REVIEW PAPER

SUGARCANE BIOTECHNOLOGY - EMERGING TRENDS

M.C. Gopinathan*, P.V. Naik and R. Pushpavalli

Abstract

The acceptance and adoption level of transgenic crops among farmers and consumers is increasing at unprecedented rates making it the fastest adopted technology in the history of agriculture. Emerging trends in transgenic research and field trials also indicate expression of second generation output traits such as omega-3 oil or pro-Vitamin A, drought & salt tolerance and Nitrogen intake efficiency in large number of the crops. Further recent success of third generation transgenic traits related to non-food, high value industrial and pharmaceutical products are expected to create new avenues of opportunities and challenges for industrial crops, for "biopharming'. Last two decades biotechnology work in sugarcane resulted in partial sequencing of complex polyploidy genome, identifying large number of developmental genes and production of transgenic plants for large number of traits. However, no transgenic sugarcane has been commercialized so far. The lack of reliable transformation system, efficient gene promoters, transgene silencing, limited knowledge of polyploidy genome architecture and field instability are the major scientific or technological limiting factors. Multiple utilities of sugarcane as a feed stock for production of sugar, power, alcohol, bio-fuels, biopolymers and bio-pharmaceuticals are creating new competitive landscape for sugarcane transgenic across the world. To capture these emerging opportunities major biotech companies are entering into strategic partnerships and alliances with traditional sugar research institutes and sugar companies at national and international level. But there are many challenges ahead for regulatory authorities and governments, especially in the areas of safety testing, regulation, and food labeling to obtain public acceptance and usher benefits of sugarcane transgenic to larger sections of the society.

Keywords *Sugarcane, Sugar, Biotechnology, Transgenic, Bio- refinery*

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Introduction

Worldwide, there is an increasing trend towards planting of transgenic crops or genetically modified crops. According to James (2010) status report, accumulated hectares of transgenic crops planted from 1996 to 2010 across the world crossed one billion hectares. This eighty seven fold increase from 1.7 million hectares to 147 million hectares in fifteen years of its first commercial introduction makes it the fastest adopted crop technology in the agricultural history. This scenario is also reflected in increased number of countries (29) growing transgenic crops and providing regulatory approvals (30). Ten countries (USA, Brazil, Argentina, India, Canada, China, Paraguay, Pakistan, South Africa and Uruguay) grew more than one million hectares of transgenic crops. In terms of acreage planted Soybean (73.3 million hectares), Maize (46.8 million hectares), Cotton (21.0 million hectares) and Canola (7 million hectares) were the four major transgenic crops. Herbicide tolerance has continued to be the dominant transgenic trait among six crops occupying 89.3 million hectares. Stacked multiple gene products (pest and herbicide resistance) are becoming important feature of new product introductions. Biotech Maize with eight genes coding for several pest resistant and herbicide tolerant traits, named Smartstax™, was also released in USA and Canada. The success of the sugar beet RR® in USA and in Canada and its acceptability by food consumers opened up a positive regulatory framework for transgenic sugar cane. The summary of field trials approved data from APHIS (2011) for emerging products indicate that, there is increasing interest by the biotech firms in range extension of existing genes to other crops and also to develop second and third generation new product

traits, resulting to new class of transgenic for commercialization in future. This also indicated the development of more than 2000 genes representing 1000 traits for transformation into large number of crops.

The outlook for biotech crops, in the second decade of its commercialization, is projected to be doubled in terms of crop area and potential for number of farmers in adopting it. Globally commercial introduction of the second generation output traits such as high omega-3 oil in soybean or enhanced pro-Vitamin A in Golden Rice, drought & salt tolerance and Nitrogen intake efficiency in most of the crops, is expected to push biotech crops' adoption rate further. Success of third generation transgenic traits related to non-food, industrial and pharmaceutical and phytoremediation products are expected to create new avenues of opportunities and challenges for industrial crops, for "biopharming" (Osman etal., 2007 ; James 2010).

However, most of transgenic crops commercialized so far are propagated mainly through true seed as compared to the vegetative propagating crops such as sugarcane where intellectual property can be protected and economic benefits can be easily quantified. Therefore investments and involvement of seed companies or large traditional biotech companies in sugarcane research were limited so far. Further, most sugarcane industries do not have commercial seed cane production systems. Therefore, seed companies interested in biotech sugarcane need to first invest in variety development (Richard 2009). Varietal development for sugarcane was predominantly a state or public funded research function. Much of the progress in increasing crop improvement has come through conventional breeding which usually takes upto 10 to 15 years of breeding and selections (Cox and Hansen 1995; Berding et al. 1997; Hogarth et al. 1997; Snyman et al. 2008 ; Cheavegatti- Gianotto et al. 2011). Although sugarcane molecular biology research began in the 1960s with *in-vitro* plant regeneration research (Nickell 1964; Heinz and Mee1969), serious efforts to apply bio-technological tools to understand the crop and enhance its potential commenced only in the past two decade. Most of these investments for research came from academic institutions, public and private research institutes but not from traditional biotech companies. Among these include BSES, SASRI, Queensland University, University of Texas, CIRAD, CTC, consortium like ICSB, SUCEST etc. With their limited resources, most of the work was focused primarily on three areas namely, genomic sequence, identification of markers and genetic manipulation.

Further, the complexity of the sugarcane genome detracted large efforts and investments in the development of biotechnological and genetic tools for this crop. Partial sequencing of genomes, identification of markers and genetic transformation of herbicide, pest, and viral-resistant plants have been reported, but so far there has been no commercial release of transgenic sugarcane

(Lakshmanan et al. 2005; Paula 2007; Menossi et al. 2007 & 2008; Cheavegatti- Gianotto et al. 2011). However very recently, there have been serious involvements of several leading biotech players such as Monsanto, DowAgro Sciences, Syngenta, DuPont, Amyris and countries such as Brazil, Australia, US and South Africa aimed at commercial development of sugarcane transgenics, resulting to emergence of new consortia, partnerships and acquisitions at various levels.

Changing Landscape – Sugarcane Crop

Among plants, sugarcane is the most efficient converter of sunlight into chemical energy which stored in the form of sugars and fiber. Traditionally, the main products of sugarcane are sugar for food, molasses or juice for alcohol, and fiber for fuel. These three products are obtained from the harvested cane while the tops and trash are removed in the field prior to or during harvest.

Sugarcane is an economically important tropical crop and has served as a source of sugar and sweetener for hundreds of years. Sugarcane is cultivated in more than 110 countries in tropical and subtropical regions of the world. In 2011, world production of sugar was estimated to be of 172.4 million tonnes, of which 135.4 million tonnes are of sugarcane (78.6%) and 37 million tonnes are of Sugar beet. Brazil (35.8 million), India (28 million), European Union (17.53 million), China (12.6 million), Thailand (10 million), USA (7.3 million) and Australia (4.0 million) are the largest sugar producers with roughly 60% of world production (ISO 2011). Seven of these top ten sugar countries are also among top sugar consumers. Only 30% sugar production is traded globally and rest is consumed locally. Sugar production and trade plays a vital role in the economic development of these countries. Therefore, sugar business has been historically

regulated at national and political level in most of the countries (Gopinathan and Sudhakaran 2009).

