

A1. Ecological Risks of Genetically-Modified Crops

유전자 재조합 작물의 생태적 안전성

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1. Introduction

Concern is increasing over the risk of environmental contamination with genetically modified (GM) crops. Under the selection pressure of herbicide, introgression between the GM crop and wild relative could produce new biota that might disrupt ecological balances. If the transgene causes the crop to become weedy, the GM crop itself could become an environmental hazard. Although there have been many debates between scientists, governmental authorities, agricultural producers, industries and consumer groups, any outputs from those debates have not yet given a confident acceptance of GM crops in our environmental context, particularly agro-ecosystem.

Novel traits being introduced into crop plants can be classified into three main categories: herbicide resistance, pathogen/pest resistance, and food improvement parameters such as shelf-life, and the amount/composition of fatty acid, protein, vitamin or other nutritional substances. So far, herbicide resistance is the most common trait being tested and thus herbicide resistant GM crops are now the most widely cultivated worldwide, reflecting both its experimental and commercial applications. Statistics show that 39.9 million ha of GM crops were cultivated worldwide in 1999. Herbicide-resistant GM crops, including insect/herbicide-resistant GM crops, represented 78% of the GM crops in this area in 1999, as shown in Table 1. Among herbicide-resistant GM crops, herbicide resistant soybean was the most popular, grown in about 21.6 million ha in 1999, followed by herbicide-resistant oilseed rape (OSR) of 3.5 million ha.

Regarding countries cultivating commercialized GM crops, the number of countries increased from six in 1996 to twelve in 1999 with the USA ranked the first place with 28.7 million ha, followed by Argentina of 6.7 million ha and Canada of 4.0 million ha (James, 2000). Interestingly, China was ranked the fourth place with 0.3 million ha. In China from 1997 to 1999, 121 GM crop applications were approved for environmental release, field testing or

commercialization, of which cotton and rice represented 33% and 26%, respectively of approvals. By trait, insect and virus resistances represented 52% and 21% of GM traits, respectively, while herbicide resistance represented only 2%, indicating that there is a lower demand for herbicide resistance traits due to low agricultural labor costs for weed control in China. Chen (1999) predicted that within the next 10 years, about 20-50% of five of China's principal crops (rice, wheat, maize, soybean and cotton), grown on a total of 98.9 million ha, could be planted as GM crops, equivalent to a half of the global GM crop area in 1999 at 20% adoption rate for GM crop (James, 2000).

The fact that GM crop cultivation has increased very fast in the last five years indicates that the consequences of the commercial release of GM crops will be realized whether they are better or worse than our prediction.

Table 1. Major GM crops planted in 1999.

Crop	Global area of GM crops (million ha)	% GM
Herbicide-resistant soybean	21.6	54
Bt maize	7.5	19
Herbicide-resistant oilseed rape	3.5	9
Bt/Herbicide-resistant maize	2.1	5
Herbicide-resistant cotton	1.6	4
Herbicide-resistant maize	1.5	4
Bt cotton	1.3	3
Bt/Herbicide-resistant cotton	0.8	2
Total	39.9	100

Source: James (2000). Bt, *Bacillus thuringiensis*.

2. Risk classification of GM crops

The risks associated with GM crops could be classified into four categories: (i) the toxicity or allergenic effects of transgenes on humans, animals or beneficial organisms; (ii) the stability of transgenes; (iii) gene flow within and between species, and to other organisms; and (iv) GM crops themselves

becoming weedy. The above risks are direct impacts of GM crops, while there are also some indirect impacts of GM crops.

