / Technology	Mechanism / Mode of Action	Breeding & Management Impact	References
Morphological	Okra / Super-Okra Leaves	Open canopy, reduced leaf surface area	Deters landing and microclimate for oviposition	Cheema et al. (2022)
	Glabrous (Smooth) Leaves	Lower trichome density on abaxial surface	Prevents 1st instar crawler movement & anchorage	Singh et al. (2022)
Biochemical	Gossypol & Phenolics	Secondary metabolites disrupting gut physiology	Causes nutritional deficiency & nymph mortality	Chamani et al. (2025b)
	Defense Enzymes	Peroxidase, Chitinase, Proteinase Inhibitors	Inhibits insect digestive proteases	Divekar et al. (2023)
Breeding Tools	Marker-Assisted Selection (MAS)	QTL tracking for trichome & leaf morphology	Accelerates early-stage selection without phenotyping	P. Upadhyay et al. (2025)
	Genomic Selection (GS)	Genome-wide marker prediction models	Captures polygenic small-effect resistance loci	Dinesh Kumar et al. (2025b)
	Speed Breeding	Extended photoperiods & controlled environment	Achieves 4-6 generations/year for fast line delivery	Samantara et al. (2022)
Biotechnology	RNA Interference (RNAi)	Host-induced gene silencing (HIGS) via dsRNA	Species-specific silencing of vital whitefly genes	Salum et al. (2026)
	CRISPR/Cas9 Editing	Targeted gene knockouts (e.g., susceptibility genes)	Precision resistance enhancement in elite cultivars	Sheri et al. (2025)

7. Integration of Host Plant Resistance into Integrated Pest Management (IPM)

Host plant resistance is most effective when integrated into a broader pest management strategy rather than used alone (Abubakar et al., 2022). Resistant cultivars can lessen insecticide use, thus helping biological control agents thrive

(Zou et al., 2024). Coupling resistant varieties with cultural practices like timed planting, balanced fertilization, intercropping, and weed removal improves pest management efficacy (Jalloh et al., 2025). Biological control agents flourish under lower pest densities, and careful application of selective insecticides can prevent economic damage while delaying insecticide resistance in whiteflies (Sharma & Muniappan, 2021). This integrated approach is key to sustainable whitefly management in cotton production systems (Tauseef et al., 2025).

The integration of host plant resistance with biological control is particularly effective because resistant cultivars maintain pest populations below economic thresholds, allowing natural enemies to exert additional mortality pressure on surviving pests (Abubakar et al., 2022). Furthermore, the use of resistant cultivars reduces the frequency of insecticide applications, thereby conserving natural enemy populations and promoting overall ecosystem health (Zou et al., 2024). Cultural practices such as crop rotation, intercropping with non-host crops, and weed management can significantly reduce whitefly populations by disrupting their life cycle and removing alternative hosts (Jalloh et al., 2025). The strategic combination of these practices has been shown to reduce whitefly populations by up to 60–70% compared to sole reliance on insecticides, leading to increased profitability and reduced environmental impact (Tauseef et al., 2025)

8. Challenges and Future Prospects

Despite significant advancements in breeding and biotechnology, several challenges persist in developing durable whitefly resistance in cotton (Amjad et al., 2026). The polygenic nature of resistance complicates achieving high levels of resistance due to the need for multiple mechanisms, as whitefly populations are genetically diverse and capable of rapid adaptation. Additionally, trade-offs between resistance traits and agronomic qualities hinder breeding efforts (Mundt, 2018). The genetic complexity of whitefly resistance, involving multiple quantitative trait loci (QTLs) with small to moderate effects, makes it difficult to achieve durable resistance through conventional breeding alone (Amjad et al., 2026). Furthermore, the rapid evolutionary potential of whitefly populations, driven by their high reproductive rate and genetic diversity, allows them to overcome single-gene resistance relatively quickly, necessitating the stacking of multiple resistance mechanisms (Mundt, 2018).

Future strategies must focus on integrating genomics and gene-editing technologies to identify and deploy resistance genes more efficiently (Razzaq et al., 2021). The use of advanced genomic tools such as genome-wide association studies (GWAS), genomic selection (GS), and CRISPR/Cas-based gene editing can accelerate the identification and deployment of resistance genes (Razzaq et al., 2021). Emphasizing the pyramiding of resistance mechanisms through marker-assisted selection and understanding whitefly population dynamics are critical for developing durable resistance (Wang et al., 2023). Collaborative research and responsible biotechnological deployment are crucial for delivering sustainable, resistant cotton varieties (Gao et al., 2024). The integration of multiple resistance genes (gene pyramiding) has been shown to enhance the durability of resistance by reducing the probability of the pest evolving to overcome all resistance mechanisms simultaneously (Mundt, 2018). Under-standing the genetic basis of whitefly adaptation and resistance evolution is essential for designing effective and durable resistance strategies (Wang et al., 2023).

Furthermore, the development of whitefly-resistant cotton varieties must consider the complex interactions between resistance traits and other agronomically important characteristics such as yield and fiber quality (Gao et al., 2024). Trade-offs between resistance and yield can be minimized through careful breeding strategies and the use of molecular markers to select for favorable combinations of traits (Razzaq et al., 2021). The integration of multi-omics approaches, including genomics, transcriptomics, and metabolomics, can provide a comprehensive understanding of the molecular mechanisms underlying whitefly resistance and facilitate the identification of key regulatory genes and metabolic pathways that can be targeted for genetic improvement (Amjad et al., 2026). The adoption of sustainable agricultural practices, including the use of resistant cultivars as a component of integrated pest management (IPM) programs, is essential for reducing reliance on synthetic insecticides and promoting long-term pest control (Gao et al., 2024).

9. Conclusions

Whitefly is a significant pest in cotton production, causing direct damage through phloem feeding and transmitting harmful begomoviruses like Cotton Leaf Curl Virus (Sani et al., 2020). Host plant resistance through strategies such as antixenosis, antibiosis, and tolerance provides a sustainable approach to pest

management when combined with practices in Integrated Pest Management (IPM) (Zhou et al., 2024). Advances in identifying resistant germplasm and genetic tools are promising, with methods like RNAi and CRISPR/Cas enabling more precise improvements (Pagnotta, 2025). The integration of these biotechnological tools with conventional breeding approaches offers significant potential for developing whitefly-resistant cotton cultivars (Pagnotta, 2025). However, the genetic complexity of resistance and whitefly adaptability require ongoing research to ensure durable resistance and the sustainability of cotton farming globally (Phogat et al., 2026).

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PHYSIOLOGICAL AND BIOCHEMICAL CHANGES INDUCED BY SALINITY STRESS IN COTTON: IMPLICATIONS FOR OXIDATIVE STRESS AND OSMOTIC ADJUSTMENT

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Abstract

Worldwide, soil salinity is emerging as a major constraint on agricultural output, affecting roughly one-fifth of irrigated cropland and an estimated 1125 million hectares overall. This problem is especially acute in arid and semi-arid zones such as Pakistan, where high evaporation rates, scarce rainfall, poor drainage, and reliance on low-quality irrigation water together drive salt build-up in the soil. Consequently, cotton yields frequently fall short of their genetic ceiling. Cotton (*Gossypium* spp.) is generally regarded as moderately salt-tolerant, with a reported threshold near 7.7 dS m^{-1}, yet rising salinity still substantially compromises growth, yield, and fiber quality. These outcomes stem jointly from osmotic stress, ion toxicity, nutrient imbalance, and oxidative injury. Under saline conditions, excess Na^+ and Cl^- accumulating in plant tissues upset ionic balance by suppressing K^+ uptake and raising the Na^+/K^+ ratio, while also triggering shortages of essential nutrients such as Mg^{2+}, Fe, and Cu. Photosynthetic capacity likewise declines, largely because of restricted leaf expansion, stomatal closure, and structural disruption of chloroplasts. These shifts reduce internal CO_2 supply, impair PSII function, and lower chlorophyll and carotenoid levels, which in turn curbs assimilate production and reduces boll number, fiber development, and final yield. Salinity stress additionally boosts the generation of reactive oxygen species (ROS), capable of damaging lipids, proteins, and nucleic acids. Genotypes with greater salt tolerance, however, tend to rely on more robust antioxidant defenses, comprising enzymes such as superoxide dismutase (SOD), catalase (CAT), peroxidases (POD), and ascorbate peroxidase (APX), together

with non-enzymatic antioxidants including ascorbate, glutathione, carotenoids, tocopherols, and flavonoids. Cotton plants also build up compatible solutes such as proline, glycine betaine, and soluble sugars that help sustain cellular osmotic balance, stabilize membranes and proteins, and partly offset oxidative stress. Osmotic adjustment and redox regulation are, notably, tightly linked processes even though they are frequently studied in isolation. Bringing together a clearer picture of ion homeostasis, osmolyte build-up, and antioxidant regulation aided by physiological markers and modern breeding tools such as genomic selection will be essential for developing cotton cultivars better suited to increasingly saline growing conditions.

