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Heat Stress: A Review on Mitigating Breeding Strategies to Reduce Its Effects on Cotton Plant

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Abstract

The need for cotton fibers is rising as the world's population expands, but the production of cotton is challenged by an unpredictably high temperature brought on by quickly shifting climatic circumstances. The stress caused by high temperatures is a significant barrier to global agricultural productivity. Therefore, the creation of cotton cultivars that are thermo-stable is becoming more and more popular. Initiating cotton breeding programs to preserve lint output without compromising its quality under high temperature stress circumstances requires an understanding of the impacts of heat stress on different phases of plant growth and development as well as its tolerance mechanism. The development of improved cultivars by conventional breeding, the use of molecular markers and transgenic technologies, or the use of genome editing techniques to get desired traits are all alternatives that cotton breeders should take into account. The probable consequences of heat stress on cotton plants, tolerance mechanisms, and potential breeding techniques are covered in this review paper.

Keywords: breeding; genetics; molecular; upland cotton; heat stress; climate change

Introduction

Gossypium hirsutum, or upland cotton, is a versatile cash crop. The primary output of this crop is cotton lint, which the textile industry uses to make clothes. With an output of 118.2 million bales and a cultivation area of around 34.1 million hectares, cotton is farmed in more than 35 nations. With an annual production of 6.6 million tons of cotton, China is the world's largest cotton grower and producer. India, United States, Brazil, Australia, Turkey, Pakistan and Uzbekistan are other major cotton-producing nations. Pakistan ranked 7th in terms of cotton production in 2022-23 (Table 1).

The cotton crop encounters a number of issues from sowing through harvesting, such as insect pest infestations, diseases, heat, drought, cold, and salt pressures, trash while picking, and post-harvest management issues (Zhu et. al., 2013; Downes et. al., 2016; Van der Sluijs and Hunter 2016; Zahra et. al., 2021). Each of them results in a considerable decrease in cotton fiber output and quality. Therefore, in order to comprehend these issues, thorough research on each component is needed. The current topic is focused on abiotic stress caused by high temperatures and on reducing losses as a result.

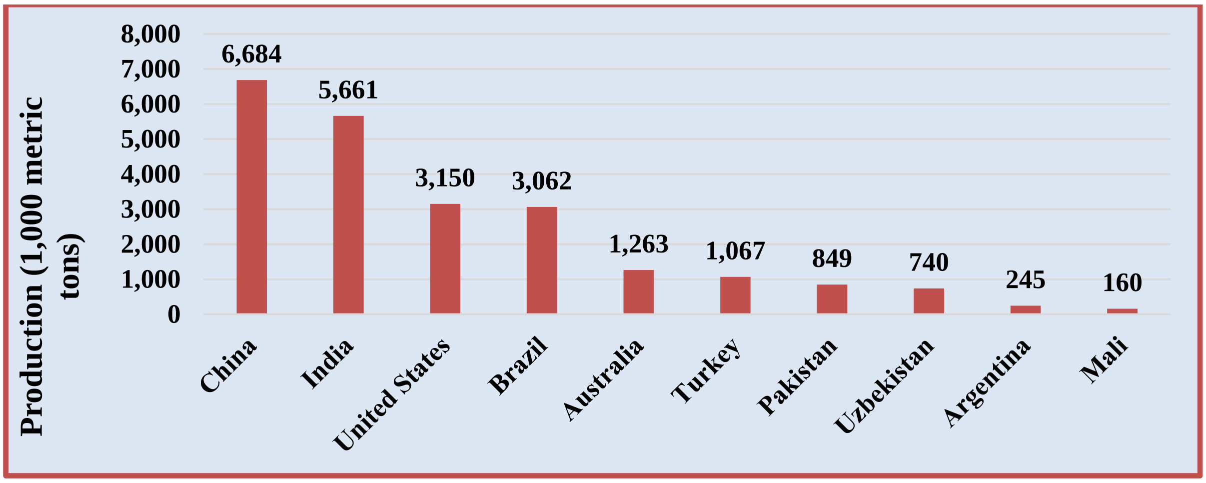

Figure 1: Global production of cotton (country wise, 2022-23)

High temperature stress is another name for heat stress. It is among the variables that restrict agricultural productivity. Heat stress is characterized as a situation when the temperature is high enough for a long enough time to permanently harm plant growth and function (Hall 2001). Plants experience "high temperature stress" when the maximum temperature suddenly rises by 5-7 °C for a few days while the ambient lowest temperature also rises. The required temperature varies depending on the species, as well as the age of the plant, the period and intensity of exposure, the air or soil temperature, and the temperature during the day and night. As a result, a certain temperature cannot be regarded as the critical level for heat stress. In general, compared to warm season crops, cold season plant species are more susceptible to heat stress (Hall 2001; Dhyani et. al., 2013). Moreover, the degree of temperature tolerance varies among plants of the same species that have evolved to various climatic zones. For instance, when temperatures rise over 38 °C, cotton cultivated in China and the United States is seen to be under heat stress, yet in Pakistan and India, this temperature is regarded as the ideal range and temperatures beyond 46 °C are deemed to be excessive heat (Phillips 2012; Raza and Ahmad 2015; Zahid et. al., 2016).