Since the introduction of the Kyoto Protocol in 1997, a worldwide concern about climate change and its impact on global warming has motivated unprecedented discussions on energy sustainability (Cox et al.2000; Hansen et al. 2005; Wigley 2005; Matsuoka et al. 2009). According to World Energy Outlook (2010), the current energy supplies are unsustainable from environmental, economic and societal stand points. Climate change threatens water, food production, human health and the quality of land on a global scale. A global effort to develop sustainable energy sources is urgent in order to both preserve the natural resources and mitigate the effects of CO_2 emissions (Fischer et al. 2008; IPCC 2010). However, the Brazilian example of producing sugarcane based liquid fuel for transportation for the last thirty years has shown a significant contribution to the world's energy needs, and at the same time, contribution to reduce the CO_2 and other greenhouse gas emissions (OCC 2006; Matsuoka et al. 2009). Mandate for blending bio-fuels have been enacted in at least thirty seven countries (Martinot 2007; Gopinathan and Sudhakaran 2009; Matsuoka et al. 2009). Ethanol production from sugarcane is the lowest cost process and is competitive with gasoline today. From the existing 4000 cane factories spread over the tropical and sub-tropical land, if effectively utilized for the production of sugar and ethanol fuel, the demand of bio-fuel (10% mix in gasoline and 3% mix in diesel) can be met. Such energy alternative reduces CO_2 emissions significantly, by displacing fossil fuels and promotes sustainable development through creation of millions of direct and indirect employment. In addition, it opens an opportunity for negative CO_2 emission when coupled with carbon dioxide capture and storage. Another important aspect of sugarcane biomass source is its significant potential to generate surplus electricity using bagasse and improving boiler efficiency. This alternative is well known for a long time, but only with the implementation of new energy policies allowing the operation of independent power producers, and thus, commercialization of electricity, became a frequent option in sugarcane mills all over the world. Co-generation usually provides greater levels of energy per unit of biomass, compared to other systems of power generation and also reduce carbon dioxide emissions. In 2009, it is estimated that 1783 million tonnes of cane

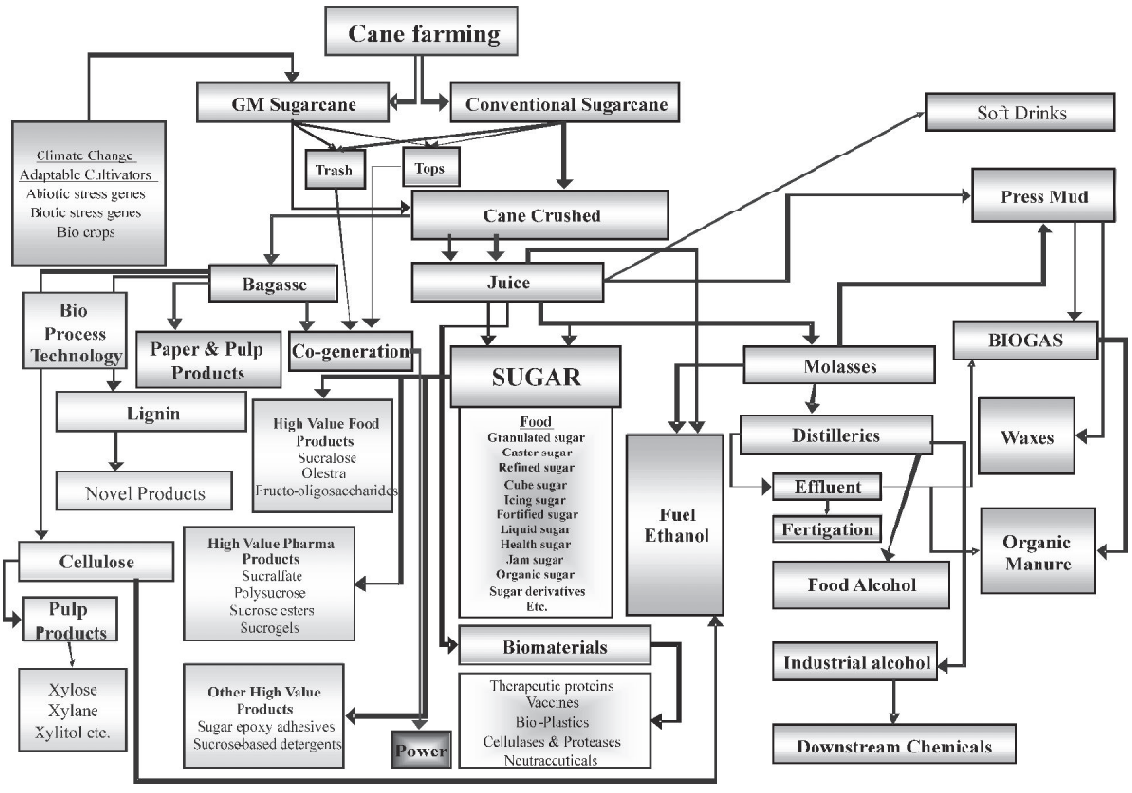

Fig. 1 Bio-Sugar Refinery of Future

was produced from 21 million hectares of land which constituted 22.4 % of total world agriculture production (FAO 2010). After the extraction of sugar, it provides 450 million tones of biomass in the form of bagasse excluding tops and leaves of the plant which is traditionally burnt or discarded during the harvest. This biomass is substantial when compared with the sugarcane biomass of 2400 million tonnes / yr from all cereals covering more than 600 million ha of land (Roberto, 2006). As per the estimate of WADE (2004), sugarcane bagasse has the potential to generate 135029 GWh per year which can make meaningful contribution to energy balance of sugarcane growing countries. Recently, there has been also an increased interest in using bagasse for processes such as paper production, as a dietary fiber, as a wood substitute, and in the synthesis of carbon fibers (Pandey et al. 2000 ; Han and Wu 2004; Paiva et al. 2004; Sangnark and Noomhorn 2004; Sun et al 2004; Cheavegatti- Gianotto et al. 2011; Manners and Casu 2011). It is expected that, developments in enzymatic and hydrolytic processes which allows fermentation of cellulose and hemi-cellulose from bagasse, will soon be scaled up for ethanol production from pilot scale to commercial level, turning sugarcane into most efficient crop for energy production.

