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THE ICAC RECORDER

International
Cotton
Advisory
Committee

Technical
Information Section

VOL. XXXI No. 4
DECEMBER 2013

Update on Cotton Production Research

Contents

Introduction

Dr. Ibrokhim Abdurakhmonov was honored as the 2013 ICAC Researcher of the Year at the 72nd Plenary Meeting of the ICAC held in Cartagena, Colombia from September 29 to October 4, 2013. Dr. Abdurakhmonov was awarded a special trophy and a certificate from the ICAC. He made a presentation on his vision for future directions in cotton research in the field of biotechnology during a Meeting of the Committee on Cotton Production Research of the ICAC on October 2, 2013.

Improvements in cotton agronomy and breeding for high yielding locally adaptive varieties have contributed immensely to increases in yields. But, the recent stagnation and even declines in yields are of concern to researchers, producers and consumers. Expanding threats from both abiotic and biotic stresses, including global warming and the narrowing genetic base of commercialized cotton cultivars, are adding to these concerns. Breeders are being prompted to develop novel biotech cultivars superior to those currently available. Considerable work has already been done to develop large genomics molecular markers, genetic mapping of complex traits, and decoding of the entire cotton genome sequence. According to Dr. Abdurakhmonov, the Uzbekistan government has invested heavily in the development of genetic engineering and genomics research. The application of novel genomics tools and biotechnologies provide a basis to boost yields and other agronomic properties of cotton. Read more in the first article.

The second article is on Long Term Trends in the Cost of Cotton Production in the world. The ICAC carries out a survey of the cost of producing cotton every three years. Thirty-two countries, accounting for almost 90% of world cotton production, participated in the 2013 cost of production survey. Some countries provided data for multiple regions, so the total number of entries is 53; 28 rainfed entries and 25 irrigated entries. Only the long-term trends in world costs are reported in the current article; comparisons among countries will be presented in the March 2014 issue of the *ICAC RECORDER*. The data for 2012/13 showed that the average net cost of production in the world rose to US$1.50 per kilogram; an increase of 28 cents compared to the average net cost of production in 2009/10. The 23% rise in the cost of production in this triennium was greater than the increase between 2006/07 and 2009/10. The main reason for the increase was an increase in the cost of weeding. The use of herbicides is still not common in many countries, while the cost of labor and cultivation are increasing everywhere. Long-term trends in individual inputs are also discussed in the article. The full report, '*Cost of Production of Raw Cotton, September 2013*,' can be requested at <https://www.icac.org/login?url=%2Fpubdetail.php%3Fid%3DPUB00000480>. Previous reports are also available from the ICAC.

2013 Technical Seminar Papers

The 2013 Technical Seminar was held on the topic "Overcoming the Period of No Growth in Yields." Seven presentations were made, including reports from the International Cotton Researchers Association (ICRA), Organizing Committee of the World Cotton Research Conference-6 (to be held in Brazil in 2016) and report of the Round Table for Biotechnology in Cotton. The ICAC Secretariat presented a report on standardization of planting seed nomenclature. The publication is available free at <https://www.icac.org/cotton_info/publications/tech_seminar/pub_tech_seminar/tis2013.pdf>.

ICAC Approved Nomenclature in Planting Seed

The 72nd Plenary Meeting of the ICAC approved a recommendation from the Secretariat to use a standard nomenclature in identifying cotton seed by category. This nomenclature does not cover issues of seed purity, germination and certification standards, which are left to national discretion. The national cotton industries are urged to adopt these recommendations. The nomenclature is:

Breeder Seed - Seed produced or supplied by breeders for multiplication purposes only. Seed may be multiplied by

The ICAC RECORDER (ISSN 1022-6303) is published four times a year by the Secretariat of the International Cotton Advisory Committee, 1629 K Street, NW, Suite 702, Washington, DC 20006-1636, USA. Editor: M. Rafiq Chaudhry <rafiq@icac.org>. Subscription rate: $205.00 hard copy, $170.00 electronic version.

the public or private sectors, but under the supervision of a breeder. Breeder supervision will constitute certification, if required.

Foundation Seed - Seed produced from Breeder Seed. Foundation seed will be used to multiply varieties to supply growers with certified planting seed. External certification by a neutral agency or self-certification will be mandatory. Purity must be at least 99%.

Certified Seed - Seed produced from Foundation Seed that will be supplied to growers.

Registered Seed - Seed produced from Certified Seed by a farmer under his/her own supervision for self-planting or to supply to other farmers. It should carry the label 'not certified.'

Commercial Seed – Seed not recommended for planting. Commercial seed will usually go to oil production or livestock feed.

Meeting of the Southern and Eastern African Cotton Forum - SEACF

The 12th Meeting of the Southern and Eastern African Cotton Forum (SEACF) will be held in Maputo, Mozambique from June 16-20, 2014. The meeting is open to all cotton researchers, public and private. The Mozambique Institute of Cotton (Instituto de Algodão de Moçambique - IAM) will host the meeting. For any additional details contact the organizers Graham J. Thompson <GThompson@arc.agric.za> and Lawrence Malinga <lawrencem@arc.agric.za>.

Role of Genomic Studies in Boosting Yield

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Abstract

Yield declines in cotton production, coming from both intrinsic and operational decreases, are a concern for producers, consumers and researchers. Yield improvement with agronomic properties such as early-maturity and superior fiber quality is the priority target of cotton breeders and cotton researchers worldwide. Contemporary cotton breeding has contributed enormously to developing high-yielding and early-maturing varieties with improved fiber quality. Other agro-technologies have also contributed to greatly improved cotton yields over the past century. The emergence of "biotech crops," adopted worldwide by cotton farmers, has further added to cotton yield gains and has brought significant economic benefits for global cotton farming. However, expanding threats from both abiotic and biotic stresses, including global warming and the narrowing genetic base of commercialized cotton cultivars, generate significant concerns and are prompting breeders to develop novel cultivars that are superior to the current (traditionally-bred or genetically engineered) ones. To address this, with the development of 21st century "omics' sciences, a considerable amount of efforts have been made to develop large genetic and genomics resources for cotton through the characterization of novel genes of agronomic importance, the development of molecular marker resources and genetic mapping of complex traits, the development of better germplasm and populations, and the decoding of the entire cotton genome sequence. These efforts led to the development of novel breeding approaches, such as marker-assisted selection, genomic selection, virtual breeding and new-generation transgenomics tools such as RNAi, which are being widely applied in order to improve cotton quality and boost yields. The objective of this paper is to revisit the current and projected status of cotton yields, causes of yield declines, and efforts, successes, failures and possible future solutions with application of modern "omics" technologies that may boost cotton production worldwide. The efforts and achievements ongoing in Uzbekistan will be briefly detailed.

Key words: *Gossypium*; cotton; cotton yield status; cotton yield decline; biotech cotton; cotton genomics; marker-assisted selection; cotton genome sequencing; Uzbekistan's biotechnology.

Introduction

World agriculture, designed to supply the human diet, clothing, and pharmaceutical products, presently cultivates around 2,000 plant species (Abdurakhmonov, 2012) on around 1.55 billion ha to fulfill human needs. Despite this, product deficiencies still exist widely and will become more common with the global human population increasing to ~9 billion by 2050 (Hubert *et al.*, 2010), whereby ~1 billion people may experience product deficiencies and hunger (http://theconversation.edu.au/time-to-modify-the-gm-debate-210). This danger is prompted by 1) a gradual decrease in cultivated land because of degradation, desertification, urban sprawl, mining, toxic pollution and rising sea levels, 2) declining yields of agricultural crops due to decreased genetic diversity and increased threats of biotic and abiotic factors, and 3) reductions in yield due to climate change that will decrease soil water availability, increase heat stress of plants, and alter crop development cycles.

Soil salinity and drought stress account for large reductions in the yields of a wide variety of crops worldwide. The area affected by salinity is very large (estimated around 320 million ha), and is rapidly increasing due to increased irrigation, farming practices in arid zones, and global warming. At the same time, due to globalization and technological advances, there are urgent concerns for world agricultural production to provide bio-safety/bio-security for the world's leading crop species and safeguard them from biotic (phytopathogens, pests, and invasive species) threats (Sharma *et al*., 2008). For example, biological threats from harmful organisms in agricultural practices cost over $1.4 trillion in crop damage, about 2% of global gross domestic product (GDP). The crop losses are even more severe in developing countries (Sharma *et al*., 2008). This prompts researchers and scientific communities, rather than overlooking crop bio-security issues, to develop bio-secure agricultural programs and to establish an innovative strategy for regional, national and global bio-security threats (Sharma *et al*., 2008).

Main Causes and Factors for Declining Yields

Cotton yield declines can be associated with many indirect factors such as cotton prices, food security, and other complex policy factors, which are outside of the scope of this paper. Genetic and environmental factors affecting cotton yields are discussed here. The decrease in *intrinsic* yield, which is the highest yield obtained under ideal farming situations (Gruian-Sherman, 2009), can be attributed to 1) shrinking genetic diversity of commercial cotton cultivars; 2) challenging and limited use of heterosis in cotton production (only a few countries like India and China use it); and 3) limitations of traditional breeding to rapidly breed a productive plant architecture (e.g. with erect, compact, short internodes, more bolls and fruiting branches, etc.,) with a developed root system, short or medium vegetation, decreased photorespiration, increased photosynthesis, and nutrient utilization capacity.

