## **Concepts for Coexistence**

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FINAL REPORT
VIENNA, SEPTEMBER 17, 2003

COMMISSIONED BY THE FEDERAL MINISTRY OF HEALTH AND WOMEN

This report is a summarised, updated English version of the report: GMO-free Cultivation Areas: Concepts for Coexistence - Conception and Analysis of Scenarios and Implementation Steps. In the light of the recommendations by European Commission on coexistence of July 2003, the mayor part of this report has been completely revised.

The calculation and interpretation of the data for the simulation models of "Overall Upper Austria" and "Wels Land" were carried out in cooperation with **DI Andreas Bartel**, Institute for Landscape Architecture and Landscape Management, University of Agricultural Sciences, Vienna.

### Acknowledgments:

I would like to thank the following persons and institutions for their cooperation: For the speedy and uncomplicated provision of data: DI Otto Hofer, Agriculture, Forestry, Environment and Water Management; Bernhard Gibitz from DORIS (Digital Upper Austrian Zoning Information System); the Upper Austrian Nature Protection Authority, the Provincial Government of Upper Austria, Department for Hydraulic Engineering, as well as the Federal Office of Metrology and Surveying; for comments, information and references: Dr. Josef Hoppichler (Federal Institute for Less-Favored and Mountainous Areas); Dr. Helmut Gaugitsch (Federal Environment Agency Vienna; for cooperation in data gathering and comments on reading of the manuscript: and financing the bodies commissioning the study, the Upper Austrian Environment Academy, the Environmental Authority of the Province of Upper Austria and the Federal Ministry for Social Security and Generations. For financing the update and English translation of the study I would like to thank Federal Ministry of Health and Women. For reading and editing the manuscript: I would like to thank DI Anna Hartl; and Mag. Bettina Jakl-Dresel for the English translation.

"WHILE THE DUTY OF PREVENTING DAMAGE TO THE ENVIRONMENT IS BASED ON A KNOWN RISK, THE NOTION OF PRECAUTION IS BASED ON LACK OF CERTAINTY."

(OECD 2000)

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### SUMMARY

Based on the European Commission recommendations on coexistence, there is a need to regulate coexistence crop specifically and step by step, from management at the farm level to management at the neighbourhood level up to measures with region-wide dimension. Based on analyses on cross-pollination, seed dispersal, seed viability in the soil and in accordance with the European Commission recommendation, cumulative effects as well as the small-scale structure of Austrian Agriculture, there will be a need to regulate the cultivation of GM maize and GM oil seed rape at a regional level. In some regions, the agricultural structure may allow management of maize at the neighbourhood level, although a detailed analyses to identify such regions was not the subject of this study. While voluntary regions will not provide a proper conflict solution potential. legally binding regions (GM regions or GM-free regions) will be necessary. From a management perspective, only minimum isolation distances would serve as a practically feasible decision tool. Hedges, for example, are very difficult to handle in the management of coexistence, as exact knowledge on the efficient reduction of the rate of crosspollination at certain distances is lacking. Varieties with different flowering time will not provide sufficient security to avoiding cross-pollination. A DIGITAL CULTIVATION PARCEL MAP is proposed by the author to serve as a core instrument in the way to handle a complex information flow ,which has to consider farmers' crop rotation plans and cultivation type (organic, conventional, GMO) for each parcel on a annual basis. Furthermore, the establishment of a regulation on "GMO cultivation and processing" is proposed. It should be based on the EC Regulation 2092/91 for organic farming, in order to get a full separation of GMO cultivation and processing to achieve full traceability as required by the adopted EC Regulation on traceability and labelling as well as EC Regulation 178/2002. Besides these recommendations on the management of coexistence, the author points out that there are huge gaps of knowledge in the underlying basic science of risk assessment of GMOs, e.g. with regard to cell biology, genetics). Moreover, the current practice of risk assessment does not provide sufficient information to conclude that GMOs for food and feed use are safe. GMO crops which have the potential for outcrossing should not be approved because there would be no (or at least very limited) options for mitigation measures in the case of error, which cannot be ruled out for risk assessment methods. Besides the problems with coexistence, an approval of GMOs would still be premature.

### **Preliminary Remark**

Important Limitation of this REPORT

The question of coexistence of organic and conventional non-GM production with the cultivation of GMOs" touches upon very diverse areas of science and social policy. These are:

Focus of this study on biological science

- The level of biological science, including questions of outcrossing distances (by airborne/wind pollen transport or distribution by insects) and the dispersal of transgenic seed (by birds, other animals, etc.)
- The level of quality assurance, which continuously investigates whether the framework conditions set are met, and whether the framework conditions are practically applicable in the sense of monitoring, or whether

adjustments should be made.

- The level of social science, which investigates the aims society develops regarding the future direction of agriculture, and which attempts to clarify what a fair balance of interests (subsidies and other measures) between various different interest groups might look like.
- The legal level, which looks into how a regulatory approach might be integrated into existing legal frameworks without undermining prevailing rules and regulations.
- The political level, which, bearing in mind cost-benefit considerations, makes decisions as to values and sets goals on the basis of findings of biology and social science.

It is obviously not possible to cover all aspects within the scope of the present study, as this would exceed by far the financial framework and the time available.

The focus of this work is on the level of biological science and that of quality assurance. Political as well as legal aspects have only been touched upon and therefore need further extensive analysis.

# 1 EUROPEAN COMMISSION RECOMMENDATION ON COEXISTENCE

On July 23, 2003 the European Commission published recommendations on coexistence. Therein, the "European Commission considers that measures for co-existence should be developed and implemented by the Member States. European Commission should support and advise Member States in this process by issuing guidelines for addressing co-existence."

This report analyses the recommendations made by the European Commission. Based on this analysis and on data of cross-pollination as well as in consideration of the small scale structure of Austrian agricultural cultivation area, the author gives some recommendations for possible solutions for the problem of coexistence.

### 1.1 Introduction

In its introduction, the European Commission summarises the problem of coexistence as follows:

"The cultivation of genetically modified organisms (GMOs) in the EU is likely to have implications for the organisation of agricultural production. On the one hand, the possibility of the adventitious (unintended) presence of genetically modified (GM) crops in non-GM crops, and vice versa, raises the question as to how producer choice for the different production types can be ensured. In principle, farmers should be able to cultivate the types of agricultural crops they choose - be it GM crops, conventional or organic crops. None of these forms of agriculture should be excluded in the EU. On the other hand, the issue is also linked to consumer choice. To provide European consumers with a real choice between GM food and non-GM food, there should not only be a traceability and labelling system that functions properly, but also an agricultural sector that can provide the different types of goods. The ability of the food industry to deliver a high degree of consumer choice goes hand in hand with the ability of the agricultural sector to maintain different production systems. Co-existence refers to the ability of farmers to make a practical choice between conventional, organic and GM-

crop production, in compliance with the legal obligations for labelling and/or purity standards."

### 1.2 ECONOMIC ASPECTS OF CO-EXISTENCE VERSUS ENVIRONMENTAL AND HEALTH ASPECTS

The European Commission considers the problem of coexistence solely as an economic but not an environmental or health issue.

"It is important to make a clear distinction between the economic aspects of coexistence and the environmental and health aspects dealt with under Directive 2001/18/EC on the deliberate release of GMOs into the environment. Since only authorised GMOs can be cultivated in the EU, and the environmental and health aspects are already covered by Directive 2001/18/EC, the pending issues still to be addressed in the context of co-existence concern the economic aspects associated with the admixture of GM and non-GM crops" (EC-COMMISSION 2003, 1.2).

This conclusion by the European Commission has to be challenged on account of the following reasons.

### 1.2.1 THE PRACTICAL SITUATION OF RISK ASSESSMENT OF GMOS

From a legal perspective, all authorized GMOs are safe. From the current practical perspective, this is not guaranteed by the current way risk assessment of GMOs is undertaken. The core assessment of health effects of GMOs is based on the principle of substantial equivalence. This principle, however, lacks a scientific basis. It is a hypothesis that has never been tested by way of a large number of controlled experiments. Thus, this hypothesis has been made a decision tool without scientific justification. MILLSTONE et al. (1999) points out::

"The relationship between genetics, chemical composition and toxicological risk remains unknown. Relying on the concept of 'substantial equivalence' is therefore just wishful thinking: it is tantamount to pretending to have adequate grounds to judge whether or not products are safe."

Furthermore, a review (SPÖK et al. 2002) of the current practice of risk assessment of 11 out of 28 applications for placing on the market of pending and authorized GMOs demonstrated that:

- "In general, toxicological information is rather a minor part of the dossiers.
- Differences in the intended use of the GMP do not affect the extent of the toxicological evaluations.
- Most toxicity tests are displayed as summaries or are just references to literature and can therefore not be verified and reviewed. Internal references are often used improperly.
- Apparently, toxicological tests were carried out rather sporadically, most likely in cases of Bt-plants, as Bt-toxins had already been approved before as insecticide in some countries.
- Data on the toxicity of the whole GMP are not provided in any dossier.
- Toxicological acceptance is often justified by three arguments: low toxicity of the gene product, substantial equivalence of the GMP to their conventional counterparts, and low exposure.
- Potentially toxic effects resulting as a secondary effect from the gene insertion are not considered in any case.
- Most of the toxicological testing was not carried out in compliance with quality assurance programs such as Good Laboratory Practise (GLP).
- GMP are very often declared as being safe just by assumption based reasoning.
- Furthermore these assumptions are sometimes not easily or not at all verifiable.
- Risk assessment procedures which are carried out in a systematic way consisting of a hazard assessment of the GMP on the one hand and of an analysis of exposure on the other hand, are lacking in the dossiers."

In the absence of a thorough risk assessment of GMOs, it is not far to conclude that consumers are unwillingly part of a great food gamble (DIAMOND 2001).

### 1.2.2 THE CASE OF UNCERTAINTY

Decisions regarding risks are always preliminary (uncertain) and there is no way to prove that a product is safe. Scientist can only assume that a product is safe after a thorough risk assessment. A review on 14 case studies like BCP, asbestos, and others, where risk assessment had failed to protect human health and/or the environment, shows clearly that we have to admit that risk assessment has a significant probability of error (EEA 2001). GEE (2003) concludes that there are far more examples which were not covered in the study, and that in general science produces more false negative risk assessments (a risk was not identified although there was a risk) than false positive risk assessments (risk assessment concluded a product is unsafe although the product was safe). There are several published reports on unintended effects like a 20 % increased lignin content in RR soybeans expression of glyphosphate (EPSPS) )resistance) or increased vitamin B6-content, (+50%) in rice with transgene for higher lignin content (20%), expression of soybean glycinin (KUIPER et al. 2001, table 6).

Besides this general view on uncertainty and precaution in risk assessment, new evidence from

Man is only attributed 40.000 genes, as opposed to previously assumed 142,000 genes. cell biology and genetics clearly demonstrates our limited understanding of the role of DNA, RNA and protein in living organism. The HUMAN GENOME PROJECT revealed that instead of the assumed 142,000 genes, man has a mere 30,000 – 40,000 genes. In a New York Times article of February 19, 2001, entitled "Humbled by the Genome's Mysteries," evolutionary biologist and Harvard University Professor of Zoology Jay Gould questioned how it might be possible that e.g. the very simply constructed worm *C. elegans*, with a mere 939 cells, has 19,000 genes, while humans, with their much more complex functions (metabolism, immune system, brain)

have only 30,000 to 40,000 genes. He postulated that human complexity could not be explained with the old view of life being embedded in a DOGMA of "DNA makes RNA makes PROTEIN." Human complexity is thus not achieved by genes but by combinations and interactions within the cell and the organism. The interaction of genes, however, cannot be derived from the knowledge of one gene alone (cf.Textbox 1). Furthermore analyses of non coding DNA (junk DNA) suggests the existence of a hidden class of abundant regulatory elements (e.g. FLAM 1994, MONK 1995, SHCHERBAK 2003, HARE und PALUMBI 2003).

### Textbox 1: Lack of understanding in cell biology

"The collapse of the doctrine of one gene for one protein, and one direction of causal flow from basic codes to elaborate totality, marks the failure of reductionism for the complex system that we call biology - and for two major reasons.

First, the key to complexity is not more genes, but more combinations and interactions generated by fewer units of code - and many of these interactions (as emergent properties, to use the technical jargon) must be explained at the level of their appearance, for they cannot be predicted from the separate underlying parts alone. So organisms must be explained as organisms, and not as a summation of genes (GOULD 2001).

<sup>&</sup>lt;sup>9</sup> The one-gene-one-protein hypothesis has no longer been valid for some time. Nor can the concept of one-gene-one-enzyme be applied any longer. "A concept of general validity that integrates the diverse characteristics of the hereditary material in a uniform and easy-to-handle pattern, can no longer be formulated today (HENNIG 2001, 457ff).

Potential patterns of explaining the small number of genes and the high number of proteins are found in the different forms of splicing - the cutting out of uncoded sequences within the mRNA synthesis and/or mRNA editing. From a gene consisting of 6 exons, for example, different but mutually related proteins are expressed. In cells of the parathyroid, the exons 1-4 are linked by splicing, with the concomitant loss of 5 and 6. Exon 4, in turn, is spliced from neurons. Thus, proteins of a gene are coded that are active in the parathyroid gland, in hypothalamics cells, in sensory gangliocytes of the spinal cord as well as in other areas of the spinal cord (KNIPPERS 2001, 412ff).

"We are in a data-rich environment, but the fact is we are information poor, You look at biological systems with much more complexity than before" Peter Sorger, an associate professor of biology at MIT

"We really have a poor understanding of what a gene actually does and where and when it should do it. You can understand the entire genome and [still] understand less than 1 percent about what is going on in a cell." Eric Neumann, vice president of bioinformatics at Beyond Genomics Inc.(DODGE 2003).

What is true for basic research is particularly true and of special significance for the risk assessment of GMOs. Risk assessment is based on the current knowledge of genetics, cell biology, molecular ecology and ecology. The risk assessment of human health effects is only as good as the underlying knowledge of the basic interactions in the cell and in an organism. Gaps in the underlying science lead to gaps in the risk assessment as well.

In view of numerous uncertainties regarding GMO risk assessment, it is possible for important risk aspects to remain unobserved. In the light of precaution, the author concludes that the release of GMOs into the environment and the approval of GMOs for food and feed is still too premature. The quote from OECD (2000) very adequately describes the link between precaution and uncertainty. "While the duty of preventing damage to the environment is based on a known risk, the notion of precaution is based on lack of certainty."

# 2 ANALYSIS OF CAUSES AND INTERRELATIONSHIPS REGARDING GMO-CONTAMINATION

Several institutions and persons - have dealt intensively with questions of pollen reach of selected crops and coexistence over the past three years INGRAM 2000, TREU und EMBERLIN 2000, SCP 2001, EASTHAM und SWEET 2002, IPTS (EDS.) 2002, BECK et al. 2002, TOLSTRUP et al. 2003.

### 2.1 BIOLOGICAL CONTAMINATION WITH POLLEN

### 2.1.1 POLLEN TRANSPORT AND FERTILISATION

### 2.1.1.1 CROSS-POLLINATORS AND SELF-POLLINATORS

Self-fertilisers: plants whose cross-fertilisation rate does not exceed an average of 3 - 4 % In flowering and fertilisation behavior, we distinguish between crossfertilisers and self-fertilisers. **Self-fertilising species** (e.g. wheat, barley, soybean, oat) are species whose **naturally occurring self-fertilisation rate does not exceed 3 - 4 % on average** (ODENBACH 1997, 106f). Species whose cross-fertilisation rate exceeds 3 - 4 %, are classified as cross-fertilisers (e.g. maize, rye, oilseed rape (depending on the variety)). In many species, both forms of fertilisation occur simultaneously, i.e. the flowers of one inflorescence may be partly self-fertilised and partly cross-

fertilised. The relationship between self-fertilisation rate and cross-fertilisation rate may fluctuate considerably within one species (depending on the variety, and, to a certain extent, on the climatic conditions at the flowering time), which sometimes renders an unequivocal classification difficult.)

### 2.1.1.2 POLLEN DISPERSAL

The way of pollen dispersal determines to a large extent the maximum reach of vertical gene transfer in genetically modified plants. Along with wind and insects (flies, ants, honeybees, bumblebees), birds and mammals are potential pollinators. With some plants, pollination occurs in water, which means that water also has to be borne in mind as a vector for various water plants. With regard to the species we are dealing with herein (oilseed rape, maize, soybean), pollen transfer is limited to insects and wind. A total separation of the various ways of

fertilisation is not feasible. Sometimes, several forms of fertilisation occur simultaneously. Oilseed rape, for example, is to a great extent self-fertilising, depending on the variety; crossfertilisation occurs mainly by way of insects but also wind. Oilseed rape pollen (which may also cause allergies) can be detected in the air in areas remote from the production areas (HEMMER et al. 1997). What is more, exclusive fertilisation by way of wind does not mean that the pollen is avoided by insects. While maize (monoecious - male and female flowers are physically separated on the plant) is exclusively wind-pollinated, maize pollen (as opposed to the female floral organs of maize that do not secrete nectar) is an attractive and in certain locations the most important late pollen source for bees.

### 2.1.1.3 PROBLEMS OF METHODOLOGY IN MEASURING THE REACH OF HYBRIDIZATION

### 2.1.1.3.1 INFLUENCE OF MEASUREMENT TECHNIQUE

Gene flow is the transport of genetic information between individuals, populations, and species. Particularly in plants, we distinguish between "potential" and "actual" gene flow. **Potential gene flow** is the transport of pollen or seeds (diaspores) as a function of the distance (i.e. the dispersal distance of pollen and seeds). **Actual gene flow** is the height of fertilisation rates (with pollen) as a function of the distance from a source (RAYBOULD und CLARKE 1999). Obviously, not every pollen grain leads to successful fertilisation and not every seed produces plants capable of reproduction. This is why the actual gene flow differs considerably from the potential gene flow (RAYBOULD und CLARKE 1999).

There are direct and indirect methods of measuring gene flow. The most common form is the

Thus, the actual gene flow may differ considerably from the potential gene flow.

direct measurement of the pollen and seed transport with respective traps that allow an estimate of the potential gene flow. Volumetric pollen traps (measurement of the amount of pollen per m³ of air and unit of time, as a rule 24 hours) (e.g. TIMMONS et al. 1995) are distinguished from pollen sinks (e.g. SQUIRE et al. 1999). Already at this stage, however, it becomes evident that the choice of the suitable methods may have a decisive influence on the result. SQUIRE et al. (1999, 60 f) point out that the result of pollen measurements (pollen trap or measurement of the pollen load on a defined area) may differ considerably depending on the method chosen. Pollen concentration

measurements with sinks at greater distances render higher pollen concentrations than measurements with pollen traps. For an estimate of the potential reach of gene flow between crops (in particular of the gene flow from GMOs to organically cultivated crops), both methods should be applied, particularly as it is necessary to further strengthen the hypotheses for gene flow on the landscape level as well as to further back the conclusions to be derived thereof (SQUIRE et al. 1999).

