

Weed management in cotton: The potential of GM crops

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ABSTRACT

In recent times, biotechnology has been widely used for crop improvement. Today, about 2 billion hectares of global area is planted with genetically modified (GM) crops. In India, the first GM crop to be introduced was Bt cotton. The current acreage planted with Bt cotton is 93% of the total cotton acreage. However, the average yield is lower than that of other countries suggesting an opportunity to increase yield further. One of the major factors affecting yield is weed competition which reduces yield by 50 to 85%. Effective weed control is achieved by Integrated Weed Management (IWM) which includes adoption of transgenic herbicide tolerant crops (HTCs). The major transgenic HTCs grown in the world are soybean, cotton, corn and canola and the yield increase due to effective weed management is significant. In cotton, glyphosate and glufosinate tolerant systems have been used successfully across the globe and are being tested at the moment in India. Over reliance on single MOA (mode of action) rather than a diversified IWM system with multiple, complementary herbicide MOAs can lead to emergence of herbicide tolerant weeds. Therefore, there is a need to use diversified management practices for sustainable weed control in cotton.

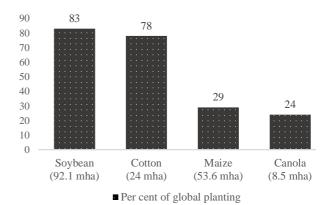
Keywords: Cotton, Genetically modified, Glufosinate, Glyphosate, Herbicide, Integrated weed management, Yield

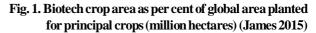
Global population is on the rise and has reached 7 billion. In India alone, the population is 1.2 billion. However, the area under cultivation is on the decrease and the current food production will not meet the growing demand. There is an immediate need to increase crop productivity to meet this demand. This necessitates the use of better seeds, better hybrids, germplasm, improved agronomic practices and novel technologies for enhancing crop productivity and yield. Biotechnology has opened the doors to improving productivity with the introduction of genetically modified crops. Ever since the first release of biotech crops, there has been an increase in the rate of adoption of these crops globally. In 2015, 28 countries planted biotech crops and it is estimated that more than half the world's population lives in these 28 countries (James, 2015). Soybean and cotton are the major biotech crops to be widely cultivated (Fig. 1).

In India, cotton is an important commercial crop supporting the livelihood of about 7.7 million farmers. Cotton occupies an area of 12.25 million ha of which

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11.6 million ha (94%) is genetically modified cotton (Bt cotton) (Choudhary and Gaur, 2015). India is the second largest exporter of cotton (FICCI report, 2012). In the last seven decades that cotton has been grown, production and productivity have steadily increased. However, in the last few years it appears to have reached a plateau. Current production is about 39 million bales (Choudhary and Gaur, 2015). The low production/productivity is attributed to lack of appropriate micronutrient and fertilizer management, prevalence of sucking pests, weeds, small farm area and more of the cotton acreage being grown in drought prone regions (GAIN report 2013). The current average yield of 500 kg/ha is significantly lower than other countries like Brazil (1393 kg/ha) (The Crop Site 2013), China (1311 kg/ha) and USA (900 kg/ha) (FICCI report 2012). There is an increasing demand for cotton fiber, both for local consumption (27.5 million 170 kg bales) as well as export to other countries. Further, consumption of cotton seed oil for domestic use is increasing as it is economical and the keeping quality is also better (Economic Times 2013). This huge gap in the production and demand opens up tremendous opportunities to increase yield (GAIN Report 2013, Economic Times 2014).

Yield in cotton is dependent on the climatic conditions, rainfall pattern, weed competition and incidence of pests and diseases. Weeds are a potential problem in cotton cultivation and reduce yield by 50 to 85% depending upon the nature and intensity (Jain *et al.* 1981). Weeds also enhance production costs posing an income risk to the farmers (Frisvold *et al.* 2009).Weed management therefore assumes prime importance. This review focuses on the different methods for weed management and gives a brief insight into the current technology available.

