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Herbicide Resistant Crops and Impact of their Use

Ministry of Environment and Energy, Denmark
Danish Environmental Protection Agency

Miljø- og Energiministeriet Miljøstyrelsen

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Nr. 363: Herbicide Resistant Crops and Impact of their Use

Herbicide Resistant Crops and Impact of their Use

The result of a 3 month investigation based on literature and available statistics summarizes the state of the art of herbicide resistant crops and impact of their use. Until now only 4 marketing release permits have been given in Europe, which means that Europe has no large scale growing experience. In order to make predictions as precise as possible, environmental impact and changes of cultivation methods have to be evaluated separately for each new release application.

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1997

Herbicide Resistant Crops and Impact of their Use

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Danish Environmental Protection Agency

The Danish Environmental Protection Agency will, when opportunity offers, publish reports and contributions relating to environmental research and development projects financed via the Danish EPA.

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The reports are, however, published because the Danish EPA finds that the studies represent a valuable contribution to the debate on environmental policy in Denmark.

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Forord

Denne rapport er resultatet af et 3 måneders udredningsarbejde. Arbejdet blev udført af videnskabelig assistent Bine Bjerregaard fra den Kongelige Veterinær- og Landbohøjskole, Institut for Jordbrugsvidenskab, Sektion for Ukrudtslære i perioden fra september til og med november 1996. Professor Jens C. Streibig var projektleder. Styregruppen for projektet bestod af Gitte S. Poulsen og Juliane Albjerg fra Miljøstyrelsen, samt Jens C. Streibig og Kathrine H. Madsen fra Den Kongelige Veterinær- og Landbohøjskole. Forfatterne vil gerne benytte lejligheden til at takke Miljøstyrelsen for finansiering af projektet.

Summary

The present report summarizes the state of the art of herbicide resistant crops and impact of their use.
Until now, only four marketing release permits for herbicide resistant plants have been given in Europe, but the numerous release permits for experimental use and the release permits from the USA may give us an idea of which plant/herbicide constructions will be the next to appear on the market.
None of the four herbicide resistant plants with a marketing release permit (Bromoxynil tolerant tobacco, glufosinate tolerant oilseed rape, glyphosate tolerant soybean, partial glufosinate tolerant chicory) are yet commercially available (November 1996). This means that Europe has no large scale growing experience with genetically modified herbicide resistant plants. The few publications that exist are on small scale controlled experiments.
The most discussed subjects concerning the release of herbicide resistant plants are whether their introduction will cause:

- herbicide use to increase or decrease
- problems with herbicide resistant volunteer plants
- problems with herbicide resistant crop/weed hybrids

There does not seem to be clear answers to these questions. Well funded arguments exist both for and against the different potential risks. From the material collected for this report it seems that introduction of genetically modified herbicide resistant plants could cause:

- a decrease in herbicide amounts, compared to present levels, for most crops, if managed correctly
- a shift to herbicides considered less environmentally suspect
- herbicide resistant weeds and volunteer plants though the extent of the problem differs in various crops
- pleiotropic effects

In order to make predictions as precise as possible, environmental impact and changes of cultivation methods have to be evaluated separately for each new release application.

Dansk sammendrag

Denne rapport er en status over herbicidresistente afgrøder og de konsekvenser det vil medføre at introducere dem i landbruget.
Der er indtil nu kun givet fire tilladelser til markedsføring af herbicidresistente afgrøder i Europa, men det store antal tilladelser til forsøgsudsætninger og markedsføringstilladelser fra USA kan give et billede af hvilke herbicid resistente planter, der vil komme på markedet i Europa i den nærmeste fremtid.
Ingen af de fire planter, der har fået marketingstilladelse (Bromoxynil tolerant tobak, glufosinate tolerant raps, glyphosate tolerant soyabønne, delvis glufosinate tolerant julesalat), er i handlen endnu (november 1996). Det betyder, at der i Europa ikke er erfaringer med dyrkning af herbicidresistente afgrøder i stor skala. De få publikationer, der findes, omhandler små kontrollerede forsøg.
De mest omdiskuterede emner vedrørende indførelsen af herbicid-resistente afgrøder er om afgrøderne:

- vil få herbicidforbruget til at stige eller falde
- vil skabe problemer med herbicid-resistente spildplanter
- vil skabe problemer med herbicid-resistente afgrøde/ukrudts hybrider

Der synes ikke at være entydige svar på disse spørgsmål. Der er velfunderede argumenter både for og imod de forskellige potentielle problemer. Det indsamlede materiale peger i retning af, at en introduktion af genetisk modificerede herbicidresistente planter kunne resultere i:

- nedgang i herbicid forbruget, sammenlignet med nuværende niveau, for de fleste afgrøder, hvis de dyrkes korrekt
- et skift til herbicider der regnes for mindre miljøbelastende
- herbicid-resistent ukrudt og spildplanter, selvom problemets omfang er ikke lige stort for alle afgrødetyper
- pleiotropiske effekter

For at gøre forudsigelserne så præcise som muligt må man evaluere indflydelsen på miljøet og ændringer i dyrkningsmønstre separat for hver enkel markedsføringsansøgning.

1 Introduction

In 1995 the European Commission established an ad-hoc group to assess the environmental effects of the use of herbicide resistant plants. One reason was the ongoing discussion whether long term consequences are covered under Directive 90/220/EEC on the deliberate release into the environment of genetically modified organisms or under Directive 91/414/EEC concerning the marketing of plant protection products. The ad-hoc group found that further work was needed to investigate relevant environmental issues.
This report is the result of a cooperation between the Danish Environmental Protection Agency and the Royal Veterinary and Agricultural University, Denmark. The aim is to summarize state of the art of herbicide resistant crops and the impact of their use.

Definitions

In this report we define <u>G</u>enetically <u>M</u>odified **<u>Herbicide resistant</u>** <u>P</u>lants (GMHPs) as plants, that have been genetically engineered to contain a gene, that confer resistance to a herbicide. The GMHPs are only resistant to one herbicide or a group of herbicides having exactly the same mode of action. Unless naturally tolerant the plant shows no resistance to herbicides with other mechanisms of action. Most crops are naturally tolerant to certain herbicide groups without being genetically modified.
The herbicide-resistance in GMHPs can be caused by either metabolic detoxification or by alterations of the target site for the herbicide that prevents the herbicide from binding to specific enzymes.

The terms tolerant and resistant are often used interchangeably. Resistance, to be specific, is a tolerance to very high doses of a herbicide. It could be argued that the term tolerance should be used in all cases, but in order to differentiate between the levels of tolerance the term resistance is often used as tolerance to high doses of herbicide. In this report the term herbicide resistant will refer to plants that have been made insensitive to a herbicide by biotechnology. Non- genetically modified herbicide resistant plants will be referred to as naturally tolerant plants.

The herbicide debate

The first petitions for marketing release of GMHPs have caused a great deal of debate in many European countries. For some people, the debate has been concentrated on the question whether genetically engineered plants in general are safe and ethically acceptable. For others, the focus has been on the potential environmental and health problems caused by herbicides.
In recent years herbicide residues have been found in groundwater, surfacewater, rainwater, air, snow and fog (Müller *et al.* 1996). This has caused a great deal of debate about the cultivation methods used in European agriculture. Some countries specifically aim on a low-pesticide agriculture. In Denmark it has been politically decided that pesticide use should be reduced by 25% from 1985 to 1990 and by another 25% from 1990 to 1997. In Austria 10% of agriculture is independent of synthetic pesticides, and public and politicians are in favor of supporting a development towards more organic farming.
The question is whether GMHPs are a step towards an agricultural practice a particular country aims to obtain.

A multitude of arguments are heard for and against the introduction of GMHPs. Common for all the arguments is that few of them are based on facts.

Contents

This report aims at summarizing some of the most common arguments for and against GMHPs and evaluating the experimental work that has been done to verify or reject the arguments.
The main subjects of the report are:

- status on marketing of GMHPs in EU

  impact on cultivation methods and herbicide use

- risk assessment in connection with volunteers and gene transfer from herbicide resistant crops to related weeds

- other potential problems: Multiple resistance, reduced biodiversity and pleiotropic effects of inserted genes

The report is written as a review based on information retrieval with help from the competent authorities in the member countries and supplemented with information published in international scientific
journals and information published by companies and authorities. The report focuses on the situation in the EU. Since GMHPs are not commercially available in the EU most experiences with GMHPs come from the USA and Canada where GMHPs are already available. The American and Canadian experiences are included in order to try to predict the future situation in the EU.
The reports on risk assessment of GMHPs are few. Development of new GMHPs has been explosive during the last years and long term risk assessment studies have lagged behind. This is partly because ecological experiments are very time consuming. The studies are not always ideal. In the USA most risk assessment studies are performed by the biotechnology companies placing them in a position of conflicting interests. In Europe risk assessment studies are performed both by biotechnology companies and by public scientific institutions. In Germany, there have been difficulties with anti-biotechnology activists who have destroyed test fields, thus making it impossible to perform the much needed risk assessment studies (Parker and Bartsch 1996, Abbott 1996).
The report only deals with genetically modified *herbicide resistant* plants and a general discussion of genetically modified plants will not be covered. Problems unique to countries outside the EU (e.g. insufficient food production, low cost of labour versus herbicides etc.) will not be discussed. Ethical questions, social consequences, economic benefits, etc. will not be discussed. For these subjects see e.g. Comstock 1989, Goldburg 1990, Radosevich *et al.* 1992.
At the end of the report there will be a tentative evaluation of the long-term consequences of the use of GMHPs together with proposals for future work needed to enlighten the potential risks of marketing release of GMHPs.