High Temperature Stress and Cotton Plant Growth

The growth, development, and production of various field crops are being negatively impacted by high temperatures in dry and semi-arid parts of the world (Challinor et. al., 2005). Nearly every stage of a cotton plant's life can be harmed by heat stress, however it has been observed that cotton's reproductive phases are more vulnerable to high temperatures than its vegetative development stages (Snider et. al., 2009). The production potential of crop plants is influenced by both daytime and nighttime temperatures, although high nighttime temperatures have a greater negative impact on yield and plant health (Khan et. al., 2020). Figure 2 depicts the detrimental impact of high day/night temperatures on various plant stages.

Figure 2: Key effects of high temperature on Cotton plant growth and development

Mitigation Strategies for Heat Stress in Cotton

a) Conventional Breeding

Breeding stress tolerance requires evaluating the germplasm. To find genotypes that are resistant to heat stress from the available gene pool, numerous experiments have been carried out. Due to their unique traits that domesticated cultivars lack, the use of crop wild cousins is increasingly growing in popularity in plant breeding. Most of these innovative characteristics are connected to biotic and abiotic environmental stress. To have a diverse gene pool, it is advised to screen related wild species and relatives (Majeed et al., 2021). The rapidly developing technologies in plant sciences have made it possible to transfer genes among many species, as is discussed below, even though gene transfer from wild to cultivated species encounters many difficulties and is not always possible without recombinant DNA technology (Raza et al., 2021). The process of transferring a suitable gene and trait to a desirable genotype or purifying the identified plant through selection follows the identification of a suitable gene and trait. The most popular traditional breeding techniques in cotton for this aim include single plant selection, bulk selection, and pedigree selection (Percy, 2003; Tokatlidis et al., 2011).

b) Molecular and Biotechnological Approaches

In recent years, numerous genetic markers connected to various abiotic stressors have been discovered in diverse crops. Using this approach, multiple QTLs with excellent impacts on the HS at various plant growth stages have been extensively identified and characterized (Foolad, 2005). In-depth analysis of breeding material at the phenotypic and genotypic levels is provided by contemporary breeding programs for the quick production of novel kinds. Although previously widely used isozymes and other marker systems were based on protein polymorphism, DNA markers are now often referred to as molecular markers. New options have opened up for investigating genetic diversity, detecting, and improving commercially valuable crop features with the adoption of DNA marker technologies in plant breeding (Preetha and Raveendren, 2008). Breeders now have access to powerful new techniques for identifying complicated quantitative features because to the development of molecular marker technology.

In addition, compared to conventional breeding techniques, DNA marker technology enables breeders to boost productivity, decrease expenses, and shorten the time required to produce new varieties and hybrids. In recent years, several DNA markers and genes regulating yield, quality, and tolerance to biotic and abiotic challenges have been discovered and mapped for many crop species (Zhang et al., 2013; Han et al., 2019; Wang et al., 2019). The majority of the qualities (yield, boll size, fiber traits, etc.) are not selected until the crop has reached maturity when using conventional methods of selection. The development of molecular markers has made it simple to directly select genotypes for complex features, such as stress tolerance traits, even at early growth stages, saving time and labor. Many molecular markers have been found in cotton that are linked to particular genes responsible for tolerance, such as those for salt tolerance (Saeed et al. 2014; Wang et al. 2014a, b), drought tolerance (Hassan et al. 2015; Rodriguez-Uribe et al. 2014), and heat stress tolerance (Demrel et al. 2015). In order to test genotypes for salinity, drought, and heat tolerance at an early stage of plant development, cotton breeders use a number of markers (Chen et al. 2015; Demrel et al. 2015; Rodriguez-Uribe et al. 2014). The physiological traits that these markers are typically correlated with include membrane thermostability, proline contents, photosynthesis activation, and evapotranspiration (Ashraf et al. 1994; Levi et al. 2009a, b). However, reproductive traits like boll retention, boll weight, and seed cotton yield are the ones that are most affected by heat and are not completely correlated with physiological traits. Real-time PCR was used to measure the expression of a few heat responsive genes in tolerant and susceptible upland cotton cultivars in order to understand the molecular mechanism of high temperature stress resistance. The genes are divided into many categories, such as heat stress transcriptional factors (HSFA1b and HSFA2), heat shock proteins (HSP101, HSP70-1, and GHSP26), calcium signaling (ANNAT8), and antioxidant activity (APX1). All genotypes showed an increase in GHSP26 levels, but only the seedlings of heat-tolerant cultivars showed several fold increases in HSP101 and HSP70-1 expression. A heat-tolerant cultivar (VH-260) had significantly higher levels of APX1 expression, which suggests that antioxidant activity contributes to heat tolerance. In cultivars that were sensitive to heat, no discernible alteration in ANNAT8 expression was found. In contrast to heat susceptible accessions, the expression of HSFA2 and HSFA1b was several fold higher in the leaves and ovaries of heat resistant accessions (Zhang et al., 2016).