Keywords: Salinity stress; Cotton (*Gossypium hirsutum*); Reactive oxygen species; Antioxidant defense; Osmotic adjustment; Ion homeostasis; Proline; Glycine betaine; Photosynthesis; Salt tolerance.

1. Introduction

The genus *Gossypium*, part of the Malvaceae family, contains more than 50 recognized species, yet only four species, *G. herbaceum*, *G. barbadense*, *G. hirsutum*, and *G. arboreum,* are cultivated at scale for fiber, animal feed, biofuel, and oil (Rafiq et al., 2024). Given its economic weight in Pakistan and its strong export earnings, the crop is often referred to locally as “white gold.” Pakistan ranks among the world's leading cotton producers, although national yields still fall well short of their potential (Ali et al., 2014); in 2022, globally, the five largest producers are India, China, the USA, Brazil, and Pakistan (Maryum et al., 2022). Soil salinity, driven largely by irrigation-borne salt accumulation, remains a persistent constraint on agricultural productivity. Countries most affected tend to lie in dry or semi-dry belts, where long-term use of poor-quality groundwater for irrigation has raised soil salinity, a trend reinforced by low rainfall; consequently, arid and semi-arid regions account for most of the world's salt-affected land (Shahid et al., 2018). Within Pakistan specifically, the dry-to-semi-dry climate marked by strong evaporation and scarce precipitation drives salt accumulation, compounded by inadequate irrigation and drainage practices that produce waterlogging and prevent salts from being flushed from the root zone; together, these factors degrade soil quality and depress crop output in affected areas (Syed et al., 2021). Roughly one-fifth of irrigated land currently carries excess salt (Qamer et al., 2021), and estimates put the global extent of salt-affected land at around 1,125 million hectares (Hossain, 2019);

projections suggest that if salinization continues, 50% of cultivable lands could be lost by 2050 (Hossain, 2019).

Cotton is regarded as moderately salt-tolerant, with a threshold near 7.7 dS m^{-1} (Sharif et al., 2019), yet growth and yield still suffer considerably once salinity exceeds this level. Salt stress generates osmotic imbalance (Sairam and Tyagi, 2004) alongside ionic toxicity and nutrient disturbances (Munns, 2010), and together these disruptions impair the plant's metabolic machinery, slowing growth and reducing yield (Guo et al., 2022). Beyond cotton, salt stress constrains the development of many crops through combined ionic, osmotic, and oxidative effects (SIKDER et al., 2020; Zafar et al., 2020). Elevated soil salt levels give rise to a range of problems affecting agricultural output, including nutrient imbalance, membrane injury, enzyme inhibition, and reduced photosynthesis, and can halt key biochemical and physiological processes altogether, ultimately arresting growth or causing plant death (Ibrahim et al., 2019).

High soil salinity also disturbs nutrient equilibrium within the plant, most notably by depressing potassium (K^+) levels while promoting sodium (Na^+) uptake; excess sodium competes with potassium acquisition and can even drive potassium leakage from cells, pushing the Na^+/K^+ ratio upward and undermining nutrient uptake and overall plant health (Muhammad et al., 2025). An elevated external Na^+ concentration establishes a gradient that pulls sodium into the cell, and as it accumulates, it becomes toxic, disturbing normal cellular metabolism; the cell's attempt to expel this excess sodium can itself damage root tissue and weaken the plant's capacity to maintain internal stability (Farooq, 2019). Salt tolerance largely reflects how effectively a plant manages sodium through restricted uptake at the root, efficient internal transport, and safe storage with vacuolar sequestration of sodium serving as a central mechanism that limits cytosolic toxicity and preserves normal metabolism (Hasegawa et al., 2000; Tester & Davenport, 2003). Certain species further restrict sodium accumulation in shoot tissue via selective transport and exclusion mechanisms; relative to salt-sensitive plants, tolerant genotypes are generally better at limiting sodium entry, distributing ions across tissues, and sequestering sodium in vacuoles, all of which favor survival under saline conditions (Flowers & Colmer, 2008; Shabala & Mackay, 2011).

Elevated soil salinity impairs plant growth, broadly restricting seedling establishment, limiting leaf and root development, and disrupting reproductive

processes with the result that cotton fiber quality declines and overall production falls substantially (Dong, 2012). In cotton specifically, salinity-driven oxidative damage arises largely from excess generation of reactive oxygen species (ROS) (Czégeňy et al., 2014). Elevated ROS levels under salt stress inflict widespread cellular injury: these reactive molecules attack the plasma membrane and oxidize key biomolecules carbohydrates, proteins, lipids, and DNA thereby disrupting normal cell function (Farooq et al., 2021). Their high reactivity also lets ROS interfere with metabolic activity by inflicting oxidative damage on lipids, proteins, and DNA (Zafar et al., 2022a). Salt-tolerant plants counter this partly through more efficient antioxidant machinery that clears excess ROS, alongside their capacity to manage ion balance and water status (Farooq et al., 2021). To cope with oxidative stress, plants deploy both enzymatic and non-enzymatic antioxidant defenses: the enzymatic arm includes peroxidase (POD), catalase (CAT), and superoxide dismutase (SOD), while non-enzymatic components such as carotenoids, tocopherols, flavonoids, and ascorbic acid provide further protection against oxidative injury (Manan et al., 2022; Shokat et al., 2020). Elevated activity of antioxidants such as POD, CAT, SOD, total soluble proteins (TSP), and carotenoids is commonly associated with stronger salt tolerance (Zafar et al., 2021). Overall, salinity stress compromises cotton growth, yield, and fiber quality by disrupting core physiological processes, curbing vegetative traits such as plant height, leaf area, and root growth as a consequence of osmotic imbalance and ion toxicity; as salinity intensifies, yield losses grow markedly through reductions in boll number and boll weight, driven largely by impaired photosynthesis and altered nutrient distribution (Zafar et al., 2021).

Alongside osmotic adjustment, maintaining ionic balance is essential for salinity tolerance in plants. Because excess sodium (Na^+) disrupts normal cellular activity, plants regulate the extent of sodium entry, sequester it within vacuoles, and work to keep potassium (K^+) levels above those of sodium, which helps preserve cell function and membrane integrity (Sharif et al., 2019; Maryum et al., 2022). Salt stress additionally drives substantial accumulation of reactive oxygen species (ROS), which can damage lipids, proteins, and DNA. In response, cotton plants activate antioxidant systems including superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD), together with non-enzymatic antioxidants

that jointly scavenge ROS and strengthen the plant's capacity to withstand saline conditions (Maryum et al., 2022). Under salt stress, plants must simultaneously contend with ionic toxicity and additional stressors such as osmotic and oxidative stress, all of which slow growth; salt exposure can disrupt normal physiological processes to the point of reducing yield and quality or even causing plant death (Parida & Das, 2005; Liang et al., 2018). Substantial ROS production during salt stress causes considerable damage within plant cells: these highly reactive species can break down the plasma membrane and damage major biomolecules, carbohydrates, proteins, lipids, and DNA, impairing normal cellular function (Farooq et al., 2021). Given their reactivity, ROS also disrupt plant metabolism by inflicting oxidative damage on essential cellular components (Zafar et al., 2022b). How well a plant withstands the harmful effects of NaCl depends largely on its genetic background and the efficiency of physiological mechanisms such as restricting sodium entry, adjusting osmotic balance, and building tissue-level tolerance; because the effectiveness of these responses differs across species, cultivars, and developmental stages, salt tolerance should be viewed as a complex, dynamic trait (Zafar et al., 2024).