Decreases in operational yields, which are greatly dependent on environmental influence (Gruian-Sherman, 2009), can be due to 1) again, lack of genetic diversity; 2) biotic (insect, fungal, bacterial, and viral invasions) and abiotic (salt and drought as well as heat eradications) stresses including global warming; and 3) no or limited use of commonly-practiced, efficient, standardized and widely proven agro-technologies to rapidly cope with environmental changes, and to sustain production (e.g. high density planting, drip irrigation, and integrated nutrient and pest management strategies. Of these, at least two are major global concerns for cotton researchers and producers. Firstly, the narrow genetic base of the cotton germplasm, because of a genetic bottleneck derived from historic domestication events and selection (Iqbal *et al*., 2001), caused recent cotton yield and quality declines (Cotton Incorporated, USA). These declines were due to the vulnerability of genetically uniform cultivars to potentially new biotic and abiotic stresses, as well as to the lack of genotypic potential or existence of fewer alleles responsible for yield traits (Abdurakhmonov, 2007; Abdurakhmonov *et al*., 2012a). Second, global climate change caused by increases in atmospheric carbon dioxide (CO_2) is expected to negatively impact agriculture, including cotton (Hake, 2012). Climate change is a huge concern that may contribute to future cotton yield declines. Increased levels of CO_2 may increase fiber yield and water use efficiency (Hake, 2012), and the fertilization effect of increased CO_2 should increase cotton yields by 10%. However, the subsequent temperature increase, projected to be ~2-3°C by 2050, will cancel out much of the potential yield gain mentioned above (Hake, 2012). Global warming will negatively affect (i) nitrate utilization, (ii) bring more drought and heat stress, especially for rain-fed cotton, (iii) cause abnormalities in pollen development affecting fertilization of ovules and reducing boll retention, (iv) accelerate population growth and geographic expansion of cotton insects, and (v) increase extreme rain events and flooding (Hake, 2012).

Approaches to Increase Yields

The widening of the genetic diversity of currently grown cotton cultivars is very important because of the impact on both the intrinsic and operational yield of cotton. Genetic diversity can by increased through combining, introducing or pyramiding new genetic variants that provide better adaption of cultivars to environmental stresses. Wider genetic diversity has the potential to protect crops from massive new pathogens and pest epidemics or sudden environment changes, and thus create an opportunity to further improve yield potential and crop productivity (van Esbroeck *et al*., 1998; van Esbroeck *et al*., 1999; Abdurakhmonov, 2007, Abdurakhmonov *et al*., 2012a). Toward this goal, the 21st century's "omics" science and innovative genomics tools are considered the most promising approaches, in combination with contemporary cotton breeding knowledge and strategies (Abdurakhmonov, 2012; Kumpatla *et al*., 2012). These include (1) accelerated development and success of transgenic, cisgenic and intragenic biotech crop technologies (Waltz, 2012; Lusser *et al*., 2012; Miller *et al*., 2011) with complex effects targeted to improve the *intrinsic* yield in cotton, and (2) decoding of cotton genomes (Chen *et al*., 2007; Wang *et al*. 2012; Paterson *et al*., 2013), and mapping and characterization of the genetic basis of complex traits (as referred to quantitative trait loci-QTLs) that provide better exploitation of existing genetic diversity of cotton germplasm and gene pools and; widening of the genetic diversity of commercialized cotton cultivars using modern marker-assisted selection (MAS), marker-assisted backcross selection (MABS) and genomic selection (GS) programs (Kumapatla *et al*., 2012).

Transgenomic Technologies and Biotech Cotton: Its Role, Success and Perspectives in Cotton Yield Improvement

The first biotech cottons developed using transgenomic tools were the genetically engineered (GE) insect resistant (Bt-cotton) and herbicide tolerant (HT-cotton) cultivars developed to minimize weed control costs, and insect infestation that severely affected productivity. Several toxin producing *Cry* genes from the bacterium *Bacillus thuringiensis* (Bt), notably affecting the larvae of moths *Helicoverpa* ssp. and harmless to other forms of life, were genetically inserted into the cotton genome to produce insect resistant cultivars (Zhang, 2013). Similarly, HT-cottons were developed through introducing the EPSPS gene providing tolerance to the herbicide glyphosate, or with a BXN gene providing tolerance to the herbicide bromoxynil (Zhang, 2013, Saha, 2011). These two transgenic cottons have been widely commercialized over the last 17 years, and the cultivation of genetically engineered crops worldwide increased from 1.7 million hectares in 1996 to 170 million hectares in 2012 in 28 countries (ICAC report, 2013). Being the third largest biotech crop, biotech cotton is currently grown on more than 66% of world cotton area (ICAC report, 2012).

The transgenic technologies, commercialized over the past several years, undoubtedly increased the income earned from cotton worldwide (FICCI report, 2012). Farmers have benefited from biotech cotton cultivation because of decreased insecticide use, reduced energy use, decreased tillage helping to reduce soil erosion, and an overall increase in operational yields. For instance, the national cotton lint yield in India rose to 554 kg/ha in 2006/07, compared to pre-Bt cotton farming with yields of 300 kg/ha during 1993-2001 (Whitefield, 2003; FICCI report, 2012; Jha, 2013). Indian cotton yields have declined in recent years, but this is thought to be connected with the cultivation of cotton in non-optimal conditions (Jha, 2013) as area has expanded. Generally, the contribution of Bt cotton varieties in boosting yields in India can be questioned due to selection and cultivation biases, such as (1) selection of successful farmers as early growers of Bt-cotton, (2) farmers taking special care of Bt-plots, and (3) short-term practices that make comparisons problematic (Stone, 2010).

A recent report compiled by Gruian-Sherman (2009), a senior scientist in the Union of Concerned Scientists (UCS) Food and Environment Program (www. ucsusa.org), based on 11,275 approved field trials for GE crops, including a large number of Bt (3,630) and HT (4,626) trials that covered more than 20 years of research and 13 years of commercialization of GE varieties, concluded that biotechnology "has done little to increase overall crop yields" with the modest aggregated success of Bt-crops. No biotech cultivars have boosted intrinsic yield of any crop with marginal operational yield gains (3-4%). The significant portion of yield increases (24-25%) observed during the 20th century was not the result of GE technologies, but the result of contemporary breeding (Gruian-Sherman 2009). According to the report, a detailed analysis of approved field trials of other transgenic traits intended to boost yields of agricultural crops such as bacterial resistance (139 trials), fungal resistance (713 trials), nematode resistance (51 trials), virus resistance (884 trials), abiotic stress tolerance (583 trials) and yield traits (652) showed limited success (Gruian-Sherman, 2009) in increasing yield components on a crop-wide basis in both national and worldwide levels.

The failure or limited success of currently available or tested GE technologies to improve yields may be due to (1) gradually loosing the 'early-stage' proven transgenic effects (in the cases of Bt and HT) because of the development of tolerance by biotic agents (resistant genotypes to Bt or herbicide tolerant weeds) resulting in more aggressive invasions in crop populations, (2) growth of secondary pests and aggressive pathogens, (3) distribution of existing management practices from "weedy volunteers" (4) non-optimal agricultural farming of GE crops that differ from conventional crops, (5) gradual loss in seed quality of GE crops due to contamination from out-crossing and off-types, and/or (6) through the generation of an epigenetic transgene silencing process that might be unrecognized and not removed in large field plots.

A decrease in the yields of GE crops in subsequent agricultural practices could also come from the introduction of transgenic traits into cultivars that are poorly adapted to local farming conditions. This is especially true with the Bt trait that must be introduced into a local cultivar background through several backcrosses. Often times, conventionally bred and approved local cotton cultivars with earlier crop maturity and desired plant architecture, i.e., more compact and erect types, have been found unsuitable for Bt introgression. Consequently, the varieties used with Bt traits have a lower yield potential but are nevertheless chosen by farmers because they require fewer pesticide applications (ICAC report, 2013).

Despite these facts, transgenic technologies will play a prominent role in improving crop productivity through (1) discovery and application of more novel gene variants of transgenic traits (e.g. new variants of Bt genes or novel candidate genes for insect and disease resistance, such as protease, amylase inhibitors, etc.), (2) use of a combination of different variations for transgenes of interest (i.e., gene stacking), and (3) development of novel GE technologies with more complex genetic effects, affecting many genetic pathways and causing multiple gene interactions compared to the GE crops currently grown, and having fewer interactions with other traits in each plant genome.

The latter case is under primary consideration and focus by researchers who are working to develop GE crops with increased intrinsic and operational yields. These efforts target genes involved with complex genetic and biochemical pathways, affecting light perception and photosynthetic rate, plant architecture and organogenesis, better development of root systems, better nutrient assimilation and water use efficiency, and improved tolerance to abiotic stresses. It is not the objective of this paper to review all details of individual genes that are currently used. Nevertheless, speaking broadly, the genes being used with more complex genetic effects include 1) photosynthetic genes, 2) transcription factors, 3) light perception genes, 4) genes from cell cycle machinery, 5) signal transduction factors, 6) plant hormones, and 7)

Table 1. Examples of Candidate Genes Used in Plant Biotechnology for Increased Crop Productivity

Genes	**Effect**	**Side effects**	**References**
APETALA2 (AP2)	Milder mutations increase flower production, seed size and total plant biomass	Strong mutations can cause abnormal flowers; change in seed fatty acid content; maturity of plants, change in plant phytohormone and defense gene expressions.	Jofuku *et al.*, 2005; Ohto et al., 2005; Ogawa *et al.,* 2007.
FASCIATED EAR2 (FEA2)	Controls branching and seed number		Taguchi-Shiobara *et al.,* 2001.
β-subunit of farnesyltransferase (ERA1/FTA)	Increased yield through drought tolerance	Disease susceptibility, delayed growth, floral organ abnormalities	Wang *et al.,* 2005; Goritschnig *et al.*, 2008; Wang *et al.*, 2009;
PHYTOCHROME B (Arabidopsis)	Overall yield increase (tuber and seed cotton), plant biomass and leaf index	Semi-dwarfism, decrease in apical dominance, and increase in boll size	Thiele *et al.,* 1999; Rao *et al.*, 2011
PHYTOCHROME A1 (Cotton)	Increased seed mass, boll number, boll size, row seed cotton yield, plant growth and biomass, root system-oriented and fiber quality	Slight lint percentage and fiber per seed decrease	Abdurakhmonov *et al.*, 2012c; Abdurakhmonov *et al.,* 2013 in press
Plant sterols/steroids and brassinosteroids	Improved plant biomass, vegetative growth, photosynthetic rate	Not known	Vriet *et al.*, 2012 (review).
Sucrose synthase (Sus)	Improved fiber yield, seed set, leaf expansion	Not known	Xu *et al.*, 2012; Jiang *et al.*, 2012.
Indole-3-acetic acid (IAA)	Lint percentage	Fiber fineness	Zhang *et al.*, 2011
Isopentenyltransferase (IPT)	Grain yield through drought tolerance	Increased expression of brassinosteroid-related genes and repression of jasmonate-related genes	Peleg *et al.,* 2011
Sucrose non-fermenting-1 (SNF1)-related protein kinases (SnRKs)	SnRKs improve yield.	Improved salinity tolerance, responses to nutritional stress and disease, and the regulation of carbon metabolism.	Coello *et al.*, 2011
miR156/156b, miR319, miR396	2-fold increase in the number of flowering shoots and seed yield.	Enhanced levels of seed lutein and beta-carotene	Wei *et al.*, 2010; Rojas *et al.*, 2010
OsSPL14 (SQUAMOSA PROMOTER BINDING PROTEIN-LIKE 14, also known as IPA1)	Promotes panicle branching and higher grain yield.	Not known	Miura *et al.,* 2010
AtNHX1 (that encodes a vacuolar sodium/proton antiporter)	Increased photosynthetic rate, plant biomass, and more fibers with superior quality.	Salt tolerance	He *et. al.*, 2005
Gossypium hirsutum casein kinase (GhCKI) gene	Increased yield through heat tolerance	Not known	Min *et al.,* 2013