2 Background

In the EU only four GMHPs (November 1996) have received an approval for marketing release under directive 90/220/EEC (see Table 2.4). A great number of experimental releases (Table 2.3) indicate which plants will most likely be on the market in the near future - provided the companies will obtain marketing release permissions from the authorities. In the USA and Canada GMHPs have been on the market for a few years. The first GMHPs in the USA were glyphosate resistant soybean and bromoxynil resistant cotton that appeared in 1994.
The first herbicide resistant crop to be placed on the market was atrazine resistant canola in Canada that appeared in 1984 (Hall *et al.* 1996). The canola was not genetically modified, but rather the result of repeated backcrossing of a naturally atrazine tolerant *Brassica rapa* to *Brassica napus*. The plants had very limited success because of a reduced photosynthetic activity resulting in yield reductions of up to 20-30% (Mallory-Smith and Eberlein 1996).
Since then the herbicide resistance research has changed significantly. The search is now concentrated on environmentally less malign herbicides, such as glyphosate, glufosinate and the sulfonylurea group - although some bromoxynil GMHPs have also been developed. Side effects such as yield penalties have to be very low or non-existent, because a more effective weed management only rarely compensate for a severe yield penalty, in terms of economic output.

Field release applications in the USA

In the USA, 28.4% of all field release permits of genetically modified plants concerns herbicide resistant plants. This number is followed closely by improved product quality (25.4%) and insect resistance (23.4%). Numbers are calculated over a period from 1987 to 1995. During the same period the most frequent crop to be genetically modified was corn (39%) followed by tomato (15%), soybean (12%), potato (11%) and cotton (9%). The most common companion herbicides in GMHPs are glyphosate and glufosinate (see table 2.1 and 2.2).

Table 2.1
*Release permits for GMHPs in the USA from March to August 1996 (experimental release)**

Glufosinate	6
Glyphosate	3
Sulfonylurea-group	4

*For a full list see appendix table 6.2

Table 2.2
*Petitions for GMHPs in the USA from June 1993 to September 1996 (marketing release)**

Glyphosate	3
Glufosinate	5
Sulfonylurea	1
Bromoxynil	1 (1993)

*For a full list see appendix table 6.3. Eight of these petitions have already been approved, one had been withdrawn and one is still under review by the competent authority.

Field release applications in EU

In the EU, 40% of release applications for genetically modified plants in 1996 were for herbicide-resistance, a much larger percentage than in the USA.
In 16 of the 113 release applications for herbicide resistant plants in 1996 the herbicide-resistance was coupled to insect resistance (Statistics from the Danish Nature and Forest Agency based on circulated summary notification information for release of genetically modified higher plants, SNIFs). In those cases the herbicide-resistance is probably only intended as a marker, used during the development of the GMHPs to select transformed plants. The main trait is the insect resistance.
The most frequent plant to be modified for herbicide-resistance was oilseed rape (39%) followed by maize (35%) and beet (including fodder- and sugar-beet) (19%) (Statistics from the Danish Nature and Forest Agency). The most common companion herbicides in GMHPs are glyphosate and glufosinate (see Table 2.3 and 2.4)

Table 2.3
*Release applications for GMHPs in the EU from August 1995 to September 1996 (experimental release)**

Glufosinate	82 (59%)
Glyphosate	40 (30%)
Oxynils	11 (8%)
Sulfonylurea	1
Asulam	1
Multi resistance	3

*For a full list see appendix table 6.1

Table 2.4
Permissions for marketing release of GMHPs in the EU

Bromoxynil tolerant tobacco
Glufosinate tolerant oilseed rape
Glyphosate tolerant soybean
Partial glufosinate tolerant chicory

(four more applications are being evaluated: two Maize/Glufosinate and two Oilseed rape/Glufosinate)

As mentioned in the introduction only four GMHPs have received a marketing release permit in the EU (Table 2.4). None of these are at present (November 1996) commercially available. A glyphosate resistant sugarbeet from Denmark is expected to be commercially available in 1998 (Axel Jørgensen, Monsanto, Pers. comm.). At the Brighton Crop Protection Conference 1995, G. Marchall gave an estimate on the anticipated commercial availability of GMHPs in Europe based on experimental releases (see Table 2.5).

Table 2.5
Estimate on the anticipated commercial availability of GMHPs in Europe (Modified from Marchall 1995)

Crop	Glufosinate	Glyphosate
Oilseed rape	1997-98	1999-2000
Sugar beet	2001	1998
Wheat	>2000	>2000

2.1 Regulation of the release of genetically modified plants in EU member states

Release of genetically modified plants, including GMHPs, in the EU requires a release permit from the competent authorities. The release petition, in the shape of a technical dossier, is send to the competent authorities in one of the member states. The competent authority will then pass the dossier on to the European Commission, which will distribute it to all member states. The member states will in turn advice and recommend on the case. In the case of *experimental* release permits the country presented with the petition is empowered to make the final judgement concerning the release. In the case of *commercial* releases the final approval must come from the Commission. If the member states, after scrutinizing the scientific basis, disagree, which they often do, the case is decided upon by a qualified majority in the Commission. For a region to reject the decision of the Commission, they must present a detailed scientific explanation supporting their position (Nickson and Fuchs 1994, Waters 1996).
Procedures and attitudes in the different member states vary considerably (Swift Community Risk Evaluation Network, Hill 1995). In France a permit for experimental release is obtainable within a month at minimal cost (the procedure is partially public), in the UK it may take up to 6 month for a permit (the procedure is fully public) and the cost is £2000. In Germany it may take 6 months or more (the procedure is fully public), the cost is 50,000 DM and the general political situation has until recently been hostile to biotechnology (Loyd-Evans and Barfoot 1996). A full overview of the procedures of the member states concerning the release permits is collected in the Heemskerk report (1994) and is also available from ProBio Partners information on Biotechnology from the Swift Community Risk Evaluation Network, via the internet
(http://www.noord.bart.nl/~biotech/screen.html).

In the following an overview is given on the arguments and objections from the member states concerning the commercial release of GMHP.

The Danish competent authorities have objected to the placing on the Community market of all the GMHP's notified so far, due to lack of information on the seed package that the seed is genetically modified. This point of view has also been taken by Austria and Sweden. In case of genetically modified herbicide resistant oil-seed rape, the objections raised by these three member states have in addition been due to uncertainty concerning the consequences for the herbicide use. Oilseed rape is a crop that in itself acts as a weed in other crops and is closely related to wild weeds. Outcrossing of genes between oil seed rape and wild relatives can take place. The expansion of resistance to related weeds - and especially combinations of resistance - will make it more difficult to exclude weeds with a minimum use of herbicides, and may consequently influence the long-term use of herbicides. This argument has also been raised by Italy and Spain. Germany has objected to the placing on the Community market of several GMHP's due to inadequate labelling. Germany finds it important that it is stated on the label that the use of the herbicide on the GMHP is a new indication area and requires registration according to the plant protection regulation. The reason for this labelling is that the use of a particular herbicide on a GMHP may lead to new metabolites, the safety of which must be assessed in connection with the registration (Juliane Albjerg, the Danish Environmental Protection Agency, Pers. Comm.).

3 Potential risks and benefits

Weeds

Weeds have been known, and feared, since man first began to cultivate the land. In this context we define weeds as unwanted plants. Earlier the scope of weed management was a total eradication of the weeds instead of a reduction of the weed infestation to an acceptable level. Today however, the beneficial effects of a low level of weeds has been recognised. The weeds serve as soil cover preventing erosion and as food and shelter for beneficial organisms that help combatting different pests, thereby lowering the need for pesticides (Müller *et al.* 1996).
The yield increase following total weed eradication is often marginal, but todays farmers need the margain profit gained by the higher yields obtained by the use of high levels of low cost herbicides.

