

**Integrating genomic tools and traditional breeding for climate-resilient cotton: A comprehensive review**^a Muhammad Talha Ramzan*, ^b Laiba Razaq, ^c Xiaoqing Zhang, ^d Muhammad Saad Zia, ^e Usama Yaseen, ^f Muhammad Usama Munir Chaudhary, ^g Muhammad Jaffer Ali, ^h Muhammad Ammad khan^a Department of Agronomy, University of Agriculture Faisalabad, Punjab-Pakistan,^b Institute of Horticultural Sciences, University of Agriculture Faisalabad, Punjab-Pakistan,^c College of Life Sciences and Technology, Huazhong Agricultural University, Wuhan-China,^d Department of Soil Science, University of Agriculture, Faisalabad Sub-Campus Burewala, Punjab-Pakistan^e Department of Soil Science and Land Resources, Faculty of Agriculture, Universitas Padjadjaran, Jalan Ir. Soekarno Km. 21, Jatinangor, Sumedang 45363, Indonesia,^f School of Human Sciences, London Metropolitan University, London, UK,^g Department of Plant Breeding and Genetics, College of Agriculture Sargodha University, Punjab-Pakistan.*Corresponding Author's Email Address: ramzantalha037@gmail.com

ABSTRACT

Review Process: Peer review

Cotton (*Gossypium hirsutum* L.) is a globally important fibre and cash crop, playing a crucial role in the agricultural and industrial economy. Climate change, including high and low temperatures, drought, and chilling stress, poses significant challenges to maintaining yield stability and fibre quality. The development of climate-resilient cultivars has become essential to ensure sustainable cotton production and secure the livelihoods of millions of farmers worldwide. This review presents both traditional and modern breeding strategies designed to enhance cotton's resilience and productivity. It begins with an overview of traditional approaches such as mass and progeny selection. Modern tools like genome selection focus on traits associated with abiotic stress tolerance, fibre quality, and seed oil enhancement. Criteria for selection in breeding programs are discussed in detail, followed by the identification of candidate genes that can serve as targets for genome editing technologies. The integration of traditional breeding methods with modern genomic tools offers promising pathways for developing climate-resilient and high-yielding cotton varieties. Candidate genes for fibre yield, oil quality, and stress tolerance have been identified, paving the way for targeted genetic improvements. These advancements are vital for ensuring long-term cotton sustainability aimed at changing climatic conditions. The primary aim of this review is to provide a comprehensive synthesis of the advancements in cultivar selection strategies for cotton, with a special focus on breeding for climate resilience and yield stability.

Keywords: Climate-resilience cotton cultivars, yield stability, fiber quality, genome selection, abiotic stress tolerance.

INTRODUCTION: Cotton is one of the oldest crops to be cultivated and is primarily used to produce raw materials for the textile industry. Over 103 million tonnes of textile fibres were used in 2019, and cotton fibre accounted for almost 24% of the global market. The U.S. Foreign Agriculture Service has reported that the world cotton production in 2023/24 was 112.9 million 480 lb. bales (25.3 MMT), which is a marginal decrease of more than 2% from the 2022/23 production of 116.3 million 480 bales (24.7 MMT). However, its production is increasingly threatened by the multifaceted impacts of climate change. Rising global temperatures, erratic and declining precipitation, intensified drought periods, and the shifting dynamics of pests and diseases pose severe risks to cotton yield and fibre quality (Attia *et al.*, 2021). These climatic stressors not only destabilize traditional farming systems but also reduce the economic viability of cotton cultivation in marginal environments. In this context, breeding for climate-resilient cultivars is no longer a choice but a necessity. Ensuring sustainable yield stability under varying environmental conditions is central to safeguarding the livelihoods of cotton growers and maintaining global supply chains (Razaq *et al.*, 2021).

This review presents a critical investigation into the traditional and the modern breeding techniques used in the cotton enhancement programs. It starts with the traditional methods like mass and progeny selection which played significant roles in the earlier development of the cotton varieties. The review then takes a turn into recent methods of genomic selection in which high-throughput genotyping and phenotyping technologies are used to more accurately estimate breeding values. Moreover, the review identifies the important target genes that can be future targets of genome-editing that can be used to improve the tolerance to abiotic stresses, enhancing the fibre traits and increasing the quality and yield of cotton seed oil (Sattar *et al.*, 2021). The difference between traditional and genomic breeding methods is significant in terms of their contribution towards the adoption of climate-smart cotton cultivars, especially to the challenges of heat, drought, and salinity, as well as pests. The traditional breeding is based on phenotypic selection, hybridization, and recurrent selection, and in the past resulted in varieties that yielded better and had improved fiber quality under stressful environments. Nevertheless, they are labor-intensive and may have their limitations in terms of limited genetic diversity (Mubarik *et al.*, 2020). Conversely, genomic breeding can use high-throughput sequencing, genome-wide association studies (GWAS), marker-assisted selection (MAS), and genomic selection (GS) to enable breeders to precisely and efficiently identify and introgress climate resilience traits (Ashraf *et al.*, 2018). Genomic

platforms also accelerated the mapping of quantitative traits locus (QTLs) that are linked with stress tolerance and fiber improvement, and CRISPR/Cas and transgenic methods enable them to modify stress responsive genes directly (Sheri *et al.*, 2025). In contrast to traditional techniques, where reliance is made on prolonged breeding cycles and field tests, current genomic breeding offers the use of bioinformatics and omics data to progressing innovative designs of cultivars to be resilient to climate change (Khan *et al.*, 2025).

