

Challenges and Opportunities in Cotton Production Research

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Global cotton production has increased over the years, with significant increases in the last 5 years. During 2007/08 a total of 26.3 million metric tons were produced. But, the share of synthetic fibers, consumed at the end use level, overtook cotton in 1995 and continues to increase. The share of cotton in global textile consumption is decreasing and has reached 39% in 2008. This is concerning. The issue relates mainly to rising production costs and stagnating prices. If cotton production costs can be reduced significantly using sustainable low cost production techniques, cotton can claim its due share in textiles again.

Some recent interesting research developments

The distance between output of fundamental science and its application for technology development, has narrowed significantly in recent times. Some very exciting developments have been happening in science over the recent immediate past especially in agricultural sciences. Molecular sciences have pervaded almost all fields of agricultural science leading to exciting breakthroughs, especially in cotton improvement for resistance to biotic and abiotic stresses, herbicide resistance and fiber quality enhancement. Several new concepts in pest management have emerged that have great potential to change the way insects pests, nematodes and pathogens can be managed in a highly precise manner with least effects on non-target flora and fauna. In recent times, new technologies are being invented at a greater frequency than ever before. Insect resistant transgenic crops, RNA interference (RNAi), mutated genes to overcome insect resistance, molecular signaling, *Wolbachia* based control, pheromone and pesticide precision application technologies, nanotechnology, molecular analysis of genetic diversity in crops, allele mining, gene mining, availability of markers for economically important traits, pests, pathogens and organisms of biological control etc., have been signaling a new era in crop improvement.

Insect resistant biotech cotton that silences gossypol

Bollworms survive on cotton because they have an enzyme called P450 monooxygenase CYP6AE14 which digests gossypol. The new biotech cotton expresses dsRNA of the enzyme. When bollworm eats the dsRNA the enzyme is silenced and undigested gossypols remain in the stomach that kills the insect. The cytochrome p450 *cyp6AE14* genes of the cotton bollworm were silenced to disable the bollworm from feeding on gossypol in cotton plants (Mao *et al.*, 2007). The technology has immense potential in pest management that can be sophisticated to the extent of being extremely specific for the

control of target pests alone. The RNAi technology is in the forefront of all the 'state of art' technologies for pest management. Ever since the publication in Nature, 1998 and the noble prize awarded to Drs. Andrew Fire and Craig Mello in 2006, for their discovery of dsRNA based silencing of specific genes through RNAi (RNA interference), the technology has fired the imagination of researchers all over the world.

Prospects of developing low gossypol seed varieties through biotech cotton

Low gossypol seed can be possible through biotech cotton expressing *CYP6AE14* genes from pink bollworm and *Helicoverpa* to be expressed in seeds. The gene sequences are known and seed specific promoters are available. These can be used to develop low gossypol seed varieties.

RNAi-mediated elimination of toxic gossypol from cottonseed

Recently, Keerti Rathore and his team at the Texas A&M University, USA utilized RNA interference to inhibit the expression of the δ -cadinene synthase gene in a seed-specific manner, thereby disrupting a key step in the biosynthesis of gossypol in cotton. Compared to an average gossypol value of 10 $\mu\text{g}/\text{mg}$ in wild-type seeds, seeds from RNAi lines showed values as low as 0.2 $\mu\text{g}/\text{mg}$. Importantly, the levels of gossypol and related terpenoids that are derived from the same pathway were not diminished in the foliage and floral parts of mature plants and thus remain available for plant defense against insects and diseases. Further, they reported that the germinating, RNAi seedlings are capable of launching terpenoid-based defense pathway when challenged with a pathogen. Thus, the silenced state of the δ -cadinene synthase gene that existed in the seed, does not leave a residual effect that can interfere with the normal functioning of the cotton seedling during germination.

Designing genes to kill Bt resistant bollworms

Mutated cadherin alleles in Cry1Ac resistant *H. armigera* insects from field population were found to confer resistance. In an extremely useful study, Soberon *et al.* (2007) showed that susceptibility to the Bt toxin Cry1Ab was reduced by cadherin gene silencing with RNA interference in *Manduca sexta*, confirming cadherin's role in Bt toxicity. Native Cry1A toxins required cadherin to form oligomers, but modified Cry1A toxins lacking one alpha-helix did not. The modified toxins killed cadherin-silenced *M. sexta* and Bt-resistant *Pectinophora gossypiella* that had cadherin deletion mutations. The author suggested that cadherin promotes Bt toxicity by facilitating toxin

oligomerization and demonstrate that the modified Bt toxins may be useful against pests resistant to standard Bt toxins.