Herbicide development

Traditionally, new herbicides were found by screening a multitude of synthetic compounds for their weed killing effect. The scope was to find a substance that would kill most plants and leave the crop unharmed. These substances are called selective herbicides. Other substances kill almost all plants and are called non-selective herbicides, although plants do show different susceptibility towards the substances.
The development of GMHPs has changed the scope of potential herbicide screening. It is now possible to look for an effective plant-killing, environmentally not too harsh substance and later genetically modify particular plants to become resistant to the substance (Burnside 1992, Dekker and Duke 1995).

Experiences from growing GMHPs

Much of our experience with growing herbicide resistant plants come from plants that are naturally tolerant to a selective herbicide or has been altered through traditional breeding methods to be herbicide resistant. Since plants that have been genetically modified to be herbicide resistant are not essentially different from plants that have been altered through traditional breeding, the experience gained from decades of growing naturally herbicide tolerant plants can be used in evaluating the potential problems appearing when growing GMHPs.
Few studies have been made to monitor the impact of GMHPs on methods of cultivation and herbicide use. The few field experiments reported are mostly aimed at showing whether GMHPs survive treatment with high doses of their companion herbicide and whether the yield is unchanged. From these experiences it seems that GMHPs do perform very well after herbicide treatment and that yield levels have not changed (De Greef *et al.* 1989, C. Haldrup, Landskontoret for planteavl, S. Bisgård, Danisco, Pers. Comm., L. Christopherson 1996, Canola Guide 1996).
Variations in habitats and cultivation methods vary considerably from field to field - even when looking at the same crop in the same country. It is therefore difficult to predict the impact a GMHPs will have when introduced in agriculture. In order to make predictions as precise as possible, each GMHP must be evaluated in the management system where it is to be used, even when discussing the same gene/crop (Gressel 1996)

3.1 Impacts on cultivation methods and herbicide use

One of the most common arguments used both by proponents of GMHPs and by those in opposition is the impact on the amount of herbicides used in agriculture. The proponents claim that the introduction of GMHPs will lead to a decreased amount of herbicide necessary for weed management together with a shift towards environmentally less malign herbicides. Those in opposition argue that the introduction of GMHPs will lead to an increased dependence on herbicides and to an increase in the amount of herbicide used (Beaumont 1993). They also argue that all herbicides are environmentally suspect and that it is therefore irrelevant to speak about more or less malign herbicides (Goldburg *et al.* 1990).

3.1.1 Quantity of herbicides

In the developed countries herbicides are used in virtually all major agronomic crops (Duke 1996). Development of GMHPs would therefore have no significant impact on the percentage of cropland treated with herbicides. They can, however, have a great impact on the amount of herbicide used on the treated area.

Low dose herbicides

A shift to new efficient low dose herbicides like sulfonyurea would mean a decrease in the volume of herbicide used, but not in the "plant killing effect" (Goldburg *et al.* 1990). Provided the non-target toxicity and soil mobility of these herbicides is the same or even less than that of traditional herbicides this would be a desirable shift considering problems like groundwater contamination. The shift would be of no consequence to non-target plants though, meaning that side effects such as reduced biodiversity would not decrease. The shift would also be of no consequence to the rate of development of herbicide resistant weeds (Darmency 1996).

Pre-emergence/post-emergence herbicides

In many crops, pre-emergence herbicides are routinely applied before the severity of the weed problem is known. This is due to a lack of effective post-emergence herbicides that are safe for the crops. Genetically engineered crops resistant to effective post-emergence herbicides (most GMHPs are resistant to postemergence herbicides) would allow the farmer to postpone herbicide spraying until he knows how serious the problem is and to adjust the amount of herbicide in response to the weed problem (Darmency 1996). Whether the farmers will exploit this possibility is another question. Baumont (1993) argues that pre-emergence and post-emergence herbicides control different kinds of weeds. This will make it impossible to substitute one for the other. He also argues that farmers are adverse to taking risks with their crops and will therefore continue the use of pre-emergence herbicides to obtain margain profits.

Others state that the availability of post-emergence herbicides could encourage the farmer to try alternative, cultural or mechanical, weed management methods, because the existence of an effective post-emergence herbicide would serve as an insurance if the alternative methods should fail (Dunwell and Paul 1990, Duke *et al.* 1991, Burnside 1992, Wyse 1992, Burnside 1996).

More effective herbicides

Many of the herbicides used with GMHPs are effective non-selective herbicides. The use of GMHPs resistant to these herbicides may cause a shift from a mixture of herbicides with less range to the use of a single herbicide effective against a much broader spectrum of weeds. This is likely to cause a decrease in herbicide use (Dunwell and Paul 1990).

Excess spraying due to increased tolerance

In some crops spray drift or carry over of herbicides from another crop may damage a less tolerant crop considerably. This is a severe problem in soybean crops growing near glyphosate treated fields. Such crop damages encourage the farmer to use less herbicide than he would have prefered to use for a more efficient weed control. The development of GMHP varieties of the susceptible crop, in this case soybean, would allow the farmer to use elevated levels of herbicides in other crops without fearing the drift or carry over of herbicide to the former susceptible crop, since it would now tolerate the herbicide. *E.g.* in a corn/soybean rotation the potential availability of an atrazine resistant soybean would allow the farmer to use three times the usual amount of atrazine in the preceeding corn crop without fearing damages to the soybean crop. This would result in an increase of herbicide use (Boumont 1993).

Likewise the level of susceptibility of non-transgenic crops to a selective herbicide, present the farmer with an upper limit of herbicide to be used in a particular crop. Crop plants, genetically engineered to resist high rates of herbicide, will no longer naturally keep herbicide levels low, but in contrary encourage the farmer to increase herbicide levels (Harrison 1992, Darmency 1996, Goldburg and Hopkins 1996). The cost of the herbicide will then be the limiting factor.

If GMHPs are developed with resistance to an inexpensive herbicide the farmer will be tempted to use excess amounts of the herbicide in order to manage weeds more efficiently. One of the advantages often mentioned about glyphosate is that it is relatively inexpensive (Delannay *et al.* 1995).

New herbicides for susceptible crops

Some low-acreage crops (e.g. lettuce) are susceptible to almost all herbicides and it has consequently been impossible to use herbicides in these crops. Generation of herbicide resistant variaties of these crops will make it possible to use herbicides where no herbicides were used before (Harrison 1992, Burnside 1992). This will of course increase the amount of herbicide used. Since genetically modifying a plant, obtaining the marketing permission and registering the herbicide for use in the crop is very costly, it is not likely that many low-acreage crops will be genetically engineered to contain herbicide-resistance in the near future. This will probably change when transformation procedures have been standardized to a level where it will be economically profitable to transform low-acreage crops (Ruckenbauer 1996).

Development of alternative methods

The existence of highly effective seed/herbicide packages that will appear with the commercialisation of GMHPs could make it less attractive to exploit the possibilities for new low dose herbicides and alternatives to the use of herbicides in weed management (Harrison 1992). This would not affect the amount of herbicide used directly, but it would slow the path towards low pesticide level agriculture.

Most predictions of changes in herbicide levels after introduction of GMHPs are compared with the present level of herbicide use. Many European contries try to lower the amount of herbicides used in agriculture. If GMHPs were not introduced, alternative methods for decrease of herbicide levels would certainly be an object of intense investigation, resulting in a decreased herbicide use. It could therefore be misleading to compare the level of herbicide use after the introduction of GMHPs with todays levels, since the amount would probably decrease with the application of alternative cultivation methods.

An estimate of herbicide use

Madsen (Madsen and Streibig 1994, Madsen 1994) gave an estimate of future herbicide use in different Danish crops (winter/spring oilseed rape, winter/spring cereals, potatoes, beets, peas and maize) if GMHPs should become widely used. She found that the amount of herbicide needed to control normal weed infestations in most of the examined crop rotations would decrease compared to the present levels of herbicide use. One exception was oilseed rape (both winter and spring varieties) where herbicide levels did not seem to decrease considerably. Most of these estimates were made on an entirely hypothetical basis and aspects such as cost of herbicide, temptations to overuse, problems with herbicide resistant weeds etc. were not included. In a study from Monsanto Europe (Anonymous 1996) it was shown that cultivation of glyphosate resistant soybeans would decrease herbicide use from an average of 1.13 kg ai/ha of traditional herbicides to 0.74 kg ai/ha of glyphosate.

3.1.2 Environmental impact of herbicides

In the debate about herbicide resistant plants it is often claimed by the proponents that herbicide resistant crops will bring a shift in herbicide use towards environmentally less malign herbicides (Dunwell and Paul 1990, Shaner 1993, Burnside 1992, Sten Bisgård Pers. comm.). It is true that most research for GMHPs is focused on herbicides considered environmentally less suspect, but some research is also being done with herbicides considered environmentally malign (Goldburg *et al.* 1990, Goldburg and Hopkins 1996). In Europe approximately 90% of experimental release applications concerning GMHPs were for plant/herbicide combinations involving glyphosate or glufosinate which are considered environmentally less suspect.