Impact of weeds on cotton

Cotton is a long duration crop and typically takes about 140-160 days to complete its life cycle. Throughout the growth cycle it is exposed to weeds and the competition therein. Every crop has a critical period of weed control (CPWC) which refers to the minimum time period during which the crop must be weed free. In cotton, the CPWC is the first 15 to 60 days (Ayyadurai and Poonguzhalan 2011). Maximum yield can be derived when there is at least 95% weed control (Sharma 2008).

Weeds compete for available resources like sunlight, water, nutrients and space. In fields infested by weeds, the top soil is drier compared to weed free plots and this is attributed to a higher extent of water removal from the top 15cm of soil. Smooth pig weed (Amaranthus hybridus) reduced soil moisture content from depths of 122 to 183 cm to a greater extent than cotton. Weed competition after the first and second irrigation cycles reduces yield to an extent of 20 per cent. In terms of competition for nutrients, weeds deplete the soil by removing 5-6 times Nitrogen, 5-12 times Phosphorous and 2-5 times Potassium than cotton crop thus reducing yields by 54-85 per cent (Information from ikisan http://www.ikisan.com/ Cropper cent20Specific/Eng/links/knt_cotton Weedpercent20Management.shtml). Grassy weeds, which grow in the cotton rows or which get blown into cotton are difficult to remove and stain the lint reducing fiber quality (Charles and Roberts 2013).

Weeds also serve as hosts for insect pests (Table 1) and diseases resulting in increased production costs ultimately reducing yield. It has been shown that weeds could also release allelopathic chemicals suppressing growth of cotton (Riffle et al, 1987). However, the impact of weeds on growth and yield is dependent on the type of weeds, the extent of spread and their duration during the crop growth period (Chiunnuswamy and Chinnagounder 2013).

 Table 1. Weeds in cotton serving as alternate hosts to different pests of cotton

Weed species serving as	Insect pests
alternate hosts	
Datura ferox, Lantana camera,	American boll worm
Nicandra physaloides	(Helicoverpa armigera)
Abutilon spp., Sida spp.,	Spotted bollworm
Hibiscus panduraeformis,	(<i>Êrias</i> spp.)
Urena lobata, Chorchorus sp.	
Hibiscus esculentus, Hibiscus	Pink bollworm
panduraeformis, Abutilon	(Pectinophora
indicum.	gossypiella)
Malva parviflora, Hibiscus	Shoot weevil
spp., <i>Urena lobata</i>	(Alcidodes affaber)

Based on the soil and climatic conditions weed flora are diverse with the major categories being grasses and broad-leaved weeds. The most prevalent weeds across cotton fields in India are *Cyanodon dactylon*, *Cyperus rotundus*, *Panicum ripens*, *Euphorbia* sp. and *Trianthema potulacstrum*. Weeds specific to different cotton growing regions in India (Table 2) has been documented by Nagrare *et al.*(2011).

 Table 2. List of weeds specific to different cotton growing areas in India

Region/State	Weeds	
Tamil Nadu	Dactyloctenium aegyptium	
Karnataka	Abutilon indicum, Panicum isachne,	
	Bracharia romosa, Bracharia cruciformis,	
	Euphorbia geniculata, Tridax procumbens,	
	Flavaria australasica and Setaria sp.	
	Digitaria marginata, and Amarathus sp.	
Andhra Pradesh	Corchorus olitorius, Abutilon indicum and	
	Sida acuta	
Punjab	Silene conoidea and Sphenoclea zeylanica	
Haryana	Trianthema portulacastrum,	
	Helianthus,Cyamopsis tetragonoloba	
Maharashtra	Trianthema portulacastrum, Digera	
	muricata, Taraxacum officinale, Euphorbia	
	sp., Abutilon indicum	
Gujarat	Trianthema portulacastrum, Digera	
	muricata, Taraxacum officinale, Euphorbia	
	sp., Abutilon indicum	

In a field study conducted at Regional Agricultural Research Station, Lam, Guntur appli-cation of post- emergence herbicides increased yield by 66-75% compared to unsprayed plots. Weed density and weed dry matter was significantly reduced in the sprayed plots (Bharathi *et al.* 2011). A combination of pre- and post-emergent herbicides resulted in 96.8% weed control. Seed cotton yield increased from 22.98 to 38.25% compared to untreated controls (Shaik *et al.* 2006).