The technological advances in the field of energy, process engineering, automobiles, biotechnology, information technology, fiscal incentives, supports for bio-fuels and carbon credit under Kyota protocol is creating a new competitive landscape for sugarcane farming and sugar and byproduct production. Recent research results and pilot plant studies in biotechnological and bio-process engineering demonstrate that the biomass produced can be tailored, in a processing factory, into various industrial raw materials with a wide range of products similar to petroleum refiners - Fig 1 (Allen et al. 1997; Rogers etal. 2001; Lakshmanan et al. 2005; Cheavegatti-Gianotto et al.2011; Manners and Casu 2011). For more than a century, petroleum refinery has been a most important part of the world economy. Many usable commodities are derived from an adjustable conversion of petroleum. New technologies, new policy frame work and increasing environmental awareness of the public are ushering in a new materials base for the 21st century. It is called “Carbohydrate economy”. Many polymeric materials conventionally derived from petrochemicals can now also be produced utilizing “Carbohydrate economy”.

Status of Sugarcane Biotechnology

Genomics

Last two decades of biotechnology work in sugarcane can be classified into, genomic sequence, gene identification and genetic manipulation. Sugarcane belongs to the genus *Saccharum L.* and modern commercial cultivars is the product of inter specific hybrids (Price 1965; Arceneaux 1967; Roach 1972) of *Saccharum officinarum* (Noble clones ($2n$ = 70-122), *S. sinense* [Chinese clones ($2n$ = 104-128)], *S. barberi* [North Indian clones ($2n$ = 60-140)], and *S. spontaneum* ($2n$ = 36-128) and rarely with *Saccharum robustum* ($2n$ = 66-170). These hybrids are high polyploids and aneuploids and their ploidy levels range from 5x to 14x (x = 5, 6, 8, 10, 12, or 14) and chromosomal mosaic is also has been reported (Lu et al.1994). From the modern cultivars chromosome numbers varies between too – 120 contain average 100–120 (D’Hont 2005). The basic genome size ranges from 760 to 926 Mbp, which is twice the size of the rice genome (389 Mbp) and same as sorghum’s 760 Mbp (D’Hont 2001).

In recent years, considerable progress has been made in mapping of the sugarcane genome and its progenitors (Aitken et al. 2005; Watt et al. 2010). The original genetic maps for sugarcane and its wild relatives were based on isozymes (Glaszmann et al. 1989), ribosomal RNA (Glaszmann et al.1990), mitochondria and chloroplast genes (D’Hont et al.1993; Al-Janabi et al. 1993; Mudge et al. 1996; Nair et al.1999) and Restriction Fragment Length Polymorphisms -RFLP(Grivet et al. 1996; Lu et al.1994; Da Silva et al. 1995; Ming et al. 1998; Ming et al. 2002b), while the most recently developed genetic maps have used markers such as Simple Sequence Repeats – SSRs (Cordeiro et al. 2003; Selvi et al. 2003; Chen et al. 2009). Amplified Fragment Length Polymorphisms - AFLPs (Hoarau et al. 2001; Rossi et al. 2003; Aitken et al. 2005), Single Nucleotide Polymorphism - SNPs (Grivet et al. 2003; Pinto et al. 2006 ; McIntyre et al. 2006) and Diversity Array Technology - DArT markers that can be analyzed with higher throughput (Heller et al. 2011). These sugarcane maps contain more than one thousand markers and are large when compared to those of most other crop species. Because of the genomic complexity of sugarcane, these maps are still incomplete. Several researchers have used molecular mapping of sugarcane in conjunction with

phenotypic data to localize Quantitative Trait Loci (QTLs). In sugarcane, QTLs usually explain only a small proportion of the variation for the trait, typically less than 10% (Cordeiro et al.2006 ; Oliveira et al. 2007) . Both micro- and macro-arrays are being used for the identification of genes expressed specifically in stems, roots, culms , leaves, disease resistance, and those involved in carbohydrate metabolism (Ulian, 2000; Grivet and Arruda 2002 ; Casu et al. 2005; Juliana et al. 2009).

The last decade has seen large-scale partial sequencing of anonymous cDNA clones from cDNA libraries and their subsequent identification of putative clones through homology searches of public databases for a wide range of gene products. This approach, commonly referred to as Expressed Sequence Tag (EST) analysis, has been extensively applied in large-scale cDNA sequencing projects for a variety of plant and animal species including humans (Adams et al.1991& 1992; McCombie et al. 1992; Sasaki et al. 1992; Hofte et al. 1993; Keith et al. 1993; Newman et al. 1994). In 1992 an International Consortium for Sugarcane Biotechnology(ICSB) was formed by a small group of sugarcane scientists that pledged to freely share technologies and information, to invest in building their own institutional biotechnology infrastructure for sugarcane genomics . Now nineteen -year-old ICSB comprises nineteen sugarcane research organizations from thirteen countries. ICSB has helped each of its members become more proficient in biotechnology and take advantage ofsharing information and technologies that were developed for sugarcane through their collaborative efforts.

In 1998, sugarcane genomics received great attention in Brazil after the formation of a sharing network to sequence and analyzes the sugarcane transcriptome (Arruda 2001). The network, called SUCEST (for Sugarcane Expressed Sequence Tags [ESTs]), produced a database of around 300,000 ESTs from the collection of cDNA libraries from different organs and tissues of sugarcane sampled at different developmental stages or conditions (Vettore et al. 2001). In 2003, a large amount of DNA sequence information for sugarcane was released into the public domain as Expressed Sequence Tags (ESTs) derived from many cDNA libraries. The majority of these ESTs originated from a genomics program in Brazil with programs in Australia and the USA being the next largest contributors-Table 1 (Casu et al. 2001; Carson and Botha 2002; Carson et al. 2002; Casu et al. 2003; Vettore et al. 2003; Ma et al. 2004; Bower et al.,2005). The Brazilian sugarcane EST project collection -SUCEST (Carson et al. 2002) generated 236,916 ESTs, which were organized into 43,141 putative unique sugarcane transcripts (26,803 contigs and 16,338 singletons) referred to as Sugarcane Assembled Sequences (SASs). The complete genome sequence of a sugarcane cultivar is not yet available.