Drift of non-selective herbicide out of the GM crop field could cause significant damage to neighboring non-GM crops and endanger some wild plants growing nearby non-cropping area. This problem of non-selective herbicide drift could be serious, particularly in the area of diversified small-scale farming. Expanding of GM crop cultivation may also bring about the risk of increased public cost of managing herbicide-resistant weeds at roadsides and parks and other areas. Unwanted gene pollution from a GM crop to nearby non-GM crops, by genes such as antibiotic-resistance genes or *Bacillus thuringiensis* (*Bt*) toxin gene, could create conflict between the farmer who does not want GM crops and the farmer planting GM crops or the company selling GM seeds. Use of so-called 'terminator gene' (Jury, 1998) would bring about a monopolization of a market by a company that sells GM crop seeds and chemicals. These may be considered as future social risks.

Insect-tolerant GM crops express δ -endotoxin genes derived from the *Bacillus thuringiensis*. The GM crop produces δ -endotoxin (Cry protein) in every cell of the plant throughout the growing season and leaves the residues in the soil.

Although the Bt toxins in the environment are biodegradable and safe, the continued and elevated presence of Bt toxins is of concern. Thomas and Ellar (1983) found that native δ -endotoxin crystals of BtI had no detectable toxicity in *in vitro* and *in vivo* systems tested, but alkali-solubilized crystal caused hemolysis of rat, mouse, sheep, horse and human erythrocytes. Intravenous or subcutaneous administration of the alkali-soluble δ -endotoxin to mice at a dose rate of 15~30 g/g body weight resulted in rapid paralysis followed by death. The crystalline toxins exist as protoxins which become solubilized in the alkaline gut contents of dipteran, lepidopteran and coleopteran insect larvae and undergo proteolytic cleavage to form active toxins, but would be degraded in the human stomach as they are not acid stable (Drobniewski, 1994). While *Bt* caused in rare incidences wound and eye infection to human and bovine mastitis. *Bacillus* species are numerous and widely distributed in nature, particularly in soil. Among them 12 species have caused human disease. *Bacillus cereus*, which quite often causes food poisoning, *Bacillus anthracis*, which causes anthrax to warm-blooded animals, and *Bacillus thuringiensis* are very closely related in taxonomy and share in common many biochemical characteristics. A dry form of anthrax spores is a conventional biological weapon. Thus, the potential risk to humans of *Bacillus thuringiensis* used for biological controls and Bt toxins produced by GM crops cannot be completely

eliminated.

Increased Bt toxin levels in the environment constitutes a high selection pressure for the development of Bt-toxin resistant insects. Another concern about GM crops expressing Bt toxins is that suboptimal production of toxins might result in an increased risk of pests developing Bt resistance (Daniell, 1999). Hence, a strategy called 'refuge' has been put forward recently by companies to delay or prevent appearance of insects resistant to Bt toxin. Refuge involves setting aside blocks of non-GM crops planted among the large acreage of Bt toxin-containing crops. The strategy assumes insect resistance to Bt toxin is genetically recessive. When the refuge is present, insects surviving Bt exposure do not need to hunt for rare Bt survivors for sex; they can limp over to the refuge and mate with insects bearing dominant genes. The offspring of such mating will all be sensitive to Bt toxin, but will bear recessive genes for Bt tolerance. The refuge delays the appearance of Bt-resistant insect populations (Cummins, 1998). However, dominant inheritance of Bt resistance has been observed in Colorado potato beetle (Rahardja and Whalon, 1995). That pest is a scourge of many vegetable crops (Cummins, 1998). Also, a recent study of a resistant strain of pink boll worm larvae on Bt cotton shows developmental asynchrony, which favors assortative mating among resistant moths emerging from Bt crops, and generates a disproportionately high

Agricultural crops support not only pest insect but also beneficial insects which feed on these herbivores and which play an important role in the regulation of pest population. Pest-resistant GM crops can thus affect natural enemies by severely depleting their supply of prey or hosts. The δ -endotoxin can kill pest insects but also non-target insects such as monarch butterflies, and beneficial invertebrates which are natural enemies of pests (Losey *et al.*, 1999). Cowgill *et al.* (1999) suggested that aphids feeding nematode-tolerant GM crops damage various natural enemies of aphids by exposing them to a nematode growth inhibitor. When highly tolerant crops are grown on a large scale, the abundance of some natural enemies may also decline due to prey depletion.