Weed management techniques

Weed management is an integral component of sustainable farming systems and comes with a cost. In a typical cotton growing season of 140-160 days, weeding is done at least three times, 15-20 days after sowing (the early-leaf stage), after 35-40 days (before square formation) and 55-60 days (before flowering). The practices adopted should prevent weed interference with the main crop, reduce the extent of weed seeds in the soil, be non injurious to the main crop, not reduce the lint quality and should be economical and sustainable. The timing of weed control and the execution of weed management practices also play an important role (Prabhu *et al* 2012).

In India, most often manual weeding practices are followed and the average cost incurred by a farmer is about 32% of his total production cost (Gandhi and Namboodiri 2009). The first manual weeding requires at least 10 laborers an acre and the number goes up to 15 to 20 for the next two weeding. For weeding alone, a farmer incurs a cost of ₹ 10,000 (Rajeswari and Charyulu1996, Seed News 2011). Given this challenge and expense, other weed management strategies need to be employed. A brief insight on the five general weed management strategies practiced in cotton (Ashigh *et al.* 2012) is given below.

Prevention: This involves preventing weeds from entering the fields by control before they set seed and planting certified seed. Tillage and harvesting equipment should be clean of weeds when moving between fields.

Cultural practices: This focuses on agronomic practices that include crop rotation, appropriate fertilization, spacing, use of cover crops and date of planting which favors cotton growth. Rotating crops suppresses weeds due to variation in the specific host. Cotton grown in narrow spacing (25 inch rows) requires a shorter weed free maintenance period compared to that grown with broad spacing (40 inch rows). Cover crops suppress weeds by preventing their germination. **Mechanical**: This is a non selective option and implies the use of mechanical tools like rotary weeders, disks, hoes or mechanical choppers. This is an efficient technique for annual weeds. Three to four intercultivations (hoeing) should be taken at 15 day intervals 30 days after sowing. It helps in keeping the plot weed free and maintains soil moisture.

Chemical control: This is the most popular method used and a number of herbicides are available. This requires skilled labor as the appropriate herbicides have to be applied in the right quantities and at the right time. Herbicides are classified as pre-emergence and post-emergence applications (Table 3).

 Table 3. Herbicides commonly used in weed control in cotton

Time of ap	plication	Herbicide
Pre-plant	Pre-plant incorporated	Pendimethalin, Trifluralin
	Pre-plant burn down	Thifensulfuron-methyl, tribenuron-methyl
Post-plant	Pre- emergent	Diuron, Fluometuron, pyrithiobac-sodium
	Post- emergent	Clethodim, fluazifop-p-butyl, metolachlor, oxyfluorfen, pyrithiobac-sodium, sethoxydim

A single technique does not provide the complete solution to weed management. Integrated Weed Management (IWM) which is a combination of strategies offers solution to this daunting problem. IWM involves good seedbed preparation, manual weeding, crop rotation, optimum plant population, intercultivation and herbicide use. IWM practices should be selected based on the soil profile, climatic conditions, crop rotation practices and most importantly the weed species prevalent in the farm. IWM is advantageous as it uses multiple practices (cultural, chemical, mechanical) to manage weeds in an economical and sustainable manner (Farrell and Johnson 2005).

A combination of weed management techniques were employed by Chinnuswamy and Chinnagounder (2013). Their results suggest that manual weeding at 25 and 45 DAS (days after sowing) or application of pendimethalin as a pre-emergent spray 3 DAS improved yield of seed cotton. Alternatively, power weeding 25 DAS and manual weeding at 45 DAS was also found to be promising. Pre-emergent application of pendimethalin controlled grassy and broad leaved weeds and sedges. In addition, a second application at 45 DAS along with two manual weeding gave good weed control and resulted in higher seed cotton yield (Manikandan 2011). In another experiment conducted in *Kharif* season in Raichur, pre-emergent application of pendimethalin with post-emergent application of quizalofop-ethyl, inter-cultivation and manual weeding at 60 DAS resulted in significantly higher seed cotton yields and higher returns (Prabhu *et al* 2012).