small RNA and microRNA genes (Rojas *et al*., 2010). Table 1 summarizes some examples of complex genes used in plant biotechnology, including genes used in cotton transformation. Readers are encouraged to address Edgerton (2009), Rojas *et al*. (2010) and Vriet *et al*. (2012) for detailed discussion of additional genes. Novel genes that are being used for cotton biotechnology have been discussed in the recent report of the Round Table for Biotechnology in Cotton (ICAC, 2013).

Although the side effects (positive or detrimental, see Table 1 on previous page) of using complex gene effects in GE crop development may prevent future commercialization of these new generation GE crops, the fundamental knowledge gained in the genomics era of the 21st Century suggests the possibility of significant yield increases using these new research results and efforts. Success in future GE crop development requires (i) a better understanding of genetic interactions and physiological consequences of modification of genes with multiple effects, (ii) optimization of multiple effects of "candidate genes" in GE development with reduced side effects (with detrimental and harmful impacts), and (iii) the conduct of detailed field trials without the selection and cultivation biases (Stone, 2010) mentioned above. Future efforts also require exploiting a new generation of transgenomics (RNAi, e.g. Mao *et al*., 2007; Saha, 2011; Fire *et al*., 1998; Smith *et al*., 2000; Waterhouse and Helliwell, 2003), synthetic anti-sense oligonucleotide (Higuchi *et al*., 2009), artificial miR (Zhou *et al*., 2013) and a new generation of genome editing such as zinc fingers (Waltz, 2012; Lusser *et al*., 2012) and use of a transcription activator like endonuclease (TALEN; Miller *et al*., 2011) technologies to generate more exact and conserved function of transgenic traits in GE crops.

Marker-assisted Selection: Its Role, Success and Perspectives in Cotton Yield Improvement

As an alternative approach being developed by the genomics research community as a result of decoding entire crop genomes and the development of sufficient genomic resources during the 21st century "omics" era, is molecular breeding or so called marker-assisted selection (MAS). Being distinct from GE approaches, MAS has great potential to improve both intrinsic and operational yields of crop species (Edgerton, 2009; Gruian-Sherman, 2009) including cotton (Abdurakhmonov *et al*., 2011ab; Saha, 2011; FICCI, 2012; ICAC, 2013).

Marker-assisted selection (MAS) is based on identification and tracking of genomic regions in introgression programs using DNA markers and quantitative trait loci (QTL), or association mapping results to select and further breed plant genotypes with minimal genomic fragment introgression; this allows only desirable alleles of interest to be transferred and minimizes the 'linkage' drag effects that negatively affect non-targeted but agronomically important traits (Zeven *et al*., 1983; Tanksley *et al*., 1989; Young and Tanksley, 1989; Abdurakhmonov, 2002; Abdurakhmonov *et al*., 2011ab). In this regard, DNA markers linked to the genomic regions of interest serve as an important tool, enabling breeders to conduct early-stage selection of the best plant(s) on the basis of genotype rather than phenotype (de Vicente and Tanksley, 1993; Abdurakhmonov *et al*., 2011ab). MAS provides many advantages over conventional breeding, a characteristic that is widely discussed by many review papers (Collard and Mackill, 2008; Kumpatla *et al*., 2012).

The most important requirement to conduct a MAS program is the availability of a sufficient number of polymorphic marker panels evenly covering the target genome and associated with a trait of interest, mapping details with flanking loci, the extent of linkage disequilibrium blocks, donor genotypes bearing the QTL of interest, and breeders ability and capacity to use available molecular resources. User-friendly automated genotyping platforms are vital to perform large-scale MAS programs (Collard and Mackill, 2008; Abdurakhmonov *et al*., 2011ab; Kumpatla *et al*., 2012).

The MAS approach is considered to be an efficient breeding tool to improve crops. According to Google Scholar, as of September 15, 2013, there were about 42,200 articles/documents containing the keyword "marker-assisted selection" although many of them related to describing the future perspectives of MAS. MAS articles targeting improved yield traits or discussing yield traits resulted in 33,000 hits. The PubMed search resulted in 2,256 scientific publications with the keyword "marker-assisted selection." Some early successful applications of MAS were improvements in maize (Ragot *et al*., 2007) and soybeans (Cahill and Schmidt, 2004; Crosbie *et al*., 2003; Kumpatla *et al*., 2012), and MAS has been limited to the major crops for which reference sequences are available, e.g., rice and maize (Chen *et al*., 2010; Zheng *et al*., 2008; Kumpatla *et al*., 2012).

Cotton is the world's leading cash crop but lags behind other major crops for marker-assisted selection (MAS) due to limited polymorphisms and 'a genetic bottleneck' through historic domestication (Iqbal *et al*., 2001; Abdurakhmonov *et al*., 2008a). MAS programs are in their early experimental phases in cotton, and MAS platforms are being developed for fiber quality traits (Zhang *et al*., 2003; Abdurakhmonov *et al*., 2008a; Abdurakhmonov *et al*., 2009; Chen *et al*., 2009; Lacape *et al*., 2009; Lacape *et al*., 2010), early maturity (Li *et al*., 2013), yield potential (Abdurakhmonov *et al*., 2007; Qin *et al*., 2008; Wang *et al*., 2007), abiotic (Saranga *et al*., 2001; Levi *et al*., 2011) and biotic stress tolerance traits (Wang *et al*., 2006 Wang *et al*., 2009; Fang *et al*., 2010; Ulloa *et al*., 2011; Ulloa *et al*., 2013); however, there has been only limited success in utilization of MAS for complex traits in cotton (Abdurakhmonov *et al*., 2011a).

Large-scale genomics resources have been developed, and significant advances in cotton genomics have been made (Chen *et al*., 2007; Zhang *et al*., 2008; Wang *et al*., 2012; Paterson *et al*., 2013; Wang *et al*., 2013) for fostering cotton MAS programs that should facilitate better and more rapid cotton improvement programs worldwide, with the potential to raise yields. For instance, recently a researcher released a set of 17 disomic alien chromosome substitution (CS-B) lines through hypoaneuploid-based backcross chromosome substitution lines in a near-isogenic genetic background of TM-1 (Stelly *et al*., 2005). Researchers documented the chromosomal effects (Wu *et al*., 2006) on agronomic properties, including increased yield and fiber quality using these CS-B lines (Saha *et al*., 2006; Jenkins *et al*., 2006). The chromosome substitution lines serve as a new tool for both MAS and conventional breeding programs to rapidly and efficiently improve genetic diversity and yield potential in Upland cotton (Jenkins *et al*., 2006).

Cotton Genome Sequencing and Re-sequencing Efforts

The recent completion of sequencing of a diploid cotton *Gossypium raimondii* genome with the draft (Wang *et al*., 2012) and the first "gold-standard" versions (Paterson *et al*., 2013) has provided a golden opportunity to study orthologous and paralogous genes and gene families in allotetraploid cotton. The completed genome sequencing for ancestral cotton, being the basis for near-future sequencing tetraploid genomes of commercial importance, further sequencing and re-sequencing of cotton genomes (Page *et al*., 2013; Rai *et al*., 2013) foster (1) the development of a whole genome map of DNA markers (Wang *et al*., 2013), (2) development of 'candidate' gene markers based on single nucleotide polymorphisms (SNP), (3) genome-wide association studies to identify biologically meaningful variations (Abdurakhmonov and Abdukarimov, 2008), (4) clearer understanding of transcriptome, proteome and metabolome complexes conditioning the complex traits like yield and quality, (5) allele mining efforts, and (6) understanding the genome-wide gene interactions and epigenetic regulations of agronomic traits in cotton genomes with commercial importance. These will be vital steps to enhance cotton genetic engineering as well as marker-assisted selection programs to develop novel GE technologies (RNAi, TALEN and Zinc fingers) and design more complex MAS programs including genomic selection (GS) and virtual breeding platforms. Unlike MAS, the comprehensive information on all possible loci, haplotypes and marker effects across the entire genome is used in genomic selection to estimate trait effects coming from the genomic regions (Meuwissen *et al*., 2001; Kumpatla *et al*., 2012); thus, increasing the efficiency of breeding for the interested trait. Therefore, modern biotechnology and "omics"-science based breeding with the ability to exploit contemporary breeding tools and agro-technologies have great power to boost cotton yields and provide sustainable cotton production in the future.

Cotton in Uzbekistan

Cotton is one of the priority agricultural cash crops in Uzbekistan and has a significant impact on annual income in the country by providing work and supporting livelihoods in rural areas. Uzbekistan accounts for ~4% of world cotton production and 8% of world exports, and Uzbekistan is the sixth largest cotton producer and the second largest cotton exporter in the world.