By means of **genetic markers**, which, like pollen traps, are suitable for estimating the reach of gene flow between crops, the actual gene flow is measured directly (RAYBOULD und CLARKE 1999).

Estimating the gene flow from crops to wild plant populations is possible in the form of an **indirect estimate of the actual gene flow via ascertaining the variation of allele frequencies**. A great variation of allele frequencies are indicative of low gene flow, while great correlation in the allele frequencies indicate high gene flow. Both direct and indirect measurements of gene flow, if applied exclusively, do not render a complete picture of the gene flow. Best results are achieved if both methods are applied simultaneously (RAYBOULD und CLARKE 1999).

### 2.1.1.3.2 INFLUENCE OF THE CULTIVATION AREA ON MEASURING DISTANCE

Along with the measurement techniques, the experimental design exerts considerable influence

Measuring technique and experimental design exert considerable influence on the final result on the result. Many experiments limit themselves to distances between 100 m and 400 m (SIMPSON et al. 1999, SCHEFFLER et al. 1993, SCHEFFLER et al. 1995, DOWNEY 1999). Thus, the quality of the findings regarding maximum reach is significantly limited by the way the experiment is designed. The few experiments that made measurements

at greater distances up to 4,000 m (SQUIRE et al. 1999, THOMPSON et al. 1999) actually detected gene flow at these distances.

Along with the maximum distances considered in the experimental design, the size of the area of the pollen source has a strong influence on the final result. The hybridization frequency turns out to ultimately correspond to the ratio of pollen availability (of the plant's own or foreign origin). In small experimental plots with a large non-GM isolation track, outcrosses of the non-GM plants on the isolation track are higher on the experimental plot than outcrosses of GM-plants of the research-size plot into non-GM plants on the isolation track (cf. SQUIRE et al. 1999,58f). This is why the pollen dispersal distances found in experiments with small research plots are only approximate values. Numerous values of maximum outcrossing distances had to be upwardly corrected.

For oilseed rape, outcrossing frequencies were initially, as a consequence of the small size of the experimental design (research-size plots with diameters of 9 m), estimated at 0.4 % at 3 m, 0.02 % at 12 m, and 0.0003 % at 47 m (SCHEFFLER et al. 1993). Later works with an experimental plot of  $400 \, \text{m}^2$ , found outcrossing rates for oilseed rape of 0.0156 % at 200 m and 0.0038 % at 400 m. These figures were below the level of tolerance for seed production, which is why the recommendation of the authors of the study was that isolation distances of 200 - 400 m would be sufficient (SCHEFFLER et al. 1995).

Works (THOMPSON et al. 1999, TIMMONS et al. 1996b1996b) with larger fields (up to 10 ha), outcrossing distances of up to 4 km have been found in experiments (details cf. 2.3.1). Similar experiences were made with alfalfa (*Medicago sativa*), for which the outcrossing frequency of large fields was 10 times higher than that of small research-size plots, and significant hybridization was detected across distances of ca. 1,000 m (AMAND et al. 2000).

### 2.1.1.3.3 INFLUENCE OF THE RECEPTOR PLANT

An equally very frequent direct way of measuring gene flow in oilseed rape is the use of male sterile receptor plants, measuring the number of fertilised seeds (e.g. TIMMONS et al. 1995, THOMPSON et al. 1999). As in this method, self-fertilisation as well as fertilisation by neighboring plants is suppressed and the hybridization frequency eventually corresponds to the ratio of pollen available (of the plants' own or foreign origin), this form of measurement produces higher values in terms of reach and hybridization than with male fertile oilseed rape plants (SIMPSON et al. 1999, 81f). However, it is a sound indicator of the actual theoretical potential of hybridization.

### 2.1.2 WIND-POLLINATION

Several criteria influence the maximum distance of a successful fertilisation by pollen transport:

- Potential and absolute pollen concentration in the field
- viability of pollen
- transport dynamics of pollen

Details shall be discussed in the following chapters.

### 2.1.2.1 POLLEN CONCENTRATION IN THE AIR

The most important factors of airborne pollen concentration are:

- strategy of generative reproduction
- pollen production per plant
- cultivation area

The potential and absolute pollen concentration in the air above a field depends primarily on the **strategy of generative reproduction**. Closely related to this concept is the plant's **open-floweredness** (**chasmogamy**), which in cross-pollinators is much more frequent than in self-pollinators. Wind-pollination, as opposed to pollination by insects, is not very precise. The probability for a single pollen grain to be wind-borne exactly onto the stigma of a plant of the same species (and not, for example, onto a leave or the stem of the plant), is very low. In order for a successful wind-fertilisation to occur, there has to be a considerable surplus of pollen. Wind-pollinators thus mostly produce several tens of millions of pollen grains per plant. Maize produces approximately 14 - 50 million of pollen grains per plant and approximately 30 - 50 times more than wheat (TREU und EMBERLIN

Many
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2000, 6f). Rye produces 8 - 35 times more pollen per anther than wheat (HORAK et al. 1980)

Chasmogamous nature and amount of pollen production determine the pollen concentration in the field and the air layer immediately above the field. The airborne pollen share of self-fertilisers (in particular remote from agricultural cultivation areas) is significantly lower than that of cross-fertilisers.

The weight of pollen is another significant factor for the extent and distance of pollen dispersal. Light pollen (e.g. rye pollen) weight leads to significantly higher concentrations in the air remote from agricultural

areas than heavier pollen weight (e.g. maize pollen) (HORAK et al. 1980).

As the pollen concentration frequently (although not always) declines continuously with the distance to the field, the maximum transport distance of relevant pollen densities increases directly in proportion to the level of the initial pollen concentration (and thus to the size of the cultivation area).

### 2.1.2.2 POLLEN VIABILITY

The theoretically global reach of pollen transfer that might result in fertilisation is limited in particular by the longevity of the pollen. In general, pollen is viable only for a short period of

Key factors of pollen viability are:

- duration of biological fertilisation capacity of the pollen
- meteorological factors

time. The fertilising capacity may last between a few hours and several days, depending on the climatic situation. Under natural conditions, maize pollen, for example, is viable between 24 hours and several days. In an experiment with maize pollen stored in bags, the pollen, when exposed to direct solar irradiation as well as at maximum temperatures of 96 °F (= 35.56 °C) was viable for 3 hours only. When shaded by plants as well as at maximum temperatures of 86 °F (= 30 °C), the pollen had a life of 30 hours (EMBERLIN et al. 1999). In contrast, LUNA et al. (2001) detected a maximum viability of maize pollen of 2 hours .

Depending on the pollen, the climatic factors exerting an unfavorable or favorable influence on pollen longevity (direct solar

irradiation, humidity, temperature) vary. In general, low humidity as well as direct solar irradiation have a negative impact on pollen viability. The fertilising capacity of oat pollen lessens considerably as soon as after 15 minutes when it is exposed to direct solar irradiation (HOFFMANN et al. 1985, 102f).

## 2.1.2.3 TRANSPORT DYNAMICS

The microclimate prevailing in the layer near the ground has a significant influence on the

Key factors of influence:

wind force

pollen dispersal rate. The convection occurring on account of the warming-up of the soil may contribute to carrying pollen into higher layers of air, which are generally characterized by greater wind velocities than layers close to the ground.

wind directionturbulences

But also actively flying insects make use of thermal up-currents in order to get into higher strata. The monarch butterfly in the United States, for example, uses thermal up-currents in order to reach layers

of air adequate for carrying it during its long-distance migration (WESTBROOK und ISARD 1999).

Depending on the pollen dispersal distance, we distinguish between three types:

- Part (ca. 50 %, LAVIGNE et al. 1998 1998) of the pollen does not reach the local thermal currents and will sink in the immediate surroundings (up to 3 m) of the plant (type: local pollen).
- Another part is seized by local thermal air currents that carry it further (several meters to several kilometers). Such small-scale convection cells rarely last longer than 20 to 30

minutes, the average wind velocity in these cells being 0.5 to 1 m/sec (TREU und EMBERLIN 2000, 2f) (type: **regional pollen**).

 Part of the pollen is seized by large-scale atmospheric circulation, taking pollen across regions and even across the globe. The major part of the pollen will sink in the evening or at night, when temperatures start to cool down (TREU und EMBERLIN 2000, 2f). Pollen that

Part of the pollen is seized by large-scale atmospheric circulation, taking pollen across regions and even across the globe. does not sink in the following night is as a rule carried away on a global scale. Thus, pollen is ubiquitous in the atmosphere (though in clearly differing concentrations) and, among other purposes, serves as crystallization nucleus for rain drops (WESTBROOK und ISARD 1999, 271f). The distances pollen is able to travel are considerable and have time and again made for surprising findings. In Swiss glaciers and lakes, for example, pollen from North Africa and the Mediterranean region was found (KJELLSSON et al. 1997, 58f) (type: ubiquitous pollen). Owing to the fact that pollen is viable only for short periods of time (a few hours up to several days, in extremely rare cases; average maximum longevity is around 24 hours), it may be assumed that ubiquitous pollen that does not sink in the first night after leaving the anthers, is irrelevant in terms of fertilisation.

The distribution of the overall quantity of pollen released from the anthers among the three pollen types is difficult to predict, considering that it is strongly dependent upon microclimatic conditions at the moment of bursting of the anthers.

As a rule, the overall quantity of the pollen released from the anthers decreases with the distance from the pollen source. This is true for both regional and ubiquitous pollen. The decline of the pollen concentration is directly in proportion to the pollen source. The course of the curve is negatively exponential and progresses asymptotically toward zero (TREU und EMBERLIN 2000, 3f). The detailed course of the curve depends upon the plant species, the initial pollen concentration, as well as microclimatic conditions.

Along with the initial concentration (cf. below), wind conditions considerably influence the pollen concentration at greater distances. Pollen, spores, seeds (diaspores), microorganisms, etc., are as a rule dispersely scattered by wind, with the concentration decreasing as a rule continuously with the distance. However, pollen may also, if wind conditions are favorable, be dispersed across long distances without a significant decline in concentration (WESTBROOK und ISARD 1999, 164f). GIDINGS et al. conclude in their theoretical model examinations of pollen dispersal in genetically modified grass varieties that the pollen concentration does not always decrease linearly with the distance (GIDDINGS et al. 1997).

GAGE et al. (1999) devised a scale linking the spatial dispersal by wind with temporal dimensions.

Micro-scale

• In the range of seconds to minutes, the spatial dispersal by wind is in the range of 1 mm<sup>2</sup> to 1 km<sup>2</sup>.

Meso-scale

- In the range of minutes to hours, wind dispersal is between 1- 10 km<sup>2</sup>.
- In the range of hours to days, wind dispersal is between 1-100 km<sup>2</sup>.
- In the range of days to weeks, wind dispersal is between 100-1,000 km<sup>2</sup>.

(GAGE et al. 1999, table 1)

As pollen retains its fertilising capacity several minutes to several days, with an average longevity of 24 hours, a dispersal of viable pollen of up to 100 km<sup>2</sup> is to be expected. On the basis of the data compiled above, this marks the "maximum" theoretical potential for successful fertilisation. The fact that pollen can be detected at distances far greater than that has already been mentioned. Thus, the potential for a dispersal of 100 km certainly exists. However, the

pollen concentration<sup>2</sup> at these distances seems to be so low that the probability for fertilisation to occur is greatly reduced or extremely small. LUNA et al. (2001) assume 32 km to be the

Pollen is as a rule dispersely scattered by wind, i.e. the concentration decreases continually with the distance. However, pollen may also, if wind conditions are favorable, be dispersed across long distances without a significant decline in concentration.

maximum theoretical dispoersal distance for viable pollen assuming a 2 h viability of pollen grain. With a clear decrease in pollen concentration, the probability of a pollen grain to land exactly on the stigma of a receptor plant decreases continually. Adventitious occurrences are, however, at least theoretically possible. The author does not have concrete statements as to the probability of such possible "lucky incidents" at his disposal. Such incidents may occur with a probability of one contaminated maize grain on the maize area of Austria per year. In any case, such incidents are clearly below the discussed limits of 0.1 % in Austria and 0,9 % in EU. Although the wind direction has a significant impact on the dynamics of pollen dispersal, even small-scale air circulation must not be omitted. As a rule, fertilising rates are higher in the direction of the prevailing wind than in the averse direction. WAGNER and ALLARD are quoting trials in which the prevailing wind direction had no significant influence on the number of fertilisations in the immediate surroundings (WAGNER und ALLARD 1991. Fertilisation rates were about the same in all

spatial directions, with fertilisation being slightly, though not significantly, higher in the direction of the wind.

### 2.1.3 INSECT-POLLINATION

In most agricultural crops (e.g. maize, rye), pollen is wind-dispersed. Oilseed rape pollen, however, is distributed, along with wind-dispersal, also by insects, e.g. honey-bees (Apis mellifera) or bumble-bees (Bombus terrestris). Fruit trees are almost exclusively insectfertilised. As opposed to pollen transfer by wind, pollen distribution by insects is much more precise. With low amounts of pollen, a very efficient fertilisation rate is achieved. This is why for an estimate of maximum reach of fertilisation through insects, the specific pollen biology (weight and number of pollen) of the plant as well as the initial pollen concentration on the field edge are of minor importance. Significant for an assessment of hybridization distances is the specific behavior of insects and their foraging of pollen and nectar. Here, it becomes obvious that detailed studies allowing first projections on the maximum reach of fertilisation exist on the European honey-bee (Apis mellifera) only. Many investigations deal with fundamental subjects such as orientation, movement across the field, planting activity. Only few experiments have been carried out with a GMO-focus - the estimation of the gene flow from synthetic transgenes into non-transgenic crops or wild plants. And yet, it is particularly this new focus that brings about entirely new findings that modify the existing status of knowledge. The following is a brief overview of the most relevant events related to this subject.

Many different insect species are able to transfer pollen. In most cases, host plant and pollinator (the insect, mammal, etc. carrying out the fertilisation) are closely adapted to each other. The most important group of pollinators is the family of bees (Apidae, insect order of Hymenoptera). Within this family, there are numerous genera, such as *Apis* (honey-bee) and *Bombus* (bumble-bee) as well as several genera of wild bees. Globally, 25,000 species of wild bees have been described (DELAPLANE und MAYER 2000, 19f), the largest part being solitary bees. The best investigated and most frequently bred species is *Apis mellifera* (European honey-bee). However, some solitary bee species are also used specifically for commercial orchards and vegetable farms (DELAPLANE und MAYER 2000).

Bumble-bees (*Bombus spp.*, family Apidae) are mostly found in regions of moderate climate. There are 400 bumble-bee species worldwide (DELAPLANE und MAYER 2000, 63f). The bumble-bee *B. terrestris* is bred and "artificially" reproduced. Bumble-bee colonies are mainly used in vegetable farming under glass covers. As opposed to bees, which prefer forage

<sup>&</sup>lt;sup>2</sup> Pollen concentration in the atmosphere or in areas remote from agricultural activities is very low. On the Swedish mainland, for example 18,000 pollen grains per 100 m<sup>3</sup> were measured, as opposed to a mere 0.7 above the Atlantic (ERDTMANN 1943, quoted in TREU and EMBERLIN 2000, 2f)

sources in the immediate surroundings of the hive, bumble-bees forage on honey plants preferably at a distance of 50 to 600 m from their nest (DELAPLANE und MAYER 2000, 13f).

As with estimates of the reach of hybridization by wind, in the area of insects, there are gaps of knowledge and a lack of comprehensive understanding of interrelationships.

But also syrphids and sawflies are capable of pollen transfer. On as well as in the immediate surroundings of an oilseed rape research-size plot area in Germany, 49 syrphid species, 94 bee species and 96 sawfly species were detected (SAURE et al. 2000).

The maximum and average foraging distances have been well documented only for honey-bees. In principle, bees prefer forage sources in close proximity to the hive. The average distances covered during foraging are approximately 2 km. For a good forage source, however, bees fly over greater distances. Here, the upper limit seems to be 5 to 7 km. Merely the queen as well as drones may cover even greater distances of over 30 km for mating flights. Queen and drones, however, do not participate in the search for pollen and

nectar, and thus do not contribute to the fertilisation of crops.

As with wind-pollination, the maximum flight distances of insects are of merely theoretical value for an estimate of the maximum transfer distances of pollen. Regarding the distribution of pollen by insects, the insects' behavior in foraging for nectar and pollen is the key factor.

As with estimating distances of hybridization by wind, in the field of insects, there are gaps of knowledge and a lack of comprehensive understanding of interrelationships. Depending on the setup and the focus of the experiment, results may vary considerably and thus lead to differing conclusions.

When we look at individual bees in a field, it becomes evident that notwithstanding a theoretical reach of several kilometers, very short distances are covered from plant to plant. Most of the pollen is deposited onto plants of the same species in the immediate surroundings. A small share of the pollen is carried ca. 20 to 40 plants further (CRESSWELL et al. 1995) cf. CRESSWELL 1997.

The detailed research of RAMSAY et al. (1999) demonstrates that observing bees in the field

Pollen transfer from bee to bee in the hive

only reveals part of the findings. RAMSAY analyzed pollen loads that bees carried into the hive. Even though in the pollen load, as a rule, the pollen of one plant species prevailed, pollen loads gathered from several plant species were found. Also, along with a main component (99 %), traces of other plant species can be detected (RAMSAY et al. 1999, table 2). A

large-scale distribution of pollen beyond the nearest 40 plants is thus very likely.

In addition, those pollen grains that are not part of the load on the bees' hind legs but are deposited in the hair coat and the wings are of importance. RAMSAY et al. (1999) counted 60,000 oilseed rape pollen grains in the hair coat of one bee. These grains seem to be responsible for the transfer of pollen from bee to bee in the hive, which is why in terms of dispersal distances, the foraging radius of bees around the hive (2 km on average) gains greater significance.

This is confirmed by research by RAMSAY et al. (1999) with 5 visually pollen-free bees caught on the alighting board of the hive, located near a field with transgenic oilseed rape, which were placed onto male sterile oilseed rape plants. Three of the five bees were able to carry out

Three in five visually pollen-free bees were able to fertilise male sterile oilseed rape, 20 % of the seeds were transgenic.

successful fertilisation. 12 of the overall 62 seeds were transgenic. This is a clear indication of the multifold nature of pollen share in a bee's hair coat (RAMSAY et al. 1999).