Despite considerable research on salt stress in cotton, important gaps remain in understanding how oxidative stress and osmotic adjustment operate together. Much of the existing work examines ROS accumulation and osmolyte responses as separate phenomena rather than probing their interaction under saline conditions. Even where key antioxidant enzymes and compatible solutes have been characterized, it remains unclear how these components coordinate to sustain cellular balance and support growth under stress. Translating such findings into improved breeding strategies for salt tolerance also remains limited, underscoring the need for more integrated approaches linking physiological stress responses to genetic improvement in cotton. As soil salinity continues to expand and its consequences for crop performance grow more severe, clarifying how plants respond to salt stress becomes increasingly important. In cotton, salinity acts through osmotic stress, ion imbalance, and oxidative damage, collectively driving poor growth and reduced yield; nonetheless, plants possess adaptive mechanisms, including antioxidant defense and osmotic adjustment, that partially offset these effects. This review therefore synthesizes current understanding of the physiological and biochemical basis of cotton's response

to salinity, with particular attention to the mechanisms underlying oxidative stress regulation and osmotic adjustment. This review aims to: synthesize existing knowledge on how salt stress affects cotton physiology and biochemistry and examine the mechanisms of oxidative stress, including ROS generation and the plant's defensive response; and assess osmotic adjustment mechanisms and their contribution to salt stress tolerance in cotton.

2. Physiological Responses of Cotton to Salinity Stress

Salinity stress imposes multiple constraints on cotton growth by disrupting fundamental physiological processes. The most immediate and visible effects appear in the photosynthetic system, where osmotic imbalance, reduced leaf expansion, stomatal closure, and damage to the photosynthetic machinery collectively limit carbon assimilation. Understanding these physiological responses is essential because they form the foundation upon which later biochemical adaptations operate and ultimately determine yield performance under saline conditions.

2.1 Photosynthetic Performance under Salinity Stress

As a central driver of plant productivity, photosynthesis depends on stomatal regulation of CO_2 and water exchange together with metabolic activity within mesophyll cells. Salinity disturbs this system, and photosynthetic efficiency falls as a result. Elevated NaCl generates osmotic stress that restricts cell expansion and narrows stomatal opening, thereby curbing photosynthetic activity in cotton. A reduction in leaf area is typically among the earliest visible responses to salinity; during early stress stages, this loss of leaf size from limited cell expansion tends to outweigh the accompanying decline in photosynthetic rate per unit leaf area (Shabala and Lew, 2002). Salinity-induced declines in photosynthesis are further linked to reductions in ribulose-1,5-bisphosphate (RuBP) carboxylase efficiency, constrained RuBP pool sizes and regeneration capacity, and increased sensitivity of photosystem II, as demonstrated in studies on salt-sensitive C3 species like Phaseolus vulgaris (Seemann and Sharkey, 1986). Salt stress also disturbs photosynthetic biochemistry by altering chloroplast lamellar structure and undermining chloroplast integrity, lowering photosystem efficiency in cotton. Stomatal closure under saline conditions compounds this by reducing internal CO_2 concentration, which further constrains enzyme activity and overall photosynthetic performance. Declining photosynthetic

capacity is frequently associated with reduced chlorophyll content and structural alterations in chlorophyll-protein complexes (Jiang & Jiang, 2011). Rising salinity markedly lowers chlorophyll a and chlorophyll b levels in cotton cultivars, indicating impairment of the photosynthetic apparatus and disruption of chlorophyll biosynthesis and stability (Zhang et al., 2014). This drop in chlorophyll content may stem from the inhibition of key enzymes involved in pigment biosynthesis (Lee et al., 2013), which is why chlorophyll content is widely used as a physiological marker for screening salt-tolerant cotton germplasm. Salinity stress significantly disrupts photosynthetic pigment composition, wherein alterations in carotenoid biosynthesis pathways correlate with diminished photosynthetic efficiency; clear declines in carotenoid levels have been documented among cotton genotypes as salinity increases (Zhang et al., 2014).

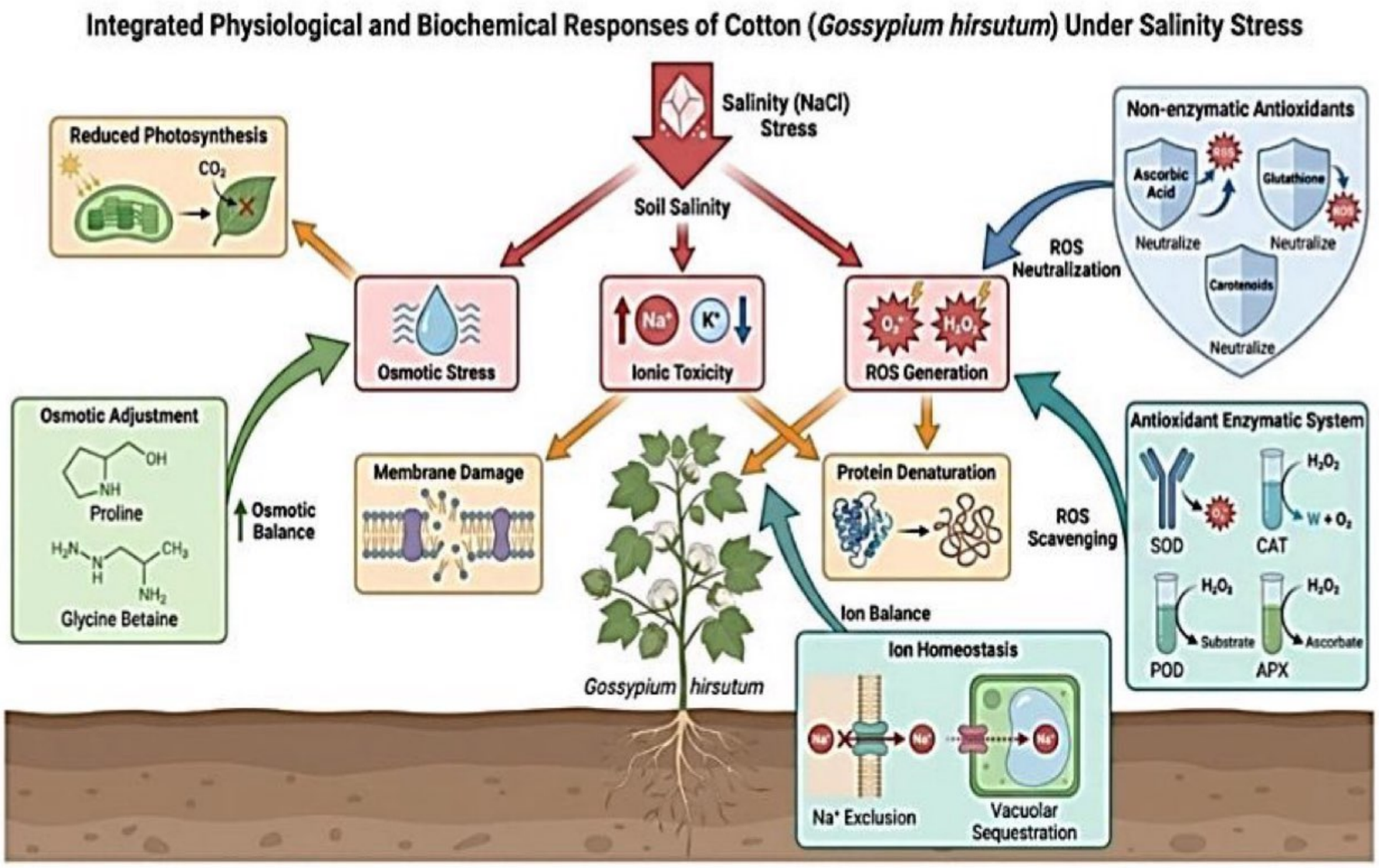


Figure 1. Salinity triggers osmotic and ionic stress in cotton, which elevates ROS levels and activates coordinated tolerance responses.