Out of 4.4 million hectares of cultivable crop land in Uzbekistan, 1.3-1.4 million ha are occupied with cotton, producing 3.5 to 4 million tons of seedcotton, and exporting cotton fiber valued at ~US$0.9 to 1.2 billion (Abdurakhmonov 2007; Campbell *et al.,* 2010). In recent seasons, Uzbekistan has usually produced between 900,000 and 1 million tons of lint. In addition, the Uzbek cotton crop is produced primarily for the export market and represented 11% of all Uzbek exports in 2012. Sustainability and bio-security of cotton production is pivotal for the Uzbekistan economy because agriculture accounts for 24-28% of total GDP of Uzbekistan (State Statistics report*, 2007: http://www.gov.uz/en/helpinfo/agriculture)*, with the cotton share being 11-17% of the country's exports (Campbell *et al*., 2010; Abdurakhmonov *et al*., 2012a).

Cotton Yield Status in Uzbekistan: Issues, Solutions and Efforts

There has been a noticeable downward trend in cotton production in Uzbekistan during the past few decades. This decline is due to both a reduction in area and lower yields caused by environmental factors. The issues associated with the decline in cotton production in Uzbekistan were addressed by Abdullaev *et al*., (2007), which is outside the scope of this paper. Although this report (Abdullaev *et al*., 2007) concluded that overall, minor yield decrease in cotton were caused by policy choices (such as food security or the quota system for production). However, environmental stresses are the main bio-security issue threatening agricultural production in Uzbekistan, including cotton, and these stresses are related to irrigation and water deficiencies, as well as soil salinization. These issues will pose significant dangers for the region (Cotton Fact Sheet, ICAC, 2011). The current water deficit may further be intensified by over 500% this century, spreading from 2 cubic kilometers in 2005 up to 13 cubic kilometers in 2050 (World Bank country note, 2010). The cotton yield in Uzbekistan was 669 kg/ha in 2011/12, 778 kg/ha in 2012/13 and 738 kg/ha in 2013/14. In the 1970s, yields in Uzbekistan were routinely above 800 kilograms per hectare.

Recent 'ominous forecast' (Sutton *et al*., 2013) for the region projects the possibility of significant temperature increases (2-3°C) for the next 50 years. "Additionally, average annual rainfall may drop by about 10 mm in the highlands and increase by 40 to 50 mm in the desert areas of the country. If no adaptation measures are taken beyond changing planting dates in response to climate change, and taking reduced water availability into account, nearly all crop yields could fall 20 to 50 percent by 2050" (World Bank report, 2013). Further, based on a vulnerability index formed by the World Bank report (World Bank country note, 2010), Uzbekistan is classified as a highly stressed region compared to most of countries in Central Asia and elsewhere.

Cotton production also suffers from pathogen attacks such as wilting disease in cotton, caused by *Fusarium oxysporum*, which is particularly dangerous for Upland cotton (*G. hirsutum*) cultivars in Uzbekistan (Marupov *et al*., 2010). Reports from 2007 to 2011 demonstrated that several other varieties of Upland cottons also became highly susceptible to *Fusarium* wilt (Marupov *et al*., 2012; Egamberdiev *et al*., 2013). Therefore, safeguarding of crop production from biotic and abiotic threats and the development of biosecure agricultural regional and national programs and strategies is vital for Uzbekistan.

In Uzbekistan, the government and research institutions put huge efforts into developing high-yielding, better quality cotton varieties using conventional breeding approaches. These efforts have produced many new cotton cultivars with increased yield and better adaptation for local farming environments (see Abdukarimov *et al*., 2003; Abdurakhmonov, 2007; Ibragimov *et al*., 2008) Abdurakhmonov *et al*., 2012a; Abdullaev *et al*., 2013). This paper is focused on cotton biotechnology and genomics efforts to address efforts at boosting yields.

Cotton Genomics and Biotechnology Research

History and the Main Factors for the Development of Biotechnology

Considering the prominent role of cotton in the economy of Uzbekistan, Mr. Islam Karimov, President of the Republic of Uzbekistan, signed a decree right after the independence of the country in 1992 to establish the Institute of Genetics and Plant Experimental Biology under the Academy of Sciences of Uzbekistan. The Uzbekistan government invested a significant amount of foreign currency in the development of genetic engineering and the genomics of cotton, and in training qualified scientists in related disciplines. During the past 20 years, the scientists have developed an effective research environment, build a well-equipped modern laboratory for genomics and biotechnology research, and they have established wide international scientific networks of collaborators.

The "Center for Genomic Technologies" was established in 2008 within the Institute of Genetics and Plant Experimental Biology. Scientists targeted applied issues confronting Uzbek cotton farmers, as well as basic problems of cotton biotechnology and genomics in the global arena. Further, with the purpose of enhancing cotton genomics efforts and expanding the expertise gained over the past several decades, the government of Uzbekistan has established a fully genomics-dedicated interagency research institute called the Center of Genomics and Bioinformatics (CGB) under the Academy of Sciences, Ministry of Agriculture and Water Resources and the "UzCottonIndustry" association. The CGB is the successor agency to the former "Center of Genomic Technologies" of the Institute of Genetics and Plant Experimental Biology. The Uzbekistan government invested a large amount of funding to build a modern genomics and bioinformatics facility, provide funding for cotton genomics and biotechnology basic and applied projects, and to prepare a young generation of cotton scientists and breeders.

At the same time, the Center developed strong international collaborations with leading cotton genomics and biotechnology laboratories worldwide to support world cotton research programs (Abdurakhmonov *et al*., 2012b). The Center received a number of international grants, which helped to enhance the research facilities, the equipment park and a reagent-material base as well as to promote young scientists and their training in leading research laboratories in the USA (AAAS-ASU joint report, 2012). For example, fruitful collaborations were established with Texas A&M University and USDA-ARS (at Starkville, Mississippi; College Station, Texas; Shafter, California; and Lubbock, Texas). These collaborations accelerated the cotton genomics and biotechnology of Uzbekistan, enhancing the development of genetically engineered cotton varieties, promoted technology transfer, and jointly patented newly developed products (AAAS-ASU joint report, 2012).

Genetic Mapping and Marker-Assisted Selection Efforts

Uzbek researchers participated in the development of pioneer cotton SSR marker collections (JESPR) consisting of 309 microsatellite markers (Reddy *et al*., 2001), which found wide application in cotton genome investigations worldwide, and JESPR was widely utilized in cotton genomics research in Uzbekistan.

As cotton production in many ways is connected with earliness, boll maturation and plant senescence, a natural leaf defoliation QTL was mapped using SSR markers, and a unique naturally leaf shedding Upland cotton line was developed (2n=2x=52), derived from tri-species crosses between *G. thurberi* (D_1D_1, 2n=2x=26, natural defoliant) x *G. harknessii* (D_2D_2, 2n=2x=26, wilt resistant) x *G. hirsutum* (variety Tashkent-1, AADD, 2n = 2x = 52). These QTL associated SSR markers were assigned to the short arm of chromosome 18, suggesting indirectly that gene(s) associated with natural leaf defoliation is located on this chromosome (Abdurakhmonov *et al*., 2005)

A set of unique recombinant inbred cotton lines (RILs) segregating for lint percentage, derived from the cross of lintless/fuzzless mutant and linted/fuzzy cottons, were exploited to tag QTL loci responsible for lint percentage traits using SSR markers. Multiple QTL-mapping (MQM) revealed that at least, two highly significant fiber development QTLs exists around regions TMB0471 and MGHES-31 (which explained about 23-59% of the phenotypic variation of lint percentage) and around markers MGHES-31 and TMB0366 (which accounted for 5.4-12.5% of the phenotypic variation of lint percentage). These lint-percentage-trait associated SSR markers have been located on chromosomes 12, 18, 23, and 26 using deletion analysis in aneuploid chromosome substitution lines (Abdurakhmonov *et al*., 2007a).

Considering the importance of day-neutral cotton flowering for Uzbekistan (the northernmost cotton growing country) that helped to develop cultivars with early flowering and maturation, researchers investigated a collection of photoperiod-converted radiomutants (32P), including their wild-type parental lines using SSR markers. Impact and pressure of radiomutagenesis to the SSR mutation patterns were studied, bi-parental mapping of populations via crossing the photoperiod-converted, day-neutral flowering radiomutants to the original photoperiodic wild parents were created, and consequent progenies segregated for the early flowering trait (Abdurakhmonov *et al*., 2007b) were obtained. Utilizing a collection of large SSR markers, QTL regions responsible for photoperiodic conversions and causing the day-neutral flowering after the mutation were mapped. These QTL regions were localized on chromosome 5 of cotton (Kushanov *et al*., 2010; Kushanov *et al*., unpublished).

Due to the emerging threat of *Fusarium* wilt disease in Uzbekistan (Marupov *et al.,* 2012), Uzbek cotton germplasm resources were evaluated for *Fusarium oxysporum* sp. *Vasinfectum* (FOV) resistance, a race/genotype distribution of FOV fungi in Uzbekistan was studied (Egamberdiev *et al*., 2013), including frequently occurring races and their pathogenecity (Egamberdiev *et al*., unpublished) were identified, and a bi-parental mapping population segregating for FOV resistance was created in order to map QTL loci controlling FOV resistance in cotton. Several QTL loci contributing for FOV resistance in cotton were mapped using SSR markers (Abdullaev *et al*., 2010; Abdullaev *et al*., 2013, unpublished).

A large number of *G. hirsutum* varieties and exotic accessions from Uzbek germplasm were characterized for both fiber quality characteristics and molecular diversity within the framework of international collaborative projects (Abdurakhmonov *et al*., 2004; Abdurakhmonov *et al*., 2006; Abdurakhmonov *et al*., 2008a; Abdurakhmonov *et al*., 2009; Abdurakhmonov *et al*., 2012). Researchers estimated the extent of genome-wide linkage disequilibrium (LD) and association mapping of fiber quality traits using >200 microsatellite markers in narrow and broad-based *G. hirsutum* germplasm grown in two diverse environments, Uzbekistan and Mexico. The existence of agronomically useful genetic diversity within a large cotton germplasm collection was demonstrated. Researchers found the level of LD between SSR sites in broad-based exotic germplasm (11-12%) was greater than in narrow-based cultivar germplasm (4-9%) groups. A genome-wide average of LD extended up to genetic distance of 25 cM and ~5-6 cM at $r^2>0.2$ in variety germplasm at the significance threshold ($r^2>0.1$). A genome-wide average of LD declines within the genetic distance at <10 cM in the landrace stocks germplasm and >30 cM in photoperiodic variety germplasm. Genome wide LD at $r^2>0.2$ was reduced on average to ~1-2 cM in the landrace stock germplasm and 6-8 cM in variety germplasm, providing evidence of the potential for association mapping of agronomically important traits in cotton. These results suggest that linkage, selective sweeps, inbreeding and genetic drift are the potential LD-generating factors in cotton (Abdurakhmonov *et al*., 2008a; Abdurakhmonov *et al*., 2009).