This review described the need to combine traditional and genomic breeding approaches in creating climate-smart cotton cultivars. Traditional and modern selection combined with the most advanced genomics and gene-editing tools is a revolutionary tool to increase the robustness and the reliability of cotton to perform against a wide range of environmental stresses (Sinha *et al.*, 2021). This cross-pollinating breeding system offers a great potential for harmonizing cotton production to the future climatic scenario, thereby, facilitating ecological sustainability as well as economic profitability. The primary aim of this review is to provide a comprehensive synthesis of the advancements in cultivar selection strategies for cotton, with a special focus on breeding for climate resilience and yield stability. Ultimately, this review aspires to guide the development of next-generation cotton cultivars capable of thriving in unpredictable and adverse climatic scenarios.

Traditional cotton breeding for crop improvement: Over the past fifty years, the basic strategies of traditional cotton (*Gossypium hirsutum* L.) breeding have remained the same, while substantial improvements have been made in the techniques and procedures. The traditional method of cotton breeding continues by crossing diverse parent plants, selecting early offspring, and evaluating the resulting varieties, with a focus on prioritizing higher lint yield and improving yield stability through traits like growth, resistance, and maturity (Khan *et al.*, 2022). The innovation of new fibre testing methods has made it easier for breeders to prioritize fibre quality and resolve the traditional negative link between yield and fibre quality. Genetic advancements in traditional cotton breeding will progress as breeders improve and apply novel techniques and strategies (Zhang *et al.*, 2022). In Pakistan, most cotton varieties cultivated in fields have been developed using traditional breeding methods, including crossbreeding, selection, and pedigree-based approaches. Cultivar development occurs in four distinct phases: assessing farmer needs and identifying gaps in existing varieties, crossing the selected parent plants to combine their genes and produce a hybrid with desirable traits, selecting the best progeny from early generations, gradually focusing the selection on elite

lines, and assessing the value of new genotype in comparison to existing varieties (Gudi *et al.*, 2024). For improving cotton crops, the main strategies in traditional cotton breeding include:

Selection: Selection is a breeding method that involves selecting and increasing specific genotypes from a segregating population after hybridization, considering both physical traits and environmental conditions. The causes of variation include inherent mutation, intra-population segregation, and natural crossbreeding. The common selection methods include pedigree, mass, and bulk selection to identify the preferred genotypes. Monogenic and oligogenic traits can be enhanced through traditional breeding, while enhancing polygenic traits using these methods is more challenging. Biotic stresses in cotton can be overcome through intervarietal hybridization including traits such as the lack of nectaries, smooth leaves, and elevated gossypol content in the pistil and some wild diploid varieties are also tolerant to pink bollworm (Han *et al.*, 2023).

Criteria for selection in a cotton-breeding program: Traditional breeding strategies focus on enhancing crop yield, improving fibre quality, ensuring tolerance to biotic and abiotic stresses, promoting early maturity, and improving plant traits, but don't involve exotic gene detection. The selection of cotton plants depends on various factors, including:

Adaptive response to climate change: Crop production declines due to climate-induced factors such as temperature fluctuations, rainfall irregularities, and pest threats, particularly in regions with underdeveloped technology. Traditional breeding methods and modern biotechnological solutions, including hybridization, selection, and transgenic, help sustain cotton productivity and quality (Zhang *et al.*, 2022).

Adaptive tolerance to stress: During all growth stages, cotton plants are affected by abiotic environmental stresses and biotic threats from pathogens, from seed emergence to maturity. Crop production declines of up to 50% are caused by abiotic challenges, including water, heat, and salt stress. Cotton yield is reduced by plant pathogens such as bacteria, fungi, viruses, nematodes, and oomycetes, leading to a global agricultural loss of \$30-50 billion annually. Traditional breeding methods, along with the integration of stress-tolerant genes, are used to overcome these stresses (Zenda *et al.*, 2021).

Crop yield and key components: Cotton yield selection is often based on physical traits, but harvested yield data provides a more accurate method for selecting advanced cotton lines, as it reflects the influence of genetics and the environment. Seeds constitute 60% of the weight in fully developed cotton bolls. Even a minor increase in fibre per seed can lead to a significant improvement in yield. The seed index measures yield and seed surface area (Zhao, 2025). Fibre traits are less affected by environmental variations compared to lint yield. Cotton fibre is formed from seed epidermal cells and is valued for its quality, which is influenced by environmental and storage factors. Tools like HVI and AFIS are used to measure fibre quality. A rapid Q score to assess fibre length, uniformity, and micronaire efficiently (Mollaee *et al.*, 2019).