A combination of weed management techniques has proven effective in controlling weeds. An additional option available to the farmers which should be considered is the adoption of genetically modified herbicide-tolerant crops which are now a component of Integrated Weed Management systems in many regions across the globe (Duke 1999).

Herbicide tolerant cotton

Genetic modification has enabled development of herbicide tolerant crops which are now cultivated widely in different countries across the globe. The first herbicide tolerant crop released was soybean followed by cotton and corn (Green, 2012). From then on, the acreage under herbicide tolerant crops has grown tremendously with soybean, cotton and corn occupying maximum area. This technology has now expanded to a number of other crops of commercial importance. In the United States, acreage under herbicide tolerant cotton expanded from 10% in 1997 to about 91% in 2014 while soybean area increased from 17% to 94%. Adoption of herbicide tolerant corn was slow in the initial years but the current figures stand at 89% (Fig. 2) (USDA, ERS, 2015). In India, insect tolerant cotton (Bt cotton) is the only bioengineered crop to be cultivated.

In cotton, glyphosate or glufosinate are the most commonly used herbicide systems. Glyphosate is a nonselective, broad-spectrum foliar herbicide known to control more than 300 weed species. It controls a spectrum of weeds ranging from annuals, perennials, sedges and broad-leaved plants (Green and Owen 2011). Glyphosate functions by inhibiting EPSPS (5enolpyruvylshikimate 3-phosphate synthase) a key enzyme in the shikimate pathway. The strategy used to introduce glyphosate resistance in crops is overexpression of an insensitive form of EPSPS (such as the EPSPS enzyme derived from Agrobacterium tumifaciens strain CP4 or the engineered Zm-2mEPSPS enzyme). Plants expressing such glyphosate insensitive EPSPS proteins are tolerant to commercial applications of glyphosate herbicide. The commercial product Roundup Ready® (RR) has this technology (Table 4) (Dill et al. 2008).

 Table 4. Commercially available transgenic herbicide tolerant cotton

Herbicide Resistance trait	Gene source	Trait
Glyphosate	Cp4 epsps	MON1445
	Two cp4 epsps	MON88913
	Zm-2mepsps	GHB614
Glufosinate	bar	LLCotton25

Glufosinate is also a nonselective, broadspectrum foliar herbicide impacting growth of more than 120 broad- leaved and grassy weed species. However, as it is a contact herbicide it cannot be used to effectively control perennials (Heap 2010).

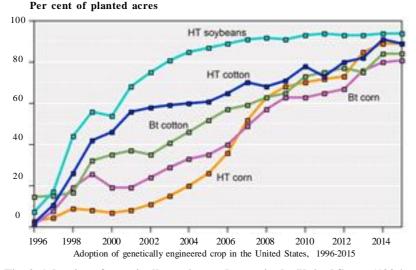


Fig. 2. Adoption of genetically engineered crops in the United States (1996-2015). Data for each crop category includes varieties with both HT and *Bacillus thuringiensis* (Bt) (stacked) traits (USDA, ERS 2015)

Data for crop category include varieties with both HT and Bt (stacked) traits. Source: USDA, Economic Research Service using data from fernandez-Cornejo and Mebride (2002) for the years 1996-99 and USDA, National Agricultural Statistics, June Agricultural Survey for the years 2000-2015

Glufosinate functions by inhibiting glutamine synthetase a key enzyme catalyzing the conversion of glutamate to glutamine in the nitrogen metabolism pathway (Senseman 2007). Glufosinate tolerance is the result of introducing either the pat or bar genes which were isolated from *Streptomyces viridochromogenes* and *Streptomyces hygroscopicus*, respectively. Both genes code for enzymes that inactivate glufosinate by acetylation (Mullner *et al.* 1993). Liberty Link® cotton employs the bar gene from *S. hygroscopicus* (Table 4).