2.2 Stomatal Conductance under Salinity Conditions

Stomatal conductance drops sharply under salinity stress, restricting gas exchange and, in turn, photosynthetic efficiency (Muhammad et al., 2024). Beyond this functional shift, salinity also reshapes stomatal structure, with studies reporting raised stomatal density while shrinking individual stomata, which further limits CO_2 diffusion into the leaf

(Kader et al., 2020). The resulting decline in photosynthesis is generally attributed to a combination of stomatal and non-stomatal causes: stomatal closure restricts CO_2 supply, while direct injury to the photosynthetic apparatus also plays a substantial role (Zhang et al., 2014).

Just how much each factor contributes is still debated. Several reports indicate that non-stomatal limitations gain importance as salinity stress persists (Bhattacharya et al., 2021; Wang et al., 2023). Salinity, for instance, can impair the photochemical efficiency of photosystem II (PSII), particularly its oxygen-evolving complex, through degradation of the associated proteins (Zhang et al., 2013). Responses also differ markedly among species: sensitive crops such as beans show rapid growth suppression even under moderate salinity, whereas cotton sustains growth more effectively, reflecting its comparatively greater tolerance (Tejera et al., 2005).

2.3 Salinity-Induced Changes in Chlorophyll Content

Salinity stress significantly affects several key photosynthetic parameters in cotton, including stomatal conductance, chlorophyll (Chl) content, and fluorescence (Hatam et al., 2020). Because chlorophyll is the principal light-harvesting pigment, it directly sets the ceiling on photosynthetic efficiency; its decline under salinity therefore restricts light absorption and energy capture, ultimately impairing plant growth (Kreslavski et al., 2018; Saleem et al., 2016). Shifts in Chl content correlate closely with reduced plant height, altered carbon exchange rate, and changes in fluorescence parameters, particularly minimal fluorescence F_0 (Turley, 2011). Salinity-driven stomatal closure can also raise leaf temperature through reduced transpiration, offering a further indicator of stress. Damage to photosystem II (PSII) under salinity likewise alters fluorescence signatures, making fluorescence a dependable tool for gauging stress impacts on photosynthesis (Khalid et al., 2023), though sustained photosynthesis does not necessarily translate into better yield or fiber quality (Khan et al., 2018).

3. Biochemical Adaptations of Cotton under Salinity Stress

Beyond the physiological disturbances, cotton plants undergo a range of biochemical adjustments that help them cope with salinity. These adaptations include the regulation of inorganic ion uptake and compartmentation, accumulation of compatible osmolytes, and activation of both enzymatic and non-enzymatic antioxidant systems. Together,

these biochemical mechanisms work to maintain cellular homeostasis, protect vital macromolecules, and mitigate the oxidative damage that accompanies salt stress.

3.1 Inorganic Ion Accumulation and Imbalance under Salinity

Accumulation of inorganic ions, chiefly Na^+ and Cl^- , under salinity stress strongly shapes plant physiological responses and drives ion toxicity. Because excess Na^+ disrupts cellular metabolism by competing with K^+ for binding at key enzymatic sites, maintaining ion homeostasis is essential; the Na^+/K^+ ratio is accordingly used as a common indicator of salt tolerance, with tolerant genotypes limiting Na^+ build-up while sustaining adequate K^+ levels. Redistributing ions internally, particularly shifting Na^+ from younger to older leaves, also helps shield actively growing tissue from toxicity (Chen et al., 2020). Further Na^+ accumulation compounds stress by lowering cellular water availability and worsening ionic toxicity. In cotton, rising salinity raises Na^+ and Cl^- concentrations in leaf tissue and drives a marked fall in the K^+/Na^+ ratio (Pervaiz et al., 2007; Zhang et al., 2014).

Salt-tolerant genotypes counter these effects through efficient root-level Na^+ exclusion and tightly regulated ion transport that preserves ionic balance, whereas sensitive genotypes regulate Na^+ poorly, leading to severe ionic imbalance and compromised physiological function (Pervaiz et al., 2007). Salinity also reshapes the broader nutrient profile: N, Zn, and Mn tend to increase, while Ca^{2+}, P, and S remain comparatively stable, and essential nutrients such as K^+, Mg^{2+}, Fe, and Cu decline substantially (Higbie et al., 2010). This ion imbalance further limits nutrient acquisition and translocation because Na^+/Cl^- compete with essential ions such as Ca^{2+}, NO_3^-, and K^+, ultimately depressing tissue concentrations of Ca^{2+}, Mg^{2+}, K^+, N, and P (Mansour et al., 2005). Even where leaf S, Ca^{2+}, and K^+ levels appear relatively stable under saline conditions, this stability often masks reduced K^+/Ca^{2+} and K^+/Na^+ ratios, pointing to an underlying disturbance in ion homeostasis and nutrient balance (Abd Ella and Shalaby, 1993).

3.2 Osmotic Adjustment and Protective Role of Osmolytes

Salinity-driven osmotic imbalance lowers cellular water potential and turgor, which makes osmotic adjustment a prerequisite for normal cell function. Plants respond by building up compatible solutes, such as proline, glycine betaine, sugars, and polyols, that reduce osmotic potential and preserve cellular homeostasis

(Fang and Xiong, 2015; Singh et al., 2015). Beyond their osmotic role, these osmolytes also stabilize proteins and membranes and guard cells against ionic and oxidative damage (Chen and Murata, 2011; Khan et al., 2015). In cotton, greater osmolyte accumulation, whether induced through exogenous treatment or genetic regulation, enhances stress tolerance by sustaining water status, limiting membrane injury, and supporting photosynthetic function (Noreen et al., 2013; Lv et al., 2007; Zhang et al., 2015).

3.3 Role of Proline and Glycine Betaine in Salinity Tolerance

As a strongly hydrophilic amino acid, proline is central to cotton's adaptation to salinity because of its capacity to bind water molecules and preserve cellular hydration (Zhang et al., 2019). This property allows proline to stabilize proteins and cellular structures under osmotic stress and prevent their denaturation. Salinity stress raises proline levels in cotton through both greater synthesis and slower breakdown, enabling proline to act not only as an osmoprotectant but also as an antioxidant that helps limit reactive oxygen species (ROS) (Ashraf et al., 2007). Proline additionally functions as a readily mobilized carbon and nitrogen reserve during stress recovery and metabolic readjustment (Verbruggen et al., 2008; Hayat et al., 2012).

Glycine betaine likewise contributes to salinity tolerance, functioning as an osmotic regulator while protecting membrane integrity and enzyme activity under stress (Hossain et al., 2021). Its accumulation is widespread among plant species, particularly halophytes and certain higher plants, in response to salt exposure. Reports of increased glycine betaine synthesis in salt-tolerant species underscore its contribution to maintaining cellular stability and supporting stress adaptation (Gupta et al., 2018; Hirt et al., 2003; Ilyas et al., 2022).