Early maturity selection: Selecting crops with early maturity is highly desirable. Cotton maturity is assessed visually by calculating the maturity gap in bolls and counting nodes above the white flower and cracked boll. Breeding short-cycle varieties enhances fibre quality for wheat-cotton-wheat systems (Manivannan and Cheeran Amal, 2023).

Minimized leaf trash: Breeders minimize cotton trash content by selecting genotypes with less plant hairiness, which traps debris. They focus on smoother genotypes to achieve cleaner seeds and have developed a specialized system to select low-hairiness genotypes (Riaz *et al.*, 2025).

Mass selection: Mass selection is based on physical characteristics and overall traits. It is most effective for genetic traits but relies on environmental factors and is suited for region-specific plants (Vala *et al.*, 2024).

Progeny selection: Progeny selection evaluates plants by progeny tests instead of visible traits. Seeds are sown to evaluate yield potential. Seeds from productive rows and top-performing half-sibs are combined for selection. Full-sib families developed by crossing two plants undergo trials, and the selected families are recombined using reserved seeds. The breeding program relies on progeny testing to assess offspring phenotypes for genetic selection

(Tejasree, 2024). Phenotypic assessment works for heritable traits, while environmental variation reduces accuracy. Testing multiple offspring addresses these limitations. Environmental variation in large populations is neutralized, and the offspring's performance reflects genetic value. Progeny testing helps select parents and identifies carriers of harmful recessive traits, which appear dominant but transmit recessive traits. This process enhances heritability, with backward selection relying on progeny testing (George and Darshana, 2024).

Genome selection: Genomic selection (GS), a novel MAS technique for plant breeding, is based on the examination of many DNA markers that are uniformly spaced throughout the genome. Haley and Visscher were the first to refer to the concept of genomic selection in 1998. It was three years later when researchers invented and released a GS method that was an advancement in the MAS technology to study quantitative characteristics. Crop genome sequencing has identified many single-nucleotide polymorphisms (SNPs) thereby making GS more productive in plant breeding (Roy *et al.*, 2021). With full-genome SNP chips, different crops can input their DNA polymorphisms on autopilot. The genetic profile of each individual character and each group population will be used in obtaining genomic estimated breeding values of the overall breeding species (Afzal *et al.*, 2023).

The assessment of breeding value in global animal breeding has undergone substantial modifications, particularly with the introduction of the GS approach. The development of foundational knowledge in these fields made it possible to identify genotype animals using DNA markers and sequence the genomes of the three primary agricultural animal species: sheep, pigs, and cattle. It should be mentioned that Holstein dairy cattle had the highest level of performance in the real-world use of GS (Paniza, 2024). Naturally, the advancement of GS techniques led to significant progress in plant breeding as well. GS approach in maize breeding has a larger genetic gain than its pedigree MAS technology. From rice recombinant inbred lines (RIL), possible hybrids were chosen using genomic prediction (Majeed *et al.*, 2021).

They were, based on a population of 215 breeding lines of tetraploid cotton *G. hirsutum*, with allowance of genotype x environment interaction (GxE) to inculcate probable breeding lines against fibre strength and length. The interspecific SNPs of CottonSNP63K have been applied to crosses of several varieties of cotton with *G. hirsutum* L. whereas the intraspecific SNPs are applied within the upland cotton cultivars (Waghmare, 2022). This is possible by using a cottonSNP70K chip whereby the SNPs of four salt-tolerant, as well as 4 salt-sensitive cotton cultivars, can be located (Prabhu *et al.*, 2023).

As earlier mentioned, the SNP arrays can be applied as a breakthrough in the molecular breeding method like; genomic selection (GS), Marker assisted gene pyramiding (MAGP) and marker assisted selection (MAS). Therefore, GS will be more effective than MAS in the effort of improving complex traits that have low heritability, hence a powerful method to apply in molecular breeding in the agriculture sector (Conaty *et al.*, 2022). Breeders can choose new breeding material using this strategy by considering its genetic potential. In other words, rather than waiting for two to three years of field data, the finest hybrid from the breeding population can be chosen based solely on a straightforward DNA test. Additionally, GS enhances the possibilities for simultaneously choosing multiple qualities. The main barrier to the widespread use of this technique in crop selection is the existence of one of the crucial phases for SNP analysis, specifically the high expense of genotyping (Shahzad *et al.*, 2022).

Potential target genes for future genome editing initiatives in cotton: Candidates' gene for increasing abiotic stress tolerance:

Abiotic stresses pose serious threats to the production of cotton. These pressures have a large impact on cotton's morphology and physiology, resulting in a startling 73% loss in cotton yields globally. Cotton's endurance to drought declines throughout the reproductive stage, although it still shows a moderate level of tolerance during the vegetative growth stage (Rasheed *et al.*, 2023). However, prolonged drought and salinity stress can have serious effects on the morphology and functional traits of physiology such as photosynthesis effectiveness (PE), rate of transpiration (TR), plant height (PH), and seed viability (SV) in the cotton plants. Abiotic stressors influence the fate of plants through complex interactions between many genes, transcription

factors, and cis-elements. Whole genome duplication events can be brought on by specific abiotic stressors, giving multi-gene families functional redundancy (Ammar *et al.* 2022).