3. Risks of gene flow out of GM crops

Gene flow within and between populations has an important role in maintaining population genetic structures, enabling adaptation to changing environmental circumstances, and reducing vulnerability to evolutionary hazards

Table 2. Petitions for non-regulated status of GM crops approved in the USA from 1992 to 1999.

Trait	Gene	Donor	Crop
Herbicide resistance to			
Bromoxynil	Nitrilase	<i>Klebsiella pneumoniae</i>	maize
Glyphosate	EPSPS	maize, <i>Agrobacterium sp.</i>	maize, oilseed rape, soybean, beet
Phosphinothricin	Phosphinothricin acetyl transferase	<i>Strep.viridochromogenes</i> , <i>A. tumefaciens</i> , <i>Strep.hygroscopicus</i>	cotton, maize, beet, oilseed rape, rice, soybean
Sulfonylurea	Acetolactate synthase	tobacco	cotton
Soil residues of sulfonylurea	-	-	flax
Insect resistance to			
Coleopteran	CryIIIA	Bt ₁	potato
Colorado potato beetle	CryIIIA	Bt ₁	potato
European corn borer	CryIA (b), CryIA (c)	Bt ₁₀	maize
Lepidopteran	CryIA (b), CryIA (c)	Bt ₁₀	cotton, maize, tomato
Herbicide/Insect resistance to			
Glyphosate/European corn borer	EPSPS, glyphosate oxidoreductase/CryIA (b)	<i>Agrobacterium sp.</i> , <i>Achromobacter sp.</i> ,/Bt ₁₀	maize
Bromoxynil/Lepidopteran	Nitrilase/CryIA (c)	<i>Klebsiella pneumoniae</i> /Bt ₁₀	cotton
Phosphinothricin/Male sterility	-	-	maize, oilseed rape
Phosphinothricin/Lepidopteran	-	-	maize
Virus resistance to			
CMV, WMV2, ZYMV	coat protein	CMV, WMV2	squash
PRSV	coat protein	PRSV	papaya
WMV2, ZYMV	coat protein	WMV2, ZYMV	squash
Product quality improvement			
Oil profile	ACP thioesterase, Delta-12 desaturase	California bay, soybean	oilseed rape, soybean
Fruit polygalacturonase level	Polygalacturonase, Polygalacturonase antisense	Tomato	tomato
Fruit ripening	S-adenosylmethionine transferase, ACC deaminase, ACC synthase, Polygalacturonase antisense	bacteriophage T3, tomato, <i>Pseudomonas chlororaphis</i>	tomato

Source: USDA-APHIS-PPQ-PRA Biotechnology Authorizations (as of 6/30/2000).

such as inbreeding depression and genetic drift. However, when we consider GM crops, gene flow is a different story. Escaped genes from GM crops will result in unknown consequences, as most transgenes

are transferred to the crops from other organisms. Gene flow is the movement of genes mediated by pollen flow and seed dispersal. Potential gene flow from GM crops is the movement of seed and pollen as a function of distance, while actual gene flow is the amount of fertilization in the case of pollen and establishment of reproductive individuals in the case of seeds as a function of distance from a source. These two processes (movement of pollen and seed, and fertilization and establishment, respectively) combine to move genes in space and time. Gene flow can occur within species (GM crops to the same crop species), between species (GM crops to different plant species), and from GM crops to other organisms such as microorganisms.