3.4 Enzymatic Antioxidant Defense under Oxidative Stress

Enzymatic antioxidants form a core part of cotton's defense against salinity-induced oxidative stress, working to keep ROS overproduction in check. Superoxide dismutase (SOD) provides the first line of defense, converting superoxide radicals (O_2^-) into hydrogen peroxide (H_2O_2) and oxygen (Ighodaro et al., 2018; Ibrahim et al., 2019). Several SOD isoforms exist, distinguished by their metal cofactors: Cu/Zn-SOD, Mn-SOD, and Fe-SOD, and are distributed across different cellular compartments (Corpas et al., 2006). Elevated ROS levels damage key organelles such as mitochondria,

peroxisomes, and chloroplasts, prompting plants to mobilize antioxidant defenses including SOD, peroxidases (POD), and catalase (CAT), together with non-enzymatic antioxidants that limit oxidative injury, protect cellular structures, and sustain normal cell function (Munawar et al., 2021). Salt-tolerant cotton genotypes, in particular, show markedly higher antioxidant enzyme activity under salinity stress (Zhang et al., 2014).

Once formed by SOD, hydrogen peroxide is broken down further by catalase (CAT), peroxidase (POD), and ascorbate peroxidase (APX). Catalase converts H_2O_2 efficiently into water and oxygen, curbing oxidative damage (Zou et al., 2015), while peroxidase (POD) uses H_2O_2 to oxidize phenolic compounds, contributing to cellular protection and metabolic adjustment under stress (Gross et al., 2006; Yang et al., 2023; Sacu et al., 2023). APX, meanwhile, operates within the ascorbate–glutathione (AsA–GSH) cycle, a key pathway for sustaining cellular redox balance; this cycle relies on enzymes such as monodehydroascorbate reductase (MDHAR), dehydroascorbate reductase (DHAR), and glutathione reductase (GR) to regenerate ascorbate and glutathione and keep antioxidant activity ongoing (Celi et al., 2023; Ding et al., 2023). Glutathione peroxidase (GPX) and glutathione S-transferase (GST) contribute further to ROS detoxification. Collectively, this coordinated enzymatic network plays a central role in limiting oxidative damage and strengthening salinity tolerance in cotton (Gaafar et al., 2018).

3.4.1 Non-Enzymatic Antioxidant Defense

Ascorbic acid (AsA)-mediated antioxidant protection.

As a major water-soluble antioxidant, ascorbic acid (AsA) plays a central part in limiting salinity-induced oxidative stress in cotton. Concentrated in actively growing tissues, it acts as an efficient electron donor that directly scavenges reactive oxygen species (ROS) while regenerating other antioxidants such as glutathione and tocopherols (Mittler et al., 2002; Akram et al., 2017). Higher AsA levels under salt stress help preserve cellular redox balance, protect membranes and macromolecules, and support overall plant performance.

Glutathione (GSH)-dependent redox regulation and the AsA–GSH cycle.

Glutathione (GSH), an important non-enzymatic antioxidant, works within the ascorbate–glutathione (AsA–GSH) cycle to detoxify hydrogen peroxide (H_2O_2) and maintain intracellular redox homeostasis. Within this cycle, dehydroascorbate reductase (DHAR) uses GSH to convert dehydroascorbate back into AsA, while

glutathione reductase (GR) regenerates GSH from its oxidized form (GSSG) using NADPH. Operating across multiple cellular compartments, this system keeps antioxidant regeneration and protection continuous (Jiang et al., 2020). Raised AsA and GSH levels have been linked to reduced ROS accumulation and lipid peroxidation and to stronger antioxidant enzyme activity under salinity stress.

Carotenoid-mediated photoprotection and oxidative stress mitigation.

Carotenoids (CAR) support both photosynthetic performance and stress tolerance, serving as light-harvesting pigments and potent antioxidants. During salinity stress, they protect the photosynthetic apparatus by scavenging ROS, quenching singlet oxygen, and dissipating surplus excitation energy through the xanthophyll cycle (Gao et al., 2024), thereby helping to prevent photooxidative damage and preserve chloroplast integrity. Higher carotenoid levels in cotton are linked to improved salinity tolerance through greater cellular stability and reduced oxidative damage (Das et al., 2014; Sheri et al., 2023). Tocopherols add further support by stabilizing membrane lipids and neutralizing free radicals.

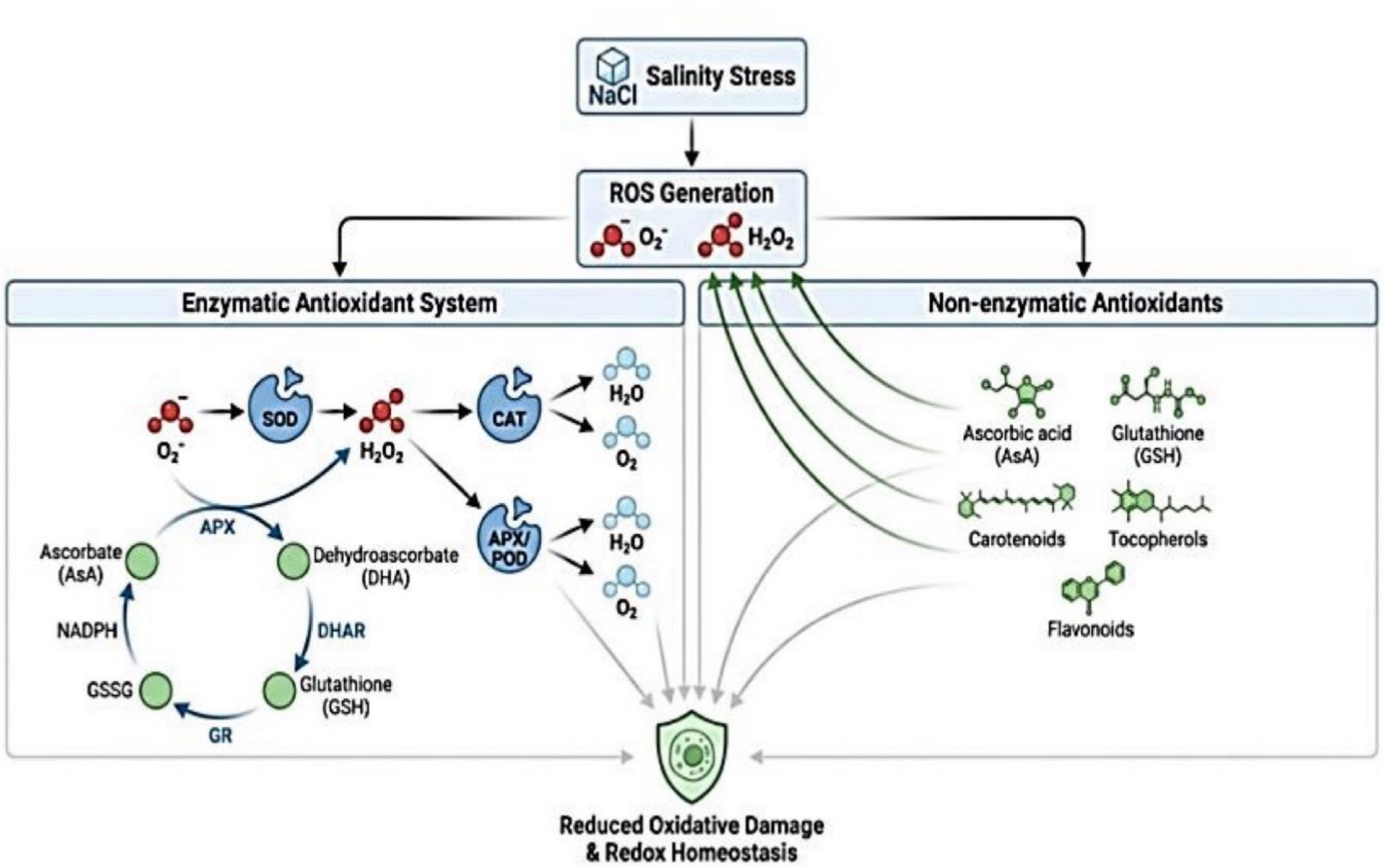


Figure 2. Enzymatic and non-enzymatic antioxidant pathways act together to detoxify ROS and preserve redox homeostasis in cotton under salinity stress.