It is essential to shed light on the intricate tolerance mechanisms and the molecular basis which governs such abiotic stimuli response i.e., salinity, drought, and other high temperatures to develop cotton genotypes tolerant to the same (Ghalib *et al.*, 2025). To reduce such adverse impacts of abiotic stresses, rising interest in the research of stress-responsive loci and genes in allotetraploid cotton has been observed over the last several years. There are several genes which either positively or negatively regulate abiotic stress tolerance and other agronomic characteristics defining the cotton product such as cotton fibre yield and quality, by encoding transcription factors, MAP-kinase signaling cascades protein, ion transporters and protective and adaptive enzymes and proteins responding to stress (Hafeez *et al.*, 2023). Demonstration of the high-rate possibility of enhancing resistance to abiotic stress with the aid of specified genome editing technologies in table 1. As an example, during a study, it was found that GhWRKY6 has adverse impact on transgenic Arabidopsis and cotton under salty and arid conditions. The roots

also were longer than the wild-type and the wrky6 loss-of-function insecticide was insensitive (Mubarik *et al.*, 2020).

Based on recent research, GhADF1 (actin depolymerizing factor 1) plays a crucial role in cotton fibre quality and drought responsiveness. The resultant effect of RNAi-mediated downregulation of GhADF1 was revealed to be greater drought tolerance, over the seed germination period, vegetative growth and during the reproductive phase in transgenic cotton. It was accompanied by the growth of root biomass accompanied by longer primary and lateral root length, larger expanded cells, decreased leaf stomatal (LS) density and leaf conductance (LC), and increased whole-plant water-use efficiency (Iqbal *et al.*, 2023). Moreover, the GhADF1-RNAi transgenic cotton lines exhibited better fibre length (5.6%) and strength (2-12%), thickening of the secondary cell wall, an enhanced fibre yield under agro-environmental conditions, and an augmented amount of cellulose (3%), in normal and stressful situations (drought) of growth (Ghalib *et al.*, 2025). Many negative regulatory genes were not the only ones which could be found in addition to protein-coding genes and transcription factors, as it has been revealed that their constitutive/over-expression leads to the overall enhancement of the resistance of the cotton plant to several types of abiotic stressors (Kumar *et al.*, 2019).

Gene	Description of function	References
Membrane attack complex/perforin gene (GhMACPF26)	Decreased expression contributed to improving tolerance against cold stress	(Chen <i>et al.</i> , 2021)
GhDRP1 (GhPEP2C)	Interfering ABA signaling and the flavonoid biosynthesis route, both decreased and increased expression levels are considered adverse regulators of drought tolerance	(Chen <i>et al.</i> , 2021)
GhPP2C43-A	Inhibiting gene function was recognized as a negative influencer of salinity tolerance	(Cui <i>et al.</i> , 2023)
GhGASA1 and GhADC2	Both increased and decreased gene expression have been confirmed to negatively influence salinity stress tolerance through gibberellic acid (GA) and (polyamine) PA signaling	(Gu <i>et al.</i> , 2021)
Homeodomain (HD)-ZIP transcription factor gene (GhHB12)	Through ABA signaling, the regulation of both drought and salinity tolerance is suppressed	(He <i>et al.</i> , 2020)
GhRaf19	ROS modulation negatively affects drought and salinity stress, but promotes cold stress resistance	(Jia <i>et al.</i> , 2016)
GhDi19-1/GhDi19-2	Increased gene expression heightens sensitivity to salt and ABA stress, indicating negative regulation	(Li <i>et al.</i> , 2010)
GhWRKY6 and GhWRKY6-like	ABA pathways inhibit drought and salt tolerance	(Li <i>et al.</i> , 2019)
GhTULP34	Osmotic stress-induced overexpression suppressed seed germination	(Li <i>et al.</i> , 2021)
GhNHX4	By increasing Na ⁺ and reducing K ⁺ levels in roots, salinity stress response is enhanced through decreased gene expression	(Ma <i>et al.</i> , 2020)
Late embryogenesis abundant protein (Lea)	Promotes drought stress tolerance in plants	(Magwanga <i>et al.</i> , 2018)
Actin depolymerization factor gene (GhADF1)	Increased gene expression boosts drought tolerance and strengthens fiber quality	(Qin <i>et al.</i> , 2022)
GhSAL1	Cold tolerance was suppressed through gene silencing via the IP ₃ -Ca ²⁺ route	(Shen <i>et al.</i> , 2023)
ORP (Oxysterol-binding protein-related proteins) GhORP_A02	Gene silencing improves drought stress response	(Tajo <i>et al.</i> , 2023)
GhWRKY33	The ABA signaling pathway suppressed the drought stress response	(Wang <i>et al.</i> , 2019)

Table 1: Potential candidate genes for cotton genome editing for abiotic stress tolerance.