RAMSAY et al. (1999) concluded that bees can visit several different plant species during a single foraging flight in their search of pollen and nectar, and that after having taken their pollen load into the hive, they carry along many pollen grains with fertilising capacity in their hair coat on their next trip. With an average foraging radius of 2 km of bees around a hive, a significant pollen transfer of 4 km is to be expected. Considering that under certain circumstances, greater distances are covered, even greater distances have to be reckoned with in terms of

the spreading of synthetic transgenes. The results of THOMPSON et al. (1999) support this hypothesis of RAMSAY et al. Despite a very low pollen density at 4 km, they detected

hybridization of male sterile oilseed rape plants and interpreted this as a result of insect-pollination.

Table 1: Overview: Increase of Area Need with Reach of Hybridization

Pollen reach in m	Buffer area need in km²	Buffer area need in ha	Hybridization frequency/ gene flow	source
			0.1 % <b>maize</b>	SCP 200115f no
100	0,03	3,14		details on source <sup>3</sup>
200	0,13	12,57		
300	0,28	28,27		
	,	ŕ	0.06 % oilseed rape male fertile but also 0.6 % at 366 m oilseed rape male fertile	EASTHAM and SWEET 2002
400	0,50	50,27	rape maie leruie	Stringham & Downey, 1978 and 1982 quoted in EASTHAM und SWEET 2002
			ca. 1 % of initial pollen concentration of <b>maize</b> , 427 m	JONES and NEWELL (1946 quoted in TREU und EMBERLIN
500	0,79	78,54		2000)
600	1,13	113,10		
700	1,54	153,94		
			0.2 % <b>maize</b>	SALMOV (1940 quoted in TREU und EMBERLIN
800	2,01	201,06		2000)
900	2,54	254,47		
1000	3,14	314,16		
1500	7,07	706,86	1.2 % oilseed rape male sterile	TIMMONS et al. 1995
2000	12,57	1.256,64	sugar-beet male sterile	SCP 200115f no details on

<sup>&</sup>lt;sup>3</sup> Most maize varieties are hybrids with male sterile seed producing plants inter-planted with rows of pollinators and surrounded by pollinators. This surrounding block of pollinators forms an effective screen against alien pollen since maize pollen is large and flow declines rapidly with distance. French and UK experiments have shown that alien cross pollination is reduced to 0.1% at distances between 20 and 50m into these male fertile barriers. Thus we can advise that current isolation distances can be used to achieve the 0.3% threshold in maize provided current seed production methods continue to be used which include the use of surrounding barriers of pollinators. While these isolation measures may be appropriate in some circumstances, seed producers will need to take account of the density of potential contaminating GM crops in a region, the flowering period of these crops in relation to the seed crop and environmental factors such as wind speed and direction, and adjust isolation measures accordingly (SCP 2001).

				source <sup>4</sup>
2500	19,63	1.963,50	0.8 % <b>oilseed rape</b> male sterile	TIMMONS et al. 1995
3000	28,27	2.827,43		
3500	38,48	3.848,45		
4000	50,27	5.026,55	oilseed rape male sterile (insect fertilisation)	THOMPSON et al. 1999, 97f.
several km			0.3 % <b>oilseed rape</b> male sterile (several kilometers)	SCP 200115f, no details on source <sup>5</sup>

### 2.2 BIOLOGICAL CONTAMINATION BY VOLUNTEERING AND ESCAPE

### 2.2.1 FUNDAMENTALS OF DIASPORE DYNAMICS

#### 2.2.1.1 SPATIAL DIASPORE DISPERSAL

For an estimate of the reach of the gene flow, the transport of seeds (diaspores) has to be

taken into account, in particular if the seeds are able to persist in the soil for several years, such as in oilseed rape.

There are only few experimental data on the dynamics of diaspore dispersal of crops.

Along with the dispersal by pollen, the dispersal of fertile seeds and their persistence in the soil is of great importance for the spatial and temporal introduction and distribution of synthetic transgenes in new gene pools. As in pollen, air, water, and animals are potential vectors for seed dispersal. But the main source of seed dispersal from field to

field is man and transport machines (see below). Distances may vary greatly and depend to a great extent on the dispersal type. For wind dispersal, weight, size, and form (granometeorochor, cystometeorochor (balloon or bladder-like diaspore), trichometeorochor (plumed diaspore), glider, etc.) are of significance. However, these factors alone can only give an approximate estimate of the dispersal, as wind direction, wind force, as well as the population density of the initial population also exert a strong influence. Ocean currents are factors of large-scale dipersal. Fruits of the leguminous plant *Entada scandens*, common in tropical regions, is carried by the Gulf Stream as far as to the coasts of England and Norway (URBANSKA 1992, 39f). The most common agricultural crops, such as maize, oilseed rape, sunflower, are not wind-dispersed but distriubted via animal feed (e.g. birds and mammals) and excretion of the diaspores with the animals' excrements (zoochory) (URBANSKA 1992, 40ff). In zoochorous plants, projections as to dispersal distances, gene flow, and population structure are particularly difficult, given that they depend to a high degree on the mobility of the animal

<sup>&</sup>lt;sup>4</sup> SCP 2001, 15f: "Comments from plant breeders suggest that doubling isolation distances of male sterile mother plants of sugar beet to 2000m will be needed to achieve the threshold for sugar beet seed, assuming that the mother plants are surrounded by non-GM pollinators."

<sup>&</sup>lt;sup>5</sup> SCP 2001, 15f: *Fertility and hybrids:* Experiments with oilseed rape varieties with reduced male fertility have shown that considerably higher levels of outcrossing can occur. Varietal associations commonly have only 20% of plants producing pollen and subsequently have a greater level of outcrossing at any given distance. In addition recent information from North America on the production of hybrid oilseed rape seed, where male sterile plants

are grown in conjunction with pollinators, has shown that these crops have been pollinated by GM crops growing at several kilometres distance. Some samples of imported hybrid rape seed contained levels in excess of 0.3% GM presence. Thus, recommendations on isolation requirements for seed crops of hybrid oilseed rape and beet (which is also commonly produced from hybridisation of male sterile mother plants) to meet 0.3% thresholds cannot currently be made.

concerned as well as the share of the seeds destroyed during digestion (URBANSKA 1992, 278f). Man has come to be the most important factor in the long-range distribution of seeds for crops (NOBLE 1989, HOLZNER 1998, pers. comm.). Man is able to surmount geographical barriers, such as the Alps, and thus significantly widen dispersal areas (URBANSKA 1992, 45f). Humans not only distribute diaspores across wide distances but also in great quantities, e.g. in the form of feed stuff. Losses occurring during shipment and transportation may thus also contribute considerably to "filling" the seed bank along transport routes. Containment of pollen dispersal is very difficult to achieve, and an effective prevention of the spreading of seeds through management measures is practically hardly feasible, as following citation shows:

"Using a chloroplastic marker and a set of nuclear microsatellite loci, the occurrence of crop-to-wild gene flow was investigated in the French sugar beet production area within a 'contact-zone' in between coastal wild populations and sugar beet fields. The results did not reveal large pollen dispersal from weed to wild beets. However, several pieces of evidence clearly show an escape of weedy lineages from fields via seed flow. Since most studies involving the assessment of transgene escape from crops to wild outcrossing relatives generally focused only on pollen dispersal, this last result was unexpected: it points out the key role of a long-lived seed bank and highlights support for transgene escape via man-mediated long-distance dispersal events" (ARNAUD et al. 2003).

### 2.2.1.2 Persistence in the Soil of Seed Banks (Diaspore Banks)

Oilseed rape can remain viable in the soil for more than 10 years. Along with the spatial component, time is also a significant factor in the dispersal of species. The spatial component may be interpreted as the search for new habitats. The formation of seed-banks of long viability serves the purpose of survival during unfavorable living conditions. In forest soils, frequently, large quantities of pioneer plant seeds are found, which, following clearing and windthrow, etc., find adequate conditions for growth and reproduction.

Just as there are different types of dispersal which exert significant influence on the average and maximum dispersal distance of seeds, different strategies may be derived from the longevity of the seeds. In general, seeds of pioneer plants are considered more persistent than those of climax communities (PARKER et al. 1989). However, the maximum persistence of individual seeds in the soil depends to a great extent on environmental conditions, with low temperatures and low water content in the soil as well as in the diaspore favoring persistence.

The reports on the maximum viability of diaspores are very diverse and partly rather spectacular. ODUM (1974 quoted in URBANSKA 1992, p. 61), reports that viable diaspores of Chenopodium album (lamb's-quarters), discovered during archaeological excavations, are estimated to be approximately 1,700 years old. These results, are, however, doubted by BAKER (1989).

The dynamics of a seed bank is not only influenced by the maximum viability of the seeds but also by seed feeders, pathogens, and soil conditions (water content, maximum and minimum temperatures, etc.). For an estimate of the dynamics of the seed bank, these factors reducing seeds in the soil are juxtaposed with additive factors such as germination, establishment and reproduction of the seeds as well as immigration (URBANSKA 1992, 65f, PARKER et al. 1989). For cereal species, the data bank of literature sources on the persistence of seeds in the soil compiled by THOMPSON et al. 1997 gives strongly diverging data. According to this data bank, maize and rye are viable in the soil for less than one year, while for wheat and barley, both short and long times of persistence in the soil are given. RUCKENBAUER and STEINER (1995) demonstrated that under favorable and artificial conditions (low water content, air exclusion, temperatures between 10 and 15 °C), oat and barley as well as some weed seeds

Burying trials by SCHLINK (SCHLINK 1998) have demonstrated that oilseed rape seeds are able to persist in the soil for more than 10 years. With all varieties, an average of 0.5 % of the seeds were viable after 10 years. Recently buried seeds showed a higher average rate of persistence than seeds allowed to afterripen for 6 weeks before being buried.

retain relatively high viability (81 % for oat) even after 110 years.

If the crop is capable of building independent populations remote from the agricultural area, the problem of seed dispersal is aggravated by the fact that these populations serve as "stepping stones" and thus may lead to a very far-reaching dispersal.

### 2.2.2 VOLUNTEERS

Before and after harvesting crops, a certain amound of grains are shed and thus find their way into the soil seed bank. Volunteer growth is the undesired emergence of these crops, which emerge from the soil seed bank in the following year(s) and are thus considered weeds. Particularly cereal and oilseed rape have a high tendency of shedding. Volunteering is best counteracted by incorporating the self-sown cereal immediately after harvesting, in order for as many seeds as possible to germinate and emerge (unless they are dormant). They are then destroyed following one further stubble cultivation (BAEUMER 1992, 354f). Nevertheless, volunteering cannot be avoided entirely. It is not only the volunteering of one crop species in another (e.g. volunteer oilseed rape in cereal) that is undesired. Volunteer growth is also problematic if it occurs in the same crop species (e.g. volunteer oilseed rape in oilseed rape), most of all if it transcends a certain level of damage and thus impairs the crop's quality and marketability.

For GM-plants with herbicide tolerance, volunteering is a difficult problem to cope with. Owing to the fact that volunteers differ only slightly from the crop, they are also herbicide resistant and can no longer be captured with total herbicides. In Canada, oilseed rape plants with resistance against two herbicides have been detected in several fields, in one case even oilseed rape plants with resistance against three total herbicides (HALL et al. 2000).

From the point of view of non-GM production, volunteering is a significant problem, as it is preceded by a long-term contamination risk. As noted above (2.2.1.2), particularly oilseed rape seeds are able to persist in the soil and retain their germinative capacity for more than 10 years, and thus also contaminate GMO-free oilseed rape plants over this period of time, or, if they are harvested along with the non-GM crop, get into the commodity flow. This is also ranked as problematic in the report of EASTHAM and SWEET (2002).

### **Textbox 2: The problem of volunteers**

Volunteering is particularly problematic in oilseed rape and sugar-beet

"Gene flow can occur to and from volunteer and feral populations which act as gene pools carrying over the contamination into subsequent crops. Management systems should be used to minimise GM seed spread on a farm and to minimise seed bank and volunteer populations. Allowing GM volunteer populations to discharge viable seed will cause a large increase in the burden for following crops (Harding & Harris, 1994) through gene exchange from volunteers to crops, and the possibility that GM volunteer plants could be harvested with the crop and passed on to the consumer. (EASTHAM und SWEET 2002, 59f)

Volunteer growth is thus a major obstacle particularly with regard to the return of a GM-production area into a GMO-free area. As opposed to switching from non-GM production to GM-production, which can be done every year without periods of transition, the return from "GMO" to "non-GMO" requires waiting periods of several years. Long waiting periods (periods of readjustment) are, however, extremely undesirable from an economic point of view, considering that there have to be several years of non-GM production without being able to market the commodity as GMO-free.

### 2.2.3 ESCAPE POTENTIAL

Some plant species, e.g. oilseed rape (PASCHER et al. 2000, PESSEL et al. 2001), have a capacity of building up independent populations outside the cultivation area. The dynamics of their spreading depends to a great extent on environmental conditions. Management methods for an effective reduction of feral plants show results at best at an early stage.

After the feral plants have become established in various ecosystems, management measures are only efficient to a limited extent. Feral GMOs constitute a reservoir with a continuous contamination potential for areas of organic or conventional GMO-free cultivation, as reflected in the following quote:

### Textbox 3: Problem of escape of oilseed rape

"Within the scope of transgenic oilseed rape cultivation, these results suggest that more studies on the dynamics of feral oilseed rape are

needed in order to assess more precisely the risks of its invasiveness and its potential impact on genetic pollution between GM fields and non-GM fields" (PESSEL et al. 2001).

### 2.3 Case Examples of Dispersal Distances of Pollen and Seeds

## 2.3.1 OILSEED RAPE (BRASSICA NAPUS)

Flowering Biology	OILSEED RAPE (Brassica napus)
Flowering Biology	Oilseed rape is a facultative self-pollinator. In case of winter oilseed rape ( <i>Brassica napus</i> ), the share of cross-fertilisation is ca. 25 %. This figure may, however, be significantly higher for individual plants and/or lines, and reach values between 0 and 100 % of cross-fertilisation (OLSON 1960 quoted in HOFFMANN et al. 1985, 294f).
Pollination Biology	Pollen transfer in oilseed rape is assumed to occur via insects. Oilseed rape is an important source of pollen and nectar for bees and bumble-bees. SAURE et al. (2000) list honey-bee, bumble-bee <i>Bombus terrestris</i> , as well as other adrena species as the most important pollinators for oilseed rape. Along with bees, other groups of insects have been observed to frequently visit its flowers and transfer pollen - syrphid species, sawflies, bibionid flies, lacewings, blossom (pollen) beetles, weevils, as well as diurnals. In addition, oilseed rape pollen is dispersed by wind, with cool and humid weather resulting in smaller amounts of pollen in the pollen traps than warm and dry weather (SAURE et al. 2000).
Pollen structure	The pollen grain of oilseed rape has typical characteristics for insect pollination: It has a size of 32 – 33 µm and is relatively heavy and sticky. Pollen longevity is ca. 24 hours up to 1 week (MESQUIDA & RENARD 1982, zit. in EASTHAM und SWEET 2002).
Pollen reach	In oilseed rape, the dispersal distances given for successful fertilisation fluctuate strongly and can be interpreted only on the basis of an exact description of the experimental design.  When male sterile oilseed rape plants were used as recipient plants (cf. description under 2.1.1.3.1), significantly greater distances were measured than with male fertile oilseed rape plants. This is attributable to the fact that the hybridization frequency eventually corresponds to the ratio of pollen availability (of the plants' own or foreign origin).  Experimental measurements of hybridization frequencies with male sterile plants (measured at male sterile oilseed rape plants whose petals were removed in order to make them less attractive to bees) ranged from 0.8 % at a distance of 2.5 km and 1.2 % at a distance of 1.5 km (TIMMONS et al. 1995) as well as 3.7 % at a distance of 360 m (TIMMONS et al. 1996a). At a distance of 360 m from an oilseed rape field of about 10 ha, the pollen concentration was about 10 % of that of the field edge. Low pollen densities (on an 11-day average 1.57 pollen grains per m³ and 24 hours, max. 27 pollen grains per m³ and hour), which, however, evidently sufficed for the

Flowering Biology	OILSEED RAPE (Brassica napus)
	fertilisation of male sterile oilseed rape plants, were measured at a distance of 2.5 kilometers.
	Similar results were gained in experiments with male sterile oilseed rape plants by THOMPSON et al. (1999). On one of the experimental plots, hybridization occurred with a certain oilseed rape genotype whose nearest pollen source was about 4 km away. As the pollen density at this site was, however, less than one pollen grain per m³ and day, this special case was in all probability a case of insect-fertilisation (THOMPSON et al. 1999, 97f). SCHEFFLER et al. (1995), too, conclude on the basis of their experiments that long-range pollen transport of oilseed rape pollen is in all probability accounted for by bees (SCHEFFLER et al. 1995). Measurements with male fertile oilseed rape plants resulted in significantly lower values, with values fluctuating strongly. They are around 0.6 % at 366 m (STRINGHAM & DOWNEY, 1978 and 1982 quoted in EASTHAM und SWEET 2002), and one tenth of that value, i.e. around 0.06 % at 400 m (SIMPSON unpublished quoted in EASTHAM und SWEET 2002, 17f). The margin of fluctuation might either be explained with the share of male sterile plants within one variety (which results in higher frequencies at the same distances), or with insect
	fertilisation (EASTHAM und SWEET 2002, 17f).  These data show that male sterile varieties and/or varieties with a high share of male sterile plants (e.g. the variety SYNERGIE with 80 % male sterile plants, EASTHAM und SWEET 2002, 18f) are clearly more affected by contamination.
Reach of passive seed dispersal	The determination of passive dispersal distances is very difficult, but is estimated at several kilometers. Factors of passive dispersal are humans (harvesting machines, transport of feed, as well as birds and mammals (cf. also 2.2.1.1). In addition, oilseed rape is capable of establishing independent populations outside agricultural cultivation areas (PESSEL et al. 2001), which, again, may serve as "bridgeheads" for further dispersal.
Persistence of seeds	Burying experimens with oilseed rape seeds demonstrate that the germinative capacity of oilseed rape seeds decreases exponentially over the first 5 years, and then decreases only slightly on a low level. Of all varieties, after 10 years, an average of 0.5 % of the seeds were still viable. Recently buried seeds had a higher average rate of persistence than seeds allowed to afterripen for 6 weeks before being buried (SCHLINK 1998). The viable persistence of oilseed rape seeds is also influenced by primary dormancy (species-specific) and secondary dormancy.  By way of targeted post-harvest management (turning-under of germinated oilseed rape seed after harvesting; refraining from intersowing catch crops, which prevents the growth and renewed seeding of oilseed rape), it is possible to reduce the probability of oilseed rape volunteers within five years to 1 volunteer oilseed rape plant per 1,000 oilseed rape plants (0.1 % or 1 plant per 100 m², which equals 100 plants/ha) (SCP 2001). In some EU member states, areas can only be used for the production of oilseed rape seed after 7 years without oilseed rape cultivation. In view of the fact that even without sowing oilseed rape, the potential for renewed seeding on account of oilseed rape volunteers is relatively high (PEKRUN et al. 1999) and the seed stock in the soil can thus be refilled, even longer interim periods are conceivable for the production of non-GM oilseed rape seed, for which demands of purity are significantly higher than for conventional seed.