c) Transgenic Approaches

Transgenic methods have also been widely employed to enhance cotton cultivars for increased resistance to the stress of high temperatures. Recently, cotton was transformed with heat shock protein 70 (AsHSP70) from Agave sisalana using the Agrobacterium-mediated transformation technique. Studies on expression revealed that altered genes expressed themselves more strongly in many plant tissues when the temperature was high. Furthermore, according to quantifiable physiological and biochemical indicators, transgenic cotton plants performed better (Batcho et al., 2021). According to a different study, over-expression of the AVP1 and OsSIZ1 genes in cotton increases lint output in comparison to wild-type cotton under conditions of combined heat and drought stress while having no negative impact on overall cotton yield in the absence of stress. Additionally, transgenic cotton plants showed photosynthetic rates that were 108% greater after heat stress and 72% higher two hours prior to it (Esmaeili et al., 22021). The role of Arabidopsis heat shock protein 101 (AtHSP101) in enhancing pollen tube elongation and increasing germination percentage in high temperatures is well known. When this protein was overexpressed in cotton (Coker-312) in comparison to non-transgenic cotton, these effects were clearly visible (Burke et al., 2015). Therefore, for increased production with a sustainable basis in the face of climate change, greater heat tolerance of reproductive systems in transgenic cotton is essential.

On the other hand, mutant plants of the Arabidopsis SUMO E3 ligase (AtSIZ1) showed greater vulnerability to high temperatures, indicating that AtSIZ1 is a crucial gene for plant heat stress response. By showing increased net photosynthetic rate and better growth and development than non-transgenic plants, the overexpression of the OsSIZ1 gene, a rice homolog of AtSIZ1, in cotton provided tolerance to both drought and heat conditions (Mishra et al., 2017). In a different study, it was discovered that the ectopic expression of the Arabidopsis stress-associated gene (AtSAP5) in transgenic cotton (Coker-312) protects a number of growth and carbon-gain-related components against high heat stress and associated drought (Hozain et al., 2012).

d) CRISPR-Cas Mediated Genome Editing

Based on the level of stress and the stress tolerance mechanism, many genes mostly control tolerance to high temperatures. Targeting a tolerance mechanism that is regulated by several genes will be more challenging. An intricate network of TFs from various families precisely controls the way plants respond to heat stress. These TFs increase the ability of plants to withstand heat stress by regulating the expression of many stress-responsive genes, either singly or in combination with other regulatory elements. Heat stress TFs and HSPs genes have been used successfully in several genetic engineering applications to induce heat stress tolerance in plants (Ali et al., 2020; Meriç et al., 2020). However, due to strict regulatory restrictions, the commercialization of lab research involving genetically modified crop plants may be delayed. Recent advances in CRISPR-Cas-mediated genome editing, an alternative to conventional transgenic methods, enable scientists to change, replace, or swap alleles as well as insert or mute gene(s) in a predetermined manner (Mubarik et al., 2016).

Numerous plant genes change their pattern of expression in response to high temperatures, either by being upregulated or downregulated. Although our knowledge of the genes that are differentially expressed in response to salt and drought stress has grown, research on the genes related to heat stress in cotton has received relatively less attention. HS126, HS128, FPGS, TH1, and IAR3 gene expression increased under high temperatures, according to research on the expression pattern of heat stress sensitive genes in cotton under long-term heat stress. The expression of the genes for ABCC3, CIPK, CTL2, LSm8, and RPS14, on the other hand, was down-regulated (Demirel et al., 2014). Therefore, employing the CRISPR-Cas system to specifically modulate these up-regulated and down-regulated genes in cotton would be an intriguing possibility to counteract the harmful effects of heat stress. Numerous HSPs and TFs linked to genes that are susceptible to heat stress have also been suggested as potential options for enhancing plant heat tolerance (Bhatnagar-Mathur et al., 2008). Understanding the precise function of these genetic regulators opens the door to the development of improved heat tolerance while retaining plant resilience in general.