Candidates' gene for increasing fiber yield and quality: Millions of farmers worldwide depend on the quality of cotton fibre, which is crucial to the global economy. Tetraploid cotton distinguishes itself from other varieties of cotton with its special qualities, such as longer fibre length (FL) and greater fibre strength (FS), which make it extremely beneficial for the textile industry. Crucial genes involved in the production of cotton fibre have recently been identified with notable success. Multiple novel genes that contribute to fibre initiation and elongation have been confirmed using molecular biology methods (Sreedasyam *et al.*, 2024). lists the numerous functional classes of these genes in Table 2. These are sucrose syntheses e.g. GhSusA1, expansions e.g. cpEXPA3, and auxin hormone biosynthesis/translocation regulation genes e.g. GhIaaM/ARF. Moreover, the family of transcription factor such as; GhKNL1, R2R3 MYB (MYB25/MYB25-like, MYB212), MYB-MIXTA-like (MML), GhHD5, GhLIM, GhWRKY, and GhADF1 and light receptors like GhPHYA1 have been discovered to play a significant role in the growth of cotton fibre. These discoveries offer the new opportunities of specific interventions and genetic augmentation to obtain the best properties and quality of cotton fibres. As an

example, drought tolerance during the critical stages of development was also observed following the RNAi-mediated downregulation of the GhADF1 in transgenic cotton plants (Han *et al.*, 2025).

Phytochromes also had the capability of expressing themselves when negative regulation of the GhPHYA1 was weakened like PHYA2, PHYC, and PHYE which enhanced favorable qualities in cotton (Abdurakhmonov *et al.*, 2014). This latter ultimately led to yielding an incredible 10-17% increase in seed cotton production and fibre quality. Another finding is that the cotton pectate lyase-like gene GhPEL76 contributes significantly to the process of fibre elongation or fibre stretch. The lengths of mature fibres were very short in GhPEL76-silencing plants compared to control plants. The dots of lint index and lint percentage have reduced in GhMYB212 upon RNAi exposure in *G. hirsutum* cv Jimian14, which is an indication of its activity to be involved in fibre quality. GhPIN3a was identified as gene that ranges on fibre initiation in the *Gossypium hirsutum* cultivar "Jimian 14." In the case of cotton, these confirmed genes offer a lot of possibilities in relation to genetic improvement methods which would enhance production, fineness and quality of

fibre. Identification and validation of such genes provides an opening to applying the latest genome editing technology to genetic enhancement strategies that might increase the attributes of cotton fibre (Han *et al.*, 2025).

Candidates' gene for increasing cotton seed oil quality and yield: Cotton seed kernel oil is the major source of edible oil after the cellulosic fibres producing approximately 1.6 times the edible oil per fibre. As the 5th largest contributor of vegetable oil in the world after soybean, palm, linseed oil, riveter rapeseed oil, cotton oil contains considerable contents of saturated fat matters such as palmitic, oleic, and linoleic (24%, 22%, and 52 %). Additionally, it is an excellent source of tocopherol, an antioxidant

with vitamin E qualities that greatly extends the products' shelf life. Therefore, the main goal of breeding efforts and genetic mapping techniques has continued to be the production of improved cotton varieties with a high seed oil content without sacrificing the fibre output and its quality. As an instance, multiple transgenic lines of the cotton GhDGAT (Diacylglycerol acyltransferase) overexpressed gene displayed a performed oil augmentation of 4.7% to 13.9%. In another research, it was found that cotton seeds have a specific gene called the gene which encodes carboxylic acid cycle (TCA) enzyme and further; it was found that GhPEP2C (phosphoenolpyruvate carboxylase) gene determines the quantity of protein and oil seed in seeds of cotton (Albahri *et al.*, 2023).

Sr. No.	Gene	Trait	References
1	Phytochrome A1 (PHYA1)	Seed cotton yield enhanced by 10–17% along with improved fibre quality	(Abdurakhmonov <i>et al.</i> , 2014)
2	MYB-MIXTA-like transcription factor	Stimulate fibre formation	(Tajo <i>et al.</i> , 2023)
3	Ubiquitin E3 ligase (GhHUB2)	Increased gene expression promoted fibre elongation and secondary wall deposition, boosting fibre length by 7.49–8.99%, while decreased gene expression suppressed it by 5.85–8.24% compared to wild-type plants	(Feng <i>et al.</i> , 2018)
4	R2R3-MYB transcription factor (GhMYB7)	Formation of fibre length and secondary wall	(Huang <i>et al.</i> , 2021)
5	Histone deacetylase (GhHDA5)	Initial development of fibre emergence and elongation	(Kumar <i>et al.</i> , 2018)
6	Actin Depolymerising Factor (GhADF1)	"RNAi targeting GhADF1 improved fibre length by 5.6% and fibre strength by 2–12%	(Qin <i>et al.</i> , 2024)
7	Pectate lyase-like gene (GhPEL76)	Extension of cotton fibre	(Sun <i>et al.</i> , 2020)
8	R2R3-MYB transcription factor (GhMYB212) and sucrose transporter gene (GhSWEET12)	RNAi targeting GhMYB212 reduced the lint index by 17.2–52.8% and lint %age by 21.8–34.9% compared to the control	(Sun <i>et al.</i> , 2019)
9	WRKY transcription factor (GhWRKY16)	Initiates fibre emergence and development	(Wang <i>et al.</i> , 2021)
10	Auxin efflux carrier protein (GhPIN3a)	Emergence of fibre cells	(Wang <i>et al.</i> , 2021)
11	Rac type GTPases (GhRAC13)	Contributed to secondary cell wall deposition in cotton fibre cells	(Zhang <i>et al.</i> , 2020)
12	Auxin response factor gene (GhARF2b)	Initial stages of fibre cell growth	(Zhang <i>et al.</i> , 2021)

Table 2: Potential genes identified for genome modification to improve cotton yield and fiber quality.