Risks of gene flow within species

The most possible gene flow can occur between GM crops and non-GM crops or their wild species. Within each crop species, there are hundreds of different cultivars bred using conventional method based on Mendelian heritance worldwide. When a GM crop is cultivated in proximity of a non-GM crop field, there will be gene exchange between them, whether it is low or high. This exchange is most possible between species belonging to Brassicaceae family, such as radish and OSR, with their high outcrossing rate (Table 3). Vigouroux *et al.* (1999) reported hybridization between bolting GM sugarbeet and weed beet occurred under field conditions. Colbach *et al.* (1999) simulated gene escape from GM OSR to volunteer OSR in time and space. Despite the predominance of non-GM OSR in the immediate locality, all sites was pollinated by a mixture of GM and non-GM sources, suggesting that the farm-to-farm spread of GM crop will be widespread. Early isolation studies in western Canada indicated an average outcrossing rate using a chlorophyll-deficient marker from large fields to small plots of 0.6 and 3.7% at 366m for *B. napus* and *B. rapa*, respectively. Although field-to-field crossing levels were low, a considerable number of hybrid seeds could remain in a field after harvest. Downey (1999) exemplified that a gene flow of 0.4% into a field yielding the Canadian average of 1,400kg/ha, with a harvest loss of 5% could result in some 70,000 outcrossed seed per ha remaining (seven seeds/m²) in the recipient field. If the hybrid is resistant to a herbicide and careful rotation and herbicide management practices are not followed, field to field gene flow can result in gene stacking. Thus, gene flow from GM to non-GM crops may

result in genetic contamination and also make its wild species weedier due to transgenes expressing resistances to herbicides or abiotic/biotic stresses.

Table 3. Gene outflow potential of some important crops.

Crop	Chromosome No. (2n) & Genome	Outcross (%)	Isolation standard (m)	Compatability with crops and relatives
Normally outcrossing crops				
Carrot	18			<i>Datura capillifolius</i> , <i>D. carota</i>
Maize	20		200	Teosinte, <i>Zea mexicana</i>
Radish	18	>85	1000	<i>Brassica oleracea</i> , <i>B. napa</i> , <i>Sinapis arvensis</i> , <i>Raphanus raphanistrum</i> , <i>R. maritimus</i> , <i>R. landra</i>
Rye	14		200	wheat, <i>Secale ancestrale</i> , <i>S. dighoricum</i> , <i>S. segetale</i> , <i>S. afghanicum</i> , <i>S. montanum</i>
Sorghum	20	30-40	300-400	wild sorghums, <i>Sorghum halepense</i> , <i>S. propinquum</i> , <i>S. bicolor</i> subsp. <i>verucilliflorum</i> ,
Squash				close wild or feral relatives, <i>Cucurbita lundelliana</i> , <i>C. martinii</i> , non-weedy congeners
Sugarbeet	18			all forms of the section <i>Beta</i>
Sunflower	34	20-75	800	<i>Helianthus tuberosus</i> , <i>H. petiolaris</i> , <i>H. agrophyllus</i> , <i>H. agrophyllus</i>
Normally inbreeding crops				
Barley	14 (28)	Variable (max :10)	0	wild forms, <i>Hordeum spontaneum</i> , <i>H. bulbosum</i> .
Cotton	52 AADD	5-40	400	wild congeners, <i>Gossypium arboreum</i> , <i>G. herbaceum</i>
Flax	30	3	0	<i>Linum africanum</i> , <i>L. angustifolium</i>
Lettuce	18	1-6	10	<i>Latuca sativa</i> , <i>L. indica (brevirostris)</i> , <i>L. serriola</i> (prickly lettuce), <i>L. virosa</i> , <i>L. saligna</i>
Oat	42	0.5-1 (max : 10)	0	<i>Avena</i> spp., <i>A. fatua</i>
Oilseed rape	38 AACC	>10	40	<i>Brassica napus</i> , <i>B. campestris (=rapa)</i> , <i>B. nigra</i> , <i>B. napella</i> , <i>B. borugeaui</i> , <i>B. cretica</i> , <i>B. montana</i>
Potato	48 AAA'A'			<i>Solanum demissu</i> , <i>S. edinense</i> , <i>S. semidemissum</i> , <i>S. curtilobum</i> , <i>S. sucrense</i> , <i>S. andigena</i> , relatives in the subsection potato
Rice	24	1-2	3	weedy rice and relatives, <i>Oryza rufipogon</i> , <i>O. nivara</i> , <i>O. glaberrima</i> , <i>O. breviculata</i> .
Soybean	40	<1	0	<i>Glycine soya</i>
Tomato	24	<2	30	<i>Lycopersicon</i> spp., tomato-like <i>Solanum</i> spp.
Wheat	42 AABBDD	Variable (max :10)	0	tetraploids, <i>Aegilops squarrosa</i> , <i>A. cylindracea</i> , <i>Secale</i> spp., <i>Hordeum</i> spp., <i>Aegilops</i> spp.