Flowering Biology	OILSEED RAPE (Brassica napus)
Hybridization partners	Along with genetic contamination on account of regular oilseed rape cultivation as well as unintended oilseed rape volunteering, genetically contaminated field associate flora (wild relatives of oilseed rape) constitutes a permanent contamination potential of agricultural crops over thousands of years and of unknown dimension in terms of spreading. The extent (the possible percentage) of the contamination potential depends on several factors (overlap of the flowering dates, nature of the oilseed rape cultivated, dynamics of spreading, flowering biology of the contaminated wild herbs, etc.) and can hardly be assessed with any degree of certainty. Currently, we know of five wild relatives of oilseed rape that are able to receive a synthetic transgene by way of hybridization (JORGENSEN 1999).
	In Austria, these are (PASCHER et al. 2000, 131ff):  • Brassica napus (wild form of oilseed rape)  • Brassica rapa (turnip rape)  • Raphanus raphanistrum (wild radish)  • Diplotaxis tenuifolia (slimleaf (perennial) wall rocket)  • Sinapis arvensis (field mustard/yellow charlock)
	And the following crops:  Brassica oleracea ("cabbage" or "kale", which refers to several cabbage species, including white cabbage, red cabbage, turnip cabbage, cauliflower)  Raphanus sativus (wild radish, including a.o. small radish, garden radish,)  Sinapis alba (white mustard)  Sinapis nigra (black mustard – rare in Austria)
	There is also a danger that "bridge building" will allow the spreading of synthetic genes also to plant species that are per se not hybridizable with oilseed rape (PASCHER et al. 2000, 132f).
CONCLUSIONS	From an ecological point of view, any genetically modified crop that is able to pass on its synthetic transgenes to wild relatives cannot be brought in line with the principle of precautionary environmental and nature protection. We currently do not know the "half-life" regarding the persistence of synthetic transgenes in natural populations. Moreover, the term "half-life" can only be applied to a limited extent - if at all - to organisms capable of reproduction (as opposed to persistent chemicals). According to the current status of knowledge, synthetic transgenes will persist in wild populations (ARRIOLA und ELLSTRAND 1997a, ELLSTRAND 2001).even if there is no selective advantage (ADAM und KÖHLER 1996). Owing to the fact that environmental hazards can never be assessed to their full extent, any damage to the environment caused by synthetic transgenes, recognized at a later date, can merely be registered. There are no management measures to go back to the status quo ante. Along with the great environmental damage potential of GM-oilseed rape, fears are justified that a genetically contaminated field associate flora might continuously contaminate crops.

## 2.3.2 MAIS (ZEA MAYS)

Flowering Biology	MAIZE (Zea mays)
Cross-pollinator	Maize is a monoecious cross-pollinator.
Flowering biology	Pollen transfer occurs mainly by wind. Maize pollen is an important pollen source for bees and bumble-bees and is also detectable in honey. Maize is monoecious, i.e. the male and the female inflorescence are totally separated on the plant. Fertilisation by bees is unlikely, in particular as the female flowers do not excrete nectar and are thus, as opposed to maize pollen, not attractive to bees.
Pollen reach	As maize pollen is relatively heavy, it sinks to a greater extent in the immediate surroundings than that of other cross-pollinators. Within the scope of the present research, no current data was found on the maximum outcrossing distances for maize. This is in accordance with the results of a study of TREU and EMBERLIN (2000), who studied the relevant literature between 1940 and 1950. SALMOV (1940 quoted in TREU und EMBERLIN 2000) found the following average hybridization events in maize: 3.3 % (10 m), 0.3 % (50 m), 0.4 % (100 m), 0.3 % (150 m), 0.5 % (200 m), 0.02 % (400 m), 0.1 % (500 m), 0.8 % (600 m), 0.2 % (700 m), 0.2 % (800 m) as well as JONES and BROOK (1950 quoted in TREU und EMBERLIN 2000): 25.4 % (0 m), 13.1 % (25 m), 6.1 % (75 m), 3.1 % (125 m), 1.6 % (200 m), 0.7 % (300 m), 0.3 % (400 m), 0.2 % (500 m).  RAYNOR et al. (1972 quoted in TREU und EMBERLIN 2000) found in their experiments with pollen traps at a distance of 60 m as much as 5 % of the pollen concentration measured 1 m away from the field edge. At a distance of ca. 427 m, JONES and NEWELL (1946 quoted in TREU und EMBERLIN 2000) found ca.1 % of the initial pollen concentration in maize. Pollen traps only give an approximate indication of potential hybridization. The results of SALMOV as well as JONES and BROOK are clearly of higher quality, on account of the fact that hybridization occurances were measured.
Reach of seed dispersal	The reach is very difficult to determine, but is estimated at several kilometers. Dispersal factors are man (harvesting machines, transport of feed) as well as birds and mammals (cf. also 2.2.1.1). As opposed to oilseed rape, maize is in Austria (Europe) not capable of establishing independent populations outside agricultural cultivation areas. In addition, maize is not perennial, which is why there is no danger of volunteering even on areas of agricultural cultivation.
Persistence of seeds	The potential of maize kernels to perennate in the soil and remain viable is considered very low. Maize kernels are thus persistent in the soil for less than one year (THOMPSON et al. 1997).
Hybridization partners	Maize has no natural hybridization partner in Austria.
CONCLUSIONS	Maize is not capable of establishing independent populations outside agricultural areas. Passive dispersal of the diaspores (by man or animals) causes short-term contamination risks. However, as maize is not perennial, the winter brings wild populations to an end, provided no renewed passive dispersal occurs. As maize is (almost) 100 % cross-pollinating and pollen can be wind-dispersed across wide distances, long-range pollen contamination is the most important risk factor in maize when it comes to the question of coexistence. The reach of successful fertilisation is up to 1 km, which may be significantly exceeded depending on the wind force and local structural conditions.

### 2.3.3 SOYBEAN (GLYCINE MAX)

Flowering Biology	SOYBEAN (Glycine max (L.))
Cross- /Self-pollinator	Soybean has a hermaphrodite flower and is to a great extent self-pollinating. The share of natural cross-pollination is around 0.5 %, although lines can be selected that have a higher tendency to cross-pollinate (HOFFMANN et al. 1985, 176f).
Pollination biology	In Europe, its pollen is only very rarely transferred by insects, as the flower excretes only little nectar (HOFFMANN et al. 1985, 176).
Pollen reach	no specific data.
Reach of passive seed dispersal	Determining the reach is very difficult, but it is estimated at several kilometers. Dispersal factors are man (harvesting machines, transport of feed), as well as birds and mammals (cf. also 2.2.1.1). As opposed to oilseed rape, soybean is not capable in Austria (Europe) of establishing inependent populations outside agricultural cultivation areas. In addition, soybean is not perennial (WAHL 1991), which is why even on areas of agricultural cultivation, there is no danger of volunteering.
Persistence of seeds	The potential of soybeans to perennate in the soil and remain viable is considered very low.
Hybridization partners	Soybean has no natural hybridization partners in Austria.
CONCLUSIONS	Soybean is not capable of establishing independent populations outside areas of agricultural cultivation. A passive dispersal of the seeds (by man or animals) leads to very low short-term contamination risks via pollen, as soybean is predominantly self-pollinating. Given that soybean, like maize, is not perennial, contaminaion with GM-soybean - provided there is no renewed passive dispersal - ends with the winter. Pollen transfer via insects is not very likely owing to its insignificant production of nectar, but may be more than 4 km in reach.

### 2.4 TECHNICAL CONTAMINATION

Technical contamination may cause a concentration of contamination.

The analysis of technical contamination with GMOs or GMO-derivatives is not part of this study. In the interest of completeness, however, works relating to this subject and the most important aspects should be mentioned.

The danger of contamination on account of technical contamination exists during the entire process (harvest and processing) and logistics chain (transportation, shipping, storage) of agricultural raw materials. An efficient segregation of non-GMO and GMO-containing raw materials is only possible in case of a local concentration of various processing steps. PASCHER et al. found oilseed rape ruderal populations on remote sites such as the Donauinsel ("Danube Island," an artificial island in Vienna) or a railway track in Lower Austria. The authors assume the deposition of excavation material

(dug earth) on the "Danube Island" and losses of seed during rail transportation, respectively, to be the causes for the emergence of these populations (PASCHER et al. 2000, 112f).

Via the production chain, contamination may be concentrated but also diluted. The SCP (Scientific Committee on Plants) of the EU Commission gives a brief overview of possible rates of concentration of foreign contamination within the processing chain (SCP 2001).

Table 2: Overview of concentration of foreign contamination with GMOs within the processing chain (SCP 2001, 81f).

Table 1. Estimated average potential rates of adventitious presence occurring at various stages during on farm production.

	Oilseed rape (fully fertile)	Maize	Sugar beet
Seed	0.3%	0.3%	0.5%
Drilling	0%	0%	0%
Cultivation	0%	0%	0%
Cross pollination	0.2%	0.2%	0%
Volunteers	0.2%	0%	0.05%
Harvesting	0.01%	0.01%	0.01%
Transport	0.05%	0.01%	0.01%
Storage	0.05%	0.05%	0.1%
% achieved	0.81%	0.57%	0.67%

**SCP 2001** did not cover contamination during the processing of food and feed. It is clear, however, that this is a major contamination point. WENK et al. (2001) give a detailed view of ways of contamination within a processing plant. GMO-traces had been detected in 20 % of the organic production when GMO feed was processed previously. An Austrian study yet to be finished is investigation ways of contamination in Austrian feed processing plants.

# 3 CHECK LIST OF ITEMS WITH POLITICAL REGULATORY NEED REGARDING COEXISTENCE

The following summary is to provide an overview of items to be regulated when it comes to regulating coexistence.

### 3.1 DETERMINATION OF PROTECTION GOALS

The determination of protection goals is of central importance, as detailed measures are derived from there. Defining protection goals and priorities for the Austrian agriculture is a purely socio-political task; deriving the necessary measures to attain the protection goals a task of both natural and socio-economic sciences. The ultimate question is: What agricultural practice is

- desirable and
- economically and ecologically viable

for Austria? In the following, some possible protection goals are listed, which individually or in various combinations may have to be considered in the context of regulating coexistence.

### 3.1.1 Possible Protection Goals

### **Protection of Organic Farming**

By now, ca. 9 % of the Austrian agricultural areas are areas of organic cultivation. By now, ca. 9 % of the Austrian agricultural areas are cultivated organically (cf. footnote 1). In terms of trade, organic farming products have been among the fastest-growing segments over the past years. This development was influenced in particular by the food scandals of the past years, e.g. BSE. The protection of organic cultivation is thus

certainly the central focus of the question of coexistence. It is certainly not only maintaining the current share of organic farmers that is important but also expanding organic farming. While organic farming is more or less stagnating in grassland areas, it is growing in arable areas by an average 8-10 % per year (MÜLLER 2000, tables 1 and 2, 19ff).

Securing the production opportunities of organic farming over a long term is not only a necessity from the consumers' point of view. It is also supported from the point of view of climate protection. The "Enquete-Commission of the Bundestag (German Lower House of Parliament) for the Protection of the Global Atmosphere" (DT.BUNDESTAG 1994) recommends a "significant expansion" and support of organic farming, in order to prevent further environmental damage and avoid, among other goals, the emission of greenhouse gases by agriculture. The contribution of organic farming to reducing stresses to the atmosphere and underground water as well as to increasing and securing food quality and the diversity of species is increasingly recognized. Among the current agricultural cultivation practices in Central Europe, organic farming is considered the practice availing the highest degree of ecologization, and thus comes closest to the principles of sustainability (SUSTAIN 1994, SRU 1985).

### **Securing of GMO-free Land Reserves**

Depending on the GM-crop admitted, a return to organic and/or GMO-free production is only possible after transition periods of several years. Along with transgenic volunteer growth, a potential enrichment with toxins might negatively effect the quality of production areas. Bt toxin, for example, which transgenic maize varieties released into the soil, was detectable in the soil over the entire observation period of 180 days (maximum duration of experiment) (STOTZKY 2001). Bt toxins, fixed to clay minerals, may continue to persist in the soil beyond this experimental period. If new scientific findings were to reveal serious health risks from the consumption of GMO-products, market turbulences similar in vehemence to those caused by the BSE crisis were to be expected. A sufficient share of GMO-free production area would provide a buffer capacity to check such fluctuations. Furthermore, once problems have been recognized, alternative technologies have to be available. A switch of production modes requires practical knowledge that is continually applied and further developed.

### Maintaining the Possibility of Epidemiological Studies (Monitoring)

Over the past years, a significant rise of various clinical syndromes (allergies, childhood cancer POLLAN et al. 1997, HJALMARS et al. 1999, SKLAR 2002, MCKINNEY et al. 2003) have been registered by way of scientific methods. It is very difficult to attribute these syndromes to respective causes. Nevertheless, such examinations are of great relevance from the point of view of preventive health care. Attributing potential factors to certain syndromes is only possible by way of epidemiological studies, which allow a statistical evaluation of differences in the way of living and possible correlations. If the freedom to chose GMO-free nourishment cannot be guaranteed, it cannot be ascertained whether or not GMO food products are potential factors in the development of clinical syndromes. As risk assessment can never give full certainty (safety) (cf.EEA 2001), monitoring measures in the sense of early warning systems are of great importance. As monitoring is an integral part of EU Directive 2001/18/EC (EU 2001), securing the option of GMO-free nourishment is a requirement also under this aspect.

Other protection goals to be considered:

- Protection of Ecologically Sensitive Areas (see HOPPICHLER 1998, HOPPICHLER 1999)
- Protection of GMO-free Seed Breeding
- Maintaining a Small-structured, Farm-centered and Sustainable agriculture
- Precautionary Principle Uncertainties of GMO Risk Assessment Wide Gaps of Knowledge in Fundamental Research (see chapter 1.2)

### 3.2 SET OF RULES

Based on the protection goals, rules have to be defined. These rules should provide clear answers for the following questions:

- How is the exchange of information regarding cultivation plans and crop rotation organized,
   e.g. which information has to be submitted by farmers to whom and at what time?
- Who is responsible for applying anti-contamination measures or obeying minimum isolation distances? (A rule as "who has the right of way" on the farmland.)
- Who bears the costs of anti-contamination measures?
- What are the minimum isolation distances for each crop?
- How is surveillance undertaken and who bears the costs?
- Who has to take liability in case of contamination beyond the admitted threshold?

One of the basic questions is who will account for the costs and be responsible for carrying out anti-contamination measures or the obeying of minimum isolation distances. Simply speaking, this means: Who has the right of way in case of conflict? Who has the right to go ahead with his or her cultivation plan and who will be required to look for alternatives, e.g. planting conventional maize varieties or an entirely different crop?

The EU-Commission gives a first hint to this kind of problem: "As a general principle, during the phase of introduction of a new production type in a region, operators (farmers) who introduce the new production type should bear the responsibility of implementing the farm management measures necessary to limit gene flow" (EC-COMMISSION 2003, 2.1.7).

While this could be considered a protection goal for existing organic farmers, this rule could result in a considerable burden for farmers (or their sons and daughters) willing to switch to organic farming in the future. Finally, it might lead to "freezing" organic cultivation area to the size of today.

Some groups have recommend establishing a legally binding regulation on "GMO cultivation and processing guidelines," such as EC Regulation 2092/91 for organic farming. This regulation on "GMO cultivation and processing" should describe how GMOs are cultivated and handled from farm to fork, to avoid contamination to other crop-systems (conventional and organic). Like organic farmers and processors, GMO farmers and processors have to ensure that no contamination occurs. This means that GMO farmers and processors have to build up their own system of GMO production, as organic farmers and processors did in the past. This requires separate harvesting, transporting, storing, processing etc. An inspection system such as that applied in organic farming has to be implemented.

Such a system could help organic and conventional GMO-free farmers to reduce costs, considering that they are already bearing the cost of coexistence. Although GMOs are not cultivated in Austria, food and feed containing GMOs (particulary soybean) is on the market. Anti-contamination measures are already handled at the expense of organic farmers, processors and inspection services. There had been significant investments by processors especially in the feed industry to exclude or minimize GMO contamination in the organic processing chain. In some cases, processors stopped producing conventional feed and switched to solely producing organic feed to ensure GMO-free organic production. Furthermore, the Austrian organic inspection services are continuously screening organic feed and food for GMO contamination. (Personal communication, Schörpf M, Organic farming association ERNTE f das Leben Lower Austria).

### 3.3 THRESHOLDS FOR CONTAMINATION

Contamination thresholds have to be established, as they play a role in determining the size of the buffer zones. Lower thresholds require larger buffer zones, higher thresholds demand smaller buffer zones. The following thresholds were established for Austria:

Table 3: Thresholds established for GMO contamination in Austria

Threshold	Valid for	Reference
0.1 %	Seed of the following species:	BMLFUW 2001
	Swede (Brassica napus L. var. napobrassica)	
	Maize (Zea mays)	
	Oilseed rape (Brassica napus)	
	Turnip rape (Brassica rapa)	

	Soybean (Glycine max)	
	Turnip (Brassica rapa L. var. rapa)	
	Tomato (Lycopersicon lycopersicon) as processing varieties	
	Chicory (Chichorium intybus L.)	
0.1 %	For food ingredients and food processing aids, feed ingredients and feed processing aids, fertilisers and soil improvers	BMSG 2001

The EU has set a 0.9 % threshold for genetically modified food and feed (EU Regulation on genetically modified food and feed, approved by the Council and the Parliament in July 2003, but not yet published), but has not yet established thresholds for adventitious contamination of seeds and organic farming products. Whether the 0.9 % threshold in the final product is also valid for organic products remains unclear. In the opinion of the European Commission, "The organic farming regulation does allow for the setting of a specific threshold for the unavoidable presence of GMOs, but no threshold has been set. In the absence of such a specific threshold, the general thresholds apply." (Original quote from EC-COMMISSION 2003, 2.2.3). Others hold an opposite opinion: Without any threshold set in EC Regulation 2092/91, the detection limit of 0.1 % should be considered the valid threshold (cf. TOLSTRUP et al. 2003). Some consider a 0.9 % tolerance level not compatible with organic consumer interests. To meet consumers interpretation of organic or GMO free products the threshold should be based on the detection limit (KC Grütter discussion remark in WENK et al. 2001, 104f).