Table 1 . The Sugarcane "transcriptome" at GenBank

Source	ESTs No.
SASRI (SASEX) ESTs Leaf roll and maturing stem	495
"Rossi" RGA ESTs:	54
Australian stem EST collection (1998-2000)	9,149
Young cane stem (YCS):	1,078
Maturing cane stem (MCS):	7,242
Methyl Jasmonate-treated roots	829
"Nogueira" cold response ESTs:	1,219
USA sugarcane ESTs:	8,125
Apex:	3,329
Stem:	2,268
Leaves:	2,396
Misc:	132
SUCEST (40 cDNA libraries)	236,916
Total	273,232

These databases allowed tagging of over 80% of the sugarcane transcriptome (Vettore et al. 2003) and have served as a tool for the identification of genes involved in tolerance to abiotic and biotic stresses, mineral nutrition, and sugar accumulation amongst others (Nogueira et al. 2003; Rossi et al. 2003; Nogueira et al. 2005;Papini et al. 2005;Calsa and Figueira 2007; Borges et al. 2007;Rocha et al. 2007). An EST survey comparing transcripts from immature and mature internodes revealed the transcripts sugar transporters showing high selectivity for sucrose (Casu et al. 2003; Rae et al. 2005; Reinders et al.2006). The large-scale analysis of gene expression in a population segregated for brix (Papini

et al.2007) using cDNA micro-arrays and transcriptome comparisons indicated a total of 125 genes were found to have expression patterns correlated with sugar content, auxin signaling and controlled sucrose accumulation. Additionally, a collection of 7409 ESTs from maturing sugarcane stems and 1089 ESTs' from immature stems analyzed by bio-informatics techniques and by cDNA micro-array methods, showed the differentially expressed genes involved in carbohydrate metabolism (Casu et al. 2001; 2003; 2004 &2005). Amongst the SUCEST sequences, dozens of orthologous genes involved in the sugarcane response to insect herbivores (Falco et al. 2001), diazothophic endophytes (Lambais 2001; de Matos et al. 2001; Vinagre et al. 2006), causal agents of smut and eyespot, were also identified.(Borras-Hidalgo et al. 2005; Rocha et al. 2007). Considerable amount of data has been also was obtained on how the plant hormone Methyl Jasmonate (MeJa) could be regulating plant defense reactions in sugarcane (Bower et al.2005; De Rosa et al. 2005). To increase the knowledge on the sugarcane responses to drought, cDNA micro-arrays were used to evaluate gene expression in water deprivation (Abe et al. 1997; Narusaka et al. 2004; Tran et al. 2004; Yamaguchi and Shinozaki 2006; Iskandar et al. 2011). Study on the evaluation of sugarcane responses to low Phosphorous (P) availability, indicated P starvation triggered oxidative stress, differential expression of several small GTPases and their regulators (Patrick 1997; Lalonde et al. 2004). Using Gene Chips from Affymetrix studies on culm maturation lead to deciphering of the developmentally regulated genes involved in cellulose synthesis, cell wall metabolism, and lignifications (da Silva and Bressiani 2005; Casu et al. 2007). Also, considerable attention has been given to the development of molecular marker technologies for sugarcane breeding and variety identification (D'Hont et al.1995; Oropeza and DeGarcia 1997; Ming et al.1998; D'Hont and Glaszman, 2001; McIntyre et al. 2001; Ming et al. 2002a; Butterfield et al. 2003; Chen et al. 2009; Maccheroni et al. 2009) and structural and functional genomics (Grivet and Arruda 2002;Da Silva et al. 2003). Molecular maps have been generated that allowed the identification of loci associated with the variation of phenotypic traits, as well as loci associated with important traits, such as sugar yield and disease resistance (Pinto et al. 2004 & 2006; Garcia et al. 2006; Oliveira et al.2007).

Genetic Manipulation

In the past decade, substantial research effort has been used to develop efficient genetic transformation systems for sugarcane (Chen et al. 1987; Bower and Birch 1992; Rathus and Birch 1992; Smith et al. 1992; Birch and Maretzki 1993; Gambley et al. 1993; Gambley et al. 1994; Birch 1997; Arencibia et al. 1998; Enriquez et al. 2000; Watt et al. 2010). Different transformation techniques such as electroporation (Rathus and Birch, 1992), Polyethylene Glycol (PEG) treatment (Chen et al. 1987) particle bombardment (Franks and Birch1991) and *Agrobacterium* mediated gene transfer (Elliott et al. 1998; Enriquez et al. 2000; Manickavasagam et al. 2004; Kalunke et al. 2009) were used to introduce marker genes in sugarcane cells and callus. To date micro projectile-mediated transformation, a technique for introducing DNA by bombarding the target tissue with DNA-coated micro-projectiles, is the most widely exploited method for sugarcane transformation (Birch 1997; Lakshmanan et al. 2005). Subsequently, micro projectile-mediated transformation of several commercially cultivated sugarcane genotypes were reported from a number of laboratories worldwide (Chowdhury and Vasil, 1992; Birch and Maretzki 1993; Birch, 1997; Joyce et al. 1998a; Joyce et al. 1998b; Nutt et al. 1999; Elliott et al. 2002; Lakshmanan et al. 2003). The applicability to a wide range of target tissues and genotypes, and the simplicity of operation, make the micro projectile approach the preferred method for sugarcane transformation (Lakshmanan et al. 2005).

For over ten years, the directed genetic modification of sugarcane has been a reality in laboratories and field trials has been conducted (Lakshmanan et al. 2005; Paula and Graham 2007; Gilbert 2009; Watt et al.2010; Neil et al. 2010; Srikanth et al. 2011). The genes that have been transferred to sugarcane can be grouped into reporter and selectable marker genes, those conferring resistance to herbicides, diseases or pests and more recently economically important traits (Table 2). During initial days of transgenic development of sugarcane, focus was mainly to transfer reporter genes or selectable markers and understand the mechanism of gene expression and genome integration. Neomycin Phosphotransferase, Glucurodinase, Hygromycin Phosphotransferase, Green fluorescent protein and Phosphinothricin acetyl transferase were the major genes

selected and transformed by various researchers -Table 2 (Bower and Birch 1992; Arencibia et al.1995 & 1998; Elliott et al. 1998; Wei et al. 2003). Subsequent to the stable expression of reporter genes, various researchers attempted to transfer herbicide resistance genes (Gallo-Meagher and Irvine 1996; Leibbrandt and Snyman 1998;Enriquez et al., 2000, Snyman 2001; Manickavasagam et al. 2004,).

Table 2. Summary of transformations of various markers and traits in sugarcane.

Trait/Transgene	**Transformation method**	**Reference**
Reporter and selection systems		
Neomycin phosphotransferase (*npt II*)	Microprojectile	Bower and Birch 1992
â-glucuronidase (*uidA*)	Microprojectile	Bower and Birch 1992
â-glucuronidase (*uidA*)	Electroporation	Arencibia et al. 1995
â-glucuronidase (*uidA*)	Agrobacterium	Arencibia et al. 1998
Hygromycin phosphotransferase (*hpt*)	Agrobacterium	Arencibia et al. 1998
Green fluorescent protein (*gfp*)	Agrobacterium	Elliott et al. 1998
Phosphinothricin acetyl transferase (*bar*) *â*-glucuronidase (*uidA*)	Agrobacterium Microprojectile	Elliott et al. 1998 Wei et al. 2003
Herbicide resistance		
Phosphinothricine acetyl transferase (*bar*)	Agrobacterium	Enriquez et al. 2000;Manickavasagam et al. 2004
Bialaphos (*bar*)	Microprojectile	Gallo-Meagher and Irvine 1996
Glufosinate ammonium (*pat*)	Microprojectile	Leibbrandt and Snyman 2003
Synthetic buster	Microprojectile	Snyman et al. 1998
Disease resistance		
Sugarcane mosiac virus (*SCMV- CP*)	Microprojectile	Joyce et al. 1998a &b
Sugarcane mosaic Virus strain E (*npt II & Ubi - cut*)	Microprojectile	Gilbert et al. 2005,
Sorghum mosiac virus (*SrMV - CP*)	Microprojectile	Ingelbrecht et al. 1999
Sugarcane yellow leaf virus (*SCYLV -CP*)	Microprojectile Gilbert et al. 2005	Rangel et al. 2003 ;
Fiji leaf gall (*FDVS9, ORF1*)	Microprojectile	McQualter et al. 2004a
Sugarcane leaf scald (*albD*)	Microprojectile	Zhang et al.1999
Leaf scald -Albicidin detoxifying enzyme(*albd*)	Homologous recombination	Zhang et al.1999
Puccinia Melanocephala (*Glucanase gene, Chitinase gene and ap24)*	Agrobacterium	Enriquez et al. 2000