Compiled from various sources: Frankel and Galun (1977), Keeler *et al.* (1996), Kwon (1972), Matsuo (1989) and Smart & Simmonds (1995), Won (1999)

Risks of gene flow between species

Of greater importance is the possibility of herbicide-resistant genes escaping to closely related weedy species. There is a risk that crops that are sexually compatible with wild relatives growing in proximity to them will receive fitness-enhancing transgenes such as herbicide- or insect-resistant genes, which could alter ecological parameters. Some examples are rice, sorghum, sugarbeet and sunflower. Other crops that have compatible relative species are also shown in Table 3.

In the case of crops with a high outcrossing rate, the largest concern in the USA and Europe has been over GM OSR, a crop with numerous wild relatives and increasing worldwide cultivation (Table 1). Significant barriers to the introgression of *Brassica napus* marker genes, as well as the herbicide-resistant genes, into the genomes of *Raphanus raphanistrum* and *Hirscheldia incana* have been found (Darmency *et al.*, 1998). However, one hybrid was obtained from each of the crosses, *B. napus* x *Erucastrum gallicum* and *B. rapa* x *E. gallicum* (Downey, 1999), indicating that the possibility of gene transfer from *B. napus* to *E. gallicum* exists, although the probability of its occurring is very low.

In the case of inbreeding crops, rice and wheat are the most widely cultivated worldwide. It is reported that the 22 species of wild rice are distributed throughout Asia, Australia, Oceania and Latin America. Two wild species, *Oryza rufipogon* and *O. nivara*, are abundant in many parts of Asia and are known to hybridize with *Oryza sativa* under natural conditions (Cohen *et al.*, 1999; Table 3). Numerous types of weedy rice also occur in Asia, derived from *O. sativa*, wild species, and hybrids between wild rices and *O. sativa*. Considering (i) the occurrence and environmental persistence of hybrids between cultivated, weedy and wild rices; (ii) the extensive area of land that may eventually be planted to GM rice; (iii) the large populations of wild and weedy rices in many rice-growing areas, it must be assumed that transgenes will escape to wild and weedy relatives. Recently, two imazamox-resistant hybrids from a cross between *Aegilops cylindrica* and imazamox-resistant wheat were discovered in a research plot. The production of this hybrid and the low level of self-fertility indicate that hybrids could serve as a bridge in the gene transfer between wheat and *A. cylindrica* in the field (Mallory-Smith *et al.*, 1999).

Risks of gene flow between higher plants and other organisms

There is a strong likelihood that transgenes conferring pest resistance will be

transferred from agronomic ecosystems into natural ecosystems, resulting in ecological risks creating more invasive weeds and affecting beneficial insects. Gene flow of the transgene to an infecting virus by recombination could lead to new viral genomes (Teycheney and Tepfer, 1999). Analysis of viral genomic sequence data often shows genetic features that can be best attributed to RNA recombination events between viruses (Revers *et al.*, 1996) or between viruses and plant RNA, indicating that RNA recombination is a normal feature of virus evolution. Thus, the use of viral sequences in transgenic plants immediately raised the questions of whether recombination could occur between viral transgene sequences and the genome of an infecting virus, and whether this could have an impact on virus evolution. Wintermantel and Schoelz (1996) showed that the inoculation of transgenic *Nicotiana bigelovii* plants generated recombinant viruses that had a distinct competitive advantage in *N. bigelovii* when compared with the parental strain, demonstrating that gene flow from GM crops to infecting viruses can occur, although at very low levels and under particular conditions.