Two diverse environments and analysis of a large sets of cotton germplasm found on the average of 20 SSR markers per specific environment (Uzbekistan and Mexico) associated with one of each of the five main fiber quality traits. The mixed liner model (MLM), considering both kinship (K) and population structure (Q) to minimize spurious associations, was used. The LD-based association mapping, (Abdurakhmonov and Abdukarimov, 2008), using the MLM approach, was found to be effective in cotton, and a number of SSR markers associated with the fiber quality traits, reported herein, provide insight into understanding environment-specific functions of genes controlling fiber development, which increases the effectiveness of cotton marker-assisted breeding programs at similar latitudes (Abdurakhmonov *et al*., 2008a; Abdurakhmonov *et al*., 2009).

Association mapping efforts in a large number of Upland cotton germplasm allowed Uzbek researchers to design an "association mapping" study to find biologically meaningful marker-trait associations for important fiber quality traits that account for population confounding effects. Several SSR markers associated with main fiber quality traits along with donor accessions were identified and selected for MAS programs of important fiber traits in Uzbekistan. As a result, researchers were able to mobilize novel haplotypes of quantitative trait loci (QTLs), "still - underutilized" in current Uzbek cotton breeding, from donor genotypes to several commercialized recipient cultivars using traditional backcrossing with the aid of trait-associated molecular markers. Testing the major fiber quality traits using high volume instruments (HVI) in MAS hybrids revealed that mobilization of the specific marker bands from donors has positively improved the trait of interest in recipient genotypes. Currently, fourth generation MAS hybrids (F_1BC_4) carrying new markers and having a better quality of fiber compared to the recipient parents are being grown and tested. Using these effective molecular markers as a breeding tool, researchers are pyramiding major fiber quality traits into a single genotype of several commercial Upland cotton cultivars of Uzbekistan (Abdurakhmonov, 2011; Abdurakhmonov *et al*., 2011ab). Thus, these genetic mapping and MAS efforts are developing in Uzbekistan that will help to address many problems associated with improving, boosting and sustaining cotton yields and quality.

Characterization of Novel Genes and Sequence Signatures

Several genes involved in cotton photomorphogenesis, flowering, and disease resistance were cloned and characterized in Uzbekistan to develop cotton biotechnology and produce Uzbekistan's own biotech cottons. For instance, two paralogous *PHYA1* and *PHYA2* genes in diploid cottons, the result of a Malvacea-specific *PHYA* gene duplication were characterized that occurred approximately 14 million years ago (MYA) - before the divergence of the A- and D-genome ancestors. One of each *PHYB*, *PHYC*, and *PHYE* genes in diploid cottons was cloned. Allotetraploid genomes retained all gene complements observed in putative diploid genome ancestors with at least four *PHYA* genes, and two genes of each *PHYB*, *PHYC* and *PHYE* in AD-genome *G. hirsutum* and *G. barbadense* (Abdurakhmonov *et al*., 2010).

Several phytochrome gene specific CAPs and dCAPs markers were developed using SNP sites for *PHYA1*/BbvI, *PHYA1*/MboI CAPs, *PHYB*/AluI dCAPs, *PHYB2*/MboI, and *PHYB2*/TaqI dCAPs that were polymorphic between *G. hirsutum* and *G. barbadense*. Cotton phytochrome A genes were found localized into chromosome 2 and 11, while cotton phytochrome B was localized into chromosome 10 using CAPS and dCAPS markers in cytogenetic stocks and linkage analysis (Abdurakhmonov *et al*., 2007c). Further, *PHYA1*-derived single nucleotide polymorphism marker was associated with fiber quality traits in an interspecific cross between Pima S-7 (*G. barbadense*) and Tamcot SP37 (*G. hirsutum*) consisting of 96 F_2 individuals (Abdurakhmonov, 2001).

In addition, the orthologs of the Arabidopsis *HY5* gene were cloned and characterized from *Gossypium* species. It was revealed that tetraploid cottons *G. hirsutum* and *G. barbadense* have at least two orthologous genes for *HY5* that were acquired from the putative diploid ancestors – *G. raimondii* (D-genome) and *G. herbaceum* (A-genome) through allopolyploidy. Each of these consensus contig sequences shared ±56% nucleotide identity and 72% amino acid similarity with the orthologous region of the Arabidopsis *HY5* gene (Abdurakhmonov, 2001; Abdurakhmonov *et al*., unpublished). The results should be useful in understanding cotton photomorphogenesis and the involvement of the light signal transduction system in fiber elongation through the genetic-developmental view.

A large number of small RNAs from 0-10 days post anthesis were cloned and annotated. Results provided the first direct evidence of siRNA/miRNA-mediated regulation of complex fiber development in cotton (Abdurakhmonov *et al*., 2008b). Using the same strategy, several small RNA/microRNA signatures from developing roots (Devor *et al*., 2009), and FOV and root knot nematode (RKN) infected, as well as salt treated cotton tissues (Shapulatov *et al*., unpublished), were annotated. These signatures currently are being used for creating biotech crops with improved pathogen resistance and salt tolerance.

Further, a family of cotton *MIC-3* genes were cloned and sequenced. The *MIC-3* gene family plays an important role in plant defense as a PR-protein. Molecular evolution of different cotton genomes was studied, and for the first time the pathogen-dependent gene duplication pattern in the cotton *MIC-3* gene was identified that completely fit with «*bait and switch*» and «*guard and decoy*» evolution models of resistant genes in plants (Buriev *et al*., 2010; Buriev *et al*., 2011). This gene and/or its regulatory elements family should have great potential to be used in GE cotton development due to its association with PR-processes, uniqueness to cotton, and tissue specific activity.

Cotton Transgenomics and Biotechnology

An efficient high-throughput cotton transformation system was established (Abdurakhmonov *et al*., 2013, in press) using modern transgenomics tools with the specific aim of developing Uzbekistan's own tissue culture derived biotech crops as well as to study the biological function of these *de novo* characterized cotton genes mentioned above. This includes RNAi (RNA interference) technology using available RNAi vector systems (Wesley *et al*., 2001; Helliwell *et al*., 2002) as well as newly developed synthetic RNAi constructs, which are highly specific to a gene or small RNA/microRNA signature of interest that reduce (knock-out) gene function (Abdukarimov *et al*., 2011).

Because of multiple gene effects in plant development, as well as its association with yield potential and productivity (Smith, 1994; Robson and Smith, 1997; Robson *et al*., 1996; Thiele *et al*., 1999; Rao *et al*., 2011), plant flowering and architecture (Fankhauser and Chory, 1997), and cotton fiber quality (Kasperbauer *et al*., 2001; Abdurakhmonov, 2001) as well as its possible involvement in salt tolerance (Datta *et al*., 2007; 2008), in regulation of nitrate reductase (Jonassen *et al*., 2008; Lillo, 2008), in cold/freezing and drought tolerance in model plant Arabidopsis (Kim *et al*., 2002; Franklin and Whitelam, 2007; Beck *et al*., 2007), in fungal disease resistance in rice (Xie *et al*., 2011), researchers targeted RNAi of the cotton phytochrome gene family and its signal transduction system for developing genetically engineered cottons for Uzbekistan. The work was done in collaboration with Texas A&M University and USDA-ARS laboratories. The phytochrome RNAi study provided the first molecular evidence of the importance of the phytochrome gene family in cotton fiber development and demonstrated the potential of phytochrome-specific RNAi, simultaneously improving several important agronomic (e.g., early maturity, high yield) and fiber quality traits (length, strength, fineness, elasticity, and uniformity) in somatically single-cell regenerated RNAi Coker 312 cotton plants (Abdurakhmonov *et al*., 2012c; Abdurakhmonov *et al*., 2013, in press).

Previous studies in model plant Arabidopsis have shown physiological consequences associated with modulation of expression of phytochromes and cross-regulatory effects as the manifestations of a compensatory regulatory network of phytochromes. Results sharply contrast with findings from Arabidopsis in which loss-of-function phyA mutations showed no increase in *PHYB* expression. *PHYA1* RNAi cotton lines with a 70% decreased level of *PHYA1* expression showed increased transcript levels for *PHYA2*, *PHYB*, *PHYC* and *PHYE*. These observations indicate that the phytochrome regulatory network of cotton may have a fundamentally different dynamic architecture than that of Arabidopsis (Abdurakhmonov *et al*., 2012c; Abdurakhmonov *et al*., 2013, in press).

Researchers showed a transfer of phytochrome-associated RNAi phenotypes from somatically regenerated RNAi Coker-312 to other 'recipient' commercial Upland cotton cultivars via sexual crosses, effectively converting recipients to the superior genotypes. Using this RNAi approach, as the first generation biotech cottons in Uzbekistan, a number of genetically modified cotton varieties with improved fiber quality (38-40 mm fiber length), improved micronaire (3.9-4.2), early maturity (earlier by 5-10 days) and higher seed cotton yield (~10-18% higher) with improved root system (two fold) have been developed (Abdurakhmonov *et al*., 2012c; Abdurakhmonov *et al*., 2013, in press). This technology demonstrates the great potential to develop superior cultivars in a globally important crop species in a short time without any adverse effects on yield (Abdurakhmonov *et al*., 2012c; Abdurakhmonov *et al*., 2013, in press). Large-scale field trials to further commercialize phytochrome derived biotech cotton varieties are being conducted for future commercialization of phytochrome gene specific RNAi varieties and to quickly boost cotton production in Uzbekistan. Similarly, biotech cottons for drought, salt, wilt (FOV) resistance traits (using RNAi approach) are being developed using characterized genes and sequence signatures in Uzbekistan. To put this in perspective, researchers will work on new genetic engineering technologies such as "Zinc-finger" and/or "TALEN" for site-specific genome editing of genes with proven RNAi effects (e.g. cotton *PHYA1*) to develop native biotech cotton cultivars for Uzbekistan farmers.