Trait/Transgene	Transformation method	Reference
Pest resistance		
Sugarcane inter node borer (*CrylAa3*)	Agrobacterium	Kalunke et al. 2009
Sugarcane Stem Borer (*Cry1A(b)*)	Microprojectile, Vector transformation	Vazquez et al. 1996 Weng et al. 2006Wu et al. 2009
Sugarcane Giant Borer (*Cry1la12synth*)	Phage transformation	Craveiro et al.2009
Top borer (Pancreatic trypsin inhibitor – aprotinin (*PTIgene*))	Microprojectile	Christy et al. 2009
Sugarcane stem borer (Cry1A)	Electroporation	Arencibia et al. 1999
Maxican rice borer & Sugarcane stem borer (Galanthus nivalis agglutinin, *gna* gene)	Microprojectile	Irvine and Mirkov 1997 Snyman et al.1998 Nutt et al.1999 Legaspi and Mirkov 2000 Setamou et al.2002 Tomov and Bernal 2003
Cane grub GNA or *pinII*	Electroporation	Nutt et al.1999
Scheloribates praeincisus – Soil Mite (Proteinase inhibitor *PI* gene)	Plasmid transformation	Simoes et al. 2008
Aprotinin – Top borer	Agrobacterium	Christy et al. 2009
Sugarcane Weevil (his - tagged cane cystatin inhibitor - HIS Cane CPI - 1 gene)	Plasmid transformation	Ribeiro et al. 2008
Metabolic engineering and alternative products		
Phosphomannose niomerane (*manA*)	Microprojectile	Jain 2005
Sucrose accumulation (*SI* gene)	Microprojectile	Ma et al. 2000Botha et al. 2001
Fructo oligosaccharide (*lsdA*)	Agrobacterium	Enriquez et al. 2000
Polyphenol oxidase (*ppo*)	Microprojectile	Vickers et al.2005
Polyhydroxybutyrate (*phaA, phaB, phaC*)	Microprojectile	Brumbley et al. 2007
β-hydroxybenzoic acid (*hchl & cpl*)	Microprojectile	McQualter et al. 2004a&b
Para - hydroxybenzoic acid (*hchl & cpl*)	Plastid transformation	McQualter et al. 2005
Sucrose-phosphate synthase (*sps*)	Microprojectile	Vickers et al.2005
Human cytokine granulocyte macrophage colony stimulating factor (*GM-CSF*)	Plasmid homologus recombination	Wang et al. 2005
Polyester poly hydroxy butyrate (*PHB*)	Plastid transformation	Purnell et al. 2007
Sorbitol produced by Sorbitol - 6 - Phosphate dehydrogenase gene (*mds6pdh*)	Plastid transformation	Chong et al. 2007
Pyrophosphate fructose 6-phosphate1 phosphotransferase	Microprojectile	Groenewaid and Botha 2008; Spracklen 2009
ADP glucose pyrophosphylrase (*AGPase*) â Amylase	Homologous Vector transformation	Ferreira et al.2008
ACC oxidase suppression (*ACO antisense gene*)	Agrobacterium	Ai Qin Wang et al. 2009

Drought & Salt Tolerance		
Proline production, Osmotic adjustment and oxidative stress (*P5CS* gene)	Heterologus transformation	Hugo et al. 2007
Trehalose synthase (*Tsase*)	Agrobacterium	Zhang et al. 2006
Drought & Salt Tolerance by Ethylene responsive factor - ERF (*SodERF3*)	Wounding & Salt stress	Trujillo et al. 2009

Further attempts were also made to transfer sugarcane plants with genes conferring resistance to a number of microbial pathogens, such as Sugarcane mosaic virus (Jyoce et al. 1998 a&b; Trujillol et al. 2009), Sorghum Mosaic Potyvirus (Ingelbrecht et al. 1999), Fiji virus and leaf gall (McQualter et al. 2004 a&b), Sugarcane yellow leaf virus (Rangel et al. 2003 ; Gilbert et al. 2005), leaf scald (Zhang et al. 1999) and leaf rust (Enriquez et al. 2000).

Considerable success was obtained in developing resistance to pests such as cane grubs, and sugarcane borer, soil mite and weevil (Vazquez et al. 1996; Snyman et al. 1998; Nutt et al. 1999; Arencibia et al. 1999; Legaspi and Mirkov 2000; Setamou et al. 2002; Tomov and Bernal 2003; Weng et al. 2006; Simoes et al. 2008; Ribeiro et al.2008;Kalunke et al. 2009; Wu et al. 2009; Craveiro et al. 2009; Christy et al.2009; Srikanth et al.2011) . For instance, transgenic lines engineered with the nicotiana proteinase inhibitor, potato proteinase inhibitor II or snowdrop lectin genes exhibited marked of antibiosis to the cane grub species (Nutt et al. 1999) and sugarcane stalk borers (Legaspi and Mirkov 2000). Remarkable tolerance to the borer - *Diatraea saccharalis* was also reported in transgenic sugarcane expressing a Bt *cry1A (b)* gene (Arencibia et al.1999), and some resistance reported with bovine pancreatic trypsin inhibitor (Christy et al.2009). Efforts are also under way to engineer sugarcane for increased accumulation of sugar (Arencibia et al. 1995;Enriquez et al.2000; Ma et al. 2000; Botha et al. 2001;Groenewaid and Botha 2008; Spracklen 2009), low color raw sugar (Vickers et al. 2005) and high-value pharmaceutical and industrial products (McQualter et al. 2004 a&b; Jain 2005; McQualter et al. 2005; Zhang et al. 2006; Wang et al. 2005;Brumbley et al. 2007; Purnell et al. 2007; Chong et al. 2007; Ferreira et al. 2008; AiQin Wang et al. 2009).