5. Conclusion

Continuous use of a single herbicide has been most responsible for herbicide resistance evolution in many cases. There have been more than 70 resistant weed species recorded since 1957 when the first incidence of herbicide resistance to 2,4-D was reported in *Commelina diffusa* and *Daucus carota*. Introduction of a GM crop resistant to a specific herbicide could result in accelerated evolution of herbicide resistance in weeds. Glyphosate use alone will clearly further engender the evolution of glyphosate resistance, and/or bring about a shift in weed spectra towards weeds that have never been controlled by glyphosate (Owen, 1997). For resistance management, Gressel (2000) proposed that one way to delay the evolution of herbicide resistance in weeds is to stack two herbicide resistant genes in a GM crop and to use a mixture of herbicides with different modes of action. This may be useful because it considerably lowers the mutation frequency for resistance in the weed. However, a recent survey has shown widespread introgression of herbicide resistance traits among the various herbicide-resistant volunteer OSR populations in western Canada, resulting in many volunteer OSR populations already containing multiple resistance to all three herbicides on which herbicide-resistant GM OSRs rely (*i.e.*, to acetolactate synthase-inhibiting

herbicides, glufosinate and glyphosate; Hall *et al.*, 2000). A regulatory system cannot prevent the re-introducing triazine-resistant cultivars, and possibly could not prevent the use of 2,4-D resistant cultivars if such were generated using known transgenes, because each case may be considered on its own merits (Gressel, 2000). This soon could become volunteer weed population of OSR in subsequent crops in rotational cycles. In addition, as shown in Tables 1 and 2, some efforts have already been made to introduce multiple traits in a single crop cultivar. If a GM crop cultivar with stacked genes for multiple resistance to herbicides and other pests is released into natural ecosystem and remains there, the GM crop cultivar itself could be a mighty weed that cannot be controlled by any herbicides available in the farmers' arsenal.

In favorable aspect to GM crops, GM technology can contribute to support increasing global populations, particularly of developing countries. This benefit seems to overwhelm potential risks when only considering present statistics, but we cannot conclude that GM crop is safe enough for us not to worry about its risks. Zero risk does not exist in many cases, but our concern is whether the risk is acceptable. So far, long-term consequences have not well been quantified. Gene flow is affected by the various factors mentioned above, so more comprehensive and systematic approaches will support the right decision for GM crops, and indeed whether any particular GM crop cultivar is acceptable. To establish better strategies for minimizing risks arising from GM crops, more studies on weed biology and ecology are essential.

References

- Chen, X. (1999) Agri-biotechnology in Asia-Oceania. In *Proc. of the Confer., Agrobiotechnology: The Financial Future for the Sector*, organized by the Financial Times, London, Oct. 22, 1999.
- Cohen, M.B., Jackson, M.T., Lu, B.R., Morin, S.R., Mortimer, A.M., Pham, J.L. and Wade, L.J. (1999) Predicting the environmental impact of transgenic outcrossing to wild and weedy rices in Asia. In *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Staffordshire. 151-158.
- Colbach, N., Meynard, J.M., Clermont-Dauphin, C. and Messean, A. (1999) GeneSys: a model of the effects of cropping system on gene flow. In *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Staffordshire. 89-94.