Conclusion

A careful review of current cotton production reports and world statistics shows that the demand for cotton fiber will likely keep increasing, but cotton production will be constrained by food security concerns associated with population growth, complex policy factors affecting a gradual decrease of cotton planted area, declining cotton yield potential associated with the narrowness of genetic diversity of cotton cultivars, and a worsening of environmental and biotic stresses. Global climate change and projected temperature increases generate alarming forecasts for even larger yield decreases, including for cotton, over the next few decades. Advances in conventional cotton breeding programs and the success of biotech cottons, widely implemented by cotton farmers in the last several decades, have resulted in significant yield gains. These yield gains contributed to the profitability and sustainability of world cotton production.

Future success in boosting cotton yields can be achieved through the development and application of new "omics"-based modern technologies. These include marker-assisted selection, genomic selection and virtual breeding, and the development of a new generation biotech cottons through modification and targeting of multiple-effect genes, improving photosynthetic rates and plant architecture with more bolls, fruiting branches and well-developed roots, resistant to environmental stress and biotic invasions, and plants with better nitrogen and water use efficiencies. In this mission, the newly completed cotton genome sequence and completion of genome sequences of commercially important allotetraploid cottons will boost the development of innovative technologies in the near future. This should help to rapidly improve yields of cotton to cover the projected needs for fiber and other associated cotton products. These challenges require more coordinated efforts, wider international collaborations, and better funding and investment. Similarly, cotton yield improvement is the priority objective and mission of the research programs in Uzbekistan. Current efforts on application of novel genomics tools and biotechnologies, in combination with contemporary breeding knowledge and expertise, will provide a basis to boost yields and other agronomic properties of Uzbek cotton.

Acknowledgments

The author is thankful to the Office of International Research Programs (OIRP) of the United States Department of Agriculture (USDA) in the frame of USDA–Former Soviet Union (FSU) cooperation programs for the funding of the Uzbekistan cotton genomics program. He acknowledges the Civilian Research and Development Foundation (CRDF), USA for project coordination. He also thanks the Uzbekistan government, Cabinet of Ministries of Uzbekistan, Academy of Sciences of Uzbekistan, Ministry of Agriculture and Water Resources of Uzbekistan, "Uzpakhtasanoat" association, association of "Oil-Fat & Food Industry" and holding company «Uzvinprom Holding» for their continuous in-house support of the research efforts. The USA collaborators and my laboratory members and students are also appreciated for their huge efforts and contributions on genomics investigations. The research team appreciates the help and support extended by the USDA Office of Transfer Technologies during the filing of a patent application for the results.

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Long Term Trends in the Cost of Cotton Production

The cost of production relative to the expected price received is the determining factor farmers consider when deciding whether or not to plant cotton and if so how much. The price of cotton is highly variable, but farmers always have a good idea of how much it will cost to produce a hectare of cotton compared to competing crops. While prices received for cotton may vary significantly from year to year, the cost of production usually does not change drastically unless yields fluctuate. Knowing the cost of production helps farmers make at least a short term plan to determine a cropping pattern including both competing and rotation crops. Increases in production costs stemming from changes in the prices of inputs and agronomic practices are relatively predictable. Input prices and the costs of farming operations tend to increase, unless an important component of input costs is replaced by a less expensive option, such as the use of insect resistant biotech cotton replacing insecticide use. But, such technological shifts are usually adopted slowly.

There are only a few countries where farmers know in advance the price they will receive with certainty. In these countries, the government, often after negotiations with ginners and farmers, fixes the price for seed cotton; farmers then decide how much area to plant. In countries where prices are announced in advance, public or private companies often supply inputs at prices negotiated with government regulators or producer organizations. Knowing the price to be received for cotton and the cost of inputs allows farmers to make decisions based on their estimates of net income, although yields are always variable.

When companies supply inputs for cotton production, they often refuse to supply similar inputs for competing crops since doing so would undermine cotton production and the likelihood of input-cost recovery. Consequently, farmers may feel pressure to produce cotton even in years when their expectations of net income are not favorable. In almost every situation in which prices paid to farmers are fixed in advance, the cost of production is estimated carefully to make certain that the farmer has an economic interest in producing cotton.

The International Cotton Advisory Committee (ICAC) has been conducting surveys of the cost of producing cotton since the 1960s, but the early surveys were conducted at irregular intervals. However, since 1992, the cost of production survey has been updated every three years. The last eight surveys have been based on the same questionnaire, making it easier to compare results from one survey to another. The same questionnaire was sent to ICAC Coordinating Agencies in ICAC member countries and to researchers or other contacts in non-member countries. Thus, the sources of information vary among countries, but the data are official. Information from non-official sources, such as private companies, have not been used. Questionnaires are sent in April/May of each survey year, and the resulting report is published in September/October before the ICAC Plenary Meeting.

The latest full report, '*Cost of Production of Raw Cotton*,' can be requested at publications@icac.org. The latest report, published in September 2013, contains data for the 2012/13 cotton production season.

The surveys are designed in such a manner that all components of the cost of production are covered in each survey. Production systems vary from completely mechanized to partially mechanized, from animal traction to manual cultivation, and sometimes even a mix of all the above. Cotton may be produced under irrigated conditions or may be entirely dependent on rainfall. Certain inputs or operations are country-specific and also depend on specific production or farming systems. This explains why the answers to all the questions asked in the survey questionnaire are not necessarily available from all countries. There is not a single country that has provided data on all the inputs and operations listed in the questionnaire, another indication that the questionnaire covers a wide variety of production practices and systems. In the most recent survey, respondents were asked to report the technology fee for insect resistant and herbicide tolerant biotech traits independently of the cost of the planting seed. Respondents were asked to report using a given unit (e.g. kg, liter, mandays, etc.,), quantities per hectare, price or cost per unit and for each item, the total cost in local currency and in US dollars. The inputs and operations covered in the survey questionnaire are on the next page.

Once the cost of production per hectare is known for each farming operation or input used in various growth stages (pre-sowing, sowing, growing, harvesting and ginning, including economic and fixed costs), the net cost for seedcotton and lint can be calculated. The respondents were also asked to report additional information under items no. 9, 10 and 11 (in the table on page 20) to calculate income from selling seedcotton, lint and commercial seed. The cost of producing a kilogram of seedcotton and the net cost of producing a kilogram of lint, i.e., total cost minus land rent and the value of commercial seed (seed after ginning), were calculated. Land rent is also excluded from the cost per kg of seedcotton.

World Average Cost of Production Per Kg of Lint and Seedcotton

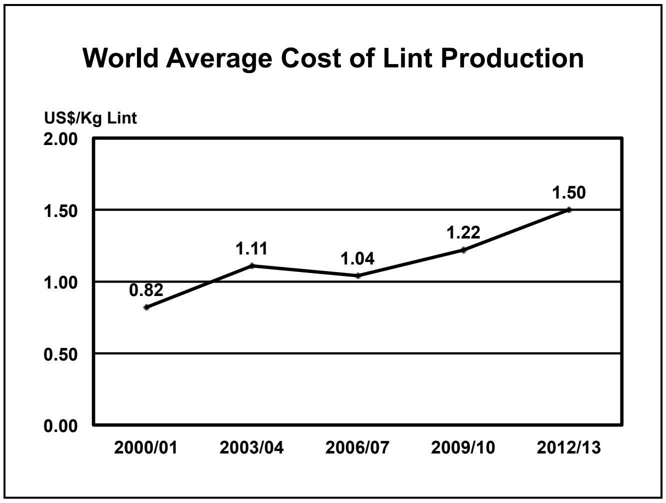
Since the same data has been collected regularly at three-year intervals since 1992, and since the sources of information have mostly been the same institutions, averages can be calculated for the costs of various inputs at regional and world levels

Cost of Producing One Hectare - 2012/13

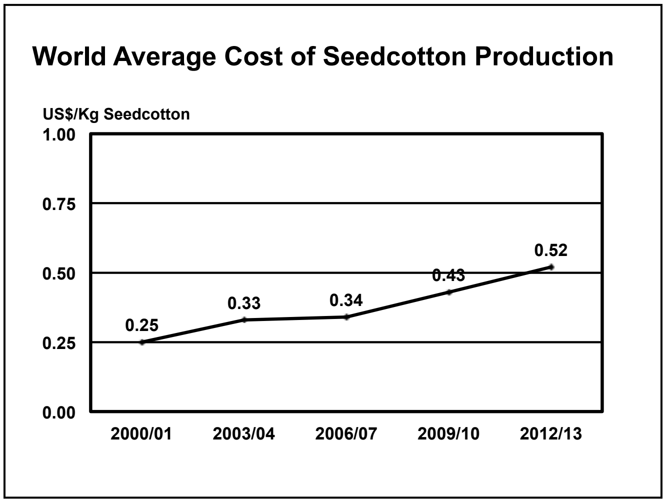
Operation/Item	Unit	Quantity per ha.	Cost or Price per unit	Cost in Local Currency	Cost in US$
1. PRE-SOWING					
Land rent for cotton	ha		1.00		
Land revenue/tax	ha		1.00		
Pre-soaking irrigation					
Ploughing					
Planking					
Other					
Sub-total					
2. SOWING					
Soaking irrigation					
Land preparation					
Seed	kg				
Seed treatment					
Herbicides (Pre-sowing)					
Fertilizer (Basal dose)					
Drilling					
Other					
Sub-total					
3. GROWING					
Thinning					
Weeding					
Hoeing					
Herbicides (Post-sowing)					
Fertilizer (Total)					
Irrigation					
Insecticides					
Tech fee for biotech (Insect resistant)	kg or ha				
Tech fee for biotech (Herbicide tolerant)	kg or ha				
Defoliation					
Other					
Sub-total					
4. HARVESTING					
Picking cost					
a. Hand picking (%)	kg				
b. Machine picking (%)					
Stick cutting/slashing	ha	1.00			
Other					
Sub-total					
Seed Cotton Costs					
5. GINNING					
Transportation to gin factory					
Ginning (Including bagging)					
Classing/grading charges					
Other					
Sub-total					
Variable Cash Costs					
6. ECONOMIC COSTS					
Management and administrative					
Interest on capital invested					
All repairs					
General farm overheads					
Other					
Sub-total					
7. FIXED COSTS					
Power supply					
Irrigation system at the farm					
Tractors					
Spray machinery					
Farm implements					
Other					
Sub-total					
8. TOTAL COST					
9. VALUE OF SEEDCOTTON	kg				
10. NET VALUE OF LINT	kg				
11. NET VALUE OF SEED	kg				