Potential of genetically-modified herbicide tolerant crops (HTC)

Effective weed control involves an Integrated Weed Management (IWM) system including herbicide tolerant crops. The area under herbicide tolerant crops is increasing over the years. In Australia, farmers growing glyphosate tolerant crops reported better control of nutgrasses and vines which could not be controlled by traditional methods. In addition, hoeing was reduced resulting in lesser production costs (Sadler 2012). Similar reports have been obtained from farmers in the United States.

Charles *et al.* (2004) reported that by growing glyphosate tolerant cotton, 7.8 million kilos of herbicide was saved while growing glufosinate tolerant cotton saved 215,000 kilos of herbicide. A survey conducted by Werth *et al.* (2006), suggests that glyphosate tolerant cotton (2.3 to 3.2 kg active ingredient per ha) but use of other pre-emergent herbicides was reduced (3.38 kg to 2.55 kg active ingredient per ha). Other reports suggest that the frequency of application and the volume of herbicide applied were impacted. While there were two additional glyphosate applications every season there was a reduction in the use of other herbicides (Preston and Roush 1998).

As reported in two studies conducted by Sankula *et al.* (2005) and Sankula (2006), it was estimated that in the United States planting Roundup Ready® cotton reduced herbicide usage by 6.3 million kg active ingredient in 2004 and 7.8 million kg active ingredient in 2005. Use of Liberty Link® cotton reduced herbicide usage by 74,000 kg active ingredient in 2004 and 215,000 kg active ingredient in 2005. In the San Joaquin valley, it is estimated that the cost savings due to RR technology varied from \$25 to \$200 an acre (Wright *et al* 2013). Adoption of herbicide tolerant crops increased the usage of glyphosate but significantly reduced use of other herbicides.

Benefits and risks of HTC technology adoption

Adoption of herbicide tolerant crops holds a lot of promise. One of the most important advantages is the environmental safety of glyphosate due to its physicochemical characteristics. favorable Glyphosate is not toxic to mammals and the tight adsorption to soil leaves no residual toxicity to the subsequent crops (Gianessi 2005). Toxicology studies of glyphosate have been conducted by Sharma and co-workers (2012). Their results suggest high soil adsorption of glyphosate, thus inhibiting its penetration to water sources. The ground ubiquity score (is the leaching potential of a herbicide vis-a-vis associated environmental pollution risk) being lesser than 1.8 classifies glyphosate as a non-leaching herbicide. Therefore, glyphosate is not a hazard to ground water contamination. Further, studies conducted by Sailaja and Satyaprasad (2006) report that glyphosate uses the glycine pathway of degradation in soil with complete degradation by the 20th day after application. Grunewald *et al.* (2001) have reported the behavior of glyphosate and AMPA (á-Amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid) in soil and water with the half life of glyphosate ranging from 11-17 days. These studies indicate the rapid degradability of glyphosate in soil thereby reducing toxicity in the soil.

Glyphosate and glufosinate being broad spectrum herbicides, the number of sprays and use of other toxic herbicides is minimized in turn reducing labor requirement (Duke 1999). Use of HTC promotes no-till or reduced till practices thereby aiding in soil conservation and reducing water pollution from nutrient and sediment run off (Knezevic 2002, Fawcett and Towery 2002, Cerdeira and Duke 2006). Lack of weeds in HTC grown areas also mitigates infestation by pests and diseases (Joel *et al.* 1995, Liu *et al.* 1998, Brookes and Barfoot 2009, Green 2012). This ensures lower cost of production and higher cost benefit ratio for the farmers.

Herbicides impose selection pressure on the weed population. When the same herbicide is used repeatedly or if herbicides with the same mechanism of action are used, weeds may develop resistance over generations (ANR publication, 2013). A number of researchers have indicated that continuous and sequential use of glyphosate in the absence of other weed management practices imposes high selection pressure on weed flora resulting in glyphosate resistant weeds (Swanton *et al.* 2000, Shaner 2000, Benbrook 2001, Owen 2008). In cotton cropping systems, RR cotton is often rotated with RR corn

resulting in RR corn becoming a volunteer weed. Non optimal crop rotation practices also result in herbicide tolerant crops turning out to be volunteer weeds which reduces yield (Owen and Zelaya 2004).