Simultaneous decrease in total proteins and increased deposition of oil content by the seeds through 5.65 and 7.3 % in the cotton RNAi-GhPEP2C transgenic plants was recorded without affecting other agronomic characteristics. Unlike linoleic acid, palmitic and oleic acid (18:1) hold a high level of oxidative stability thus becoming an adequate and viable fatty acid in the food industry. The cotton seeds RNA interference (RNAi) has caused a significant raise in the palmitic fatty acids which is beyond natural values with up to 65% of the total fatty acids in the oil by silencing a keto-acyl synthase (GhKASII) gene. The level of oil in seeds is regulated by an AP2/EREB type transcription factor: WRINKLED1 (WRI1), which is also activated by the changes in enzymes (Zhu *et al.*, 2023).

In upland cotton, the effect on oil accumulation in the development of the seeds demonstrated by ghWRI1 is shown by the elevated values in seed weight and kernel oil composition during overexpression. These have made scientists discover and prove the existence of genes that could enhance and raise the content of oil in cottonseed kernels. However, it can be difficult and time-consuming to introduce improved genes into elite cotton types through sophisticated breeding and genetic approaches. Furthermore, it is challenging to properly and efficiently manipulate specific genes due to the complex genome of upland cotton, which contains several repeating DNA sequences and gene redundancy (Mallikarjuna *et al.*, 2022). The advanced gene editing technology such as the CRISPR/Cas9 system, however, could accelerate the process. Recently, transformed cotton targeting cis-encoded cotton fatty acids synthase-2 (GhFAD2) through CRISPR/Cas methodology has generated transgenics mutant plant that has high seed oleic acid accumulation. There was about a 77-%age-point increase in oleic acid content per seed in the cotton seeds following a desirable change compared with the wild type, which was 5.58-fold faster ($p < 0.01$) and was 13.94% %. These can be used in breeding programs where a parent with the high-oleic trait in its genes is desired and is thus oriented to be crossed with commercial varieties with other desirable agronomic traits (Iqbal *et al.*, 2022).

Current efforts to improve plant architecture in cotton are limited, and there are significant limitations on the investigation of key genes. Some potential genes have been found in (figure 1) that

exhibit early flowering and determinate growth phenotypic traits, such as terminal and axillary flowers (TXandA) on the main stem, which eventually result in a more compact plant structure that makes it easier to plant more densely and produce more. The practical data obtained in these earlier experiments enhances the complex scenario of gene control in cotton plant architecture and provides possible gene targets that can be targeted using the CRISPR/Cas9 knock-out mutagenesis approach to generate the most optimal morphology of the cotton and increased yields (Pourkheirandish *et al.*, 2020).

Harnessing linkage disequilibrium (LD) to advance marker-assisted breeding in cotton: The mapping of the linkage disequilibrium (LD) in cotton genomes has become a pillar in the enhancement of accuracy and effectiveness of marker-assisted selection (MAS). The LD parameter that is a condition of non-random association of alleles at other loci directly influences the power and resolution of an association mapping that is used to determine locations of markers correlated with desired agronomic traits. Heterogeneous population of upland cotton (*Gossypium hirsutum*) showed that LD decayed at around 300 kilobases. This observation meant that breeders need a relatively dense set of molecular markers to accurately detect the loci that tend to fall in the trait interval. This review also emphasized the role of LD structure knowledge that simplified the execution of genome wide association study (GWAS), a method which has effectively been applied in the identification of loci that govern prominent traits in the breeding process and thus breeders will be able to strategically incorporate the markers in their MAS pipelines to the Chinese cotton cultivars (Huang *et al.*, 2017). SSR-based association mapping study using more than 500 Chinese upland cotton cultivars. They have been able to find LD decay patterns over a scale of around 100-300 kilobases, in an agreement with moderate LD as exhibited. They were able to detect various elite alleles which were directly connected to quality character traits of cotton fibers. Such findings are very useful as they are guiding breeders on certain genome locations where MAS may be focused on to enhance fiber mechanical properties in terms of strength, fiber length and growth (Nie *et al.*, 2016).