and compared over time. Thirty-two countries, accounting for almost 90% of world cotton production, participated in the 2012/13 cost of production survey, including Argentina, Australia, Bangladesh, Brazil, Bulgaria, Cameroon, Chad, China, Colombia, Egypt, Ethiopia, Greece, India, Iran, Israel, Kazakhstan, Kenya, Mali, Mozambique, Myanmar, Pakistan, Philippines, Senegal, South Africa, Spain, Sudan, Tanzania, Thailand, Turkey, Uganda, USA and Zimbabwe. Because some countries provided data for multiple regions, the total number of entries in 2012/13 was 53; 28 rainfed entries and 25 irrigated entries. Only the long-term trends in world costs are reported in the current article; comparisons among countries will be presented in the March 2014 issue of the *ICAC RECORDER*.

The average net cost of production in the 32 countries participating in the 2012/13 survey rose to US$1.50 per kilogram; an increase of 28 cents compared to the average net cost of production in 2009/10. The 23% rise in the cost of production in this triennium was greater than the increase between 2006/07 and 2009/10. The main reason for the increase was an increase in the cost of weeding. The use of herbicides is still not common in many countries, while the cost of labor and cultivation are increasing everywhere.


World Average Cost of Lint Production

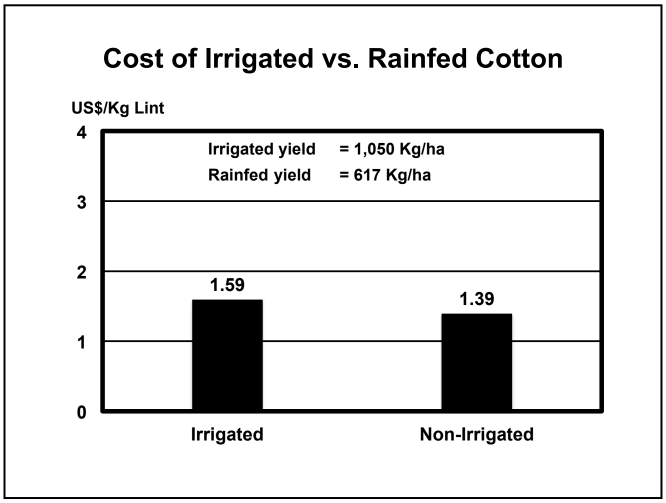
The average cost of production of seedcotton increased from US$0.43/kilogram in 2009/10 to US$0.52/kilogram in 2012/13. These cost estimates are based on the assumption that farmers are cultivating their land themselves and are not paying rent for land use. Some countries do not have a land rental market, and out of 32 countries 14 did not report land rent. In the case of countries that did report land rent, the land rent value was deducted from the total cost for calculating the cost per kg of seedcotton. The percentage increases in the cost of production per kilogram of seedcotton and lint between 2009/10 and 2012/13 are nearly the same, and the small differences in measured costs might be due to errors in measurement, especially because of a lack of data from some countries on ginning costs.


World Average Cost of Seedcotton Production

Irrigated and Rainfed Cotton

The 32 countries that participated in the 2012/13 cost of production survey planted 61% of their area under irrigated conditions and 39% under rainfed conditions. The cost of production under irrigated conditions averaged US$1.59 per kilogram of lint, compared to US$1.39 per kilogram under rainfed conditions. In other words, when rainfall is abundant enough to make irrigation unnecessary, rainfed cotton production is cheaper than irrigated cotton. Presumably, farmers choose to irrigate only in situations where irrigation is necessary.

On average, the net cost of production per hectare was US$1,658 under irrigated conditions and US$860 under rainfed conditions —about half the irrigated cost. The average yield per hectare for irrigated cotton in 2012/13 was 1,050 kilograms of lint, accounting for 73% of world production. The average rainfed yield among the reporting countries was 617 kilograms of lint per hectare, accounting for 27% of world production. When one takes into account the Cotlook A Index price for 2012/13 and the average cost and yield under irrigated verses rainfed conditions, the conclusion is that farmers are receiving more net income from irrigated cotton than from rain-fed cotton. The greater yields under irrigated conditions more than compensate for the higher cost of irrigated production over rainfed cotton production.


Cost of Irrigated vs. Rainfed Cotton

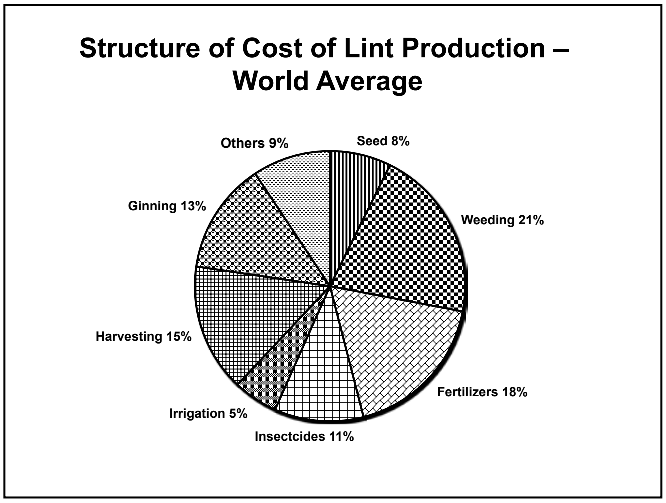
Structure of the Cost of Production

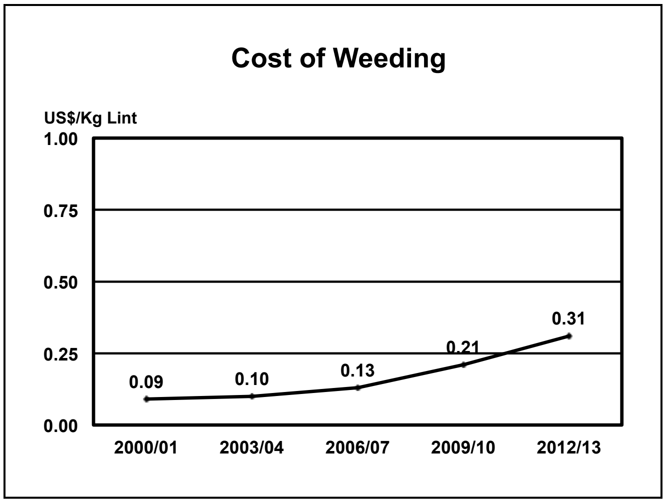
Of the net cost of lint production, 21% or about one-fifth, went to weed control, followed by 18% spent on fertilizers. The cost of insecticide use on cotton declined over the last decade and stood at only 11% of the net cost in 2012/13. Irrigation accounted for 5% of the net cost, harvesting was 15% of the net cost, and ginning was 13%. Planting seed, including the technology fees in the countries that have adopted biotech cotton, accounted for 8% of the net cost of production, or 11 cents per kilogram of lint. As with all other inputs and operations, the reported cost of irrigation represents the average cost of water per kilogram of lint produced in all the countries that participated in the survey.

In 2012/13, a typical cotton grower spent an average of US$1,332 to produce, harvest and gin the production of a single hectare of cotton. As stated in the beginning, the world average net cost of production in 2012/13 was $1.50/kg of lint. The average of the 32 countries that participated in the survey showed that farmers spent the following amounts on each input and operation in 2012/13.

Actual Net Cost Per Kg of Lint Produced

Input/Operation	Cost/kg of Lint Produced (US$)
Planting seed	0.11
Weeding	0.31
Fertilizer	0.27
Irrigation	0.08
Insecticides	0.16
Harvesting	0.23
Ginning	0.20
Others	0.14
Total:	1.50


Structure of Cost of Lint Production – World Average


Cost of Weeding

The reported cost of planting seed includes the cost of seed and seed treatments, which are given separately by at least one-third of the reporting countries. The cost of weeding includes all weeding operations, both chemical and mechanical. Harvesting includes stick cutting, thrashing and incorporation, and ginning includes the cost of transportation to the gin and classing, both seedcotton and lint. Other costs include pre-sowing operations, drilling and sowing seed, economic costs and fixed costs.

Weed Control

Proper weeding has always been critical for achieving high yields. Weeds can be removed through cultural operations such as forming ridges or loosening soil to retain water, manually, mechanically or chemically, and it is very important to remove weeds before they are able to form seeds. Manual and cultural methods are now generally less common than in the past because of farmers' inability to get rid of all weeds, the rising cost of operations and the need to return to the field for more weeding. If weeds are not removed and inputs are applied, weeds take a heavy toll on the cotton crop and almost certainly cause economic losses. Mechanical weeding is more environmentally friendly than the use of herbicides, but it is nearly impossible to get rid of weeds that are close to cotton plants or in between plants in a row using mechanical means. Mechanical implements can be used only until plants reach a certain height, and it is always a problem to eliminate climbing weeds like lily.

The use of herbicides has environmental consequences, but an increasing number of countries are adopting herbicides, albeit at a slow pace. Herbicides provide nearly perfect control for a longer time compared with mechanical control if they are applied properly. Herbicides can be used pre- or post- sowing, depending upon the field situation and the probability of eliminating weeds. Herbicides have been used for over 60 years, much longer than insecticides. Farmers must rotate classes of insecticides frequently to avoid the development of resistance and to respond to changes in pest populations in response to control methods. In contrast, the weed complex has not changed much in most countries. One of the first herbicides used in the world 2,4-D is still being used today and remains one of the most commonly used herbicides in the world.