Experts (Heissenberger UBA, pers. communication 2003, Moder ABG, pers. communication 2003) remind that a threshold of 0.1 % is only achievable if a complete segregation from farm to processing unit is undertaken. That means that GMO processing is completely separated from GMO-free or organic processing. In Austria, organic feed processing is completely separated from conventional feed processing (which contains GMO soybean) in order to get organic GMO-free feed.

It remains unclear if processing of GMOs and conventional GMO free products is possible to observe the  $0.9\,\%$  threshold.

Experts (Heissenberger UBA, pers. communication 2003, Moder ABG, pers. communication 2003) estimate that during processing, a minimum contamination of 0.5 % will take place. Processors will therefore require food or feed delivered with about 0.3 to 0.4 % contamination. The following table gives a short overview of contamination rates during the production chain.

Table 4: Overview of GMO contamination rates during the production chain

Production steps	Con- tamination rate	Remaining threshold	Con- tamination rate	Remaining threshold
Threshold in final product		0.90 (conventinal)		0.10 (organic)
Contamination during processing	0.50	0.40	0.00	0.00
Storage	0.05	0.35	0.00	0.00
Transport	0.05	0.30	0.00	0.00
Harvesting	0.05	0.25	0.05	<u>0.05</u>
Cultivation (drilling, cross-pollination, volunteers)	0.20	0.05	0.05	0.00
Seed	0.05	0.00	0.00	0.00

Table 4 clearly shows that for seed purity and cultivation together a threshold of about 0.25 % (0.05 %) in organic crops has to be met. If seed purity of 0.00 % is guaranteed, a contamination tolerance of 0.25 % during cultivation is possible. For crops without volunteer growth, cross-pollination which leads to a maximum of 0.25 % has to be considered in the calculation of

isolation distances. If there is a tolerance level for seed purity, there will be a lower tolerance level for cross-pollination. In the case of oilseed rape volunteer growth, which is assumed to contribute to 0.2 % of the contamination (cf.Table 2), the margin for cross-pollination and seed contamination is almost zero.

For an organic production with a threshold of 0.1 % in the final product, a total separation of the production chain is needed. This will mean separate harvesting, transport machines and a totally separated processing unit. In Austria, the feed processing industry already uses totally separate production units for organic feed.

### 3.4 ISOLATION DISTANCES

Based on remaining thresholds (if contamination rates through processing, storage, transport, harvesting are taken into account, cf. Table 4 above), minimum isolation distances have to be determined. Minimum isolation distances are one of the central policy decisions to be taken. Measures to ensure coexistence at the farm level, neighbourhood level, and regional level are derived from minimum isolation distances.

There is the possibility to describe minimum isolation distances for every crop, or to form two groups of crops.

- 1. Self-breeding crops which require a certain isolation distance, e.g. 60 m, and
- 2. outcrossing crops for which an isolation distance of e.g. 1,000 m is required.
- 3. Besides, a red list of crops might be drawn up which are incompatible with Austrian nature protection goals, e.g. all crops that have the potential to outcross with wild species in Austria, such as oilseed rape.
- 4. A discussion on minimum isolation distances is provided in chapter 4.4.3.

### 3.5 LIABILITY

If anti-contamination measures prove to be insufficient e.g. on account of false assumption and models, improper implementation, or neglecting of coexistence rules by farmers, a clear liability program has to be in place before GM cultivation starts.

### 3.6 SURVEILLANCE

The success of a measure ultimately lies in the extent to which the conditions set can be met to the highest percentage possible using efficient means. This requires an efficient system of checks and inspections, modelled, for example, after the quality assurance system of the Austrian Agri-Environmental Program and/or the surveillance system for organic farming. This system establishes:

- goals,
- critical control points,
- measurement density,
- measures (sanctions) in case of deviations, and
- review and monitoring of the surveillance activities.

On the basis of the data of the two previous points, it has to be examined whether:

- the measures established have been observed.
- the measures established (provided they have been observed) guarantee protection against contamination with GMO pollen, and thus whether the buffer zones have been sufficiently or insufficiently dimensioned.

Any system only works well as long as all persons involved adhere to the agreements made. Bearing this in mind, regulations (SANCTIONS) have to be devised for the case of deliberate violations of the agreements made. Beyond that, it has to be settled "WHO" will carry out the checks and surveillance, and "HOW" it is to be carried out (time-frame, details of drawing samples, etc.)

# 4 ANALYSES OF PROPOSED APPROACHES TO POSSIBLE SOLUTIONS

### 4.1 EC RECOMMENDATIONS

The European Commission recommends that every measures must be

- crop-specific
- production-specific (seed production or food/feed production)
- step by step from farm to region.

In this section, possible approaches to solving the problem of coexistence are examined on a crop by crop level as to their efficiency and practicability. Requirements to practically avoid technical contamination through harvest, transport and processing are not considered. The following analysis focuses on the long-term securing of a coexistence of ecological and conventional non-GM agricultural production and the cultivation of GM crops, with an emphasis on scientific requirements as well as on aspects of practicability. Every approach has to consider the following points:

- Pollen flow on the air current and by wind or distribution by insects
- Seed transport
- Volunteering
- Accumulation factor (through seed savings)

Based on the recommendations by the European Commission, the management options are divided into three management levels:

- Management at farm level
- Management at neighbourhood level
- Management at regional level

On a case by case basis and step by step, coexistence measures should be first applied at the farm level. If there is no sufficient reduction of contamination, measures/tools of the next management level have to be adopted.

The European Commission is referring to anti-contamination measures via genetic engineering methods (EC-COMMISSION 2003, 2.2.7). Therefore, the author gives a brief summary on the current state of the art of what is called biological containment.

# 4.2 MEASURES AT THE SEED LEVEL – GENETIC ENGINEERING APPROACHES – BIOLOGICAL CONTAINMENT

Biological containment through genetic engineering methods is considered to provide some approaches to solve the coexistence problem. According to EASTHAM und SWEET (2002), the following approaches are worth considering with regard to GMOs:

- Apomixy production of seed without fertilisation
- Cleistogamy self-pollination without opening of the flower.
- Prevention of flowering by subsequent control of flowering through application of chemical elicitors.
- Male sterility that prevents not only flowering but also pollen development.
- Plastid transformation not the DNA of the nucleus but merely the DNA of the plastids is transformed. In many cases, this allows to prevent transgenic pollen, as plastids are maternally inherited in most macrophytes.
- Sterile seed is made possible by using genes that prevent seeds' germinative capacity.

EASTHAM und SWEET (2002) unfortunately do not indicate whether and to what extent these possibilities are merely theoretical in nature, or whether they have already been further

developed. A review from DANIELL (2002) confirms the preliminary character of these approaches but states that: "As yet, however, no strategy has proved broadly applicable to all crop species, and a combination of approaches may prove most effective for engineering the next generation of GM crops (DANIELL 2002).

Male sterility a fully developed approach: EEC Directive 90/220/EEC admits the release of two male sterile oilseed rape lines, MS1, RF1 and MS1, RF2 (system for the production of hybrid seed and herbicide tolerance with phosphinothricine as effective agent - Register of Organisms Containing GMOs #06 and #07 (both so far without national admission, status: 18 April 2002) as well as one male sterile line of chicory (*Cichorium intybus L.*), Register of GMOs #04 - http://www.gentechnik.gv.at/gentechnik/set/recht\_set.html). Male sterile plants (which through respective mutants have already found their way into conventional cultivation) are required in hybrid breeding in particular for bisexual (hermaphrodite) flowers in order to prevent self-pollination.

For the purpose of profit, the use of male sterile plants as a rule does not make sense, as it may result in yield losses. Whether the restoration of male fertility through spraying with respective chemicals would be a promising approach remains to be seen. Here, too, the key issue is whether the approach entails yield losses.

**Plastid transformation**, as a further approach, is fairly advanced. Plastids are supposed to derive from endosymbiotic bacteria and thus have a transcription and translation mechanism of procaryotic origin. Thus, for example, unmodified Bt kurstaki HD73 crylA( c ) genes were not built into the nucleus of a tobacco cell but into the plastom, the genome of plastids such as chloroplasts. As plant cells contain up to 50,000 copies of one plastid, the delta-endotoxin gene existed in several thousand copies per cell in transgenic plants. Plastid transformation resulted in a very high accumulation of 3-5% soluble protein (Bt toxin) in tobacco cells and an extremely high toxicity for the insects Tobacco budworm (*Heliothis virescens*), Corn earwom (*Helicoverpa zea*) and beet armyworm (*Spodoptera exigua*). Along with the high expression of unmodified Bt toxin, this method allows to prevent position effects, as the transgenes are introduced into the plastid genome by homologous recombination. Equally, the transgenes contained in the plastids are not expressed in the pollen, and the genetic information is exclusively maternally inherited (MCBRIDE et al. 1995). This might be a containment strategy for gene flow via pollen. The containment of gene flow via seeds i.e. volunteers seems to be quite limited (HANSEN et al. 2003).

**Seed sterility**, as a further approach, is fairly advanced, but it will just help to reduce the risk of volunteers but do not cover the risk of contamination through cross-pollination.

All other approaches appear to be rather theoretical at this point.

### CONCLUSIONS: BIOLOGICAL CONTAINMENT

Biological containment faces several problems:

**Prelimary status:** Except male sterility, seed sterility and plastid trasnformation all other approaches appear to be rather theoretical at this point.

Its effect is uncertain. Seed sterility is only of advantage in volunteer control, but contamination by cross-pollination is not affected. In many approaches, the stability of expression is not fully developed. Thus, the protective effect (prevention of pollen emission) can only be guaranteed for a certain percentage. The extent of the contamination potential is uncertain, as there are presently no known results (to the knowledge of the author) of respective cultivation attempts.

**Yield.** With regard to most crops, the grain is the central unit of yield. Any measures that cause a change in flowering biology might also result in respective yield losses, which is why even technically highly developed approaches are unlikely to be put into practice from an economic point of view.

The problems of volunteer growth and airborne pollen movement can only be solved by a combination of approaches that prevent flowering and approaches that impair the seed's germinative capacity. As the individual approaches are technically insufficient at the moment, it is doubtful whether and when a combination of these approaches may be put into practice. Non of current applications which had been submitted for approval by the companies, has

"build in" anticontamination measures. Biological containment is not able to provide solutions for current coexistence problem.

#### 4.3 **MANAGEMENT AT FARM LEVEL**

#### 4.3.1 **BARRIERS**

Even though it is a frequent argument that planting barriers might be a remedy to reduce outcrossing distances, this area has not yet been subject to thorough scientific investigation (HOKANSON et al. 1997). The key issue in need of clarification is what factors allow a reliable diminishing of dispersal distances as well as a reduction of the amount of transgenic pollen. In his literature analysis, INGRAM (2000) repeatedly notes that border planting helps reduce outcrossing frequencies. According to INGRAM, the reduction of the outcrossing frequency and the maximum outcrossing distance is most effective by diluting pollen with non-transgenic pollen. Hedgerow plantings in the form of rows of trees, for example, lead to a decrease in effectiveness in shortening distances and/or reducing the frequency of hybridisation (INGRAM 2000). New proposed rules from APHIS-USDA which don't allow the use border rows on field test plots with pharmaceutical corn, seem to underline the uncertainty associated with the establishment of border rows or barriers to reduce gene flow."(APHIS-USDA 2003b).

#### 4.3.2 **DILUTION WITH NON-TRANSGENIC POLLEN**

Dilution with non-transgenic pollen by planting mixtures or rows of conventional varieties is another approach to reduce cross-pollination that is being discussed. This approach is successful primarily in wind-pollinaters and demands that:

- the flowering times of transgenic and non-transgenic varieties occur largely simultaneously
- the growth height of transgenic varieties be lower or equal to that of the non-transgenic

Admixture of the available pollen with non-GM varieties dilutes the GMO share, which decreases the statistical probability of fertilisation for the GMO share. It remains unclear if that approach will be adopted by farmers who want avoid separate workload through row planting. Dispersed mixtures may provide a practical solutions to farmers but will reduce some "benefits" of GM crop. Moreover, in herbicide tolerant crops, seed mixtures will not be applicable.

#### 4.3.3 **CROP-SPECIFIC CONCLUSIONS FOR MEASURES AT FARM LEVEL**

For maize and oilseed rape, these measures do not guarantee coexistence. Further measures at the next management level have to be implemented. The risk of contamination by cross-pollination could be sufficiently reduced for soybeans. No further requirements at a higher management level may be necessary. Yet, while soybeans are also fertilised by bees, further studies are needed to prove this hypothesis.

### MANAGEMENT AT NEIGHBOURHOOD LEVEL

#### 4.4.1 **PREFACE**

The idea of a coexistence of "organic" and "GMO" on the basis of flexible coordinated cultivation management measures is not new and has been proposed time and again since the start of the discussion on possible modes of coexistence. The EEA Report also gives mention to this concept: "Neighbouring farms should inform each other of their planting intentions in order for appropriate isolation measures to be considered." (EASTHAM und SWEET 2002, 59f). But the authors give no recommendations how conflicts could be settled with this approach.

The European Commission recommends (EC-COMMISSION 2003, 3.3):

- Voluntary clustering of fields of different farms for the cultivation of similar crop varieties (GM, conventional or organic) in a production area.
- Use of crop varieties with different flowering times.
- Arranging for differences in sowing dates in order to avoid cross-pollination during flowering.
- Coordinating crop rotations.

No details are given how farmers should come to agreements on sowing dates, use of specific varieties, clustering fields etc.

The following is a discussion of a selection of the most important proposals for cultivation management, without differentiating between measures considered meaningful only in combination with other measures, and measures considered meaningful by and in themselves.

### 4.4.2 SELECTING VARIETIES WITH DIFFERING FLOWERING DATES - DIFFERENT SOWING DATES

Different varieties and different sowing dates are dealt together because different sowing dates are in many cases associated with a different variety.

By selecting varieties with differing flowering dates, the requirements of cultivation management might, even if the crop species remain the same, be reduced to a minimum, and the farmer would not be limited in his or her selection regarding crop rotation. However, significant factors speak against this approach:

This proposal could only be put into practice if varieties were chosen that vary significantly with regard to their flowering dates, and provided that there is no **overlap of flowering** (flowering

The choice of differing varieties can neither theoretically nor practically guarantee sufficient protection against cross-contamination.

periods). In most cases, however, this is unlikely. In a cultivar evaluation trial using 12 varieties of winter oil-seed rape, for example, the maximum difference in the start of the flowering time was 4 days (SCHULZ und JAKISCH 2001). The flowering period of approximately 3 - 4 weeks, however, leads to considerable overlaps in flowering (INGRAM 2000, 25 f). Only the flowering period of winter oilseed rape varieties has as a rule ended when summer rape varieties start flowering (INGRAM 2000, 25f). But there are also exceptions from that rule (see Textbox 4). The same applies to maize - almost all varieties overlap in terms of their flowering stages (INGRAM 2000), 17f).

Owing to the fact that neither the beginning of flowering nor the flowering period can be managed, control might only be executed via the sowing date and the variety. In winter-planted crops, flowering date and sowing date are approximately 6 - 9 months apart, in spring-planted crops approximately 3 - 5 months. After sowing, environmental factors have a strong impact on the beginning of flowering and the flowering period of varieties and species. Heat and dryness make for an earlier start of flowering and a shorter flowering period (SCHULZ und JAKISCH 2001), while cold, humid weather cause flowering to begin later and prolongs the flowering period.

Conclusion: An exact steering of the flowering date as well as the prevention of overlaps of different varieties during flowering are thus hardly feasible. (See also interesting data from wild plant studies FITTER und FITTER 2002).

# Textbox 4: Winter oilseed rape and summer rape ought to be considered as one fruit in terms of coexistence

While from a normal sowing, the flowering periods of spring and winter crops are quite different; flowering times can be heavily modified by sowing date and other husbandry or environmental factors. It is therefore suggested that spring and winter types are considered as one crop for separation purposes. (INGRAM 2000, 28f).

This approach limits the farmer in his or her choice of varieties. The choice of a variety in relation to the properties of the site is one of the most important yield factors. The frequency of late frosts, for example, as well as the soil type are significant factors in selecting a certain variety. The demand of producing optimum yields only allows the farmer a small margin of variation regarding flowering dates (BAEUMER 1992, 127ff). The practical fitness of this approach (different sowing dates) remains to be seen. Weather conditions especially in the spring open only small time-windows to choose optimum sowing times for a perfect seed bed. Yet, farmers have to agree on different sowing dates several months in advance of the sowing

date to make a coordinated management process work. It seems to be very unlikely for farmers to agree in advance and limit their cultivation management to a limited range of sowing dates. Spring-varieties for soybean and maize are exclusively planted in Austria. Oilseed rape spring and winter-varieties may be planted, but as shown above, even these types have to be regarded as one crop for separation purposes.

Apart from the unresolved difficulties of finding agreement among a group of farmers, selecting varieties with differing flowering dates and selecting different sowing dates does not seem to be an adequate strategy to significantly reduce contamination by cross-pollination. In addition, this measure does not provide any regulation for problems of GMO volunteer growth. This approach might only be adopted in combination with other approaches.

### 4.4.3 ISOLATION DISTANCES

The European Commission recommends isolation distances as a tool on the farm level. Given Austria's small-scale structures, this is not feasible at farm level in most of the cases, such as maize and oilseed rape. Isolation distances at farm level may be useful for highly selfing species like wheat and barley. Soybean is considered a selfing species, but bees are likely to play a significant role in cross-pollination, which is why it remains unclear which level will apply to guarantee coexistence in soybean cultivation.

The EU recommends that isolation distances be specified as a function of the outcrossing potential of the crop. For open pollinating crops, such as oilseed rape, larger distances are required. For self-pollinating crops and plants where the harvested product is not a seed, such as beets and potatoes, shorter distances are possible. Isolation distances should minimise but not necessarily eliminate gene flow by pollen transfer. The objective is to ensure a level of adventitious presence below the tolerance threshold.

The EU Commission gives no detailed recommendation on that issue. Scientific advice on isolation distances varies significantly (e.g. cf. Table 1). The extent of isolation distances is derived from biological circumstances (pollen reach) as well as from the thresholds and a security factor potentially to be determined.

Based on previous calculation (cf. Table 4) isolation distances have to reduce cross-contamination at or below a level of 0.25 % in maize or soybeans and almost 0.00 % in oilseed rape. The percentage of contamination decreases (logarithmically) with the distance. Thus, low thresholds result in larger buffer zones. This has lead for example to the view that a threshold of 0.1 % is not practically applicable. In the following, some crop specific recommendations on isolation distances will be discussed.