Third generation transgenic plants are becoming a general platform for large scale production of wide range of recombinant proteins such as viral proteins, vaccines, antimicrobial peptides, antibodies, pharmaceuticals, and industrial compounds (Ma et al. 2000; Botha et al. 2001; McQualter et al. 2004b; McQualter et al. 2005; Wang et al. 2005; Zhang et al. 2006; Brumbley et al. 2007; Purnell et al. 2007; Chong et al.2007; Osman et al. 2007; Fereira et al. 2008; AiQin Wang et al. 2009). Sugarcane has all the features needed for a natural "bio-cane factory": it grows rapidly, has a very efficient carbon fixation pathway (C4), produces a large biomass, possesses a well-developed storage system (stem) with a large pool of stored sugar, clonally cultivated, well developed farming, production simple extraction systems in different parts of the world and generally harvested before flowering (Lakshmanan et al. 2005; Osman et al. 2007). Recently, a human pharmaceutical protein, human Granulocyte Macrophage Colony Stimulating Factor (GM-CSF) used in clinical applications for the treatment of neutropenia and aplastic anemia, was successfully produced in sugarcane (Wang et al., 2005). Sugarcane has also proved to be a model system for the production of industrial products such as poly-3-hydroxybutyrate (PHB) (Brumbley et al. 2007), p-hydroxybenzoic acid (pHBA) bacterial enzymes like Chorismate pyruvate-lyase (CPL) and 4-hydroxycinnamoyl-CoA hydratase/ lyase (HCHL) (McQualter et al. 2004b). Though the opportunities to develop sugarcane as a bio-factory are expanding, several technical hurdles remain to be solved.

Transgenic Field Trials

Initial years' of transgenic research in sugarcane was limited to laboratory. Further somaclonal variation caused by tissue culture procedures produced undesirable field characteristics in genetically transformed sugarcane that are not readily identifiable in the laboratory or greenhouse (Arencibia et al.1998). The frequency of

variants and suitability of tissue culture treatments varied between cultivars. Therefore, subsequent transformation studies focus was on agronomic analyses in the field across several generations to ensure the stability of transgenic expression. However, despite its importance, agronomic analyses of transgenic sugarcane generally have been lacking (Christy et al. 2009; Gilbert et al. 2009), or reports have focused on only a single transgenic line (Leibbrandt and Snyman 2003). Current field data are only available for sugarcane plants engineered for few traits such as sugarcane borer, Mexican rice borer, herbicide resistance, sugar color improvement and metabolic and alternate product engineering (Arencibia et al. 1999; Legaspi et al. 2000; Vickers et al.2005; Gilbert et al. 2009; Neil etal. 2010). Few of these results have lead to the testing of the plant phenotypes in large scale under field conditions for regulatory purpose in USA, Brazil, Australia and South Africa. Country wise and year wise details of transgenic traits under various regulatory purposes trials are shown in Table 3, 4 & 5. However, to date none of these products have reached commercialization stages in these countries, and field trials assessing its safety are in progress.

Evaluation of transgenic sugarcane in Brazil has been continuing since 1997 under National bio-safety commission (CTNBIO). Studies carried out by CTC and various collaborators has produced transgenic sugarcane (Snyman et al., 1998, Matsuoka et al., 2009 , Cheavegatti-Gianotto et al., 2011 and field trials have been conducted evaluating genes responsible for herbicide tolerance, virus and insect resistance, flower inhibition, and increased sucrose yield (Table 3). According to CTC, the field trials will test - three varieties of genetically modified cane. These GM plants have been modified to exhibit sucrose levels 15 % higher than those of ordinary sugar cane. However, if field trials are successful, the company may bring these plants to market by the end of the decade. Herbicide resistant transgenic sugarcane has also been tested in field trials conducted by BASF (Table 3). It is expected that in the next few years, the first commercial transgenic sugarcane variety will be available for the sugarcane growers in Brazil.

Australia's Office of the Gene Technology Regulator (OGTR) is organizing limited and controlled release sugarcane transgenic at various locations in Australia. BSES has proposed a number of control measures to restrict the dissemination and persistence of the GM plants in the environment, including monitoring of fields for volunteer plants, destruction of plant materials not required for experimentation and isolation of fields from natural waterways. In addition to the herbicide-tolerance genes, the sugar cane lines express the antibiotic resistance markers *nptII* and *bla* from *E. coli* and *gfp*

Table 3 . Field trials of transgenic sugarcane approved in Brazil by the National Biosafety Commission (CTN Bio)

Institution	Trait	No. of field trials	Year of approval
CTC	Herbicide tolerance	9	1997, 1998, 1999, 2000
	Virus resistance	3	1999, 2000
	Insect resistance	1	1999
	Flowering inhibition	1	2002
	Sucrose yield	5	2005, 2006
Alellyx SA	Virus resistance	2	2005, 2006
	Sucrose yield	6	2006, 2007, 2008
	Drought tolerance	3	2007, 2008
	Herbicide tolerance + Insect resistance	4	2007, 2008
BASF S. A.	Herbicide tolerance	4	1999, 2000, 2001, 2002

Source: CTNBIO (www.ctnbio.gov.br)

Table 4 . List of applications and licences for dealings involving intentional release (DIR) into environment

Organisation	Title of Project	Modified Trait
Bureau of Sugar Experiment Stations	Agronomic assessment of transgenic sugarcane engineered with reporter genes	Green fluorescent reporter gene
The University of Queensland	Field trial of genetically modified (GM) sugarcane expressing sucrose isomerase	Altered sugar production and antibiotic resistance
BSES Limited	Limited and Controlled Release of GM Sugarcane with altered plant architecture, enhanced water or improved nitrogen use efficiency	Altered plant architecture, enhanced water or improved nitrogen use efficiency
The University of Queensland	Limited and controlled release of sugarcane genetically modified for altered sugar production	Altered sugar production
BSES Limited	Limited and controlled release of sugarcane genetically modified for altered plant growth, enhanced drought tolerance, enhanced nitrogen use efficiency, altered sucrose accumulation, and improved cellulosic ethanol production from sugarcane biomass	Altered plant growth, enhanced drought tolerance, enhanced nitrogen use efficiency, altered sucrose accumulation, and improved cellulosic ethanol production from sugarcane biomass
BSES Limited	Limited and controlled release of sugarcane genetically modified for herbicide tolerance	Herbicide tolerance

Source: http:// www.OGTR.gov.au/ internet/OTGR/publishing risf/content

gene from jelly fish and altered genes for sugar production are the major types of transgenic under field trial.

In USA, thirty one applications, permissions were granted to carry out various stages of field release trials at various locations -Table 3 (APHIS 2011). Traits include viral resistance, pest resistance, and herbicide tolerance. Predominantly these trials are from public and academic institutions such as Department of Agriculture, Texas AM University, University of Florida, Hawaiian Agriculture centre and recently from Syngenta. Even though there are large number of trials are progressing in USA, Brazil, Australia and South Africa no clear cut plan for large scale release or commercial release of these products are known till date.