Insecticide resistant bollworms can be made susceptible

Bollworms have been found to have developed resistance to insecticides by over-expressing a few enzymes selectively that degrade insecticides. Some of the examples are, cytochrome p450 (*cyp6b7*) over expresses in pyrethroid resistant *H. armigera*; a protease over-expresses in Cry1Ac resistant *H. armigera*; esterase E9 over expresses in Methomyl resistant *H. armigera*; and esterase E5 over expresses in quinalphos resistant strains. The genes responsible for insecticide resistance can be effectively silenced through RNAi so that the insects show susceptibility to the toxins.

Biotech crops that can scare pests

It is now proven that new biotech crops that scare insects can be developed. Insects release chemicals called alarm pheromones when they are scared by their enemies. This warns their colonies to escape. New biotech crops express alarm pheromones that scare specific insects. The alarm pheromone for many species of aphids, which causes dispersion in response to attack by predators or parasitoids, consists of the sesquiterpene (*E*)-farnesene (*Ef*). High levels of expression in *Arabidopsis thaliana* plants of an *Efsynthase* gene cloned from *Mentha piperita* were used to cause emission of pure *Ef*. These plants elicited potent effects on behavior of the aphid *Myzus persicae* (alarm and repellent responses) and its parasitoid *Diaeretiella rapae* (an arrestant response). Also new lectin genes have been found to be effective against sucking pests and are being used to develop cotton crops that can resist sucking pests.

Gadgets for pest scouting

Simple gadgets can be designed to scout insect pests, without having to count any insects. Some plants have been found to help cotton fight pests. Insects make ultrasonic sounds or release pheromones or cause plants to emit ethylene that can be detected by simple gadgets for farmers to precisely detect insect infestations, even from home.

Border plants that help cotton crop fight pests

Insect injury causes signal transduction. The signal transduction pathways leading to the release of plant volatiles have been found to alert other plants in the neighborhood. The scent of jasmine reduces populations of jassids, aphids, and the *H. armigera*, and enhances populations of predators and parasitoids in cotton fields.

Development of ‘Global Cotton’

Cotton is sensitive to photoperiod and thermal conditions and does not adjust easily to new environments. Genetic engineering can help to develop cotton varieties that can grow anywhere in the world. Researchers should be able to exchange

germplasm without any restrictions for the betterment of cotton.

Developing photoperiod insensitive, biotic and abiotic stress resistant biotech cotton varieties

Cotton is sensitive to photoperiod and thermal conditions and does not adjust easily to new environments. For example, it took about 60-70 years for *G. hirsutum* and 150 years for *G. Barbadosense* to adapt to Indian climatic conditions. This clearly indicates that each of the individual cotton genotypes has a specific photoperiod and thermal requirement for optimal performance. Therefore it would be appropriate to identify the highest yielding genotypes for extremely specific geographical zones that have a common photo and thermal profile across the season. Genetic engineering can help to develop cotton varieties that can grow anywhere in the world. Manipulation of Rubisco activase can alter photoperiod and thermal sensitivity to enhance the adaptability of cotton to a wide range of environments. Drought responsive element binding proteins (DREB) *rd29A* genes for drought, high-salt & cold stress have been identified and used in several crops including cotton. Superoxide dismutases (SOD) confer chilling stress and is being explored for its utility in cotton. Biotech cotton varieties for other traits such as drought and disease (leaf curl virus) management have not yet been released commercially and have immense potential in many countries. Herbicide resistant biotech cotton in small scale production systems should find a useful place. Careful planning and design of alternative placement of intercrops are needed to avoid the direct effect of herbicide on them and to ensure that cotton does not become the sole crop in the production systems because of the new weed management biotechnology use.