Whether a herbicide is more or less environmentally suspect is difficult to establish. Many, contrasting, parameters have to be examined and the parameters are often impossible to compare (Madsen and Streibig 1994). Proponents of herbicides like to use words as "environmentally benign" and "safe" about certain herbicides. Harrison (1992) even puts it this way "By developing tolerance to *nontoxic, nonpolluting* herbicides...". Such herbicides do not exist, because they have to be toxic at least to target-plants. It is therefore important to make one thing clear: statements about the safety and environmental impact of herbicides are always made after a cost-benefit analysis of which risks we are willing to run to get the benefits from the herbicides. No herbicide is without side effects, but we are willing to run a certain risk in order to get a higher output from our agriculture. Whether the risk we are running is worth the benefits is not within the scope of this review to discuss, but it is an important aspects in the discussion of GMHPs. Many herbicides affect biochemical pathways that exist only in plants e.g. sulfonylureas, imidazolines and triazolines that inhibits the acetolate synthase enzyme and atrazine and bromoxynil that are photosynthetic inhibitors (Dekker and Duke 1995). This has been used as an argument for the safety of the herbicides (Comstock 1989), but the specialised phytotoxic effects do not necessarily mean that the herbicides are not toxic to insects, fungi, bacterias and mammals. Examining the LD_{50} (the lethal dose of oral intake for rats) it is seen that the photosynthetic inhibitor bromoxynil is one of the more toxic herbicides, interfering with the oxidative phosphorylation in the mitochondrias. For an overview of the health and environmental impacts of herbicides see Madsen and Streibig (1994). Another factor to consider is that the toxic effect of a herbicide can be modified in the joint action of herbicide mixtures (Müller *et al.* 1996).

Before it is stated whether a herbicide is safe enough for release, risk assessments have to be performed both for the pure active ingredient, for breakdown products and for the commercial product. Commercial formulations of a herbicide may contain surfactants which have a higher toxicity than the active ingredient. Mixtures of herbicides used together in the field also have to be analysed since chemicals acting in concert may have effects that do not exist in either of the chemicals alone (Goldburg *et al.* 1990).

3.1.3 Conclusion

There is no unambiguous evidence to support neither the argument that herbicide use will increase nor the opposite. Field experiments tend to show that, if managed correctly, GMHPs are likely to bring about a decrease in herbicide use in most crops, compared to levels applied today. The herbicide use will have to be monitored in the years following introduction of GMHPs in order to establish in which direction GMHPs influences the overall herbicide use. Introduction of GMHPs supports an agricultural practice that is strongly dependent of herbicides.

Most GMHPs are resistant to herbicides considered less environmentally suspect than herbicides traditionally used in those crops. The problem is to evaluate the relative toxicity of a herbicide. If GMHPs resistant to environmentally suspect herbicides are avoided and if GMHPs resistant to environmentally less suspect herbicides are used according to prescriptions from seed and herbicide companies, there will probably be a shift towards herbicides that are currrently considered less environmentally suspect.

3.2 Risk assessment in connection with volunteers and gene transfer from GMHPs to related weeds

There has been a lot of concern about the possibility that GMHPs may increase the problem of herbicide resistant weeds (Harrison 1992, Keeler *et al.* 1996). The GMHPs could themselves become weeds, if they survive as volunteers in the following crop, resistance genes could be transferred through cross pollination to related weed species and the selection of naturally herbicide tolerant weeds could be intensified by continuous use of one herbicide (Dale and Kinderlerer 1995, Keeler *et al.* 1996).

In some GMHPs, the herbicide-resistance has only been added as a selective marker during the transformation process. The main trait of these plants could be e.g. insect resistance or improved product quality. The plants are therefore intended to be grown without their companion herbicide. The use of GMHPs without the companion herbicide may involve a risk of resistance gene transfer to wild weedy species if weeds are not effectively removed by other means.

The concern is not so much the gene-transfer in itself, but rather the herbicide resistant volunteer plants and weeds becoming invasive, like some introduced exotic species.

3.2.1 Development of herbicide resistant weeds

Herbicide resistant weeds are not a new problem. Triazine resistant weeds were first found in 1970 (Bandeen *et al.* 1982). Since then over 100 herbicide resistant weed species have been found, each resistant to one or more of 14 different herbicide classes (Holt 1996, Shaner 1996).
Herbicide resistant weeds can be generated in two ways: by gene transfer from herbicide resistant crops or by selection for naturally tolerant weed biotypes.

Selection of herbicide resistant weeds

The selection for herbicide resistant weed biotypes is already a problem in most crops sprayed with selective herbicides and the problem is still increasing. A widespread use of crops resistant to herbicides with the same mode of action is likely to accelerate the development of resistant weed biotypes because of increased selection pressure (Darmency 1996). On the other hand it could be argued that GMHPs could be used in the management of herbicide resistant weeds by expanding the number of herbicides with different modes of action available in a particular crop. In this way the use of GMHPs would help decrease the selection pressure on the weeds (Shaner 1996)

Transfer of resistance genes to weeds

The mechanism of herbicide-resistance found in the weeds has in most cases been shown to be different from the mechanism in the crop plants (see Table 3.1). This indicates that the resistance has not been transferred from the crop but rather has been present at low frequencies or has been caused by mutations in the genome of the weeds. The few naturally herbicide tolerant plants in the weed population have then been selected by their higher fitness during herbicide application and have finally given rise to a whole population of herbicide resistant weeds (Sherman *et al.* 1996, Duke 1996). Some scientists use this as an argument that gene transfer from genetically modified crops to weeds is not likely to become problem.
One of the most important matters to consider when discussing the spread of genetic traits from crop plants to weeds is whether the crop plants have close relatives amongst the weeds growing nearby.
If there are no such relatives, then a spread of genes is very unlikely. If there are such relatives the risk of gene transfer depends on how closely related the two species are and how their biology match in other relations (flowering time, hight of the plant etc.). It is therefore of little use to discuss the problem of gene transfer as a general
problem. We need to perform risk assessment studies for every single crop-weed pair in order to be able to predict the possible impact of the GMHPs.
Some of the crops grown in Europe do not present a gene transfer problem, since there are no wild relatives growing in Europe, except for wild growing volunteer plants. This is true for e.g. maize, soybean and tobacco. At the other extreme we find e.g. oilseed rape that readily hybridizes with *Brassica campestris*, a common weed in oilseed rape fields (Dyer 1993, Shaner 1993, Jørgensen and Andersen 1994, Keeler 1996). In Switzerland a risk assessment study has been made concerning the most common crop plants in Switzerland placing each plant in a risk category (Ammann *et al.* 1996).

Table 3.1
Mechanisms of naturally occurring crop resistance, GMHPs resistance and evolved weed resistance (from Dyer et al. 1993).

Herbicide class	**Naturally occurring crop resistance**	**GMHPs resistance**	**Weed resistance**
Aryloxy-phenoxy-propiontes	Active site[1]	Active site	Metabolism, active site and perhaps sequestration
Bromoxynil	Metabolism	Metabolism	Active site
Cyclohexane-diones	Active site	Active site	Metabolism and active site
Glyphosate	None	Active site Metabolism	None reported
Glufosinate	Little or none	Metabolism	None reported
Imidazoli-nones	Metabolism	Active site	Active site
Sulphonyl-ureas	Metabolism	Active site	Active site
Triazines	Metabolism	Active site	Primarily active site
2,4-D	Active site	Metabolism	Metabolism

[1] Except in wheat, in which case it is resistant by metabolic degradation.

Because of the difference between the crops to consider, we have chosen to address the problems of the GMHPs that have been released for experimental purposeses in Europe during 1996, taking each crop at a time. We have chosen not to include experiments with cauliflower, wheat and poplar since the experiments were few.

Oilseed rape

Oilseed rape, *Brassica napus*, is insect pollinated and the large amounts of pollen contributes to both cross- and self-pollination. Pollen has been found at least 2 km from large sources and can probably be found even farther away. *Brassica napus* hybridizes with *B. campestris, B. napus* ssp. *rapifera, B. napella* and *B. juncea* (Poulsen and Højland 1994, Harding and Harris 1994).
The transfer of genes from oilseed rape to the common weed *Brassica campestris* has been extensively studied. Most studies have concluded, that gene transfer between the two species is possible and that it does happen in nature (Kerlan *et al.* 1992, Jørgensen and Andersen 1994, Harding and Harris 1994). This is of no surprise though since oilseed rape is a hybrid between *Brassica campestris* and *Brassica oleracea* (Høst 1982).
In a study of the natural spontaneous hybridization between oilseed rape, *Brassica napus* L., and *Brassica campestris* ssp. *campestris* L., Jørgensen and Andersen (1994) found that if the two species were mixed 1:1 *B. campestris* produced 13% hybrid seeds. If *B. campestris* was grown widely spaced within fields of oilseed rape, *B. campestris* produced up to 93% hybrid seeds and analysis of a weedy population of *B. campestris* in oilseed rape revealed 60% hybrid seeds.