Emerging Sugarcane Trans-gene Alliances

The importance of sugarcane as a principal crop for, sugar power and alcohol in Brazil attracted the attention of private investors interested in the creation of biotech companies for sugarcane. In 2002, a biotech company called Alellyx was founded with investments from Votorantim. Subsequently a sugarcane breeding company namely, Canavialis was also established by Votorantim to develop sugarcane varieties superior agronomic traits and biotech traits. Recently biotech major Monsanto, acquired these companies to strengthen its biotech program in Sugarcane. Canavialis recently signed a US$25 million deal with Cosan- one largest sugar production companies in Brazil to set up 10 research stations and develop sugar cane varieties. These companies are currently conducting several field trials for herbicide, insect resistance drought resistance, and sugar yield traits - Table 2 (Richard 2009; Grain 2009). CTC – Centro de Tecnologia Canavieira one of the largest sugar and sugarcane research centre in Brazil and Bayer crop science recently announced a co-operation agreement for combining their competencies in sugarcane breeding and biotechnology with the aim

Table 5 . List of approvals issued for intentional release of sugarcane transgenics in USA.

S. No.	Institution	Date /year	Phenotypes
1	Syngenta	07/08/2011	HT-CBI, MG-Gus Expression
2	Syngenta	05/25/2011	HT-CBI, IR-Resistant To Lepidopteran, IR-Resistant To Lepidopteran Larvae, MG-Alternate Carbon Source Utilization, MG-Fluorescent Marker
3	Texas AgriLife	04/18/2011	HT-Phosphinothricin Tolerant, IR-Mexican Rice Borer Research Resistant, VR-Sorghum Mosaic Potyvirus Resistant
4	Syngenta	03/28/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
5	Syngenta	03/14/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
6	Syngenta	03/14/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
7	Syngenta	02/15/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
8	Syngenta	02/11/2011	HT-CBI, MG-Fluorescent Marker, PQ-Altered Sugar Storage
9	Syngenta	02/06/2011	HT-CBI, IR-Resistant To Lepidopteran Larvae, MG-Fluorescent Marker, PQ-Altered Sugar Storage
10	Syngenta	02/06/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
11	Syngenta	02/04/2011	PQ-Altered Sugar Storage
12	Syngenta	01/10/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
13	Syngenta	01/10/2011	MG-Fluorescent Marker, PQ-Altered Sugar Storage
14	Syngenta	12/13/2010	MG-Fluorescent Marker, PQ-Altered Sugar Storage
15	Syngenta	12/13/2010	MG-Fluorescent Marker, PQ-Altered Sugar Storage
16	Syngenta	12/13/2010	MG-Fluorescent Marker
17	Syngenta	12/01/2010	MG-Fluorescent Marker, PQ-Altered Sugar Storage
18	Syngenta	06/01/2010	IR-Resistant To Lepidopterans
19	Syngenta	06/01/2010	HT-CBI, MG-Fluorescent Marker
20	Syngenta	05/28/2010	MG-Selectable Marker Gene, PQ-Altered Sugar Storage
21	Syngenta	05/15/2010	MG-Fluorescent Marker
22	Syngenta	03/16/2010	MG-Fluorescent Marker, MG-Fluorescent Protein Expression
23	Syngenta	03/16/2010	MG-Fluorescent Marker
24	Syngenta	03/03/2010	MG-Fluorescent Marker
25	Syngenta	03/03/2010	MG-Fluorescent Marker
26	Syngenta	03/02/2010	MG-Fluorescent Marker
27	Texas A&M	04/18/2008	HT-Phosphinothricin Tolerant, IR-Mexican Rice Borer Agricultural Resistant, VR-Sorghum Mosaic Potyvirus Resistant Experiment Station
28	Hawaii Agriculture	01/11/2002	OO-Pharmaceutical Proteins Produced Research Center
29	Texas A&M University	10/27/1997	IR-Mexican Rice Borer Resistant
30	Texas A&M University	04/21/1997	VR-Srmv Resistant
31	Texas A&M University	08/30/1995	HT-Phosphinothricin Tolerant

Source: http://www.isb.vt.edu/search-release.aspx

of bringing sugarcane growers, higher-yielding and drought-tolerant sugarcane varieties. To respond to the increasing needs of R&D in the area of Biofuels, State of Sao Paulo Research Foundation (FAPESP) also created a Bioenergy program (BIOEN) linking public and private R&D in Brazil. Biomass program of Bioen focus to integrate comprehensive research on sugarcane and other plants that can be used as biofuel sources, thus assuring Brazil's position among the leaders in the area of Bioenergy. Their research includes biomass production and processing and new paths of genetically manipulate the energy metabolism of sugarcane by creating new bio-fuel alternatives (Grice et al. 2003). In 2008, a group of 15 researchers from 4 countries (Australia, Brazil, EU and France) met in São Paulo to evaluate the feasibility of deciphering and assembling the sequence genomes of modern sugarcane cultivars in the light of new technologies, focusing on whole genome shotgun (WGS) and/or BAC insert sequencing (BAC) approaches .To pursue this, researchers agreed to form a multi-national initiative of an International Consortium for the Sugarcane Genome Sequencing (ICSGS)

The Australian sugar industry has been making substantial investment in the development of genetically modified sugarcane, with involvement of the key agencies such as BSES Ltd, the Cooperative Research Centre for Sugar Industry Innovation in Biotechnology (CRC SIIB), CSIRO Plant Industry, CSR Ltd, Queensland University of Technology, and the University of Queensland (Richard 2009). The Co-operative Research Centre for Sugar Industry Innovation through Biotechnology (CRC SIIB) and Dow Agro Sciences have recently signed research collaboration to combine technologies and capabilities to accelerate discovery and development of novel sugarcane products. US based DuPont Co. (DD) and Australia's BSES Ltd. announced a research, development and commercialization alliance to boost productivity and use of sugar cane varieties using genetic modification and other biotechnologies. The alliance, which brings together DuPont's plant biotechnology expertise with BSES' knowledge of cane breeding, cropping and milling, will focus on the development and delivery of technologies to improve planting technology and agronomic practices to boost productivity and reduce production costs.

Emerging Challenges

Despite the last two decades of advancements biotechnogical research and its commercial success in large number of crops and countries its application in sugarcane in commercial level is far from immediate reality. For the successful release of sugarcane transgenic at commercial level various scientific, regulatory, consumer and public issues need to be addressed. The lack of reliable transformation system, efficient gene promoters, transgene silencing, the limited knowledge of polyploidy genome architecture and heritability of quantitative traits field and agronomic variability are the major scientific or technological factors limiting implementation of transgenic in sugarcane. Since 1980 diverse methods such as biological, chemical and physical have been employed successfully to transfer genes to plants. Among the transformation systems micro projectile-mediated transformation and *Agro bacterium* based methods are the largely employed in sugarcane (Bower and Birch 1992; Arencibia et al. 1995; Enriquez et al. 2000; Manickavasagam et al. 2004; Lakshmanan et al. 2005). However, transgenic lines produced by both methods show large variation in clonal expression and variability in field. Tissue culture induced somaclonal variations continue to be another major impediments for successful transgenic development and considerable improvements of transformation systems are required to ensure clonal fidelity and reduce field variability (Birch 1997; Arencibia et al. 1998; Lakshmanan et al. 2003 & 2005; Gilbert 2009).