### **OIL SEED RAPE**

INGRAM (2000, 28f), for example, recommends 1.5 m for a threshold of 1 %; 11.5 m for a threshold of 0.5 % und 100 m for a threshold of 0.1 %. He makes the reservation, however, that the degree of safety is rather low at 0.1 %, given that beyond a distance of 60 m, the hybridization frequency decreases very slowly.

Some authors recommend significantly shorter minimum separation distances.

These distances can only be justified in cases of a high degree of self-fertilisation and very low bee activity. Even for rigid self-fertilisers such as barley, distances of 60 m have meanwhile come to be suggested as safe if the contamination level is to be kept below 0.1 % (WAGNER und ALLARD 1991). INGRAM (2000, 28f) also estimates that for male sterile oilseed rape varieties, 100 m of isolation distance suffice to achieve a cross-pollination rate below 1 %. He admits, however, that

exact data to this effect are not yet available. EASTHAM und SWEET (2002) recommend 100 m (10 times the recommendation of INGRAM (2000)) for oilseed rape varieties to keep cross-pollination under 0.5 %; for oilseed rape varieties with a share of male sterile plants (varietal associations, e.g. synergy), they recommend "considerably greater isolation distances from GM crops than conventional varieties," without giving concrete figures.

INGRAM (2000, 26f) recognizes that bees forage over average distances of 2 km and sometimes even further. He believes, however, that fertilisation by bees across wide distances occur only very rarely. In tests conducted by RIEGER et al. (2002) with commercial fields canola, randomness of long-distance pollination events was detected. The authors state, "We

did not observe a leptokurtic or exponential decline, as found in many small-scale studies. Instead, a more variable distribution with isolated pollination events was detected. The multiple pollinating agents (wind and insects) of canola and the large size of the source may contribute to the randomness of long-distance pollination events. Varietal differences among canola sink fields were observed (Fig. 4), but no consistent effect of wind direction on pollen-mediated gene flow was detected (data not shown). The variety of canola may be a contributing factor in random pollination events at distance. Pollination has been shown to be affected by crop variety. Varieties have differences in flowering period, which will affect pollination events over such a large scale. Another explanation for these seemingly random events may also be related to insect behavior. Roaming insects may target single plants flowering early or late in a field, resulting in sporadic pollen movement." .... "These observations, coupled with our data on long-distance pollen movement, indicate that laboratory and small-scale experiments may not necessarily predict pollination under commercial conditions. This study demonstrates that cross-pollination between commercial canola fields occurs at low frequencies but to considerable distance." Figures show that cross-pollination had occurred up to nearly 3,000 m (Fig 2) at a low frequency 0.05 % pooled by all samples of a field. Several incidents of crosspollination had been detected between 1,000 and 2,500 m (FIG 2 in RIEGER et al. 2002). Given the fact that in this study the commercial variety with ALS resistance, had been in its first year of introduction, a much higher rate of contamination has to be assumed on a longer time perspective through volunteer growth and seed dispersal by birds and man.

### Recommendation by the author on isolation distances for OILSEED RAPE:

Many recommendations for Isolation distances take only cross-pollination into account and are based on results obtained by research-plot size field experiments. Seed dispersal by machines will lead to volunteer growth which will contribute to significant long-range contamination (several km). Seed dispersal by machines is very hard to control and to manage, seed dispersal by animals is not manageable. Moreover, long range cross-pollination (RIEGER et al. 2002 and STOKSTAD 2002) of up to 4 km (THOMPSON et al. 1999) has been detected in commercial field size research. Isolation distances from 2 to 4 km are justified if a sustainable GMO free production is envisaged. Isolation distances above 1,000 m should be handled at the regional level. The author therefore recommends to choose tools at the regional level to ensure sustainable levels of cross-pollination below 0.1 or 0.0 % in GM-free oilseed rape production. Regulations for beekeepers also have to be met at the regional level.

### **MAIZE**

For maize, separation distances of 420 m are recommended (INGRAM 2000, 31f) in order to keep cross-fertilisation below 0.1 %. The preliminary recommendations of the Danish study are 200 m to achieve a 0.1 % level of cross-pollination if seed purity is high, i.e. 0.0 %. In research plots sized 0.4 ha, cross-pollination was observed at a maximum of 200 m. But up to 0.2 % cross-pollination at 800 m has been detected by JONES and BROOK (1950 quoted in TREU und EMBERLIN 2000).

Plant breeders trying to create corn seed of high genetic purity have recognized that the physical separation of different corn varieties by 200 m (660 feet) will still result in "contamination" due to cross-pollination at levels of about 0.1% (National Academy of Sciences, 2000 quoted in ELLSTRAND 2003. In order to prevent cross-pollination from transgenic corn with pharmaceutical traits, APHIS requires in a proposed rule that there will be no corn grown within 1 mile (1.6 km) of the field test site throughout the duration of any field test which involves open-pollinated corn. This establishes a physical isolation distance that is eightfold greater than the isolation distance required for the production of foundation seed (660 feet). Furthermore APHIS requires." With the establishment of isolation distances of 1 mile for open-pollinated corn and one-half mile for controlled pollination corn field tests, APHIS will not allow the use of border rows to reduce these isolation distances. APHIS believes that other methods are available and do not pose the difficulties inherent in using border rows" (APHIS-USDA 2003b). (Since August 6th 2003 field releases of transgenic plants with pharmaceutical or industrial traits require a permit by APHIS instead of just a notification (APHIS-USDA 2003a)).

Recommendation by the author on isolation distances for MAIZE:

Owing to the fact that maize cultivation in Austria is almost complete based on hybrid seeds, a cumulative effect through seed savings by farmers has not to be considered. Moreover, volunteer growth of maize is unlikely. The contamination potential thus only needs to be calculated by the risk of cross-pollination. Recommendations for isolation distances vary significantly. Given the fact that significant cross-pollination had been detected at 800 m, the author recommends 800 m as minimum isolation distance for cultivation and 1,200 m for seed propagation. It is unclear whether coexistence in maize cultivation might be resolved at the neighbourhood level. Aspects for beekeepers have to be met at the regional level.

#### MANAGEMENT AT THE REGION LEVEL

#### 5.5.1 EUROPEAN COMMISSION RECOMMENDATION: MEASURES OF REGIONAL DIMENSION

The European Commission document refers to that as follows: "Measures of a regional dimension could be considered. Such measures should apply only to specific crops whose cultivation would be incompatible with ensuring co-existence, and their geographical scale should be as limited as possible. Region-wide measures should only be considered if sufficient levels of purity cannot be achieved by other means. They will need to be justified for each crop and product type (e.g. seed versus crop production) separately. (EC-COMMISSION 2003, 2.1.5).

The European Commission refers to voluntary GMO-free regions as follows: "3.3.3. Voluntary agreements among farmers on zones of a single production type. Groups of farmers in a neighbourhood may achieve a significant reduction in the costs related to the segregation of GM and non-GM production types if they coordinate their production on the basis of voluntary agreements" (EC-COMMISSION 2003, 3.3.3). No further concepts for the establishment of GMO regions or GMO-free regions are given by European Commission.

#### 5.5.2 VOLUNTARY REGIONS: EXPERIENCE IN AUSTRIA

The European Commission recommends voluntary regions as a regional management tool. As discussed above management, measures must help to avoid conflicts and solve conflicts. "Voluntary regions" are a group of farmers in a consolidated area who favour a special cultivation system. Becoming part of this group is a decision of each farmer.

There is some experience in Austria regarding voluntary agricultural regions.

RWA (Raiffeisenware Austria), for example, is putting its stake in Austria upon non-GM maize production, including the necessary checks and controls, rather than on mass-produced commodities, with an attempt to establish as consolidated GM-free maize cultivation areas as possible. This is ultimately considered the only chance to relieve Austrian cultivation of some of the price pressure farmers will be facing with an even wider opening of the EU market for maize imports from Eastern Central Europe. RWA is supported in this initiative by the Chambers of Agriculture. Along with numerous RWA "Lagerhäuser" (large-scale stores especially dedicated to farmers' needs) in Lower Austria, Styria, and Upper Austria, several private provincial product traders and Upper Austrian "Lagerhäuser" not pertaining to RWA wish to join the initiative and establish **consolidated genetic-engineering-free** regions in their resource areas. (APA 2002)

In Austria, there are several organic regions (e.g. http://www.bioalpeadria.info/). The existing organic regions exist on a voluntary basis, i.e. within organic regions, also conventional farmers farm their land. Organic regions contribute to a common marketing of products of the same region. A consolidated region is not necessary for this concept. On the contrary, it would be very difficult to obtain a fully consolidated organic region without different production type. Voluntary GMO or GMO-free regions will face the same problem that organic regions are already facing - "How to get a consolidated area on a voluntary bases." In the province of Lower Austria a voluntary GMO free Region "Waldviertel" has been proclaimed in December 2002. But even 6 months later farmers did not formally sign binding agreements (SCHÖRPF 2003, head of the GMO free Waldviertel initiative, personal communication).

Even with subsidies, there is no guarantee to get consolidated regions. The probability of getting consolidated areas rises when the subsidy approach is bound on an Agri-Environmental Programme which has been widely adopted among Austrian farmers. Protection covering as

extensive an area as possible could be achieved by coupling the use of GMO-free seed to various subsidies under the Austrian Agri-Environmental Programme. In terms of duration, a period of time of e.g. 10 years might be foreseen regarding the coupling with subsidies under the Austrian Agri-Environmental Programme. But some problems still remain. Small areas of continuous GM-cultivation suffice to contaminate several hectares. Thus, accompanying measures have to be resorted to in order to guarantee the long-term coexistence of different farming methods.

#### 5.5.3 LEGALLY BINDING REGIONS

A possible approach is to define legally binding instead of voluntary regions . This is fully in line with European Commission's recommendation on coexistence. In the view of the Commission, "region-wide measures should only be considered if sufficient levels of purity cannot be achieved by other means".

In Austria, there is some experience with legally binding regions, such as:

- Water Protection Zones" in accordance with Austrian Water Act
- Consolidated Cultivation Areas in accordance with Seed Act of 1997
- A protection zone of four kilometers around mating station in accordance with Upper Austrian Apiculture Act.
- A protection for the entire Province of Vienna admitting exclusively the variety Carnica
  (Apis mellifera carnica). The keeping of other bee varieties (Apis mellifera mellifera) and/or
  other species (e.g. the Asian bee Apis cerana) is subject to authorization in accordance
  with Vienna Apiculture Act.

Depending on several protection goals certain areas such as the following ones might be declared free of GMO cultivation: "seed production area", "ecologically sensitive areas", "areas of special interest for organic farming."

As mentioned above, at least for oilseed rape and most likely also for maize, coexistence management has to be obtained at a regional level. While voluntary regions have a limited conflict solution potential legally binding region have to solve conflicts between different farming types in the case of oilseed rape.

# 5.6 SIMULATION MODEL CALCULATION

The calculation and evaluation of the data was carried out in cooperation with DI A. Bartel, Institute for Landscape Architecture and Landscape Management, University of Agricultural Sciences, Vienna.

It was agreed with the bodies commissioning the study to develop a graphic model in order to depict the need of protected areas for farming enterprises operating on an organic basis. The district of Wels Land, Upper Austria, was selected to serve as an example. For the district of Wels Land, a detailed calculation of the area need for agricultural purposes that have to be labelled as GMO-free was carried out on the basis of the existing digitally stored properties and crofts. The data set provided courtesy of DORIS did not entirely cover the district of Wels Land. The following data were worked into the map of "Wels Land":

- DKM (Digital parcel map) (Wels Land) DORIS OÖ, with property (plots of land) limits and property numbers (the data set was incomplete at the time the report was drafted, some partial areas were missing on account of work not yet completed.)
- Property numbers of organic farming areas of the year 2000 for Wels Land (Federal Ministry of Agriculture and Forestry, DI HOFER)

Owing to the fact that the property numbers of the data sources of the Federal Ministry of Agriculture and Forestry and DORIS had different formats, they had to be partly hand-adapted. Around every ORGANIC plot of land, separation distance circles of 500 m to 4,000 m in steps of 500 m were marked in colour. In addition, the organic farming fields were colour-marked in yellow. The map shows area need for organic farmers for growing GM-free. The analysis ends at cultivation type level. For a decision tool in accordance with the European Commission recommendations, a map providing crop-specific data, not only for organic but also for conventional farmers would be needed to meet the requirements of crop specific "region-wide measures". With regard to the update and English translation of this study, an update of this

map had not been planned. But his map should serve just as an example how to come to decisions regarding oilseed rape using a digital cadastral map as a decision tool. For this purpose following calculation has been assumed: All yellow organic plots in Wels Land are organic oil seed rape or GMO-free oil seed rape fields While in Austria the cultivation of organic oilseeds is rather low, i.e. 1,6 % of the total cultivated oilseeds area (data specific for oil seed rape was not available) in Austria (personal calculation for 2001 based on data provided by www.awi.bmlf.gv.at). A voluntary contract programme called "RAPSO" for GMO-free oilseed rape has been conducted rather successfully in Upper Austria for several years (for further details see http://www.vog.at/rapso.htm). WELS Land is the district with the lowest density of organically operating enterprises in Upper Austria. Out of a total of 24 municipalities, 10 municipalities do not have any organic farms, 13 municipalities have an organic share lower than 4 % of the municipality's agricultural area, and one municipality has an organic area share of the agricultural below 8 %. It seems realistic that organic rape and gmo-free oil seed rape (RAPSO) fields could almost reach the small amount of overall organic fields in the district Wels Land

Assuming that all yellow organic plots in Wels Land are organic oil seed rape or GMO-free oil seed rape fields (RAPSO), there will be almost no place to cultivate GMO oilseed rape in the district of Wels Land. As reflected on the map of WELS LAND (Appendix 1), even if the number of organically or GMO free oil seed rape cultivation is small, large sectors of the municipality will be claimed as protection areas. The map also clearly depicts the overlapping of areas claimed as protection zones with neighbouring municipalities. Owing to the fact that the available data set for Wels Land was incomplete and the city of Wels is located entirely within the district of Wels Land, the author is notable to make complete statements as to the size and location of potential areas on which GMO cultivation would be possible without posing a hazard to organic or GMO free areas on account of pollen contamination and seed dispersal. In addition, the adjoining municipalities of the neighbouring districts would have to be included in the evaluation.

#### 5.6.1 SMALL SCALE STRUCTURE IN AUSTRIA

The map in Appendix 2 clearly reflects the small scale of Austrian fields. Fields 80 m wide and 300 m long are quite common in Austria. Also, the various fields of individual farmers are often dispersed among other farmers' fields. Consolidated fields in terms of crop cultivation are quite rare in Austria. With regard to grassland, consolidated areas are more frequent in mountain regions. Thus, conflicts over coexistence will in many cases arise between not only two but several farmers, which is why the coordination at neighbourhood level of different field plots and different crop rotation plans (management a neighbourhood level) seems rather unrealistic from a practical point of view.

#### 5.6.2 CONCLUSIONS: REGIONAL LEVEL

There will be a need to organize coexistence at the regional level. Currently, there are no tools except legally binding GMO or GMO-free regions to solve coexistence conflicts properly. In principle, the designation of protection areas is not new and does not constitute a problem arising from GMO cultivation. What is new is the dimension caused by the problem of coexistence of organic, conventional non-GM, and GM-production. The Upper Austrian Apiculture Act, for example, implements via decree a protection zone of 4 km around a recognized mating station, in order that only a certain bee variety be used in the respective area. Any implementation of GMO-free cultivation areas to secure coexistence in the small-structured Austrian landscape has to meet, from the point of view of natural science, two minimum requirements:

- implementation of a compact area, and
- securing of this compact, consolidated area over several years.

# 5 OUTCROSSING TO WILD PLANTS THE CASE OF OIL SEED RAPE

#### 5.1 DISPERSAL DYNAMICS OF SYNTHETIC TRANSGENES

When we look at the phenomenon of "outcrossing" of crops to wild plants and/or wild plants to crops, questions of population dynamics arise along with the question of pollen reach. As opposed to crops, whose "population dynamics" is anthropogenically controlled via cultivation (cultivation frequency and density), wild plants are to a great extent beyond the influence of anthropogenic management. Gene flow between locally separated populations is determined by pollen and diaspore flow (HU und ENNOS 1999). Estimates of the dynamics of gene flow (pollen and diaspore flow) vary greatly, from low dynamics in wild barley to high dynamics in oak, and depends strongly on the reproductive type (ENNOS 1994). Exact statements as to dispersal distances of outcrossing synthetic transgenes can hardly be made. Superficial classifications via a selective disadvantage or selective advantage of a gene do not allow conclusions as to the actual dynamics of the spreading of synthetic transgenes.

A central misunderstanding in the interpretation of Darwin's theory of evolution relates to the concept of "survival "survival of the fittest." This metaphor Darwin used does not put aggressive confrontations into the foreground, nor does it mainly refer to the competitive strength in the struggle for survival (i.e. different mortality of different genotypes). Rather, it is of importance what contribution an individual (with certain alleles) contributes to the gene stock of the following generation. Selection thus consists in the differing reproductive success. It is a statistical process in which individuals with more favorable characteristics generate on average more numerous offspring than individuals with less favorable characteristics.

Reproductive success, however, depends strongly on a variety of factors and is not to be compared to competitive strength in a direct confrontation (CZIHAK et al. 1996, p. 883). WILLIAMSON (1993) believes that there is no significant correlation between the weed characteristics<sup>6</sup> listed by BAKER and the properties of the weeds listed as the 10 most important weeds in a questioning of weed experts. Considering that it is eventually not a plant's characteristics, but the mode of interaction between the environment and the plant's characteristics that accounts for dispersal tendencies in neophytes, the use of BAKER's characteristics for the purpose of a predictive risk assessment of genetically modified plants is not adequate (WILLIAMSON 1993). The change of a plant's site alone, without a change in characteristics, can turn a plant into a weed (WILLIAMSON 1993). TILMAN's experiments (1997) also point to the significance of intentional or unintentional dispersal of plants. Obviously, the diversity of plant populations frequently correlates more with chance than with competition. In an experiment of sowing 54 grass species native in the United States, more than 50 % were able to establish themselves on the experimental plots even though they had not occurred in that location before. This is why the concept of a "gene by environment" assessment is gaining ground in analogy with "step by step" assessment (several contributions to the discussion at the conference "Living Modified Organisms and the Environment - An International Conference", OECD 2001). However, not even this approach can obscure the fact how difficult it is to assess the "ecological damage potential" of a synthetic transgene. In many cases, the insertion of synthetic transgenes produces unexpected, unintended effects (pleitropic effects). In all Bt-maize varieties, for example, the lignin content is significantly raised (between 33 and 97 %) vis-a-vis that of their isogenic lines. This is why degradation rates of Bt maize were protracted as compared to the isogenic lines (STOTZKY 2001). It is also difficult to define properties of fitness per se. The problem of defining characteristics was investigated within the scope of the comprehensive SCOPE 37 Programme on the "Ecology of Biological Invasions", carried out by several institutions on several continents, on the basis of the following key questions of invasion biology:

<sup>&</sup>lt;sup>6</sup> As early as in 1965, BAKER (quoted in WILLIAMSON 1996) put together a list of plant characteristics frequently found in weeds. According to BAKER, weeds are "super generalists."