These results show that transgenes could be dispersed from oilseed rapeseed to *B. campestris* by natural hybridization.

Beet

Beet, *Beta vulgaris*, is wind and insect pollinated and is most often self-incompatible. Wind borne beet pollen can disperse up to 8 km. *Beta vulgaris* subspecies are relatively interfertile. In Northern Europe the commercially grown beet species have one wild relative, the sea beet, *Beta vulgaris* ssp. *maritima*, that grows in costal areas. Cultivated beet and sea beet can hybridize and give rise to *Beta vulgaris vulgaris* x *Beta vulgaris maritima* hybrids (Harding and Harris 1994, Højland and Pedersen 1994).
Beet can also hybridize with *B. atriplicifolia and B. macrocarpa* found in the Mediterranian area. In California hybrids between *B. vulgaris* and *B. macrocarpa* cause weed problems in sugar beet fields (Madsen 1994).
Besides this beet can hybridize with wild weed beet, which is a modified annual variety of the cultivated beet that is often growing in beet fields.
Bartsch and Pohl-Orf (1996) found that cultivated transgenic beet and wild beets readily hybridized and that some of the hybrids showed herbicide-resistance.
Since seabeet rarely occurs on arable land, gene transfer to sea beet is not considered a problem. The potential weed problem caused by *B. vulgaris* x *B. macrocarpa* hybrids has to be studied more in detail.
Gene-transfer from transgenic beet to weed beets is a potential problem.

Maize

Maize, *Zea mays*, is a wind pollinated grass native to Central and South America. Maize pollen is transported by wind at least 500 meters. Maize can cross hybridize with other cultivated maize varieties, but since there are no wild relatives in Europe the transfer of genes to wild weedy species is not likely to happen under European conditions (Harding and Harris 1994).

Soybean

Soybean, *Glycine max*, is native to china. It has no close wild relatives in Europe (Keeler *et al.* 1996).

Tobacco

Tobacco, *Nicotiana tabacum*, has no close wild relatives in Europe (Keeler *et al.* 1996).

Chicory

Chicory, *Cichcorium intybus*, is mainly insect pollinated, but 10-20% of the plants are self-compatible. Most varieties require vernalization in order to flower. *C. intybus* hybridizes with wild growing chicory and with the relative *C. endivia*, but since *C. intybus* is harvested while still vegetative, very few individuals flower and are able to hybridize. In Europe *C. endivia* is found in the Mediterranean countries, Bulgaria and the Netherlands (Pedersen and Højland 1994).

Sunflower

Sunflower, *Helianthus annuus*, is native to North America. There are no wild relatives in Europe. Sunflower can cross hybridize with other cultivated sunflower varieties (Keeler *et al.* 1996)

3.2.2 Methods to prevent the development of herbicide resistant weeds.

In order to prevent the development of herbicide resistant weeds by *selection*, the farmer should (Madsen and Streibig 1994):

1) Refrain from using the same herbicide repeatedly on the same area
2) Use herbicides with different modes of action
3) Use herbicide mixtures
4) Use mechanical weed management methods
5) Lower herbicide doses

Methods for reducing the risk of resistance-gene *transfer* from GMHPs to wild weedy species include the incorporation of a suicide gene into the GMHPs, that is triggered by cold or by the very hybridization process. The herbicide-resistance genes could be placed on organellar DNA that is not transferred with pollen (Darmency 1996). The risk of herbicide-resistance transfer is also reduced if the trait is caused by several independently inserted genes instead of one easily transfered dominant gene (Darmency 1996).

If a problem with herbicide resistant weeds develop it has probably "come to stay". A switch to a different kind of herbicide will only help as long as the new herbicide is used. If the farmer should return to the herbicide that the weeds were resistant to, he will discover that there are still herbicide resistant plants left. This is due to the seed bank in the soil. If 30% of a weed population is herbicide resistant, approximately 30% of the seeds in the soil seed bank will also be herbicide resistant. Provided there are no fitness differences between resistant and sensitive biotypes, the proportion of the two will remain the same during the years of tratment with an alternative herbicide. If this spraying stops there will still be 30% herbicide resistant weeds. So the problem has not been solved, only delayed. In this way effective herbicides could be made useless.

3.2.3 Enhanced fitness of weeds with herbicide resistance

Once stated that the transfer of herbicide-resistance genes to weeds is possible and probably also likely to happen for some crop plants the question is whether this constitutes a problem. The gene transfer in itself is not a problem unless one is concerned with the "contamination" of wild gene pools. The problem arises if the transferred genes give the weeds a selective advantage over non-transformed relatives. Crop plants that are genetically manipulated to show other traits than herbicide-resistance e.g. resistance to fungi or insects, may transfer genes to weeds that lead to higher fitness of the weed (Harding and Harris 1994). Herbicide-resistance though is dependent on the presence of the herbicide. The general opinion is that herbicide-resistance will not give weeds a higher fitness in habitats not treated with herbicides. Target site triazine resistance even confers lower fitness (Holt 1996, Gliddon 1994). According to Keeler *et al.* (1996), a lower fitness in herbicide resistant wild relatives of commercial plant varieties could also be a problem in the long run. A lower fitness could lead to the extinction of the plant species. This is a problem since many wild relatives serve as gene pools to improve the commercially grown species.

Few experiments have been performed on enhanced fitness of GMHPs and herbicide resistant weeds. One study on transgenic oilseed rape showed no selective advantage or disadvantage of transgenic plants to non-transgenic plants (Poulsen and Jensen 1995). A comparation between the fitness of seabeet, transgenic sugarbeet and their hybrids revealed no differences in fitness (Madsen 1994). Studies of the multiresistant weeds *Alopecurus myosurroides* and *Lolium rigidum* showed no differential fitness for resistant biotypes compared to sensitive biotypes (Hall *et al.* 1994).
In fields and other areas that are routinely treated with herbicides (roadsides, golf courses etc.) the problem with herbicide resistant weeds can be serious because the herbicide resistant weeds have an advantage compared to the non-modified flora of the area.

3.2.4 Effects of herbicide resistant weeds on herbicide use

The generation of herbicide resistant wild weedy plants by escape of herbicide-resistance genes or by selection present a real threat to the efficacy of herbicides as a weed control option.
Especially the appearence of weeds resistant to widely used non-selective herbicides such as glyphosate and glufosinate will leave farmers and others with a weed management problem such as loss of non-selective herbicides to "clean" fields and other areas of problematic weeds. A consequence could be a return to the application of a mixture of old environmentally harsh herbicides, provided these are still on the market (Keeler *et al.* 1996, Dyer 1993, Shaner 1992, Holt 1996, Jensen 1993).

3.2.5 Conclusion

The transfer of herbicide-resistance genes to wild weedy species is a potential problem in oilseed rape and beet. It could also be a problem in plants that are not mentioned in this report. The hybridization of GMHPs with unmodified crops of the same species growing nearby is possible for all outcrossing species that flower before harvest or are able to survive and flower the next year as volunteer plants.
There is no evidence from invasivenes and yield studies that GMHPs and herbicide resistant weeds possess an increased fitness. The risk of these plants becoming invasive is therefore small. This, however, can vary significantly between the different GMHPs, even with the same herbicide/plant combination and has to be evaluated separately from case to case.
The herbicide resistant weeds could become a major problem when several crop species on the market have been engineered to contain the same herbicide-resistance. Continued use of the same herbicide for several years will increase the selection for herbicide resistant weeds and as a consequence make weed management with the herbicide in question impossible. This could force the farmer to return to the use of older herbicide mixtures and thereby loose the benefits of GMHPs on herbicide levels and toxicity.
On non-agricultural areas the spread of weeds resistant to glyphosate and glufosinate could cause a shift from the use of non-selective herbicides to more environmentally suspect herbicide mixtures.

3.3 Risk assessment in connection with volunteer plants

A volunteer plant is a plant emerging from buried seeds or plant parts from the previous crop. Some plants are ready volunteers giving the farmer a hard time trying to get rid of them. In other crops the problem does not exist at all. Normally, crop plants that are not native to a region will seldom survive without the intensive care of the farmer. This does not always apply, considering the problems created by volunteer potatoes in the UK (Harding and Harris 1994, Dyer 1993, Shaner 1992).

Herbicide resistant volunteer plants could be a severe problem particularly in rotations between two or more crops genetically engineered to possess the same herbicide-resistance. Herbicide resistant volunteer plants would in this case be impossible to control with the companion herbicide of the GMHP. Other types of herbicides would need to be mixed with the companion herbicide of the GMHP resulting in an increased herbicide use (Darmency 1996).