- Cowgill, S.E., Coates, D. and Atkinson, H.J. (1999) Non-target effects of proteinase inhibitors expressed in potato as an anti-nematode defence. In *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Staffordshire. 203-208.
- Cummins, J. (1998) Why refuge will not work to reduce Bt resistance. *Genetically Manipulated Food News*. [http://\(jcummins@julian.uwo.ca\)](http://(jcummins@julian.uwo.ca)) Accessed 5 December 2000.
- Daniell, H.(1999) GM crops: public perception and scientific solutions. *Trends in Plant Sci.* 4: 467-469.
- Darmency, H. (1994) The impact of hybrids between genetically modified crop plants and their related species; introgression and weediness. *Molecular Ecology* 3: 31-36.
- Downey, R.K. (1999) Gene flow and rapethe Canadian experience. In *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Staffordshire. 109-116.
- Drobniewski, F. A. (1994) The safety of *Bacillus* species as insect vector control agents. *J. of Appl. Bacteriology* 76: 101-109.
- Gressel, J. (2000) Molecular biology of weed control. *Transgenic Research* 9: 355-382.
- Hall, L. M., Huffman, J. and Topinka, K. (2000) Pollen flow between tolerant canola(*Brassica napus*) is the cause of multiple resistant canola volunteers. *Weed Sci. Soc. of Am. Abstracts* 40: 117.
- James, C. (2000) *Global status of commercialized transgenic crops: 1999*. ISAAA Briefs No.17. ISAAA, Ithaca, New York.
- Jury, L. (1998) UN Aid agencies slam Monsanto's campaign. *London Independent*, July 25, 1998.
- Kwon, Y.W. and D.S. Kim. 2001. Herbicide-resistant genetically-modified crop: its risks with an emphasis on gene flow. *Weed Biology and Management* 1:42-52.
- Liu, Y.B., Tabashnik, B.E., Dennehy, T.J., Patin, A.L. and Bartlett, A.C. (1999) Development time and resistance to Bt crops. *Nature* 400: 519.
- Losey, J.E., Rayor, L.S. and Carter, M.E. (1999) Transgenic pollen harms monarch larvae. *Nature* 399: 214.
- Mallory-Smith, C.A., Snyder, J., Hansen, J.L., Wang, Z. and Zemetra, R.S. (1999) Potential for gene flow between wheat (*Triticum aestivum*) and jointed goatgrass (*Aegilops cylindrical*) in the field. In *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Staffordshire. 165-169.

- Owen, M.D.K. (1997) North American development of herbicide resistant crops. *Proc. of the BCPC-Weeds*: 955-963.
- Pyon J.Y., Kwon Y.W. and Guh J.O. 1998. Distribution, emergence and control of Korean weedy rice. In *Report of Int. Symp. on Wild and Weedy Rices in Agro-ecosystem*. Ho Chi Min city, APWSS and CLRRRI. 58-62.
- Rahardja, U. and Whalon, M. (1995) Inheritance of resistance to *Bacillus thuringiensis*. *J. Econ. Entomology* 88: 21-26.
- Revers, F., Le Gall, O., Candresse, T., Le Romancer, M. and Dunez, J. (1996) Frequent occurrence of recombinant potyvirus isolates. *J. of General Virology* 77: 1953-1956.
- Teycheney, P.Y. and Tepfer, M. (1999) Gene flow from virus-resistant transgenic crops to wild relatives or to infecting viruses. In *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Stafordshire. 191-196.
- Thomas, W. E. and Ellar, D. J. (1983) *Bacillus thuringiensis* var. *israelensis* crystal-endotoxin: effects on insect and mammalian cells *in vitro* and *in vivo*. *J. Cell. Sci.* 60: 181-197.
- USDA-APHIS-PPQ-PRA Biotechnology Authorizations (2000) Permits, Notifications and Determinations of Non-regulated Status(as of 6/30/2000).
- Vigouroux, Y., Darmency, H., de Garambe, T.G. and Richard-Molard, M. (1999) Gene flow between sugar beet and weed beet. In: *Symp. Proc. No. 72. Gene flow and Agriculture, Relevance for Transgenic Crops*. BCPC/Univ. of Keele, Stafordshire. 83-88.
- Wintermantel, W.M. and Schoelz, J.E. (1996) Isolation of recombinant viruses between cauliflower mosaic virus and a viral gene in transgenic plants under conditions of moderate selection pressure. *Virology* 223: 156-164.