With the help of CRISPR activation and interference mechanisms, CRISPR-Cas9 has been adapted and used for a variety of different purposes, including regulating gene regulation (Mubarik et al., 2021). Through the CRISPR activation mechanism, positive gene regulators connected to HSPs and stress-related TFs could be activated with high specificity. Furthermore, the CRISPR interference mechanism might be used to eliminate harmful regulators. In one work, CRISPR activator and interference systems were used to up-regulate and repress the BZR1 gene. The findings demonstrate that overexpressing the BZR1 gene increases H2O2 production and restores thermotolerance in rice, whereas plants with gene suppression exhibit impaired H2O2 apoplast production and decreased heat tolerant (Yin et al., 2020). Prior to this, there was little knowledge of MAP3Ks' functions in cotton. According to recent reports, heat stress, pathogen infection, and numerous signaling molecules all increase the expression of the MAP3K65 gene. By negatively regulating processes involved to growth and development, this gene increases susceptibility to pathogen infection and heat stress. Additionally, GhMAP3K65 silencing improved cotton's resilience to pathogen invasion and heat stress. To develop heat tolerance in cotton using the CRISPR-Cas9 genome editing method, GhMAP3K65 is a viable candidate gene to target (Zhai et al., 2020).

Conclusions and Future directions

In all phases of plant development, high temperatures have a detrimental effect on cotton output and growth. By obstructing typical plant biological processes and routes, it lowers lint production and quality. In order to create stress-tolerant cultivars for long-term cotton production under changing climatic circumstances, it is crucial to comprehend the heat tolerance mechanism and molecular characterization of associated genes. Without a doubt, the secondary structure of proteins is maintained by the heat stress TFs, HSPs, and other genes, and quick heat sensing is essential to activate the defense mechanisms against high temperature stress. Modern genome editing tools, speed breeding techniques, and various omics tools are enhancing traditional breeding methods for stress tolerance development, which could speed up the process of creating cotton cultivars with improved heat tolerance. The CRISPR-Cas system is regarded as a non-genetically modified (nGM) technique for engineering high temperature stress tolerance, enabling the development of scientific community efforts to include heat stress resistance in future cotton cultivars for all cotton-growing locations. Furthermore, the concept of speed breeding will be used to address the rising need for high-quality lint output in a fast-expanding global population. To reduce the entire growth cycle and speed up cotton breeding programs, it will facilitate rapid generation progress. The complicated cotton genome has also been successfully sequenced thanks to recent developments in sequencing technology. Following this, the use of diverse omics methods has significantly improved our comprehension of cotton physiology and the roles played by genes in response to heat stress. As a result, various omics technologies may be utilized to identify differentially expressed genes, proteins, and metabolites, which can then be used as a possible biomarker to create cotton cultivars that are resistant to high temperatures.

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Biotic Stress in Cotton; A Review on Mitigating Stress through Cotton Antibiosis Mechanism of Resistance

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Abstract

Cotton faces numerous challenges for its production, with biotic stress emerging as a key one. Biotic stress, primarily caused by living organisms (pests and pathogens) , leads to significant production losses and decreased fiber quality. To fulfill world's demand for cotton-based products and to maintain cotton productivity in this era of climate change, it is an utmost important to understand and mitigate biotic stress. Antibiosis refers to the production of biochemical compounds that cause adverse effects on growth and reproduction of pests and pathogens that feed on host plant. The paper highlights the physiological mechanism underlying cotton's antibiosis mechanism against biotic stressors. Study on biochemical factors present in cotton plant, which cause resistance against different pests and pathogens. Moreover, this review discusses strategies to enhance the antibiosis mechanism in cotton for improved resistance against biotic stresses. These strategies not only contribute to management of biotic stress in cotton plant but also contribute to minimize environmental impact

Keywords: cotton, biotic stress, antibiosis resistance, pathogens, pests, nematodes, plant defense, genetic traits, sustainable agriculture.

Introduction:

Cotton (*Gossypium spp*.) is one of the most economically important and widely cultivated crop, serving as a primary source of natural fibers for the textile industry. Cotton is one of the main cash crops of Pakistan and contributes 10% to gross domestic product (GDP) (Rehman, Abdul, et al 2015). Exports of cotton goods make up over half of the nation's total foreign exchange profits. However, cotton production faces numerous challenges and factors (biotic and abiotic) that causes negative effects on its yield and quality. Among all these, biotic factors are significant contributors to yield losses and reduced crop quality estimated up to 10 to 30% (Hussain, Mehboob *et al.*, 2023). Biotic stresses in cotton arise from various pathogens, pests, and herbivores that feed on or infect the plant, leading to a wide range of detrimental effects. To control these losses, Insecticides and pesticides are used that cause adverse effects on human health and environment like reproductive disorders, diabetes, neurological dysfuction and respiratory disorders. In this way plant protects itself from pathogens and herbivores by synthesizing specialized secondary metabolites that performs defense function due to its cytotoxicity behavior(Duisembecov *et al.*, 2017). Recently studies shows that plant allelochemicals consumed by hosts may have a negative impact on parasitoids, making biological control and plant resistance incompatible(Campbell and Duffey, 1979).