As in the case of gene transfer between crop plants and weeds the problem of herbicide resistant volunteer plants has to be evaluated from case to case. We have examined the GMHPs released for experimental purposes in 1996. Before 1996 there have also been releases involving cauliflower, wheat and poplar.

Oilseed rape

Oilseed rape, *Brassica napus*, is a ready volunteer plant. It sheds large amounts of seeds at harvest, as many as 10,000 seeds per square meter, that remains viable for a long time in the seed bank.. Volunteer oilseed rape is found in as much as 23% of winter cereal fields. Volunteer oilseed rape is easily controlled in cereal crops, but in crops like peas, bean and linseed it is a severe problem because it can not be controlled by selective herbicides (Harding and Harris 1994).

Beet

Beet, *Beta vulgaris*, is a biannual species that is normally harvested after the first year - except when it is grown for seed production. Most beets flower and sets seed the second year, but some individuals, known as bolters flower the first year and leave seeds for a long-lived seed bank that serves as a source of volunteer plants in the following crops. Pieces of the beet roots left in the field is another source of volunteer plants in milder climates.

In the beginning of the 70's a different kind of beet, known as the weed beet, emerged in Europe. The weed beet is annual and requires no vernalisation. Weed beet can be a serious weed problem in beet crops (Højland and Pedersen 1994, Harding and Harris 1994).

Maize

Maize, *Zea mays*, is not likely to become a weed problem in Europe. In northern Europe it is unlikely that maize will be able to set viable seed. Maize is considered a safe transgenic crop under European conditions as far as weedines is concerned (Hardinger and Harris 1994)

Soybean

Soybean, *Glycine max*, is not weedy in Europe (Keeler *et al.* 1996).

Tobacco

Tobacco, *Nicotiana tabacum*, is not weedy in Europe (Keeler *et al.* 1996).

Chicory

Chicory, *Cichorium intybus*, is mostly reproduced by seed, but can propagate asexually by fragmentation of the taproot. Since chicory is harvested before flowering it will not leave a seed bank that could give problems in the next crop. Roots left in the soil could give rise to volunteer plants. Chicory is a common wild plant throughout the temperate world (Pedersen and Højland 1994).

Sunflower

Sunflower, *Helianthus annuus*, is not weedy in Europe (Keeler *et al.* 1996).

3.3.1 Conclusion

Among the crops mentioned in this report, problems with volunteer plants are most likely to appear after growing oilseed rape and beet. Chicory could produce volunteer plants, but it is not reported as a problem.

Since herbicide-resistance genes do not seem to provide the plants with an increased fitness in areas not treated with herbicides the volunteer problems can be expected to be the same as the present. New problems are likely to arise when many crop species on the market are resistant to the same herbicides. This could make the management of volunteer plants difficult and could result in a return to old herbicide mixtures.

3.4 Other potential problems

3.4.1 Multiple resistance

The existence of crop plants resistant to many different types of herbicides presents us with the potential problem of multiple resistance, in both crops and weeds. Multiple herbicide-resistance has already evolved in some populations of *Alopecurus myosurroides* in Europe and is a big problem in *Lolium rigidum* in Australia. Multiple herbicide-resistance in weeds and volunteer plants reduces the weed management options for the farmer and in worst cases leaves no herbicide for chemical management of the troublesome weeds. In Australia this has led to increased use of integrated weed management (Powles *et al.* 1996).

GMHPs could further increase the problem with multiple resistance. The use of new herbicides in a given crop rotation would present the weeds with an additional selection pressure and probably confer resistance to these new herbicides too.

For volunteer plants, the problem of multiple resistance could arise if varieties of the same crop species with resistance to different herbicides were grown adjacent to each other. Cross hybridization of the two species would produce multi-resistant volunteer plants.

The ultimate consequence of multiple herbicide-resistance would be a total loss of chemical weed management options in certain crops.

Adding to the concern is the fact that the companion herbicides of GMHPs, are often non-selective herbicides that were previously used when all weeds had to be controlled in fields or other areas. The development of weeds and volunteer plants resistant to more of the non-selective herbicides would result in a switch to herbicide mixtures to replace the non-selective herbicides. This would probably increase the overall herbicide use.

3.4.2 Reduced biodiversity

One consequence of the introduction of plants resistant to non-selective herbicides could be a reduced biodiversity on the arable land. The non-selective herbicides would more efficiently remove all weeds from the fields. The reduced weed diversity could lead to a reduced biodiversity because the feed and shelter of insects, fungi and bacteria would disappear. The reduced biodiversity could result in a disturbance of symbiotic relations as e.g. mycorrhiza and a decrease in beneficial organisms that help combating pests. This would increase the use of pesticides other than herbicides (Müller *et al.* 1996). Proponents of GMHPs claim that the shift from pre-emergence to post-emergence herbicides will conserve the habitats of beneficial organisms for a longer period. Others argue that this is unlikely to be of any major importance since glyphosate and glufosinate are most effective on weeds at their early stages of development (Müller *et al.* 1996).

Most farmers see the reduced weed diversity as the very scope of weed management. Others have recognised the beneficial effects of low weed levels. The weeds serve as soil cover to prevent erosion and as food and shelter for beneficial organisms (Müller *et al.* 1996, Ruckenbauer 1996).

The decreased complexity of the agroecosystems would provide a perfect environment for the massive propagation of the few weeds surviving the herbicide treatment (Burnside 1992, Radosevich 1992).

The agroecosystem is an important habitat for wildlife because it provides food and shelter. A severely reduced biodivercity of the agroecosystems could endanger threatened species (Gerber and Young 1993).

3.4.3 Pleiotropic effects of inserted genes

A pleiotropic effect is an unforeseen effect brought about by the inserted gene i.e. an unintended side effect. This could be a yield penalty, a higher content of a toxic substance etc.

Herbicide-resistance can be achieved by different means:

1) altered target site
2) over expression of target enzyme
3) detoxification of herbicide

Each of the resistance mechanisms could lead to different pleiotropic effects. An altered target site could result in altered pathway regulation if the target site is part of a pathway and to reduced fitness due to the altered pathway regulation. Over expression of target enzyme could result in side effects from the altered protein levels. Detoxification which is a result of herbicide degradation could produce degradation products with unwanted characteristics (Dyer *et al.* 1993).

The most extensively studied pleiotropic effect has been yield penalty. Atrazine resistance is normally due to an altered target site that prevents binding of the herbicide. Unfortunately, the alteration also results in a reduced photosynthetic productivity with yield penalties up to 20-30% (Mallory-Smith and Eberlein 1996). There have also been reports of yield penalties in bromoxynil resistant potatoes.

In tomatoes a reduced and delayed flowering was reported in glyphosate resistant cultivars. Altered flowering characteristics could affect the interaction of the GMHPs with wild relatives and result in decreased or increased gene transfer between the species.

Glufosinate resistant carrots were reported to accumulate degradation products. The possible toxic effects of such products need to be examined before commercialization of the GMHPs (Dyer *et al.* 1993).

Allergenic substances could be transferred from the doner organism and/or could be increased in the receiving organism (Müller *et al.* 1996).

All transgenic plants could have their biochemistry altered in an unpredictable way that could result in accumulation of toxic substances. In one case, a genetically modified potato was found to be blight resistant due to high levels of the toxic compound solanine (Dyer *et al.* 1993). The potato variety was rapidly removed from the market.
One of the problems about pleiotropic effects is that they might only turn up in certain environmental conditions or with certain background genotypes (Gressel 1996). This makes it very difficult to assess the potential risks.

3.4.4 Conclusion

Multiple resistance is likely to develop following the increased use of new herbicides in connection with GMHPs. As is already the case in areas with multiple resistant weeds this could lead to a loss of chemical weed management options and force the farmers to use alternative non-chemical methods. The effective non-selective herbicides that are used with most GMHPs will probably result in a decreased biodiversity, not only of weeds, but possibly also of non-target organisms. Since some of these organisms are beneficial to the crop plants by combatting pests, this could result in an increased use of pesticides other than herbicides.
The most extensively studied pleiotropic effect of the herbicide-resistance genes is yield reduction. Other effects such as alteration of phenotypic characteristics have also been reported. For herbicide resistant plants the accumulation of toxic compounds have not yet been reported. More research is needed to evaluate the potential problems caused by pleiotropic effects.

4 Discussion

In this report we have outlined a number of potential problems and benefits of the introduction of GMHPs in agriculture. It is not possible to make a prediction that turns out to hold. Keeler *et al.* (1996) put it this way:

> "To predict the environmental impact of transgenic crop requires a number of assumptions none of which apply. These assumptions include:
>
> 1. the plant will always be responsibly handled
> 2. the plants will not evolve
> 3. land use patterns, e.g. agricultural and pasturage systems, will not change significantly
> 4. human uses of plants will remain the same
> 5. human evaluation of the worth of different species will remain stable
> 6. the distribution of people and the way they use the land (e.g. urban/rural) will be stable

We have chosen to make these assumptions anyway and tried to give a prediction of the impact of GMHPs on environment and agricultural practice.