Combating the recent resurgences of minor pests

Insecticide use on cotton declined significantly after the introduction of insect resistant Bt cotton. As a consequence, several minor pests have been resurfacing in the cotton ecosystems mainly in India and China. Recent reports show that new pests such as the mirid bugs and mealy bugs have been causing significant economic damage, thereby necessitating the continuance of insecticides for pest management. RNAi should be used to develop insect and disease resistant varieties. The insect resistant products developed through RNAi will give India a competitive edge over other countries that have been developing biotech-crops. Efforts should be made to identify ‘insect-species-specific’ genes present in the insect gut that are functionally important for feeding, digestion and other biological activities. There is a need to identify effective siRNAs and/or miRNAs and their targets. Gene sequences and the novel structures must be explored for their utility for crop protection through conventional or transgenic approaches for the management of cotton insect pests such as the bollworms, mealy bugs and new pests.

Biotech cotton with new genes and gene pyramids

Insect resistant biotech cotton has contributed immensely to pest management mainly by causing a significant reduction in the insecticides used to control bollworms. More importantly, farmers in developing countries are no longer stressed with impending bollworm infestations that would otherwise cause severe damage to the crop and thereby reduce production. It is important to develop biotech cotton management strategies so that the full benefits from the technology can be harnessed and the technology can be sustainable for the longest possible time. Bollworm resistance management strategies have not been followed as prescribed in many developing countries, and the problem needs to be addressed on priority basis. Alternative genes (new Cry genes, lectins, protease inhibitors, genes from nematodes etc.) and RNAi based crop protection against insect pests, should be introduced as soon as possible through biotech cotton for more effective pest management. Insect resistant biotech crops (pigeon pea, chickpea, tomato and other vegetables, etc.) that serve as alternate host plants for bollworms should be developed with genes that are not used in biotech cotton. Use of the same Cry genes in all crops will enhance the chances of resistance development in insects.

Impact of climate change on cotton production

Cotton crop productivity is sensitive to climate-induced effects like temperature, rainfall, radiation, CO₂ concentration, and changes in soil, pests and diseases. Work carried out at the Central Institute for Cotton Research (CICR), indicates that select conventional cotton varieties/hybrids are well adapted to elevated CO₂ levels due to better morphophysiological and biochemical attributes. Elevated levels of CO₂ significantly increase plant height, node number, sympodia number, leaf number, leaf area, dry matter production, reduced shedding of bud and bolls and delayed senescence of leaves. Productivity of cotton in terms of total number of bolls and weight increased significantly with an increase of 73%. Fiber quality improved significantly under elevated CO₂ atmosphere. The photosynthetic rate in cotton varieties increased by 34-45% while stomatal resistance decreased significantly. Microbial population increased in soil under elevated CO₂ atmosphere. Elevated CO₂ atmosphere of 650 ppm and temperature of 40 degrees centigrade was found to be optimum for growth of cotton plants. Although it appears that cotton crop will do better in the changed atmospheric scenario during the later part of the 21st century, studies indicate that the pest problem will be aggravated further leading to an increased use of pesticides. By and large, research in India indicates that the impact of climate change on cotton production and productivity will be favorable.

Mechanization of cotton production

Cotton production is labor intensive in almost all developing countries. Cotton production demands labor all through, start-

ing from sowing to harvesting which include several operations including inter-culturing and hand weeding. Cotton in several countries is cultivated in small scale production systems, which demand smaller machines that are affordable for small scale farmers. Several attempts are underway to develop machines for picking and other important operations in cotton cultivation in small scale production systems. Recently, a 3-row, self-propelled check row planter with a pneumatic metering mechanism was developed and evaluated in India. The field capacity was 0.50 hectare per hour with 88% field efficiency. The cost of operation was US\$4.0 per hectare, which is remarkably less than any other traditional method. Also, a self propelled inter-row cultivator was developed and tested on cotton in India. The field capacity was found to be about 0.3 hectare per hour with 48-98% field efficiency. Small-scale, two-spindle machine pickers are being developed and tested for Indian conditions. Research needs to be done to ensure that new machines are developed such that crop production operations are not stalled in rainy days, which is normally the case with labor-intensive operations.