In response to these biotic stresses, cotton has evolved a complex defense system to protect itself from damage caused by invading pathogens and pests. These mechanisms include non-preference, antibiosis and tolerance (Painter RH 1951). In this article, we will discuss antibiosis mechanism of resistance that plays a vital role in mitigating the impact of biotic stress. Antibiosis refers to the ability of the plant to adversely affect the biology and survival of pests or pathogens that feed on or interact with the plant, thereby reducing their population growth or pathogenicity. In cotton, antibiosis related with gossypol content, helicoids, tannins, flavanols and sugar contents. These contents in cotton cause detrimental effects on growth and development of insect and pests.

Understanding the intricacies of cotton's antibiosis resistance mechanism is crucial for developing sustainable and effective strategies to mitigate biotic stress and improve cotton productivity. This review aims to explore the current state of knowledge regarding cotton's antibiosis resistance mechanism, its underlying molecular and biochemical basis, and its potential applications in crop protection and pest management.

Major biotic stresses of cotton crop(*Gossypium Spp.*):

Biotic stress in cotton refers to the adverse effects that imposed on cotton plant by living organisms like insects, pests, pathogens etc. These factors are considered the most important factors that cause great reduction in cotton production. Biotic factors include insect pests, pathogens and nematodes. Losses in cotton yield due to pests may be upto 84% (Kamburova *et al.*, 2018)and due to pathogens are up to 30% (Tarazi *et al.*, 2019).

1. Insect pests:

About 130 species of insect pests attack on cotton plant (Sharma *et al.*, 1982). Insects pests of cotton are classified into two groups according to mechanism of plant damage; chewing insects and sucking-piercing insects(Tarazi *et al.*, 2019). Chewing insects feed on plant biomass; spotted bollworm (*Earias vittella*), fall armyworm (*Spodoptera frugiperda*), pink moth (*Pectinophora gossypiella*), cotton bollworm (*Helicoverpa armigera*)(Stipanovic *et al.*, 1986) and cotton leafworm (*Alabama argillacea*).

The other group that is sucking-piercing damage the plant phloem; it includes cotton aphid (*Aphis gossypii*), boll weevil (*Anthonomus grandis*), thrips (*Thrips tabaci*), cotton flea hopper (*Pseudatomoscelis seriatus*) and spider mites (*Tetranychus urticae*). These insect pests feed on plant leaves, flowers and bolls, leading to defoliation and boll damage (Tarazi *et al.*, 2019).

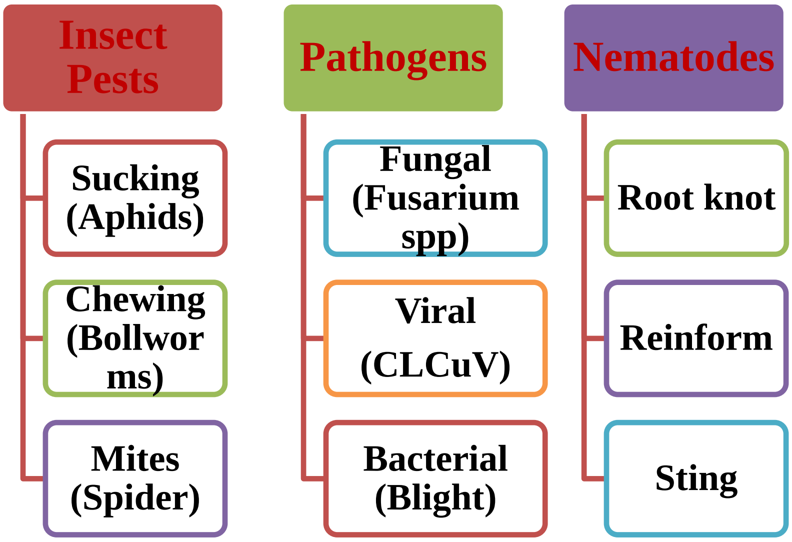

Figure 1: Major biotic stresses of cotton crop (*Gossypium Spp.*)

2. Pathogens:

Major pathogens of cotton are fungal (*Fusarium oxysporum*, *Verticillium dahliae*, and *Alternaria macrospora*), viral (cotton leaf curl virus and cotton mosaic virus, cause stunted growth and leaf curling) and bacterial blight(Sharma *et al.*, 1982). These causes wilting, chlorosis, defoliation and even plant death and eventually all these lead to yield loss

3. Nematodes:

Soil nematodes also cause significant reduction in cotton yield. Major nematodes that parasitized on cotton plant are sting nematode (*Belonolaimus longicaudatus*), root-knot nematode (*Meloidogyne incognita)* and reniform nematode (*Rotylenchulus reniformis)(Weaver, 2015)*.