Notably, high degrees of parents with desirable alleles can be used to speed up the transfer of such characteristics into elite breeding types; hence, improving cultivar performance and field uniformity in the end. The territory studied a worldwide source of upland cotton germplasm. They found that in some of the elite breeding lines their LD was longer, and explained some of this by historical selection bottlenecks and directional breeding. This extended LD implies that shortened markers might be adequate to perform successful MAS in these populations as well as being a potential limitation to mapping. Although possibility of long duration LD would make the selection of marker easy, there is a necessity to have

a reasonable and adequate genetic diversity that would facilitate further breeding and adaptability to emerging environmental stresses (Fang *et al.*, 2013). A breakthrough by directly associating LD-based association mapping with identification of marker-trait associations that are important to increase fiber yield and quality. They showed how careful LD analysis in a broad germplasm of *G. hirsutum* has allowed characterization of robust markers that can be used in MAS which can simplify breeding decisions and shift away from selection by phenotype, which is often time-consuming and environment-dependent.

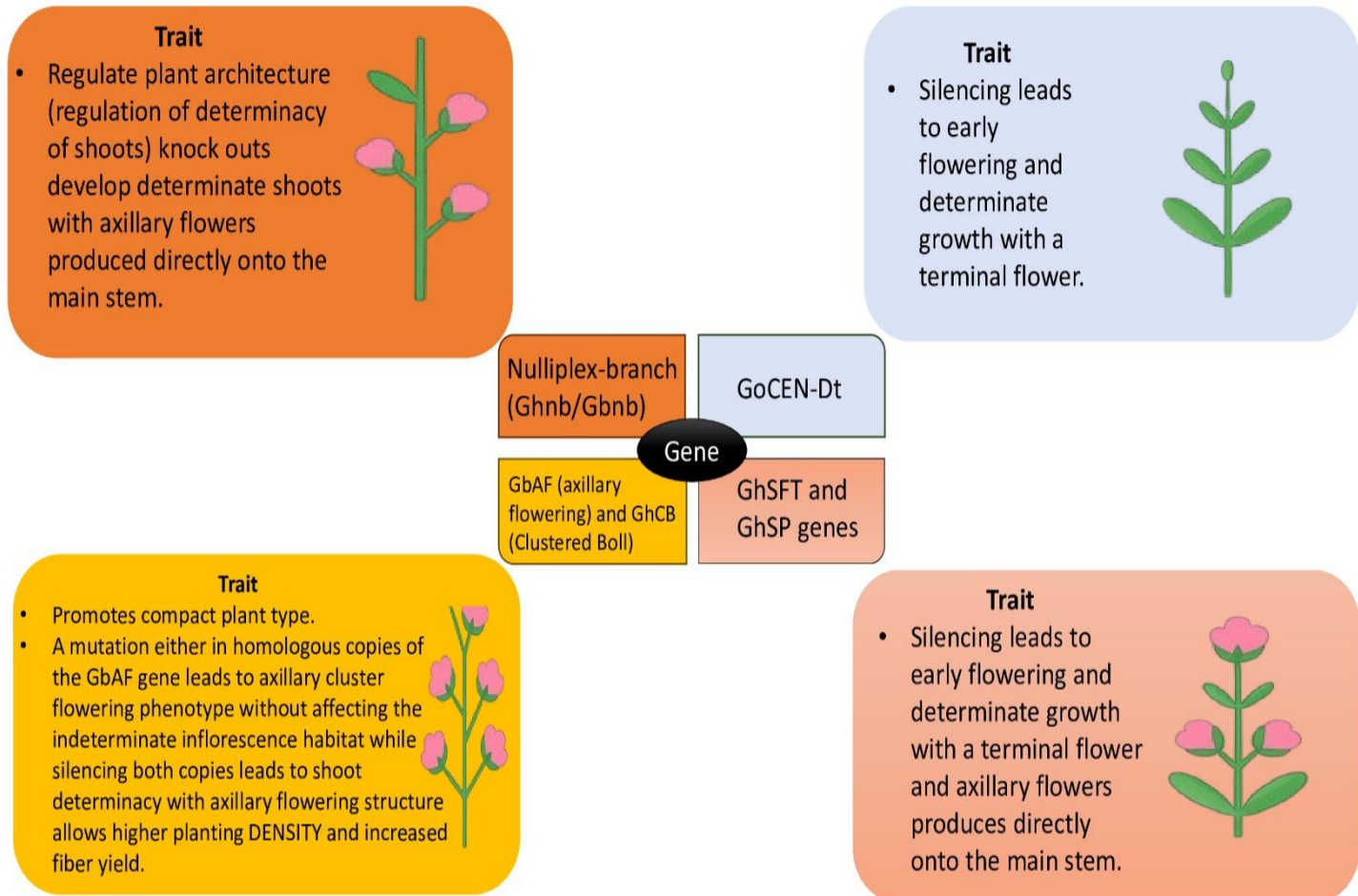


Figure 1: Potential genes for effective genome editing to improve the structure of cotton plants. There has been application of these results to produce high yielding and better-quality fiber cultivars which are more suited to be grown under various climate conditions (Abdurakhmonov *et al.*, 2009). Marker-assisted selection (MAS) in cotton is essentially influenced by the composition and pattern of linkage disequilibrium (LD) in the cotton genome. LD is the non-random connecting of alleles at dissimilar landmarks and the problem is the cornerstone of effective association mapping and genomic selection. The knowledge of LD is especially important in a complex polyploid crop such as cotton, since it determines the degree of precision with which breeders can identify and choose desirable alleles associated with important agronomic traits such as fibre quality yield and stress tolerance in cotton. Various researchers have given an insight into LD patterns in cotton. Genome-wide LD in *G. hirsutum* and was reported to have moderate LD decay generally over the order of centimorgans. This intermediate LD implies that even though the markers and causative loci are sufficiently correlating to allow MAS, uneven decay of the genome can be a problem in fine mapping. This aspect of GM influence marker density requirements on high-resolution mapping: mapping needs where decay of LD is fast required denser marker coverage whereas where decay is slower a wider-spacing of markers is possible (Abdurakhmonov *et al.*, 2009). Wide-range association studies (GWAS) to find out markers that are associated with the yield traits and drought tolerance. Their study found out that LD decays relatively faster at around 20 kb indicating high recombination's and their gingery haplotypes in the cotton lines studied. This fast degradation helps the association mapping better in its resolutions but at the same time, it raises the number of markers needed to sustain the predictive efficiency in MAS (Baytar *et al.*, 2018). Other than yield, LD analysis has proved to play a critical role in the breeding of diseases. A worldwide pool of *G. hirsutum* accessions to be used in mapping the resistance to Verticillium wilt in LD patterns. The study indicated that LD information was reliable enough to identify MAS marker, thus