Recent Progress in Enhancing Salinity Tolerance in Cotton

Recent work has centered on finding the genetic basis behind cotton's variable salt response, aiming to use that variation to breed more tolerant germplasm. Compared to other crops, relatively few salt-tolerance genes have actually been pinned down in cotton. The ones reported so far include an ERF transcription factor, a tonoplast Na+/H+ antiporter, GhMT3a, a ZFP gene, a NAC gene, DREB, and the MPK/MKK kinase pair. In one study, researchers profiled G. aridum via transcriptome sequencing and turned up 108 WRKY genes, then used RT-PCR to confirm that 26 of them were actively expressed in root tissue. Several ROS-scavenging genes (GhCAT1, GhSOD1, GhMT3a) showed elevated expression in cotton lines that tolerate salt better. Separately, a newly characterized gene, GhNHX1, appears to control a tonoplast-localized Na+/H+ antiporter and shows higher transcript levels in salt-tolerant genotypes than in sensitive ones, hinting at a role in the tolerance mechanism (Luo et al., 2013; Wu et al., 2004).

On the editing side, tools like meganucleases, TALENs, ZFNs, and CRISPR/Cas systems have become standard for reshaping plant genomes to handle stress. CRISPR-Cas9 in particular has been used to introduce targeted mutations, working through either homology-directed repair or non-homologous end joining. The associated literature illustrates how these molecular tools feed into developing salt-tolerant cotton cultivars (Shelake et al., 2022).

Table 1. Key genes associated with salinity tolerance in cotton and their functional roles.

Sr no.	Genes	Function	Refrence
1	GhDREB	Dehydration-responsive element-binding protein	(Gao et al., 2009)
2	GhERF6	ERF encoding genes	(Jin et al., 2010)
3	GhNHX1	Tonoplast Na/H antiporter	(Wu et al., 2004)
4	GhMT3a	Type 3 metallothionein protein	(Xue et al., 2009)
5	GhMPK2	Mitogen-activated protein kinase	(Lu et al., 2013)
6	GhNAC1 GhNAC4 GhNAC6	Encode NAC dom	(Meng et al., 2009)
7	GhSOD1	Superoxide dismutase	(Luo et al., 2013)

Conclusion and Future Prospects

Salinity stress constrains cotton productivity on multiple fronts at once, disrupting ionic homeostasis, cellular water balance, and redox equilibrium together. As this review shows, oxidative stress and osmotic imbalance are not separate phenomena but closely interconnected processes that jointly determine the severity of salinity-induced damage. ROS accumulation, combined with impaired osmotic regulation, drives metabolic dysfunction, weakens photosynthetic efficiency, and ultimately erodes yield. Cotton counters these pressures through coordinated antioxidant defense and osmolyte accumulation, yet how well these mechanisms perform varies strongly by genotype and is frequently inadequate under prolonged or severe stress.

A major shortcoming in current research is the limited integrative understanding of how ROS detoxification and osmotic adjustment interact. Individual components antioxidant enzymes, proline, and glycine betaine have received extensive attention, but their dynamic coordination with ion transport systems (e.g., Na^+/K^+ homeostasis) and stress signaling pathways remains poorly defined. Much of the supporting evidence also comes from controlled settings, with limited testing under field conditions where salinity interacts with other abiotic stresses.

Moving forward, research should adopt a systems-level approach to map the regulatory networks connecting oxidative stress, osmotic adjustment, and ion homeostasis in cotton. Identifying reliable physiological and biochemical markers, especially those that integrate ROS balance with osmolyte dynamics, will be key to screening salt-tolerant genotypes accurately. Genomics, transcriptomics, and QTL mapping should be leveraged strategically to dissect complex tolerance traits and channel them into elite cultivars via marker-assisted selection. Ultimately, closing the gap between mechanistic understanding and field performance will be essential for developing resilient cotton varieties able to sustain productivity as salinity pressures increase.

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EVALUATION OF COTTON VARIETIES UNDER HIGH DENSITY PLANTING SYSTEM (HDPS)WITH DIFFERENT NUTRIENT LEVELS

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Abstract

A field experiment was carried out during kharif-2 season of cotton growing year 2025-26 at three main cotton research farm, (Sreepur, Gazipur,Jagadishpur,Jashore,and Sadarpur, Dinajpur) of Cotton Development Board to study three cotton hybrid varieties under high density planting system with different nutrient levels. The experiment was laid out in factorial randomized block design. The experiment consists nine treatment combinations with three varieties viz— CB Hybrid-1, White Gold-2, and DM-4 of plant spacing 70 cm × 10 cm and three levels of inorganic fertilizer viz., 100% RDF, 125% RDF and 75% RDF. The results of the study revealed that cotton variety White Gold-2 recorded significantly higher seed cotton yield as well as net returns. Among the different levels of inorganic fertilizers, 125% RDF remarkably improved higher seed cotton yield as well as net returns.

Key Words: High density planting system (HDPS), Cotton varieties, Recommended doses fertilizer (RDF), Sed cotton Yield.

INTRODUCTION:

The increasing population and decreasing land availability pose new challenges for agriculture in Bangladesh. Our primary priority is to feed the people with safe and nutritious food. With cropping intensity nearing 200% and projected to reach 211% by 2030, continuous cropping is likely to harm land fertility and reduce crop productivity by 2050. Horizontal expansion of agricultural land is not feasible due to the shrinking cultivable area. In response, we need to explore and implement vertical expansion like high density planting system, increase yield per unit area in certain regions. Apart

from food, clothing is a fundamental necessity and cotton is the main raw material of textile industries of Bangladesh. Cotton (Gossypium hirsutum L.) belongs to the malvaceae family is the predominant fiber and multipurpose industrial crop of the whole world. In Bangladesh , cotton has an area of 47600 ha with a production of 2.24 lacs bales and productivity of 3 ton seed cotton per ha during 2024-25 and have taken a target an area of 200000 ha with a production of 19 lacs bales in 2050 (Anon 2024-25).Presently we meet only 18-20% of our country demand, but we have a big business demand, In 2025-26 marketing year importers projected to reach 8.4-8.5 million bales of raw cotton to support its massive apparel industry with huge amount of foreign currencies.

Historically cotton is a perennial crop, cotton is cultivated annually, with a growing season from mid-July to December, lasting almost six months at previously but recently growing season have prolonged seven to eight month due to climate change and lack of appropriate varieties and production technologies. The cotton farmers are not satisfied with the yield performance and duration of the existing cultivars. High density planting System (HDPS) with suitable varieties with proper fertilizer management practices may be is now understand as an alternative production system for enhancing the productivity and profitability of cotton. Cotton production with high densities is considered as a potential strategy for reducing production cost by shortening the growing season, a smaller number of cottons picking and leave the land for high value Rabi crop cultivation. Given the high demand and profitability of cash crops and the decreasing land availability, adopting a high-density planting system (HDPS) with suitable cotton varieties and fertilizer proper management is a viable approach to improving cotton yield and production in Bangladesh. High Density Planting Systems (HDPS) are commonly followed to obtain high yields with straight varieties across world, especially in the major cotton growing countries such as USA, Australia, China, Brazil, Turkey, India and Uzbekistan. High Density Planting Systems (HDPS) is more relevant to Bangladesh to establish sustainable production system. So, it was necessary to conduct experiment to know the **Evaluation of cotton varieties under high density planting system with different nutrient levels** growth and yields.