- What factor determines whether a species will become an invader or not?
- What site properties determine whether an ecological system will be prone to or resistant to invasions?

Answering these questions is indispensable for a predictive risk assessment of GMOs. Again, however, it shows how difficult it is to make reliable statements regarding the development of ecosystems. Reliable characteristics of plants and ecosystems with general validity, which would allow a predictive risk assessment, have not yet been encountered (WILLIAMSON 1996, KOWARIK 1996, SUKOPP und SUKOPP 1995, CRONK und FULLER 1995, NOBLE 1989, MOONEY und DRAKE 1989, REJMÁNEK 1989).

KOWARIK summarises the state of matters as follows:

#### **Textbox 5: Result of the SCOPE 37 Project**

"Despite several approaches within the SCOPE 37 Project on the `Ecology of Biological Invasions,` in which several continents cooperated, we have not found a satisfactory solution to develop sound prognoses on the success of a species - and thus also on its risk potential - on the basis of species-specific characteristics. At best, there is an increased probability of success in certain ecotypes for certain groups of species, e.g. in sites of disturbance for species with typical weed characteristics (in the sense of BAKER 1965), while with regard to other ecotypes, success prognoses are hardly possible. Considering that the decision as to success or failure of a species may be possible not until decades or centuries after their first introduction, it is not possible to either predict or rule out the future success or failure of a species." (translated quote from KOWARIK 1996, p. 121).

## 5.2 PROBABILITY OF ESTABLISHMENT OF SYNTHETIC TRANSGENES IN NEW GENE POOLS

Along with general considerations on the basis of invasion biology, ADAM and KÖHLER (1996) calculated the probability of establishment of synthetic transgenes in the gene pool of a natural population, using different simulation models. According to the authors, the influence on the establishment of new (natural) genes (originated by way of mutation) in a gene pool is subject to the following factors:

- mutation
- selection
- reproductive type (self-fertilising or cross-fertilising)
- adventitious genetic drift
- migration (i.e. the exchange of genes between different populations)
- competition
- coevolution (evolutionary response to the interaction with the environment).

For the calculation of the probability of establishment of synthetic transgenes in natural gene pools, the following simplifications were made:

- Only the synthetic transgene was taken into account, not the genetic changes occurring at hybridization.
- The synthetic gene is dominant vis-a-vis a recessive allele in the wild population. Considering that in nature, there is no allele equivalent to a synthetic transgene, we should speak in this respect not of "heterozygous or homozygous" alleles but rather of "hemizygous" alleles (ADAM und KÖHLER 1996).

On the basis of various different model calculations, ADAM und KÖHLER (1996) demonstrate that

- synthetic transgenes that result in a selective advantage are almost always able to become established in the recipient population;
- a synthetic transgene with an average selective disadvantage of 20 % is able to
  establish itself in the wild population after 25 generations to the extent of 70 to 90 %,
  given migration rates between 10 and 20 %;
- small populations and genetic drift equally favor the establishment in the wild population of genes disadvantaged in terms of selection.

Other calculations with different selection coefficients and hybridization rates have arrived at similar results. Only with a synthetic transgene, which causes a selective disadvantage, and with low hybridization rates (ca. 1 %), the establishment of the synthetic gene in the wild population is lower (below 5 %) (ADAM und KÖHLER 1996).

Along with these model calculations, research on genetic diversity as well as the neutrality or mutation-random-drift theory (founded by KIMURA 1987) support the assertion that synthetic transgenes will spread in the population: According to KIMURA (KIMURA 1987), the share of selection in evolution is small. Most mutations are neutral and have no selective advantage. Neutral mutations spread in a population via random-genetic-drift and result in the great intraspecific genetic variability *in situ*. Strong selection would lead to low genetic variability within species. According to LI (1997), more recent data of molecular biology support the "inadequacy of Neo-Darwinism" (LI 1997, p.432). He, too, assigns a relatively minor role to selection. Experiments with GMOs also support the simulation model calculations cited above (KLINGER und ELLSTRAND 1994, ARRIOLA und ELLSTRAND 1997b, SNOW et al. 1999). We thus have to assume that synthetic transgenes persist in natural populations (possibly over thousands of years).

#### 5.3 PRECAUTION, UNCERTAINTY AND PERSISTENCE

Genetic contamination is to be evaluated as giving rise to concern both from an ecological perspective and from the perspective of an agriculture free of GMO-contamination, given that it constitutes a reservoir with a continuous contamination potential for areas farmed in accordance with the guidelines of organic farming as well as conventional non-GM agricultural areas.

The ecological hazard consists in the long persistence of synthetic transgenes in the feral plant populations. We have to assume that synthetic transgenes are transferred from crops to wild relatives, and will manifest themselves over a long period of time in the wild plant populations as long as compatible hybridization partners exist in the specific cultivation areas. In case of damage, synthetic transgenes cannot be removed - they can be expected to persist for several thousands of years in the wild plant populations. In this case, safety and ecological soundness can no longer be guaranteed. Experience from the field of eco-toxicology of chemicals has shown that particularly persistent substances, regardless of a first assessment of their risks, have a very high ecological damage potential. As the history of the risk assessment of pesticides demonstrates (EEA 2001), a risk assessment will always have imperfections (on account of the limits of the human ability of perception and recognition). This becomes all the more serious the longer chemicals persist in the environment. KLÖPFFER formulated the idea of avoiding persistence of chemicals as follows: (KLÖPFFER 1994):

## Textbox 6: KLÖPFFER (1994) on the problem of persistent substances

"Moreover, even if an ideal testing system were possible, it would necessarily always reflect the current status of knowledge; persistent chemicals, however, remain in the environment for a very long period of time and cannot be removed, in particular in case of molecular, ubiquitous dispersal. If a damaging effect is recognized at a later point in time, the exposal cannot be stopped! The argument is thus: Persistence stands for principally never fully recognizable eco-toxicity. Persistent substances should thus in principle never be released into the environment, i.e. withdrawn from human control. Special precaution is

needed if along with persistence, the criteria of accumulation potential and mobility are met." (translated quote from KLÖPFFER 1994)

As opposed to possibilities of fighting weeds in agriculture, the possibilities of risk management of wild plants in natural ecosystems are limited and mostly of short-term effectiveness (seed stock in the soil). In view of longer persistence of synthetic transgenes in natural populations and the fact that management measures are hardly possible, the significance of the criterium of persistence for an assessment of the risks of GMOs becomes evident (i.e. a total ban on GM-plants in those regions in which they are able to hybridize successfully with wild relatives). The point is not an unconditional rejection or support of technological developments, but rather, as

SCHERINGER 1999 ... in how far learning from experience (the trial-and-error principle) is an adequate strategy to assess environmental interventions.

correctly stated by SCHERINGER (1999, 149f), the important question as to how far learning from experience (the trial-and-error principle) is an adequate strategy to assess environmental interventions. And "to what degree of seriousness of an experience the effect of learning and the use for the purpose of which we accepted to make the experience, can still be justified?"

In the sense of the criterium of persistence, the outcrossing potential per se - regardless of the properties of the foreign gene - would have to be defined as an undesired event.

#### 5.4 DIFFERENCE OF GENES AND SYNTHETIC TRANSGENES

In the discussion on uncertainties, precaution and persistence associated with transgene escape into wild populations the question arises, if there is a difference between genes inserted by conventional breeding methods and synthetic transgenes inserted by gene technology. Many scientist assume that there is no difference between this two type of genes and therefore risk of gene escape has to be compared with risks of gene escape from conventional bred crops. Table 5 gives a short overview on differences between genes and synthetic transgenes..

Table 5: Differences between genes and synthetic transgenes

	Conventional genes	Synthetic transgenes
Range of gene exchange	Conventional genes  Crosses between plants of the same varieties within species, closely related genera. Even hybridization and wide crosses cannot move genetic material much beyond these limits (i.e. within the same family)	Most GM-crops are obtained by transferring genes from bacteria, viruses into plants (e.g herbicide tolerant, insect resistant traits)
genetic elements	Genetic elements from the plant	Genetic elements from different organisms: promotor from a virus, expression sequence from a bacteria,
Recombination of genetic elements	No	Yes
base exchanges	No (mutation breeding yes)	Yes (bases exchange and truncation of originally expression sequence)
do have introns	yes	no
Difference on chromosome level	The inserted gene has a corresponding allele (homozygot or heterozygot)	The inserted gene has no corresponding allele (hemizygot)

It should be noted that the role of introns is unclear. Some scientist assume that introns may act as hidden genetic regulators (e.g. FLAM 1994, MONK 1995, SHCHERBAK 2003, HARE und PALUMBI 2003).

Some scientists argue that crops genes derived by mutations or derived by genetic engineering are equal from the point of uncertainty and persistence. From an evolutionary perspective the creation of genes by enhanced induced mutation is "just" speeding up evolution 100-10,000

times faster than it would occur naturally. The creation of synthetic transgenes in plants with combinations of virus promoters, bacterial expression sequences etc. are not known to occur as a consequence of evolutionary forces. Persistence of synthetic transgenes must be considered a more serious hazard to biodiversity than persistence of crop genes derived by induced mutations, simply because possible impacts of exotic genes may be highly uncertain.

#### 5.5 CONCLUSION TRANSGENE ESCAPE

Synthetic transgenes differ from conventional or natural genes in many aspects. Synthetic transgenes (as well as conventional genes) will stay for a long period in wild populations. The certainty about harmful effects from Synthetic transgenes is considerable low, and quite much lower than for conventional or natural genes. In the case of error in risk assessment removal of synthetic transgenes from various ecosystems is not possible. Therefore the cultivation of GM-crops which have the potential to exchange their synthetic transgenes to feral populations should not be approved.

# 6 CONCLUSIONS AND RECOMMENDATIONS

#### 6.1 CONCLUSIONS

Based on the European Commission recommendations on coexistence, there is a need to regulate coexistence crop specific and step by step, from management at farm level to management at the neighbourhood level and measures with region-wide dimension.

Based on analyses of cross-pollination, seed dispersal, seed viability in the soil and in accordance with the European Commission recommendation, cumulative effects, as well as the small scale structure of Austrian Agriculture, there will be a need to regulate the cultivation of GM maize and GM oilseed rape at a regional level. In some regions the agricultural structure may allow management of maize at the neighbourhood level. It was not subject of this update of the study to identify such regions for maize on the basis of a digital cadastral map for Austria. While voluntary regions will not provide a proper conflict solution potential, legally binding regions (GM regions or GM-free regions) will be necessary.

From a management perspective, only minimum isolation distances would serve as a practically feasible decision tool. The "choice of different varieties with different flowering times" will not serve as a management tool, because the weather is not subject to human decision-making. Yet, weather conditions have a great influence of the success on varieties of different flowering times. If the success of a tool is based on conditions beyond management, it will not serve as an appropriate management tool.

Hedges and barrier crops will significantly reduce the rate of cross-pollination. But currently it is unclear how hedges could be part of a calculation model to help reduce minimum isolation distances. From a management perspective, taking hedges into account would require a calculation on a field by field basis - an enterprise that appears to be rather labour-intensive.

#### 6.2 RECOMMENDATIONS

The problem of coexistence cannot be solved solely at the farm or neighbourhood levels. Rules have to be established to regulate coexistence and to define who is responsible for applying anti-contamination measures or obeying minimum isolation distances.

Because it is not known whether it is possible to process GMOs "side by side" with non-GMOs, production chains will have to be totally separated from the beginning. The author thus recommends a regulation on "GMO cultivation and processing" which fully addresses all issues of coexistence from farm to table. The adopted EC Regulation on Traceability and Labelling strongly supports the idea of such regulations in order to guarantee a complete tracing of GMOs from farm to table. EC Regulation 178/2002 requires full traceability from farm to fork for the whole (conventional, organic as well as GMO) food and feed production chain. The proposed regulation on "GMO cultivation and processing" should be based on the experience of EC Regulation 2092/91 for organic farming. Organic systems have shown how to avoid admixtures of crops from other cultivation types. Like organic farmers and processors. GMO farmers and processors have to ensure that no contamination is taking place. This means that GMO farmers and processors have to build up their own system of GMO production, as organic farmers and processors have done in the past. This means separate harvesting, transport, storage, processing, etc. An inspection system according to organic farming has to be implemented. This should lead to a decrease of costs for organic farmers and processors which are currently paid for applying anti-contamination measures.

The regulation on "GMO cultivation and processing" should consider following tasks to regulate cultivation:

Table 6: Tasks for a regulation on "GMO cultivation and processing"

The European Commission recommends that "As a general principle, during the phase of introduction of a new production type in a region, operators (farmers) who introduce the new production type should bear the responsibility of implementing the farm management measures necessary to limit gene flow" (EC-COMMISSION 2003, 2.1.7). While currently GMO farmers are introducing a new technology and in order to avoid conflicts by unregulated cultivation the author recommends that all GMO farmers shall be subject to approval regarding cultivation. An approval of GMO cultivation will also help to meet the requirements of EC-Directive 2001/18/EG, which requires "Member States shall also establish registers for recording the location of GMOs grown under part C, inter alia so that the possible effects of such GMOs on the environment may be monitored" (EC 2001, Article 31(3)b).	
described how this approval is to be granted.	
Based on protection goals (see 3.1), crop-specific consolidated regions for special interests (GM-free seed production or GM seed production, organic farming regions, sensitive ecological areas, Natura 2000 regions) should be declared from the beginning and integrated into the cultivation register.	
Minimum isolation distances have to determined for each specific crop, or for groups of crops.	
Based on a digital cadastral map and a cultivation register, a DIGITAL CULTIVATION PARCEL MAP has to be established. Therein, the cultivation (crop rotation) by all farmers is stored crop and year specifically. Each agricultural parcel should be assigned the following data on an annual bases:	
<ul> <li>Cultivation type (organic, conventional GMO)</li> </ul>	
Planned crop to be cultivated	
<ul> <li>Other types of protection goals such as region for seed production, nature conservation area, etc.</li> </ul>	
<ul> <li>Regional information such as private GMO cultivation and processing region, or organic region</li> </ul>	
The way how all farmers or alternative sources (Austrian subsidies programme) are to provide the relevant information needed for the DIGITAL CULTIVATION PARCEL MAP in time should be described.	
E ()	

#### Proposed approval system:

The farmer who intends to grow a GM crop asks the regulators for formal approval and provides following information.

- GM crop and specific trait in order to settle possible conflicts between and organic/conventional GMO-free farmers
- specific trait order to settle possible future conflicts between GM farmers with the same crop species but with different GM traits (e.g pharmaceutical trait vs herbicide tolerant trait).
- The field with cadastral number
- A plan for anti-contamination measures at farm or neighbourhood level for the specific field in order to avoid contamination through cross-pollination and seed dispersal (e.g. oilseed rape)
- A plan for segregation measures from drilling, harvesting and transport in order to avoid technical contamination
- A contract with an inspection body responsible for surveillance

Based on the information provided, the regulator has to decide if the minimum isolation distances apply, or if there is a possibility to reduce minimum isolation distances to a certain degree. As long no qualified data on the efficiency of anti-contamination measures at farm level, such as on hedges, exists, the minimum isolation distances should apply in all cases.

The authority in charge of the approval uses the minimum isolation distances determined in the Regulation on "GMO cultivation and processing". The regulator checks in the DIGITAL CULTIVATION PARCEL MAP if there are conventional or organic farmers who intend to cultivate the same crop species as proposed by the adjoining GM farmer. Based on the DIGITAL CULTIVATION PARCEL MAP and on minimum isolation distances for the specific crop, the regulators could easily identify areas of conflicts.

If the proposed GM field e.g. GM maize filed lies outside the minimum isolation distances to the nearest organic or conventional maize field and no other regional aspects forbid GM-cultivation, the cultivation of the GM crop should be granted. If a conventional or organic farmer has a maize field within minimum isolation distance, the approval should not be granted. In this case, the regulator should provide information regarding alternative GM crops to be used in the same cultivation period.

#### 6.3 FINAL REMARK

The proposed recommendations are meant to stimulate the discussion on coexistence with concrete rather than undetermined measures for a management of coexistence of different farming systems. While coexistence is based on, but not limited to natural science, these recommendations are based on natural science and on management experience, with a digital cadastral map as a decision tool. The DIGITAL CULTIVATION PARCEL MAP is proposed as a core instrument to identify areas of conflicts. The tool allows solving conflicts in a wellstructured way, provided minimum isolation distances are determined. The decision tree is based on European Commission recommendations: "As a general principle, during the phase of introduction of a new production type in a region, operators (farmers) who introduce the new production type should bear the responsibility of implementing the farm management measures necessary to limit gene flow" (EC-COMMISSION 2003, 2.1.7). Currently, GMO farmers are those who are introducing a new technology. At a later stage, this rule will mean freezing organic farming where any further expansion would be difficult. Besides that, this rule could also be applied if conflicts between GM famers with different traits arise in the future. In any case, every recommendation on coexistence will have to be adjusted after a certain time period, to address changed economic and scientific circumstances.

Finally, it must be noted that, besides this recommendation on coexistence, it is still too early in the author's view to approve GMOs for cultivation as well as for food and feed use. The above recommendations are based on huge gaps in the underlying basic science of risk assessment of GMOs (e.g. cell biology, genetics) and the current weakness of risk assessment (cf. chapter 1.2). Moreover, GMO crops which have the potential for outcrossing should not be approved because there would be no (or at least very limited) options for mitigation measures in the case of error (cf. EEA 2001), which cannot be ruled out with risk assessment methods.