making it easier to introgress the disease resistant alleles to form elite cultivars (Zhao *et al.*, 2014). Given LD in diverse cotton germplasm and mapping QTLs associated with drought tolerance, they can tell that combined LD knowledge to find out about robust and transferable markers in breeding under environmental stress (Dahab *et al.*, 2016). A key goal in cotton breeding-LD analysis has been useful in improving the fibre quality. LD was found in *Gossypium barbadense* (*Pima cotton*) and emphasized the possibility of exploiting the presence of LD to monitor the dispersal of high-quality alleles of fibre through the breeding population. New marker-trait linkages of lint production through utilization of LD recent patterns finally increasing MAS strength (Kumar *et al.*, 2021). The higher fibre quality cultivars based on LD-guided MAS approach. They showed how well-chosen markers inside high-LD sections may also decrease linkage unintended insertion of repugnant donor alleles-and as a result sourcing efficiency. The inclusion of LD analysis in MAS and genomic selection at the cost of a phenotype-based breeding strategy, leading to precision breeding (Darmanov *et al.*, 2022). **CONCLUSION:** Cotton production is facing risks that climate change may have consequences which are unpredictable in terms of weather patterns such as extreme temperatures and rainfall pattern. Breeding of climate-sustaining cotton species is therefore an interventional measure to ensure the maintenance of yield and fibre quality under these stresses. Traditional methods of breeding, like mass selection, progeny selection, and hybridization are very helpful but their effectiveness has been increased complemented with the modern-day methods like genome selection and marker-assisted breeding. Genome selection (GS) is a breeding method in which genomic markers throughout the whole genome are used to predict the genetic worth of crops to choose, predicting their performance without extensive field trials. This process takes advantage of DNA data to speed breeding cycles and possibly enhance genetic gain. Through these methods it is possible to target

desirable characteristics such as drought and heat tolerance and fibre quality to an amazing specificity. Moreover, stress resilience, yield improvement and seed oil-enhanced candidate gene identification and use holds much potential in fast tracking adaptation of cotton crop. It is important that the convergence of both traditional and modern methods be able to provide sustainable cotton productivity in the context of an ever-threatening climate change.

Future prospective: The future of cotton breeding is in a multidisciplinary researcher who will apply genomics, bioinformatics, gene editing (such as CRISPR/Cas9), and high-throughput phenotyping. It will also be more necessary to develop cotton types that suits agro-climatic areas. The candidate genes identified will be related to abiotic stress tolerance, fibre yield and oil content and their functional validation would hasten the rate of trait introgression. Besides, climate modeling tools can be integrated to assist in predicting future climate conditions and help in hybrid development. Interplay of geneticists, agronomists, and climate scientists will be important in coming up with strong cultivars that can sustain the demand for cotton in the world. Biotechnological tools are becoming cheaper and accessible; hence such tools can now be deployed to benefit even the smallholder farmers of cotton crop, securing the crop as economically viable and environmentally sustainable. In short, the further breeding plan must not only concentrate on productivity, resilience, sustainability, and adaptability to guarantee the sustainability of cotton production across the globe.

Declaration: We hereby certify that the text of this review has neither been published elsewhere, nor is it under consideration elsewhere, and will not be submitted elsewhere.

Certify from authors and co-authors: We certify that all co-authors have been informed and agree with the submission of this paper.

Ethics approval: Not needed for this review.

Consent for publications: Not applicable.

Conflict of interest: The authors declare that they do not have any personal relationships that can affect the work reported in this review.

Data availability statement: No new data were generated or analyzed in this study. All data discussed in this review are derived from previously published works, which are cited appropriately in the manuscript.

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