Large number of diverse promoters used successfully in other crops have also been attempted for transformation and expression in sugarcane (Rathus and Birch 1992; Gallo-Meagher and Irvine 1996; Lakshmanan et al. 2003) Their success rate in providing a stable expression in sugarcane is highly varied. Gene silencing, lack of genomic integration and varied expression have been major concern in most of the studies (Birch and Maretzki1993). In the recent past, research has been focused on increasing the range of promoters available for sugarcane transformation. Although the exact mechanism of lack of genomic integration and expression are not known in sugarcane, however, it has been postulated that it arise from defective promoters on redundant alleles in the highly polyploid genome, or

from efficient transgene silencing at both transcriptional and post-transcriptional levels (Mudge et al. 1996; Arencibia et al.1998; Ingelbrecht et al. 1999; McQualter et al. 2004 b; Vickers et al. 2005) efficient promoter system which deliver stable expression without negative yield impact on plant in diverse field and agronomic conditions are needed to ensure large scale commercialization of existing transgenic in sugarcane. Further understanding the control of trans-gene expression, stability, accumulation, and biological activity of its end products are critical. This is particularly true when attempting to produce foreign proteins in a polyploidy plant like sugarcane.

Despite the large scale adoption of biotech crops by developed and developing countries and positive recommendations from various agricultural organizations such as FAO , controversies regarding its benefits to mankind, perceived safety and ecological fate continue to be debated all over the world (Altieri 2005;Paula and Graham 2007; Lu 2008; Sherman 2009; Wikipedia 2011). The controversy is a dispute over the relative advantages and disadvantages of transgenics which involves biotechnology companies, governmental regulators, non-governmental organizations and scientists. The dispute is most intense in Japan and Europe, where public concern about GM food is higher than in other parts of the world such as the United States. This situation is unlikely to change also in the present context. Globally the benefits derived from transgenic crops are subjected to specific crop, trait and region. A survey of global impact of biotech crops for the period of 1996-2007, Brookes and Bare foot estimated reduced pesticide use by 359 million kg (-8.8 percent), and as a result, decreased the environmental impact associated with herbicide and insecticide use on the area planted to biotech crops by 17.2 % (Brookes and Barfoot 2009). They also reported substantial net economic benefits to farmers amounting to $10.1 billion in 2007, and $44.1 billion since 1996. Of the $44.1 billion, 46.5 percent ($20.5 billion) was due to increased yields and the rest to reductions in the cost of production. However organizations such as Union of Concerned scientists, certain number of pressure groups and consumer rights groups, such as the Organic Consumers Association, Greenpeace, World Wildlife Fund, and Consumers Union of Japan claim long-term health risks or the environmental risks have not yet been adequately investigated, refute data's, lack of substantial yield improvements, continue raising environmental, safety concerns and call for additional and more rigorous regulatory testing (Richard 2009; Sherman 2009; Wikipedia 2011). These groups continue to voice that the level of regulation for transgenic organisms is not proportional to their potential risk to human health or to the environment, and demand revision to the regulatory system. Further, public opinion currently appears to be biased against foods derived from transgenic crops compared to fiber or animal feed crops. However, in most of the controlled feeding trials, no toxic effects have observed. GM foods have been eaten by millions of people worldwide for over 15 years, with no reports of ill effects (Brookes and Barfoot 2009; Wikipedia; 2011). Another area of controversy is its effect on biodiversity and ecology and risk of horizontal gene transfer to wild relatives (Brookes and Barfoot 2009; Wikipedia; 2011). These issues and debates indicate controversies of transgenics crops benefits over perceived risks continued to exist in near future and no immediate solution is expected . There are many diverse and complex challenges ahead for regulatory authorities and governments, especially in the areas of safety testing, regulation, international policy and food labeling to obtain public acceptance of sugarcane transgenic as it is an important food crop.

Conclusion

Last two decades of research carried out by academic and sugarcane research Institutes from various parts of world have contributed substantially in understanding genomic structure and architecture of sugarcane. Extraordinary development of high-throughput methods for identifying and quantifying DNA, mRNA, proteins and metabolites and its application on sugarcane in recent years provided an opportunity to use, partial sequencing of genome, metabolite fingerprinting and profiling to identify desirable traits in sugarcane, from different populations. Transfer of this knowledge and technology & its close integration with breeding programs is anticipated to result in rapid translation of the sequence data and genetic manipulation of sugarcane for improvement of agronomic and productivity traits. Also, the analysis of the transcriptome in transgenic plants, altered for genes of interest, would certainly prove to be an excellent tool to unravel further sugarcane regulatory networks associated with important traits.

Genetic manipulation and production of transgenic sugarcane has been a reality in laboratory and field level. However various scientific, regulatory, public perception and acceptance issues need to be addressed to enable commercial release of sugarcane. Since the approval of Roundup Ready® sugar beets for production and for food and feed uses, in the United States and Canada, its large scale adoption by farmers and successful consumer acceptance would pave way for faster regulatory approvals for sugarcane transgenic crops. The current potential for transgenic in sugarcane lies with enhanced sugar content, pest and herbicide resistance, stress tolerance and other agronomic traits. The potential of sugarcane as a multiple feed stock for bio refineries producing diverse industrial products attracted multinational corporations of chemical pharmaceutical, seed and biotech origins to make large investments for sugarcane research. These investments are expected to play a crucial role in genetic modification of sugarcane crops by modifying to produce quantity or quality of biomass, high value pharmaceutical and industrial products. The bio- refinery of the future, in strategic alliance with manufacturing, marketing and other partners, utilizing new technologies, new chemistries, and new processes will be energy efficient and produce a range of low medium high value product whose output can be tailored to the demands of future. New generation of platform bio- sugar refineries will continue to produce sugar, transportation fuels, provide electricity and plastics, supplementing many of the current uses of fossil fuel. In addition to these, third generation bio-pharming combined with bio-refineries could also be the source of valuable co-products, such as chemicals, high value commercial and pharmaceutical products from within the sugarcane plant. Technological advancements and innovations in genomic research and transgenic will undoubtedly usher new tools to develop sugarcane as a commercially viable biological platform for crop improvement bio-pharming and establishment of integrated bio-refineries in future. Policy makers, Regulators, scientists, entrepreneurs and farmers at domestic and international level need to understand these emerging opportunities and plan and make sugar cane farming and industry a vibrant, most efficient and rewarding to all its stakeholders.

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