Antibiosis mechanism of resistance:

Antibiosis is a type of plant defense mechanism against pests, particularly insects through antimicrobial compounds. In cotton, the antibiosis mechanism involves the production of natural compounds or substances that negatively affect the development, survival, or reproduction of insect pests. These compounds are often toxic or detrimental to the pests, making them less likely to feed or reproduce on the cotton plant. The effectiveness of antibiosis resistance can be observed by studying different biological parameters like developmental period, survival, fecundity, longevity, sex ratio of pests (Jindal *et al.*, 2007). These all parameters help to determine the antibiosis level of resistance in cotton. Antibiosis is one of the key mechanisms that help protect cotton crops from insect damage.

Genetic basis of antibiosis:

Cotton researchers are keen to enhance resistance in cotton against pathogens and pests and it opens up new phenomenon of antibiosis (David *et al.*, 2018). Here antibiosis means adverse effects of host organism exerted on pathogen or pest's growth and development (Bennett *et al.*, 2012). In order to develop more sustainable and robust varieties of cotton it is crucial to understand the genetic basis of defense mechanism (Daniell *et al.*, 2016)..

I. The role of secondary metabolites

Production of various types of secondary metabolites are a contributing primary genetic factor that play vital role in antibiosis of cotton (Nikaido, 1996). Secondary metabolites are chemical compounds which have no influence on growth and development of plant rather it plays a crucial role in plant's defense mechanism (Verma and Shukla, 2015). (TAO *et al.*, 2012).

II. Defense genes in plants

An initiation of cascade of biochemical responses when cotton plant detects presence of such pests and pathogens, there is activation of a range of defense genes (Harman *et al.*, 2004). Responses due to activation of defense-related genes include toxic compound's production , defense signaling production and cell wall reinforcement. All these factors aimed to inhibit procreation and growth of invading pests and pathogens (Ab Rahman *et al.*, 2018).

III. Non-specific resistance

Antibiosis is a complex trait in cotton which is influenced by polygenes (Brinkerhoff 1970). There is a continuous spectrum of observable traits, and this type of resistance is called quantitative resistance (Wang and Chen 2017). Several genetic loci control the quantitative resistance, each locus contributes to different degree of resistance against different pathogens (Manosalva, Davidson *et al.* 2009).

Quantitative resistance is generally controlled by several genetic loci, each contributing to varying degrees of resistance against a particular pest or pathogen (Ramalingam, Vera Cruz *et al.* 2003). While this genetic complexity makes it difficult to isolate specific genes, it also allows for a wider range of resistance levels and potentially more durable efficiency against pests that are constantly developing (Kennedy 2008).

IV. Genetic and environmental interactions

Genetic bases of antibiosis are also influenced by some environmental factors. Environmental conditions can be affected on production of secondary metabolites and the expression of defense genes (Li *et al.*, 2020). Environmental factors, which influence those genes, include humidity, temperature and availability of nutrients. In order to develop cotton varieties which exhibit reliable and consistent antibiosis across diverse growing conditions, it is crucial to have understanding of gene-environment interaction (Sams, 1999).

Outcomes for pest management and cotton breeding

It unlocks new possibilities in field of cotton breeding by having a deeper knowledge of genetic basis of antibiosis (Davies, 2002). We can reduce the use chemical pesticides and can promote sustainable agriculture by identifying and selecting varieties which have good antibiosis traits, in this way we can develop varieties that are more resistant to pathogens (Bhattacharya *et al.*, 2020). A deeper understanding of the genetic basis of antibiosis in cotton opens up new possibilities for cotton breeding programs(Trapero *et al.*, 2016). By identifying and selecting for cotton varieties with enhanced antibiosis traits, Furthermore, understanding the precise defense genes and pathways involved in antibiosis can open the door to adapted genetic engineering techniques that give resistance against particular pathogens or pests(Rato *et al.*, 2021).

This defense mechanism is robust because of the interaction of secondary metabolites, defense genes, and the complexity of quantitative resistance (Kennedy, 2008). With this information, we can endeavor to develop ecologically friendly and sustainable pest management techniques that will help cotton growers and the cotton industry as a whole (Deguine *et al.*, 2021).