MATERIALS AND METHODS:

A field experiment was conducted during the Kharif 2 season in 2025 at three location namely Cotton Research, Training and Seed multiplication farm, Sreepur, Gazipur, Bangladesh, Cotton Research, Training and Seed multiplication farm Jagadishpur, Jeshore, Bangladesh and Cotton Research, Training and Seed multiplication farm, Sadarpur, Dinajpur, Bangladesh. The experiment was laid out with RCBD design with two factors where factor A: Fertilizer Doses (F1: CDB recommended dose fertilizer i.e. RDF 100%, F2: 25% less of CDB recommended fertilizer dose i.e. RDF 75%, F3: 25% more of CDB recommended fertilizer dose i.e. RDF 125%) and factor B: Variety (V1: CB Hybrid-1) V2: (White Gold-2) V3 :(DM-4)). So, total nine treatment combination with 03 replications comprises the whole experimental design. Cotton spacing (Row × Plant: 70 cm × 10 cm) will be selected for the study. These cultivars will be chosen based on their known adaptability to the region and potential for high yield in Bangladesh. The optimum time of sowing of cotton in Bangladesh is from the month of July to 15th of August. Cotton was sown in all three locations like Sreepur, Sadarpur and Jagadishpur from 31 July to 13 August respectively as per layout. Later all other management like agronomic practices, fertilizers application, crop protection measures and inter cultural operations were done as per farm standard. The soil of the experimental site were red brown in color with clay loam soil texture and low to medium nutrient condition in Sreepur, Gazipur, dark brown in color with clay loam to sandy loam soil texture and medium nutrient in Sadarpur,Dinajpur and dark grey in color with silt loam and medium nutrient in Jagadispur,Jashore. Meteorological data, including temperature, rainfall, and relative humidity, will be recorded throughout the growing season from a nearby weather station to analyze their impact on the growth and development of the cotton plants. For comparison of the performance of HDPS, ten plants were selected randomly from each plot. Then, plant height, Plant population, number of monopodial branch/plants, sympodial branch/plant, bolls/plant, single boll weight and yield, population per hectare, Ginning Out Turn were recorded from those plants. Yield and yield contributing data were analyzed using statistics 10 software by calculating Least Significant Differences (LSD) with 5% level of significance.

Per hectare Fertilizer Guidelines:

INPUTS	RECOMMENDED DOSES FERTILIZER(RDF)(kg)	RDF-25%(kg)	RDF+25%(kg)
Urea	300	225	375
TSP	285	213	356.25
MoP	400	300	500
Gypsum	160	120	200
Boron	25	18	31.25
Zinc	30	25	37.5
Magnesium	25	18	31.25
Organic Mater	2500	1875	3125

Total Distribution per plot:

INPUTS	RECOMMENDED DOSES FERTILIZER(RDF)(kg)	RDF-25%(kg)	RDF+25%(kg)
Urea	0.52	0.40	0.65
TSP	0.5	0.38	0.62
MoP	0.7	0.5	0.87
Gypsum	0.28	0.21	0.35
Boron	0.045	0.03	0.05
Zinc	0.05	0.04	0.06
Magnesium	0.04	0.03	0.05
Organic Mater	4.3	3.25	5.5

RESULTS AND DISCUSSION:

Varieties:

Plant population at harvest, plant height, days to first flower, days to first boll split, sympodial branches, single boll weight and GOT, micronaire and color grade were not significantly influenced due to different varieties (Table: 1, 2 &3) whereas the yield and quality attributing characters viz, boll number per plant, seed cotton yield and fiber length were significantly influenced by different varieties (Table:1,2 &3). Variety V2 and V3 recorded significantly higher number of bolls per plant, number of bolls per square meter, boll weight, seed cotton yield per plant, seed cotton yield, stalk yield and lint yield than V1 variety (Table:2&3). Similar observation was noticed by Ajayakumar et al. (2017) and Anbarasan et al. (2022). Quality parameters viz., ginning percentage, lint index and seed index and fiber length were also recorded significantly higher in variety V2, V3 than V1 variety (Table 2 and 3). Similar observation was noticed by Kumar et al. (2017).

Table 1: Individual effect of Variety (V) and Treatment (F) on growth and phenological trait of cotton at different location:

Factor	Plant height (cm)	Days of first flower	Days to first boll split
Location			
Sreepur	107.89 a	59.815 a	118.07 b
Sadarpur	91.02 b	59.778 a	132.67 a
Jagadispur	115.68 a	55.704 b	97.85 c
Level of Signifi	**	**	**
Variety			
V1	109.22	59.00	117.78
V2	102.43	60.22	118.56
V3	112.02	60.22	117.89
Level of Signifi	Ns	Ns	Ns
Treatment			
F1	105.41	59.11	117.78
F2	106.23	60.00	118.33
F3	112.03	60.33	118.11
Level of Signifi	ns	ns	ns

Table 2: Individual effect of Variety (V) and Treatment (F) on yeild and yield-contributing parameters of cotton at different location:

Factor	Sympodial branch per plant (no)	Boll number per plant (no)	Single boll weight (gm)	Seed cotton yield (kg/ha)	GOT %
Location					
Sreepur	10.38	10.30	5095 b	3694.5	39.50 a
Sadarpur	10.30	10.04	6.40 a	3731.2	38.77 ab
Jagadispur	9.92	10.18	6.25 a	3620.9	38.45 b
Level of Signifi	Ns	Ns	*	Ns	*
Variety					
V1	10.03	9.35 b	6.08	3491.4 b	38.60
V2	9.94	10.56 a	6.25	3847.7 a	38.78

Factor	Sympodial branch per plant (no)	Boll number per plant (no)	Single boll weight (gm)	Seed cotton yield (kg/ha)	GOT %
V3	10.64	10.60 a	6.26	3707.7 a	39.34
Level of Signifi	Ns	*	Ns	**	Ns
Treatment					
F1	9.70	8.25 b	6.01 b	3506.3 c	38.72
F2	10.20	9.18 b	6.24 ab	3670.8 b	39.33
F3	10.71	13.08 a	6.34 a	3869.5 a	38.67
Level of Signifi	ns	**		**	ns

Inorganic fertilizer:

Plant population at harvest, plant height, days to first flower and days to first boll split of cotton was not significantly influenced due to different doses of fertilizer (Table1). The yield attributing characters viz., boll number, single boll weight and seed cotton yield were significantly influenced by different doses of fertilizer but and number of sympodial branches per plant were not significantly influenced by different levels of inorganic fertilizer (Table 2). Significantly quality attributes viz, micronaire value, fiber length was observed with application of 125% RDF, but GOT and Color grade were not significantly influenced by RDF% (Table3) S Aravindhan1, R Gobi1,2*, S Sathiyamurthi3,4, A Balasubramanian 1,5, N Satheeshkumar6, P Stalin1 & K Suseendran1 noticed higher cotton yield with application of higher doses of fertilizer.

Location:

The growth attributing characters viz., plant height, days to first flower and days to first boll split were significantly influenced by different location (Table1), whereas number of sympodial branches per plant, boll number per plant and seed cotton yield did not influence significantly (Table 2). Significantly higher plant height was observed at Jagadispur Farm. These findings have indicated similarities in the vegetative growth characters of cotton in all the areas under study and conforms with the results obtained by Kumar and Ogunlela, (1991b) who reported that vegetative

Growth components similarly resemble each other under the influence of the same factor. Whereas single boll weight and GOT% were significantly

influenced by different location (Table2) and quality attributes fiber length and yellowness were significantly influenced by different location but micronaire value and redness did not influenced by location (Table3).