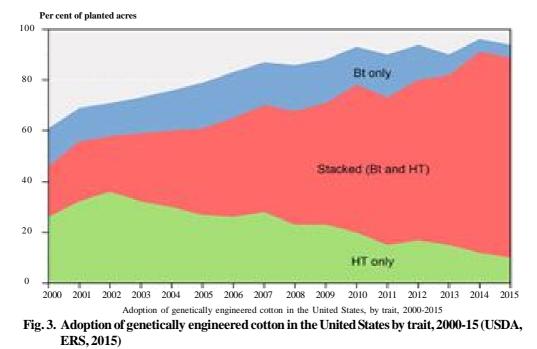
There is growing concern on development of herbicide resistant weeds. Repeated use of a single weed control technique in the absence of other weed management practices continuously over the years could have resulted in herbicide resistant weeds. As on date, 60 countries have reported herbicide tolerant weeds from about 350 unique species. The herbicides to which weeds have developed resistance are photosystem II inhibitors and ALS (Acetolactate synthase) inhibitors as they are used widely on cereals and grains (ANR publication 2013). Employing appropriate diverse management practices will reduce selection pressure from a single practice and mitigate the development of herbicide resistant weeds (Hurley et al. 2009). This includes pre-plant herbicide applications (Sosnoskie and Hanson 2013, Wright et al. 2013), deep tillage, use of mould boards or other tillage implements for tillage, crop rotation, use of residual herbicides along with glyphosate, mixing different herbicides together and herbicides with multiple modes of action (Duary 2008).

Concluding thoughts

The key to effective weed management is integration of diverse technologies like herbicides, agronomic practices and biotechnological approaches. This forms the basis for an integrated weed management program. Rather than a single tool/ technique being adopted, a suite of tools/techniques need to be utilized. A farmer now has multiple options to choose from and he needs to do so judiciously. This would be cost effective and give higher economic returns.

Perspectives for the future

The quest is on for durable and sustainable weed management practices. Herbicides with different modes of action but similar weed spectrum provide an option to combine herbicides for more durable weed management. Seeds with multiple traits like quality, disease resistance, insect protection and weed control could be developed by breeding or transformation technology. Stacking strategies also hold a lot of promise. Stacked products are available and are being used commercially. Across the globe, 13 countries planted stacked traits on an area of 51 million hectares, an increase of 4.3 million hectares compared to 2013 (James 2014). In double cropping systems comprising of soybean and wheat, stack of glyphosate and sulfonyl urea have been used effectively (Dupont Biotechnology 2007). Glyphosate and glufosinate dual stacks are also in commercial use in cotton, corn and soybean (Green 2012). In the United States, adoption of stacked traits is increasing with stacked cotton reaching 79 per cent of cotton plantings in 2015. Genetically engineered cotton including stacked traits reached 94 per cent of the total cotton acreage in 2015 (Fig. 3) (USDA, ERS, 2015).



Source: Economic Research Service using data from USDA, National Agricultural Statistics, June Agricultural Survey

Precision Agriculture with capability of imaging techniques helps identify the extent of weed infestation allowing determination of the appropriate timing and application rate of herbicides for maximized benefits (Duke 1999). Adoption of Bt cotton has been successful for Indian farmers. The cost of production of Bt cotton is higher compared to non Bt cotton under irrigated conditions. However, the output value is higher and the net profit from Bt cotton is about 56% compared to non-Bt cotton (Gandhi and Namboodiri 2009). The economic data for 16 years (1996-2012) suggests that farmers in India gained US\$14.6 billion. In addition, insecticide application reduced by 50% contributing to a sustainable environment and improved quality of life (James 2013). Given the success of Bt cotton in India, adoption of herbicide tolerant cotton would be promising and sustainable. This would also help reduce the yield gap and put farmers in India on par with other cotton growing regions of the world in terms of production and productivity. Farmers need to use this technology judiciously and include this in their IWM program.

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