Organic cotton

Biotech cotton is not eligible for certification as organic. The rapid adoption of insect resistant biotech cotton cultivars into many cotton growing agro-climatic zones reduces the benefit of organic cotton and limits the spread of organic cotton cultivation to new areas. At this juncture, it would be possible to promote organic cotton only into the desi cotton (*G. arboreum* and *G. herbaceum*) belt in India. Presently, about 0.5 million hectares are under desi cotton cultivation in India. Initially, organized organic cotton cultivation may be promoted in these areas where the input use in cotton is less and is traditionally non-chemical. Currently, Bt cotton is not permitted in the non-traditional cotton growing states – Orissa, West Bengal and the NE states that grow cotton on about 100,000 hectares. There is further scope for expanding organic cotton into these areas.

Yield and quality enhancement through molecular breeding

Plant breeders all over the world have so far subjected germplasm resources to intensive breeding, so as to enhance yield, fiber quality traits, high oil content or resistance to biotic or abiotic stresses. Such programs also inadvertently result in the narrowing of the genetic base. There is a need to take a re-look at the entire germplasm collection once again in light of the use of molecular markers and the genes that are currently available. The markers and genes identified recently for economically important traits can provide an elegant tool to convert some high yielding germplasm lines into elite cultivars. Out of the 50 cotton species, 5 are included in the primary germplasm pool, 21 as secondary, and 24 as tertiary germplasm pool, based on the relative genetic accessibility. There are several high yielding germplasm lines that are deficient in just one or two economically important traits such as fiber strength or length or susceptibility to biotic or abiotic stresses.

Useful genes can be transferred into cultivars through genetic engineering or desired traits, for which molecular markers are available that can be back-crossed into the lines through accelerated marker assisted breeding. In addition to its lint, the oil and protein portion of cottonseed also represents significant economic value. As far as possible, plant breeding programs should also ensure that the newly developed cultivars have reasonably high levels of oil and proplant-breeding

High yielding elite germplasm lines, which are inferior in only one or two of the desirable traits such as fiber quality or resistance to biotic or abiotic stresses, should be chosen as recurrent parents for marker assisted accelerated back-cross breeding methods. Another set of high yielding germplasm lines should be identified, which possess the trait of interest, and can be used as donor parents. Recently, 2,937 SSR primer pairs have been identified as highly informative which target unique genomic sequences and amplify about 4,000 unique marker loci in a tetraploid cotton genome. Chromosome-marker bins, each 20 cM in size, were constructed on the genetic linkage map containing the markers. Thus 207 marker bins were assigned for a total of about 4,140 cM, which is approximately the size of the tetraploid cotton genetic map. The markers can be used effectively to tag quantitative traits of interest in the already characterized germplasm pools and thereafter be utilized in marker assisted breeding programs for genetic enhancement of elite lines and genotypes to develop high yielding cultivars. Genes conferring strength and fineness can be identified from Ramie and utilized to enhance fiber traits in cotton through genetic transformation. Sucrose phosphate synthase and extensin genes have been shown to enhance fiber length and strength and can be further explored

Issues in fiber quality testing

Cotton fiber quality assessment through instrumentation is still a challenge. There are no rapid internationally acceptable uniform methods of testing cotton for neps, stickiness and maturity. The testing procedures are still time-consuming in many countries. There is an imminent need to invent simple and rapid testing equipment and procedures for fiber quality evaluation that can give a preliminary assessment before the

fiber can be subjected to the high volume instrument and other tests to ensure better returns for the producers.

High yields with narrow spacing and nutrient management

How can yields be increased in developing countries? Yields in developing countries mostly in Africa have been stagnating. Ultra narrow row spacing is highly popular in China, Uzbekistan and several other countries where plant populations of 100,000 to 200,000 per hectare give yields of 7,000-8,000 kilogram of seedcotton per hectare. The same approach should help other countries to identify and develop varieties through 'ideotype breeding' of compact genotypes suited for ultra narrow row spacing, with specific fiber traits for specific locations. Additionally, the compact genotypes with specific fiber traits can be converted to insect resistant biotech cotton. Such location, specific high yielding varieties will ensure sustainable production in major cotton growing countries of Asia and Africa in the future.

The need for a global institute on cotton

There are research institutes on many crops, but not on cotton. Together the governments can move forward for the betterment of cotton, perhaps only through a global institute 'International Cotton Research Institute-ICRI' that may be set up in any of the developing countries, and which addresses all our problems together without having to restrict ourselves to technologies with intellectual property rights issues, especially in Asia and Africa.

References

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