Table 3: Individual effect of Variety (V) and Treatment (F) Quality parameters of cotton at different location:

Factor	Mic (ug/inch)	Fiber length (mm)	Yellowance of the fiber	Rd (%)
Location				
Sreepur	4.64	30.63 a	6.63 b	68.22
Sadarpur	4.64	30.68 a	6.12 c	67.77
Jagadispur	4.61	30.20 b	7.29 a	67.63
Level of Signifi	Ns	**	**	Ns
Variety				
V1	4.46	28.40 c	6.60 a	67.91
V2	4.60	30.85 b	6.75 a	67.89
V3	4.65	32.26 b	6.70 a	67.82
Level of Signifi	Ns	**	Ns	Ns
Treatment				
F1	4.60 b	30.44 b	6.60	68.65 a
F2	4.68 a	29.93 c	6.74	67.70 ab
F3	4.63 ab	31.14 a	6.71 a	67.27 b
Level of Signifi	**	**	ns	*

Interaction effect:

Interaction effect of varieties and treatments, treatment and location and varieties and location were significantly influenced on seed cotton yield (Table 5). Sowing of variety V2 and RDF125% at plant spacing of 70 cm × 10 cm registered significantly higher seed cotton yield (4364.8 kg/ha) than rest of the other treatment combinations. In case of interaction effect of fertilizer and varieties, fertilizer and location, verities and location on plant height, days to first flower were not significantly influenced but days to first flowers were highly significantly influenced on varieties and location interaction (Table 4) and yield contributing characters sympodial branch per plant and

single boll weight were not significantly influenced by interaction effect while boll number per plant were significantly influenced by varieties and location interaction (Table 5) and seed cotton yield /ha also significantly influenced by fertilizer doses and varieties interaction (Table 5). Similar results were in conformity of Narayana and Aparna (2011). Interaction effect on cotton quality parameter only micronaire values were significantly influenced by fertilizer doses and varieties while fiber length and color grade were not significantly influenced (Table 6).

Based on the results, it can be concluded that V2 variety found superior in high density planting system at spacing of 70 cm × 10 cm and application of 125% RDF found to be optimum for higher seed cotton yield in interaction effect.

Table 4: Interactive effect of Treatment* Variety, Treatment*Location and Variety* Location on phenological trait of cotton at Three Cotton Research,Training and seed Multiplication Farm,Sreepur,Gazipur,Sadarpur,Dinajpur and Jagadispur,Jashore.

Treatment*Variety, Treatment*Location, Variety*Locatin	Plant height (cm)			Days to first flower			Days to first boll split		
	Trt.xVar	Trt.xLoca	vari.xLoca	Trt.xVar	Trt.xLoca	vari.xLoca	Trt.xVar	Trt.xLoca	vari.xLoca
F1xV1	100.06	105.41	109.22	57.44	59.11	59.00 bc	114.67	117.78	117.78
F1xV2	95.44	79.2	89.00	59.22	59.55	58.33 bc	115.78	132.44	130.33
F1xV3	104.54	115.61	113.24	57.55	55.55	55.77 c	117.00	97.22	97.33
F2xV1	103.56	106.23	102.43	58.11	60.00	60.22 ab	115.78	118.33	118.56
F2xV2	101.00	90.77	86.74	59.66	60.22	63.00 a	116.89	132.44	132.67
F2xV3	108.84	116.40	114.76	58.22	55.77	55.66 c	117.00	99.11	98.11
F3xV1	107.86	112.03	112.02	57.55	60.33	60.22 ab	115.00	118.11	117.89
F3xV2	107.49	103.27	97.31	60.00	59.55	58.00 bc	116.67	133.11	135.00
F3xV3	114.98	115.02	119.03	58.11	55.77	55.66 c	116.78	97.22	98.11
Level.Sig:	ns	ns	ns	ns	ns	**	ns	ns	ns

Table 5: Interactive effect of Treatment* Variety, Treatment*Location and Variety* Location on yield and yield -contributing parameters t of cotton at Three Cotton Research,Training and seed Multiplication Farm, Sreepur, Gazipur, Sadarpur, Dinajpur and Jagadispur, Jashore.

Treatment*Variety, Treatment*Location, Variety* Locatin	Sympodial branch per plant			Boll no. per plant			Single boll weight (gm)			Seed cotton yield (ton/ha)			GOT (%)		
	Trt.x Var	Trt.x Loca	vari.x Loca	Trt.x Var	Trt.xLo ca	vari.xL oca	Trt.xVar	Trt.x Loca	vari.xL oca	Trt.xVar	Trt.xLo ca	vari.xL oca	Trt.xVa r	Trt.xLo ca	vari.x Loca
F1xV1	9.51	10.76	10.72	4.74	8.68 b	9.50	5.92	5.83	5.86	3386.5d	3534.8	3401.1	38.31	38.66	39.15
F1xV2	9.51	8.87	10.12	8.46	8.04 b	9.28	5.93	6.33	6.41	3504.8cd	3619.0	3663.5	38.98	38.80	38.20
F1xV3	10.08	9.46	9.25	8.83	8.04b	9.28	6.19	5.88	5.96	3627.6bcd	3365.1	3409.5	38.86	38.70	38.51
F2xV1	10.07	10.21	9.86	8.32	8.89b	10.77	6.15	5.86	6.06	3519.4cd	3660.0	3912.3	38.95	40.42	39.26
F2xV2	9.70	10.28	10.10	7.95	9.07b	10.24	6.29	6.43	6.35	3673.6bc	3676.2	3854.0	39.44	39.24	38.57
F2xV3	10.84	10.12	9.85	11.26	9.47b	10.67	6.27	6.43	6.35	3819.4b	3676.2	3676.9	39.61	38.34	38.51
F3xV1	10.51	10.18	10.58	12.27	13.22a	10.62	6.15	6.17	5.94	3568.3bcd	3888.8	3770.1	38.54	39.41	40.07
F3xV2	10.61	11.75	10.70	15.27	13.00a	10.58	6.54	6.43	6.33	4364.8a	3898.5	3776.2	37.92	38.28	39.55
F3xV3	10.84	10.18	10.66	11.10	13.03a	10.58	6.34	6.43	6.33	3675.5bc	3821.3	3676.2	39.55	38.32	38.40
Level.Sig:	ns	ns	ns	ns	**	ns	ns	ns	ns	**	ns	ns	ns	ns	ns

Table 6: Interactive effect of Treatment* Variety, Treatment*Location and Variety* Location on fiber quality parameters of cotton at Three Cotton Research,Training and seed Multiplication Farm, Sreepur, Gazipur, Sadarpur, Dinajpur and Jagadispur, Jashore.

Treatment*Variety, Treatment*Location, Variety*Locatin	Mic (ug/inch)			Fiber length (mm)			Yellowness of the fiber			Rd (%)		
	Trt.xVar	Trt.xLoca	vari.xLoca	Trt.xVar	Trt.xLoca	vari.xLoca	Trt.xVar	Trt.xLoca	vari.xLoca	Trt.xVar	Trt.xLoca	vari.xLoca
F1xV1	4.55c	4.61	4.66	28.23	30.76	28.23	6.61	6.61	6.60	68.58	68.88	68.00
F1xV2	4.55bc	4.61	4.66	30.91	30.65	28.58	6.57	6.04	6.11	68.71	68.82	67.87
F1xV3	4.68ab	4.56	4.62	32.18	29.90	28.37	6.61	7.14	7.10	68.67	68.26	67.86
F2xV1	4.70a	4.69	4.61	27.96	29.88	30.99	6.56	6.73	6.64	68.14	67.77	68.44
F2xV2	4.67abc	4.69	4.61	30.27	30.11	30.99	6.85	6.16	6.16	67.68	67.74	67.80
F2xV3	4.67abc	4.65	4.59	31.56	29.80	30.56	6.82	7.37	7.45	67.26	67.57	67.44
F3xV1	4.69a	4.63	4.66	29.00	31.25	32.68	6.63	6.56	6.66	67.01	68.00	68.22
F3xV2	4.59abc	4.63	4.66	31.37	31.28	32.46	6.83	6.17	6.11	67.28	67.75	67.64
F3xV3	4.60abc	4.63	4.63	33.05	30.89	31.65	6.66	7.38	7.32	67.52	67.06	67.60
Level. Sig:	**	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

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