Factors that influence antibiosis:

Plant chemical defense and natural enemies are two major components that suppress the population of insect pests. Antimicrobial compounds are naturally produced in cotton plant to cause resistance against insect pests. Many secondary metabolites that are correlated with antibiosis but main are terpenoids, tannins and flavonoids and sugar contents.

a. Terpenoids

A set of closely related terpenoids produced in cotton plant (Benson *et al.*, 2001). The best known are gossypol and helicoids, major terpenoids present in dot like glands on all parts of cotton plant that cause resistance to pests including bodies (*Helicoverpa zea),* tobacco budworm (*Heliothis virescens)* and cotton bollworm (*Helicoverpa armigra).* Terpenoids present in small sub epidermal and intracellular pigment glands in all gossypium species (Gershenzon and Croteau, 1991). All exterior tissues, including the seed, contain pigment glands but mainly abundant in leaves and squares.

As antibiosis, it plays a key role in self-protection of cotton plant and acts as a natural insecticidal (Stipanovic *et al.*, 1986). High gossypol content confers resistance to bollworms, tobacco budworm and spider mites (*Altman et al., 1987*). Resistance to insects related with gossypol level; high gossypol content associated with low insect population. The amount of gossypol varies from plant to plant under different environmental conditions (Tian *et al.*, 2019). Gland density also responsible for gossypol concentration in plants (Wilson and Smith, 1976). It generally ranges 0.02-6.64% within plant parts. Cotton seed has the highest amount of gossypol, ranges 33.8-47% in upland cotton and 24.9-68.9% in Egyptian cotton seed (Stipanovic *et al.,* 1986). In wild species, Gossypium devidsonii , it is reported up to 9% (*Altman et al. 1987*).

Compared to mature leaves, immature leaves have 2.5–5 times more terpenoids (Hagenbucher *et al.*, 2013). Stigma and anthers contain about 10 times more gossypol as compared to bracts (Hedin et al., 1992a). In spotted bollworm, gossypol content has deleterious effects (Duhoon *et al.*, 1981) High content of this compound in leaves are strongly correlated with resistance to spider mites (Schuster and Kent, 1980). Glandless cotton is more susceptible to biotic stress as compared to glanded cotton (Jenkins *et al.*,1966).

b. Tannin and Flavonoid's

A large family of natural compounds called polyphenols are widely distributed in plant. Tannins, produced by plants, are stored in vacuoles and cause resistance to insects and diseases(Chan *et al.*, 1978; Schultz 1989; Wu and Guo, 2000; Gershenzon and Dudareva, 2007). They play an important role in plant defense mechanism and are toxic to insects because these compounds bind to salivary proteins and digestive enzymes that leads to inactivation of protein(Blytt *et al.*, 1988). Those insects that ingest high amount of tannin may lose their weight and eventually die.

Flavanols are subclass of polyphenols that are present in cotton plant and act as a natural insecticide (Hedin et al., 1992). It has repellent effects against aphids , leaf miners , bollworms , lygus bugs and spider mites (El-Zik and Thaxton, 1989; Narayanan, 1991). It varies from 0.3 to 1.2% in current cultivars (*Altman et al. 1987*). High concentration of tannins in leaves confers resistance to jassids and in burs associated with bollworm tolerance (Kadapa *et al.*, 1983). High concentration of condensed tannins is present in young leaves as compared to old (Howell *et al.*, 1976). Cotton use flavonoids as defense strategy to produce poison for reducing the growth of lepidopteran insects' larvae(Hanny 1980).

c. Sugar contents

Sugar content in cotton is negatively correlated with resistance to insects (Luo *et al.*, 2011). Low levels of glucose, fructose and sucrose in anthers cause resistance to bollworm and boll weevil (Hedin and McCarty, 1990).

Strategies to enhance antibiosis resistance:

Cotton is a major crop across the world and a vital source of textile fiber and used as raw material for many products. Along with other challenges, pests and diseases impose a serious challenge in cotton productivity. In order to ensure sustainable agriculture implementing effective strategies for enhancing antibiosis resistance in cotton is a major concern.