Summary of conclusions

Arguments for and against the different potential problems have been closely studied in the report. The following is a summary of the conclusions presented in the text:

1) GMHPs will probably result in a decrease in herbicide use IF MANAGED ACCORDING TO APPROVED OR RECOMMENDED USE

2) GMHPs will probably lead to a partial shift to herbicides currently concdered less environmentally suspect

3) GMHPs do not support a development towards decreased dependence of herbicides

4) transfer of herbicide-resistance genes to weed species is possible for outcrossing species with close weedy relatives

5) GMHP volunteer plants can not be controlled with the GMHP companion herbicide. This reduces the volunteer plant management options and increases problems with plants that are already today problematic volunteers

6) most GMHPs do not seem to have an altered fitness outside arable land and are therefore not likely to become more invasive than untransformed plants, however, this needs further investigation.

7) multiple resistance will most likely appear in both weeds and volunteer plants and could become a weed management problem

8) introduction of GMHPs could cause a reduced biodiversity

9) there could be pleiotropic effects of the herbicide-resistance genes in GMHPs

Goals for the development of agricultural practice

Evaluation of the potential problems in connection with the introduction of GMHPs is strongly dependent on the goals a country has for the development in agricultural practice and technology (van den Daele 1995). If the goal is to increase the percentage of organic farming, as in Austria, GMHPs will be easily rejected because the genetic manipulation of the plants is incompatible with organic farming principals. If the goal is a reduction in herbicide use, as in Denmark, the problem is more complex. There are a multitude of arguments for and against the view that GMHPs might decrease the rate of herbicide use. Since most of the arguments are based on assumptions rather than experiments it is mostly a matter of belives which arguments to trust. Another goal could be an increased production rate. This would make most of the arguments against the introduction of GMHPs useless.

Most European countries want to preserve the present profit levels in agriculture and at the same time reduce the impact of agriculture on the environment. Having this goal in mind all new inventions concerning weed management methods are evaluated according to two questions: What is the gain/loss in yield and profit? Which impact does the invention have on the environment?

Influence of GMHPs on the fulfillment of the goals for agriculture

In terms of profit and yield, the introduction of GMHPs will probably be considered a gain. The higher price of seeds will most likely be counteracted by cheaper weed management. The yield of the GMHPs does not seem to change.

If we look at the impact on the environment the picture changes a bit. For most crops GMHPs are likely to cause a decrease in herbicide use, if managed correctly, and a shift towards herbicides considered less environmentally suspect - provided no resistant weeds develop. On the other hand, the GMHPs could also give rise to a multitude of weed management problems following the development of (multiple) herbicide resistant weeds and volunteer plants. Adding to this are other potential problems such as reduced biodiversity and pleiotropic effects of inserted genes, that will have to be studied more in detail in order to make conclusions about the consequences.

It is likely that some herbicides will be found in the groundwater, herbicide-resistance genes from GMHPs could be transfered to closely related weeds, herbicide resistant volunteer plants could become a problem, multiple resistance could develop, problems with reduced biodiversity and pleiotropic effects could appear. The question is whether these potential problems are acceptable in order to gain the possible benefits (van den Daele 1995).

Suggestions for the management of GMHPs

According to the potential problems outlined in this report, there are some questions that we find it important to consider when evaluating an application for marketing release of a GMHP:

- Is the herbicide in question less environmentally suspect than herbicides traditionally used in the same crop?

- Does introduction of the GMHP result in a decreased herbicide use in the given crop?

- Is herbicide-resistance the main trait of the GMHP?

- Does the GMHP have close relatives among the weeds normally found in the area where it is grown? and if yes, does the GMHP contain barrier to genetransfer (suicide genes, male sterility, transgenes on organellar DNA).

- Is the crop that the GMHP originates from a problematic volunteer plant?

- Are there any pleiotropic effects of the herbicide-resistance gene?

In Norway it is not enough to prove the absense of negative effects. The GMHP also has to be an improvement compared to traditional varieties of the same crop.
Permissions for marketing release of GMHPs could be given as "conditional registrations". This means that if the promises made by the herbicide and seed producing companies (lower herbicide use, herbicides that do not end up in ground water etc.) turn out not to be true, the permission can be cancelled. This has been done in the USA for the registration of acetochlor use in GMHPs and for genetically modified crops containing a gene from *Bacillus turingiensis*. The strategy would also be useful in connection with GMHPs (Goldburg and Hopkins 1996). The companies could also get permissions on the condition that they pay for water and environment tests needed to monitor the environmental fate of herbicides, herbicide breakdown products and herbicide-resistance genes.

5 Future prospects

The conclusions in this report leave a number of uncertainties about the impact of GMHPs on the environment. Some questions have been answered to a satisfying degree, e.g. that it is quite certain that gene transfer between oilseed rape and weedy *B. campestris* is possible and does happen under European conditions, whereas for other crops like soybean and maize this is highly unlikely. Whether these hybrids are problematic remain obscure. For other questions a prediction of the possible development has been made. The problem about predictions is that, as reliable as they may be, they are only qualified guesses based on the available knowledge. In order to minimize the degree of uncertainty that is always connected to predictions we suggest that those predictions, some of which are based on models, should be verified with experiences from the first years of commercially growing GMHPs. A monitoring approach is strongly advocated by other researchers (Kareiva *et al.* 1996).
Some key issues to be further investigated are listed below.

Herbicide use

We need to know more about the effect of GMHPs on herbicide use. The unanswered questions in connection with this includes which herbicide doses are used in GMHP crops, the number of applications used and the timing of the applications.
This knowledge will make it possible to validate models predicting the potential increase or decrease in herbicide use following the introduction of GMHPs. The data will be available in a few years as pesticide statistics and herbicide labels on recomended use.

Herbicide resistant weed

We also need to know more about the effect of GMHPs on the weed population. We know that a transfer of the herbicide-resistance genes from some crops to some weed species is possible, but we have little knowledge of the effect this will have on weed populations. The time horizon for weed and volunteer plant problems is e.g. still an open question. Will it be a problem now or in 30 years? Questions about the invasiveness of herbicide resistant weeds and volunteer plants need to be studied more in detail.
The effect of different crop rotation systems is another important aspect to consider. We know e.g. that a continous growing of one crop which is sprayed with the same herbicide for years is likely to increase the development of herbicide resistant weeds as is seen in many places.

Pleiotropic effects

Pleiotropic effects of the inserted genes are a potential risk in al genetically modified plants. Although it seems that herbicide-resistance does not bring about any important pleiotropic effects we have to remember that the knowledge comes from small scale experiments. There could be new effects under the different field conditions the plants will be submitted to after commercial release.
There could also be new pleiotropic effects showing up in weed species following a gene transfer from GMHPs.

Monitoring and models

GMHPs are easily monitored. They require herbicides in order to obtain a selective advantage from the inserted trait and are therefore not likely to have an increased invasiveness. The inserted trait also makes it easy to screen a large numbers of individuals for possible gene transfer. Based on a relatively simple procedure we could therefore obtain knowledge about gene transfer, weed development in GMHP fields and invasiveness of herbicide resistant weeds and volunteer plants.

Together with the statistics on changes in herbicide use, these data can be used to validate and refine models of the impact of GMHPs, that are already being developed today (Ongoing work at the Agricultural University, Department of Weed Science, Denmark).

6 Appendix

Table 6.1
Release applications (experimental release) of GMHPs in Europe august 1995 to September 1996

Plant	Herbicide	Country	Date
Eucalyptus grandis	Glyphosate	UK	030895
Brassica napus	Oxynil	UK	030895
Brassica napus	Glufosinate	UK	030895
Brassica napus	Glufosinate	UK	100895
Brassica napus	Glufosinate	D	210895
Brassica napus	Glufosinate	D	210895
Brassica napus	Glufosinate	D	210895
Brassica napus	Glyphosate	B	310895
Brassica napus	Glufosinate	F	110995
Zea mays	Glufosinate	NL	260995
Brassica napus	Glyphosate	UK	260995
Brassica napus	Glyphosate	UK	061095
Dianthus caryophyl	Sulfonylurea	NL	201095
Cichorium intybus	Glufosinate	IT	201095
Cichorium intybus	Glufosinate	IT	201095
Beta vulgaris	Glyphosate	DK	150296
Beta vulgaris	Glyphosate	DK	150196
Beta vulgaris	Glyphosate	DK	150196
Beta vulgaris	Glyphosate	DK	150196
Beta vulgaris	Glufosinate	IT	020296
Brassica napus	Glufosinate Male sterile	UK	020296
Beta vulgaris	Glufosinate	UK	020296
Beta vulgaris	Glyphosate	UK	020296