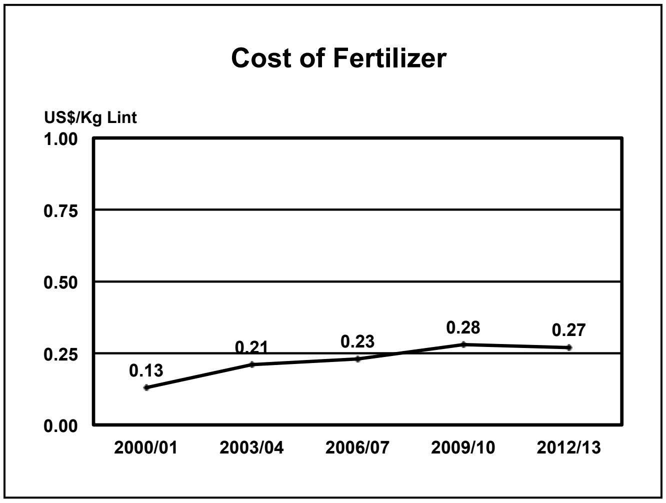
Herbicide tolerant cotton was commercialized in the 1990s, one year before insect resistant cotton was released. Even though herbicide tolerant cotton varieties have been available for almost two decades, most countries that were not using herbicides in the 1990s are still not using herbicide tolerant biotech cotton on large areas. Herbicide tolerant biotech cotton led to increased use of herbicides in the countries that were already using herbicides. However, as the current survey documents, the cost of weed control is rising, therefore concerns about weed control costs are rising. Nine US cents were spent per kilogram of lint production in 2000/01, compared to 21 cents per kilogram of lint in 2009/10, and 31 cents per kilogram of lint in 2012/13.

Weed control costs are rising because of the higher costs associated with field operations and because farmers are placing a greater emphasis on weed control in order to raise yields. Higher weed control costs are encouraging research into less expensive weed control means that are effective and provide control for a longer time.

Fertilizer Costs

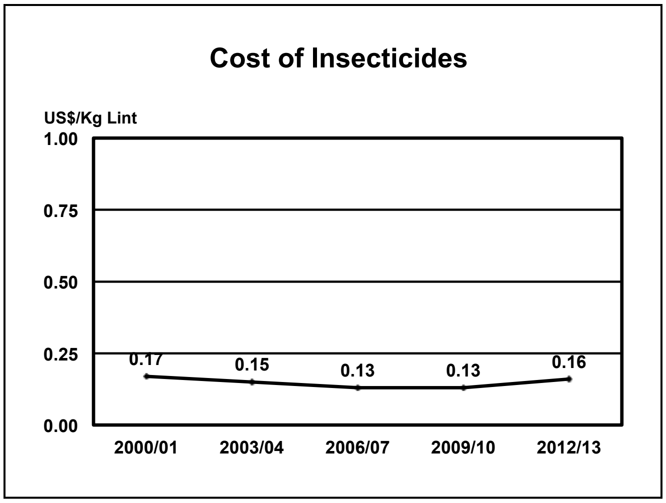
Fertilizer use has always been a function of the cost-benefit ratio, and that ratio was extremely high for most farmers when synthetic inorganic fertilizers were introduced. In the early years of fertilizer use, farmers were able to correct nutrient deficiencies in soils in accordance with the actual needs of plants in each field by using synthetic fertilizers. However, the introduction of synthetic fertilizers has tempted farmers to overlook organic fertilizers because they are slow in action, they are required in voluminous quantities and because it is very difficult to closely match the nutrient needs of plants with organic fertilizers. However, the reduced application of organic fertilizer has led to deterioration in soil texture in many areas, particularly a lack of organic matter and micronutrients, and this deterioration reduces the benefits of inorganic fertilizers. Consequently, the cost/benefit ratio on the addition of inorganic fertilizer is not nearly so positive anymore.

The cost of acquiring and applying fertilizers more than doubled in the nine years from 2000/01 to 2009/10. In 2000/01, the typical cotton grower spent an average of 13 cents on fertilizer to produce a kilogram of cotton lint, compared with 28 cents in 2009/10 and 27 cents in 2012/13. The data indicate that farmers are no longer increasing the quantity of fertilizers used per hectare of cotton, and they are finding ways to maximize the benefits of the nutrients that have already been applied.



Insecticide Use

The third most important component of the cost of production is insecticide use. Weeding, fertilizers and insecticides formed 50% of the net cost per kilogram of lint in 2012/13. In 2000/01, the average farmer spent 17 cents on insecticide use to produce a kilogram of lint, compared to 9 cents on weeding and 13 cents on fertilizers.



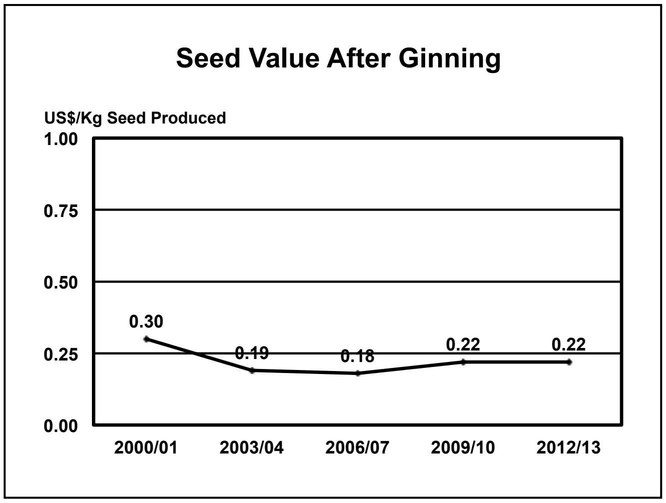
While the cost of weeding has been continuously on the rise and the cost of fertilizers increased until 2009/10, the cost of insecticides and their application has been on the decline. Based on the average of the 32 countries that participated in the current survey, a cotton grower spent 16 cents on insecticides in 2012/13 to produce a kilogram of lint —almost the same as in 2000/01 but much lower as a percentage of the net cost.

There are many factors responsible for the decline in insect control costs. The adoption of insect resistant biotech cotton undoubtedly reduced the need for insecticides. The cost of insect control operations may have remained the same since 2000/01, but the biotechnology fee is included in the costs of planting seed rather than being accounted for as a form of insecticide. Countries suffered because of their heavy reliance on insecticide use, and the negative consequences of insecticide use are better understood now than when they were introduced and broadly encouraged in the 1960s and 1970s.

Apart from the severe consequences of over use of insecticides and the more complete understanding of their chemistry, the cost of insecticides also played a role in encouraging a reduction in use. Insecticides were often subsidized and promoted by governments before it was realized that insecticides are not a viable long-term solution to insect management. The development of resistance was not foreseen prior to the introduction of insecticides. With the improved understanding of the consequences of insecticide use, confidence in non-chemical control measures has grown over time.

Value of Commercial Seed

Cotton growers may sell their product as seedcotton or they may have it ginned and sell it as two separate items: lint and cotton seed. When seedcotton is sold, it may be sold directly to a ginner or to a middleman for ginning. In this situation, the ginner is responsible for all ginning costs, which he recovers from selling seed. The ginner also owns the lint. In countries where average farm size is larger, custom ginning is popular and farmers pay for ginning and retain ownership of both the



lint and the seed. Even if custom ginning is not popular in a given country, a farmer selling seedcotton is receiving an implicit price for the lint and for the seed, although the prices are not separately identified. The value of seed after ginning, which is a substantial amount in many countries, has been deducted from the total cost in order to determine the net cost per kilogram of lint. Thus, the value of seed has a significant impact on the net cost of production per kilogram of lint.

On average, 1,498 kg of commercial seed/ha was produced in 2012/13 in the 32 countries that participated in the current survey, and the value of that seed per hectare averaged US$328, or US$0.22 per kg.

In the five surveys conducted in the last 12 years (one every three years), the value of seed was the highest in 2000/01 (30 cents per kilogram of seed), thus lowering the net cost of production to only US$0.82 per kilogram of lint. The long-term trend shows that commercial seed has usually been sold at around 20 US cents per kg. However, there is a great deal of variation in prices for commercial seed from country to country.

The value of cotton seed in 2000/01 was unusually high, perhaps due to tight supplies caused by a reduction in planted area.

Consequences of Escalating Costs

The cost of production of seedcotton has been increasing for the last 12 years, and the increase has had repercussions. Cotton production must continue to be an economically viable choice for farmers if they are going to continue to produce. The economic viability of cotton is directly related to the net cost of production, the net cost of producing competing crops and prices for cotton and competing crops. However, given that prices of cotton and competing crops are highly variable, the cost of production is the most significant factor affecting farmers' choices of whether or not to plant cotton. If the cost of production of cotton continues to increase without proportional increases in the price of cotton, it is going to have many impacts on the cotton sector.

- Farmers' income will suffer and a big shift in the location of cotton production may take place. Countries and areas where producing cotton is expensive will shift to other crops, while low cost producing areas or countries may or may not compensate for the shortfall in supply.
- In countries where the cost of production is already higher than market prices and farmers are continuing to produce cotton because of government support, those government support programs will have to come up with additional funding for cotton producers.
- Consolidation of farming operations is one of the means to lower the cost of production. In the USA, the average size of a US cotton farm in 1946 was about 7 hectares. Average farm size increased and stabilized around 180 hectares, which is probably an economical unit in terms of a cost of production threshold under the practices currently followed in the USA. Similarly, the size of gins has increased, and the number of gins has declined. Rising production costs will not only trigger a drive to identify ways of lowering the costs of production, but will also bring pressure to bear on researchers to invent methods that are less expensive to implement and will reduce input use.
- If higher costs lead to reduced supplies and higher prices of cotton, demand would be negatively affected.
- If rising costs force farmers to become focused on immediate reductions in the cost of production, researchers and farmers will look for quick fixes rather than investigating and adopting technologies with potential long term beneficial impacts.