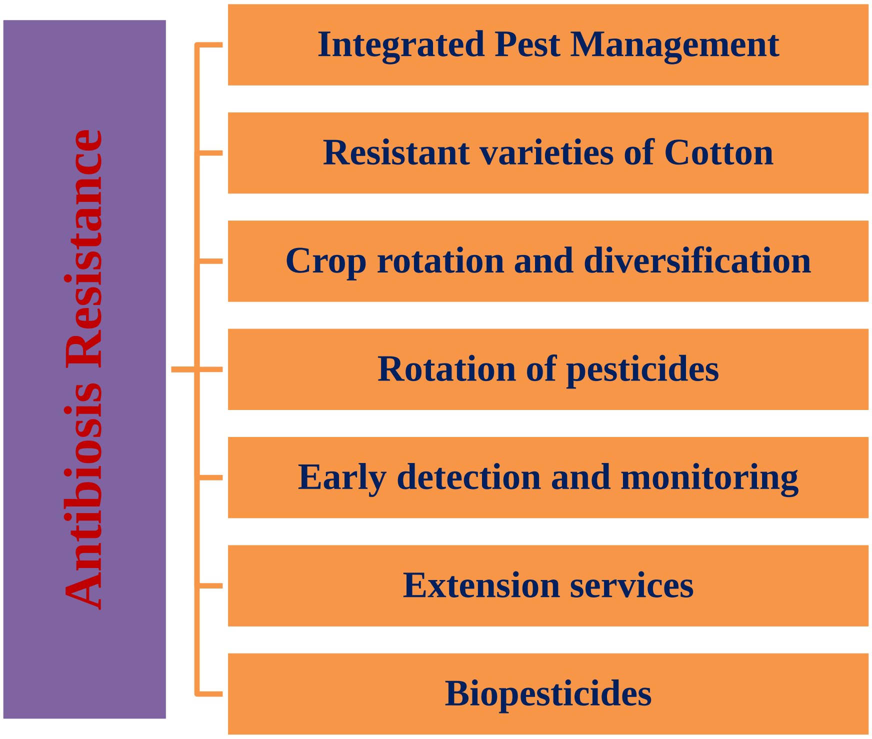

Figure 2: Key strategies to enhance antibiosis resistance in Cotton

1. Integrated pest management (IPM)

IPM include cultural and biological practices by controlling chemical measures to control pests in environment friendly way (Meissle *et al.*, 2010). IPM concern with use of beneficial insects, promoting natural predators, conserving favorable insects, use of resistant cotton varieties, minimizing the use of pesticides thus reducing the selection pressure in pests for antibiosis resistance (Kennedy, 2008) .

2. Resistant varieties of cotton

By advanced biotechnological techniques and conventional breeding methods we can transfer genes that can make cotton plant resistant to specific pests and pathogens (Kennedy, 2008). We can reduce pest damage by using these resistant cotton varieties as a powerful tool and can limit usage of chemical interventions and mitigate the risk of resistance development (Corkley *et al.*, 2022).

3. Crop rotation and diversification

Practices like crop rotation and diversification can disrupt pest life cycle and make environment unfavorable for them to survive continuously (Kremen and Miles, 2012). Farmer can break reproductive cycle of pest by alternating cotton cultivation with pest resistance crops and non-host crops (Trenbath, 1993). In this way we can reduce the population of pests and pathogens and eventually the probability of resistance development (Ingwell and Preisser, 2011).

4. Rotation of pesticides

Using adequate amount and rotating different types of pesticides with distinction in their mode of action are essential in pest management strategies (Demirozer *et al.*, 2012). Indiscriminate and excessive use of such chemicals can enhance the development of antibiosis resistance (Lee *et al.*, 2001). Farmers can slow down the antibiosis resistance development by rotating different classes of pesticides, in this way we can maintain effectiveness of chemical control (Karlsson Green *et al.*, 2020).

5. Early detection and monitoring

It is crucial to identify pest population and their susceptibility level to different pesticides. It can be done by regular monitoring of cotton field (Horowitz *et al.*, 1994). If we timely detect the pests, we can make targeted interventions and minimize their spreading and impact on resistance (Corbel and N'Guessan, 2013). These measures include pheromone traps , trap crops , and modern remote sensing technologies can help in detecting pest pressure precisely (Zijlstra *et al.*, 2011).

6. Extension services

It is vital to promote the adoption of best field practices among cotton farmers by disseminating knowledge about them (Rodriguez *et al.*, 2009). Agricultural extension services must collaborate with Government and research institutions to keep farmer up-to-date to information about integrated pest management, resistance management, and importance of sustainable agriculture (Lefebvre *et al.*, 2015). We can make farmers more adoptive to such practices to enhance antibiosis resistance by empowering them with knowledge (Haque *et al.*, 2020).

7. Bio pesticides

Biological control agents and use of natural sources, such as parasitoids and predatory insects come under the umbrella of bio pesticides. These sources are alternative of chemical pesticides (Van Lenteren *et al.*, 2020).These biological agents have minimum effect on beneficial organisms and environment and effectively target specific pest (Kumar *et al.*, 2008). We can reduce the selection pressure for antibiosis resistance by incorporating bio pesticides in our pest management strategies (Zhu *et al.*, 2016).A multifaceted challenge that requires cooperation of policy makers, cotton researchers and industry stakeholders to enhance antibiosis resistance in cotton. By adopting above-mentioned strategies, we can cope with these issues and can ensure sustainable agriculture.

Conclusions:

Antibiosis is very important as a defense mechanism against biotic stresses in cotton. It investigates the genetic underpinnings and influencing elements of this crucial resistance mechanism, offering insightful information to stakeholders and researchers. In order to improve cotton resilience and maintain a healthy and prosperous cotton business, the study emphasizes the necessity for ongoing research and development in this field.

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