Plant	Herbicide	Country	Date
Zea mays	Glufosinate Quality change	B	230296
Zea mays	Glufosinate Insect resistance	F	230296
Beta vulgaris	Glyphosate	F	230296
Beta vulgaris	Glufosinate	F	230296
Beta vulgaris	Glyphosate	F	230296
Zea mays	Glufosinate	F	230296
Zea mays	Glufosinate	F	230296
Zea mays	Glufosinate Insect resistance	F	230296
Brassica napus	Glufosinate Male sterility	F	230296
Beta vulgaris	Glufosinate	F	230296
Beta vulgaris	Glufosinate	F	230296
Beta vulgaris	Glyphosate	F	230296
Beta vulgaris	Glyphosate	F	230296
Beta vulgaris	Glufosinate	D	230296
Zea mays	Glufosinate	D	230296
Brassica napus	Glufosinate	D	230296
Beta vulgaris	Glufosinate	D	230296
Zea mays	Glufosinate	NL	230296
Helianthus annuus	Glufosinate Glyphosate Fungal resist. Male sterility	NL	230296
Beta vulgaris	Glyphosate	SE	230296
Beta vulgaris	Glufosinate	SE	230296
Beta vulgaris	Glyphosate	UK	230296
Zea mays	Glufosinate Insect resistance	F	270296
Zea mays	Glufosinate	ES	120396

Plant	Herbicide	Country	Date
Brassica napus	Glufosinate Male sterility	SE	120396
Beta vulgaris	Glufosinate	ES	210396
Beta vulgaris	Glyphosate	ES	210396
Brassica napus	Glufosinate Male sterility	UK	280396
Beta vulgaris	Glyphosate	UK	280396
Zea mays	Glufosinate	UK	280396
Zea mays	Glyphosate Insect resistance	UK	280396
Zea mays	Glufosinate Insect resistance	IT	280396
Beta vulgaris	Glufosinate	UK	180496
Brassica napus	Glufosinate	UK	180496
Cichorium intybus	Glufosinate Male sterility	BE	180496
Zea mays	Glufosinate	F	180496
Brassica napus	Glufosinate	F	180496
Zea mays	Glufosinate	F	180496
Brassica napus	Oxynil	F	180496
Nicotiana tabacum	Asulam	F	180496
Beta vulgaris	Glufosinate Glyphosate Product quality Virus resistance	F	180496
Beta vulgaris	Glufosinate	F	180496
Zea mays	Glufosinate	F	180496
Zea mays	Glufosinate	F	180496
Zea mays	Glufosinate	F	180496
Cichorium intybus	Glufosinate	AT	240496

Plant	Herbicide	Country	Date
Zea mays	Glufosinate	IT	240496
Zea mays	Glufosinate	IT	240496
Zea mays	Glyphosate Insect resistance	IT	240496
Zea mays	Glyphosate Insect resistance	IT	240496
Beta vulgaris	Glyphosate	IT	240496
Zea mays	Glyphosate Insect resistance	IT	240496
Zea mays	Glufosinate	IT	240496
Zea mays	Glufosinate	IT	240496
Glycine max	Glyphosate	IT	240496
Brassica napus	Glufosinate Male sterility	F	100596
Zea mays	Glyphosate Insect resistance	F	210596
Zea mays	Glyphosate Insect resistance	F	210596
Zea mays	Glyphosate Insect resistance	F	210596
Zea mays	Glyphosate Insect resistance	F	210596
Zea mays	Glyphosate Insect resistance	F	210596
Nicotiana tabacum	Oxynil	F	210596
Zea mays	Glufosinate Insect resistance	F	210596
Zea mays	Glufosinate	UK	210596
Zea mays	Glyphosate Insect resistance	F	100696
Glycine max	Glyphosate	F	100696
Zea mays	Glufosinate	F	100696

Plant	Herbicide	Country	Date
Zea mays	Glufosinate	F	100696
Brassica napus	Glufosinate Male sterility	SE	140696
Brassica napus	Glufosinate Male sterility	SE	140696
Zea mays	Glufosinate	F	280696
Zea mays	Glufosinate	F	280696
Brassica napus	Glufosinate	D	280696
Brassica napus	Glufosinate	D	280696
Brassica napus	Glufosinate	D	280696
Zea mays	Glufosinate Insect resistance	IT	280696
Zea mays	Glufosinate Insect resistance	IT	280696
Brassica napus	Glufosinate	D	280696
Brassica napus	Glufosinate	D	280696
Brassica napus	Glufosinate	D	280696
Zea mays	Glufosinate	F	280696
Zea mays	Glufosinate	F	280696
Zea mays	Glyphosate	F	120796
Brassica napus	Oxynil	F	170796
Brassica napus	Oxynil	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Bromoxynil Glufosinate Glyphosate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Glufosinate	F	170796
Brassica napus	Oxynil	F	170796

Plant	Herbicide	Country	Date
Brassica napus	Oxynil	F	170796
Brassica napus	Glufosinate	F	170796
Brassica napus	Glufosinate	F	170796
Zea mays	Glufosinate	F	170796
Zea mays	Glufosinate	F	170796
Zea mays	Glufosinate	B	170796
Zea mays	Glufosinate	B	170796
Brassica napus	Glyphosate	D	170796
Brassica napus	Glufosinate	F	170796
Brassica napus	Glufosinate	F	170796
Zea mays	Glufosinate	F	170796
Zea mays	Glufosinate	F	170796
Brassica napus	Oxynil	F	170796
Brassica napus	Oxynil	F	170796
Brassica napus	Glufosinate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Glyphosate Bromoxynil Glufosinate	F	170796
Brassica napus	Glyphosate	F	170796
Brassica napus	Oxynil	F	170796
Brassica napus	Oxynil	F	170796
Brassica napus	Glyphosate	UK	190896
Brassica napus	Glufosinate	UK	190896
Brassica napus	Glufosinate	UK	220896
Brassica napus	Glufosinate	SE	220896

Country: UK=United Kingdom, DK=Denmark, F=France, B=Belgium, NL=The Netherlands, ES=Spain, SE= Sweden, AT= Austria, D=Germany

Date: The date the application was received by the National Forest and Nature Agency, Denmark.
Plant: *Brassica napus*=oilseed rape, *Beta vulgaris*=beet, *Zea mays*=maize, *Nicotiana tabacum*=tobacco, *Glycine max*= soybean

Table 6.2
Applications for release permits (experimental releases) of GMHPs in the USA from march to august 1996

Plant	**Herbicide**	**Date**
Rice	Glufosinate	220396
Beet	Glyphosate	80496
Oilseed rape	Sulfonylurea	110496
Cichorium intybus	Sulfonylurea	110496
Arabidipsis thaliana	Sulfonylurea	110496
Tobacco	Sulfonylurea	110496
Rice	Glufosinate	80596
Sweet potato	Glufosinate	40696
Wheat	Glyphosate	140696
Beet	Glufosinate	160796
Oilseed rape	Glyphosate	290796
Oilseed rape	Glufosinate	080896
Brassica oleracea	Glufosinate	120896

Date: The day the application was received by the competent authorities.
Plant: *Brassica oleracea*= cabbage, *Cichorium intybus*= Chicory

Table 6.3
Release petitions (marketing release) of GMHPs in the USA from 1993 to 1996

Plant	**Herbicide**	**Date**
Cotton	Bromoxynil	150793
Soybean	Glyphosate	150993
Corn	Glufosinate	231294
Cotton	Glyphosate	140295
Corn	Glufosinate	250595
Corn	Glufosinate	160895
Cotton	Sulfonylurea	130995
Soybean	Glufosinate	201195 (Withdrawn)
Soybean	Glufosinate	080396
Corn	Glyphosate	300896

All petitions were approved except one that was withdrawn and the last one that is still (Nov 96) under rewiew of the competent authority.

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Abstract:
The result of a 3 month investigation based on literature and available statistics summarizes the state of the art of herbicide resistant crops and impact of their use. Until now only 4 marketing release permits have been given in Europe, which means that Europe has no large scale growing experience. In order to make predictions as precise as possible, environmental impact and changes of cultivation methods have to be evaluated separately for each new release application.

Terms:
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Resumé:
Resultatet af en 3 måneders undersøgelse af litteratur og tilgængelig statistik giver status over herbicidresistente afgrøder og de konsekvenser, det vil medføre at introducere dem i landbruget. Da der indtil nu kun er givet 4 tilladelser til markedsføring af herbicidresistente afgrøder i Europa, er der ikke erfaringer med dyrkning i stor skala. For at kunne forudsige konsekvenserne så præcist som muligt, må man evaluere indflydelsen på miljøet og ændringer i dyrkningsmønstre separat for hver markedsføringsansøgning.

Emneord:
genmodificerede planter; afgrøder; resistens; pesticider; godkendelser; USA; EU; konsekvensanalyser

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