# 7 LITERATURE

- 1. Adam KD, Köhler WH (1996) Evolutionary genetic considerations on the goal and risks in releasing transgenic crops. In: Tomiuk J, Wöhrman K, Sentker A (Hg.) Transgenic Organisms: Biological and Social Implications. pp. 59-80, Birkhäuser, Basel.
- 2. Amand PCS, Skinner DZ, Peaden RN (2000) *Risk of alfalfa transgene dissemination and scale-dependent effects*. Theoretical and Applied Genetics **101**(1-2): 107-114.
- 3. APA (2002) RWA Initiative for Certified GMO-free Austrian Maize- Domestic maize can only justify higher prices through quality assets. APA news of 28 March 2002 OTS0125 5 WI 0962 AIZ001 CI 28.March 02.
- APHIS-USDA (2003a) Introductions of Plants Genetically Engineered to Produce Industrial Compounds. 46434-46436, APHIS Animal and Plant Health, Inspection Service, USDA. 7 CFR Part 340 [Docket No. 03-038-1], Federal Register / Vol. 68, No. 151 / Wednesday, August 6, 2003 / Rules and Regulations (see http://www.aphis.usda.gov/brs/#perm\_not).
- 5. APHIS-USDA (2003b) *Proposed Rules: Field Testing of Plants Engineered To Produce Pharmaceutical and Industrial Compounds*. 11337-11340, APHIS Animal and Plant Health, Inspection Service, USDA. 7 CFR Part 340 [Docket No. 03-031-1], Federal Register / Vol. 68, No. 46 / Monday, March 10, 2003 (see http://www.aphis.usda.gov/brs/#perm\_not).
- 6. Arnaud JF, Viard F, Delescluse M, Cuguen J (2003) Evidence for gene flow via seed dispersal from crop to wild relatives in Beta vulgaris (Chenopodiaceae): consequences for the release of genetically modified crop species with weedy lineages. Proceedings of the Royal Society of London Series B: Biological Sciences 270(1524): 1565-1571.
- 7. Arriola PE, Ellstrand NC (1997a) *Fitness of interspecific hybrids in the genus Sorghum:*Persistence of crop genes in wild populations. Ecological Applications **7**(2): 512-518.
- 8. Arriola P, Ellstrand NC (1997b) *Fitness of interspecific hybrids in the genus Sorghum:* persistence of crop genes in wild populations. Ecological Applications **7**: 512-518.
- 9. Baeumer K (1992) Allgemeiner Pflanzenbau 3.Auflage. UTB Ulmer.
- 10. Baker HG (1989) Some Aspects of the Natural History of Seed Banks. In: Leck MA, Parker VT, Simpson RL (Hg.) Ecology of soil seed banks. pp. 9-21, Academic press.
- 11. Beck A, Brauner R, Hermanowski R, Mäder R, Meier J, Nowack K, Tapesser B, Wilbois KP (2002) Bleibt in Deutschland bei zunehmenden Einsatz der Gentechnik in Landwirtschaft und Lebensmittelproduktion die Wahlfreiheit auf GVO-unbelastete Nahrung erhalten? Forschungsinstitut f biologischen Landbau Berlin e.V. (FIBL-Berlin), Öko-Institut e.V. im Auftrag von BUND (Frieds of the earth Germany).
- 12. BMLFUW (2001) Verordnung 478 des Bundesministers für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft über die Verunreinigung von Saatgut mit gentechnisch verönderten Organismen und die Kennzeichnung von GVO-Sorten und Saatgut von GVO-Sorten (Saatgut-Gentechnik-Verordnung). BGBL II/478/2001 vom 21.12.2001.
- 13. BMSG (2001) Beschluss betreffend "Festlegung von Schwellenwerten für zufällige, unvermeidbare Verunreinigungen mit gentechnisch veränderten Organismen und deren Derivaten" zur Verordnung (EG) Nr. 2092/91, Biologische Landwirtschaft. Das Bundesministerium für soziale Sicherheit und Generationen gibt nach Befassung des Plenums der Kommission zur Herausgabe des Österreichischen Lebensmittelbuches (Codexkommission) die nachstehende in der UK BIO ausgearbeitete "Festlegung von Schwellenwerten für zufällige, unvermeidbare Verunreinigungen mit gentechnisch veränderten Organismen und deren Derivaten"GZ.32.046/72-IX/B/1b/01 vom 18.12.2001.
- 14. Cresswell JE, Bassom AP, Bell SA, Collins SJ, Kelly TB (1995) *Predicted pollen dispersal by honey-bees and three species of bumble-bees foraging on oil-seed rape: A comparison of three models.* Functional-Ecology. **9**: 892-841.
- 15. Cresswell JE (1997) Spatial heterogeneity, pollinator behaviour and pollinator-mediated gene flow: Bumblebee movements in variously aggregated rows of oil-seed rape.

  Oikos **78**: 546-556.
- 16. Cronk QCB, Fuller JF (1995) *Plant Invaders*. Chapman & Hall.
- 17. Daniell H (2002) *Molecular strategies for gene containment in transgenic crops*. Nature Biotechnology **20**(6): 581-586.

- 18. Delaplane KS, Mayer DF (2000) Crop pollination by Bees. CAB International.
- 19. Diamond E (2001) *The food gamble*. Report of FOE UK on the safety of GM food, edited by Helen .
- 20. DODGE J (2003) Data glut. The Boston Globe , The Boston Globe, USA, by John Dodge, http://www.boston.com/dailyglobe2/055/business/Data\_glut+.shtml DATE: Feb 24, 2003
- 21. Downey RK (1999) Gene flow and rape the Canadian experience. In: Lutman PJW (Hg.)

  Gene Flow and Agriculture: Relevance for Transgenic Crops. pp. 109-116, British Crop

  Protection Council.
- 22. Enquete-Kommission des Bundestages zum Schutz der Erdatmosphäre (Eds.)Dt.Bundestag (1994) *Schutz der Grünen Erde*. Economica Verlag, Bonn.
- 23. Eastham K, Sweet J (2002) Genetically modified organisms (GMOs): The significance of gene flow through pollen transfer. Report, Environmental issue report No 28, A review and interpretation of published literature and recent/current research from the ESF 'Assessing the Impact of GM Plants' (AIGM) programme for the European Science Foundation and the European Environment Agency.
- 24. EC (2001) Directive 2001/18/EC of the European Parliament and of the Council of 12 March 2001 on the deliberate release into the environment of genetically modified organisms and repealing Council Directive 90/220/EEC. Community legislation in force Document 301L0018 Official Journal L 106, 17/04/2001 P. 0001 0039.
- 25. EC-Commission (2003) Commission Recommendation of 23 July 2003 on guidelines for the development of national strategies and best practices to ensure the co-existence of genetically modified crops with conventional and organic farming. COMMISSION OF THE EUROPEAN COMMUNITIES Brussels, 23 July 2003, C(2003).
- 26. EEA (2001) Late lessons from early warnings: the precautionary principle 1896-2000.

  Environmental issue report No 22, European Environment Agency (EEA).
- 27. Ellstrand NC (2001) When transgenes wander, should we worry? Plant Physiol **125**(4): 1543-1545.
- 28. Ellstrand NC (2003) Going to "Great Lengths" to Prevent the Escape of Genes That Produce Specialty Chemicals. Plant Physiology **132**(4): 1770.
- 29. Emberlin J, Adams-Groom B, Tidmarsh J (1999) *The dispersal of maize (Zea mays) pollen.*A report commissioned by the Soil Association: National Pollen Research Unit,
  University College Worcester, UK.
- 30. Ennos RA (1994) Estimating the relative rates of pollen and seed migration among plant populations. Heredity **72**: 250-259.
- 31. EU (2001) Richtlinie 2001/18/EG des Europäischen Parlaments und des Rates vom 12.

  März 2001 über die absichtliche Freisetzung genetisch veränderter Organismen in die Umwelt und zur Aufhebung der Richtlinie 90/220/EWG des Rates. Erklärung der Kommission Amtsblatt Nr. L 106 vom 17/04/2001 S. 0001 0039.
- 32. Fitter AH, Fitter RSR (2002) *Rapid Changes in Flowering Time in British Plants*. Science **296**(5573): 1689-1691.
- 33. Flam F (1994) Hints of a language. Science **266**(5189): 1320.
- 34. Gage SH, Isard SA, Colunga M (1999) *Ecological scaling of aerobiological dispersal processes*. Agricultural and Forest Meteorology **97**: 249-261.
- 35. Gee D (2003) late leesons from early warnings mfm.
  europa.eu.int/comm/health/ph\_determinants/
  environment/EMF/Conf24\_26feb2003/gee.pdf,
  http://europa.eu.int/comm/health/ph\_determinants/environment/EMF/conf24\_26feb200
  3/gee.pdf.
- 36. Giddings GD, Hamilton NRS, Hayward MD (1997) *The release of genetically modified grasses .2. The influence of wind direction on pollen dispersal.* Theoretical and Applied Genetics **94**(8): 1007-1014.
- 37. Gould S (2001) *Humbled by the genome's mysteries*. The New York Times , Opinion 19 02 2001 .
- 38. Hall L, Topinka K, Huffman J, Davis L, Good A (2000) *Pollen flow between herbicide-resistant Brassica napus is the cause of multiple-resistant B. napus volunteers.* Weed Science **48**: 688-694.
- 39. Hansen LB, Siegismund HR, Jorgensen RB (2003) *Progressive introgression between Brassica napus (oilseed rape) and B. rapa.* Heredity **91**(3): 276-283.

- 40. Hare MP, Palumbi SR (2003) *High Intron Sequence Conservation Across Three Mammalian Orders Suggests Functional Constraints*. Molecular Biology and Evolution **20**(6): 969-978.
- 41. Hemmer W, Focke M, Wantke F, Jager S, Gotz M, Jarisch R (1997) *Oilseed rape pollen is a potentially relevant allergen*. Clin Exp Allergy **27**(2): 156-161.
- 42. Hennig W (2001) Genetik. 3. überarbeitete und erweiterte Auflage Springer Verlag 853pp.
- 43. Hjalmars U, Kulldorff M, Wahlqvist Y, Lannering B (1999) *Increased incidence rates but no space-time clustering of childhood astrocytoma in Sweden, 1973-1992: a population-based study of pediatric brain tumors.* Cancer **85**(9): 2077-2090.
- 44. Hoffmann W, Mudra P, Plarre W (1985) *Lehrbuch der Züchtung landwirtschaftlicher Kulturpflanzen*. Verlag Paul Parey.
- 45. Hokanson SC, Grumet R, Hancock JF (1997) Effect of border rows and trap/donor ratios on pollen-mediated gene movement. Ecological Applications **7**(3): 1075-1081.
- 46. Hoppichler J (1998) *GVO-freie ökologische sensible Gebiete*. 89pp, Studie im Auftrag des Bundesministeriums für Frauenangelegenheiten und Verbraucherschutz .
- 47. Hoppichler J (1999) ExpertInnenbefragung zur Bewertung und Evaluation "GVO-freier ökologisch sensibler Gebiete". 89pp, Forschungsberichte 10/99, Studie im Auftrag des Bundeskanzleramt Sektion VI.
- 48. Horak F, Jager S, Skoda-Turk R (1980) *The relevance of cereal grain pollen in hayfever* . Wien.Klin.Wochenschr.Suppl **117**: 34-35.
- 49. Hu XS, Ennos RA (1999) *Impacts of seed and pollen flow on population genetic structure for plant genomes with three contrasting modes of inheritance.* Genetics **152**(1): 441-450.
- 50. Ingram J (2000) Report on the separation distances required to ensure cross-pollination is below specified limits in non-seed crops of sugar beet, maize and oilseed rape. National Institute of Agricultural Botany, Cambridge UK. published by Ministry of Agriculture Fisheries and Food - UK (MAFF).
- 51. IPTS (Eds.) (2002) Scenarios for co-existence of genetically modified, conventional and organic crops in European agriculture. A synthesis report prepared by Anne-Katrin Bock, Karine Lheureux, Monique Libeau-Dulos, Hans Nilsagård, Emilio Rodriguez-Cerezo (IPTS JRC).
- Jorgensen RB (1999) *Gene flow from oilseed rape (Brassica napus) to related species*. In: Lutman PJW (Hg.) *Gene Flow and Agriculture: Relevance for Transgenic Crops*. pp. 117-123, British Crop Protection Council.
- 53. Kimura M (1987) Die Neutralitätstheorie der molekularen Evolution. Parey, Berlin [u.a.].
- 54. Kjellsson G, Simonsen V, Ammann K (1997) *Methods for risk assessment of transgenic plants. 2. Pollination, gene-transfer and population impacts.* Birkhäuser Verlag.
- 55. Klinger T, Ellstrand NC (1994) *Engineered genes in wild populations: fitness of weed-crop hybrids of radish, Raphanus sativus L.* Ecological Applications **4**: 117-120.
- 56. Klöpffer W (1994) Kriterien zur Umweltbewertung von Einzelstoffen und Stoffgruppen. UWSF-Z.Umweltchem.Ökotox. **6**(2): 61-63.
- 57. Knippers J (2001) *Molekular Genetik*. 8. neu bearbeitete Auflage, Thieme Verlag, 586pp.
- 58. Kowarik I (1996) *Auswirkungen von Neophyten auf Ökosysteme und deren Bewertung*. Texte Umweltbundesamt Berlin **58/96**: 119-155.
- 59. Kuiper HA, Kleter GA, Noteborn HP, Kok EJ (2001) Assessment of the food safety issues related to genetically modified foods. Plant J. **27**(6): 503-528.
- 60. Lavigne C, Klein EK, Vallee P, Pierre J, Godelle B, Renard M (1998) *A pollen-dispersal* experiment with transgenic oilseed rape. Estimation of the average pollen dispersal of an individual plant within a field. Theoretical and Applied Genetics **96**: 886-896.
- 61. Li W-H (1997) Molecular evolution. Sinauer associates, inc.
- 62. Luna V, Figueroa M, Baltazar M, Gomez L, Townsend R, Schoper JB (2001) *Maize Pollen Longevity and Distance Isolation Requirements for Effective Pollen Control.* Crop Science **41**(5): 1551.
- 63. McBride KE, Svab Z, Schaaf DJ, Hogan PS, Stalker DM, Maliga P (1995) Amplification of a chimeric bacillus gene in chloroplast leads to an extraordinary level of an insecticidal protein in tobacco. Bio/Technology **13**: 362-365.
- 64. McKinney PA, Feltbower RG, Parslow RC, Lewis IJ, Glaser AW, Kinsey SE (2003) *Patterns of childhood cancer by ethnic group in Bradford, UK 1974-1997*. Eur J Cancer **39**(1): 92-97.
- 65. Millstone E, Brunner E, Mayer S (1999) Beyond 'substantial equivalence'. Nature **401**(6753): 525-526.

- 66. Monk M (1995) Epigenetic programming of differential gene expression in development and evolution. Developmental Genetics **17**(3): 188-197.
- 67. Mooney HA, Drake JA (1989) *Biological Invasions: a SCOPE Program Overview*. In: Drake JA, Mooney HA, di Castri F, Groves RH, Kruger FJ, Rejmánek M, Williamson M (Hg.) *Biological Invasions: a Global Perspective*. pp. 491-506, John Wiley & Sons.
- 68. Müller W (2000) Die Problematik der genetischen Verschmutzung hinsichtlich des Aspektes der Sortenreinheit von Kulturpflanzen im Ökologischen Landbau in Österreich. Forschungsberichte 9/2000 Bundesministeriums f. Soziale Sicherheit und Generationen Sektion IX
- 69. Noble IR (1989) Attributes of Invaders and the Invading Process: Terrestrial and Vascular Plants. In: Drake JA, Mooney HA, di Castri F, Groves RH, Kruger FJ, Rejmánek M, Williamson M (Hg.) Biological Invasions: A Global Perspective. pp. 301-313, John Wiley & Sons.
- 70. Odenbach W (1997) Biologie von Fortpflanzung und Vermehrung Geschlechtliche Fortpflanzung. In: Odenbach W (Hg.) Biologische Grundlagen der Pflanzenzüchtung. pp. 97-119, Parey, Berlin.
- 71. OECD (2000) Uncertainty and Precaution: Implications for Trade and Environment. Joint Working Party on Trade and Environment, Organisation for Economic Co-operation and Development, COM/ENV/TD(2000)114/REV1 For Official Use 04-May-2001.
- 72. Parker VT, Simpson RL, Leck MA (1989) *Pattern and process in the dynamics of seed banks*. In: Leck MA, Parker VT, Simpson RL (Hg.) *Ecology of soil seed banks*. pp. 367-384, Academic press.
- 73. Pascher K, Macalka-Kampfer S, Reiner H (2000) *Vegetationsökologische und genetische Grundlagen für die Risikobeurteilung von Freisetzungen von transgenem Raps und Vorschläge für ein Monitoring*. Bundesministerium f. soziale Sicherheit und Generationen, Forschungsberichte 7/2000.
- 74. Pekrun C, Lane PW, Lutman PJW (1999) *Modelling the potential for gene escape in oilseed rape via the soil seedbank: its relevance for genetically modified cultivars.* In: Lutman PJW (Hg.) *Gene Flow and Agriculture: Relevance for Transgenic Crops.* pp. 101-106, British Crop Protection Council.
- 75. Pessel F, Lecomte J, Emeriau V, Krouti M, Messean A, Gouyon P (2001) *Persistence of oilseed rape (Brassica napus L.) outside of cultivated fields.* Theoretical and Applied Genetics **102**(6/7): 841-846.
- 76. Pollan M, Lopez-Abente G, Ardanaz E, Moreo P, Moreno C, Vergara A, Aragones N (1997) Childhood cancer incidence in Zaragoza and Navarre (Spain): 1973-1987. Eur J Cancer 33(4): 616-623.
- 77. Ramsay G, Thompson CE, Neilson S, Mackay GR (1999) *Honeybees as vectors of GM oilseed rape pollen*. In: Lutman PJW (Hg.) *Gene Flow and Agriculture: Relevance for Transgenic Crops*. pp. 209-214, British Crop Protection Council.
- 78. Raybould AF, Clarke RT (1999) *Defining and measuring gene flow.* In: Lutman PJW (Hg.) *Gene Flow and Agriculture: Relevance for Transgenic Crops.* pp. 41-48, British Crop Protection Council.
- 79. Rejmánek M (1989) *Invasibility of Plant Communities*. In: Drake JA, Mooney HA, di Castri F, Groves RH, Kruger FJ, Rejmánek M, Williamson M (Hg.) *Biological Invasions: A Global Perspective*. pp. 369-388, John Wiley & Sons.
- 80. Rieger MA, Lamond M, Preston C, Powles SB, Roush RT (2002) *Pollen-Mediated Movement of Herbicide Resistance Between Commercial Canola Fields*. Science **296**(5577): 2386-2388.
- 81. Ruckenbauer P, Steiner AM (1995) *Die agronomischen Eigenschaften des Nachbaues* eines 110 Jahre alten Hafers die Wiener Probe von 1877. Die Bodenkultur **46**(4): 293-302.
- 82. Saure C, Kühne S, Hommel B (2000) Bewertung der insekten- und windbedingten Pollenübertragungen von gentechnisch verändertem Raps auf artverwandte Kreuzblütler. Mitt.Biol.Bundesanst.Land-Forstwirtsch. 376: 157, 52. Deutsche Pflanzenschutztagung, Freising Weihenstephan.
- 83. Scheffler JA, Parkinson R, Dale PJ (1995) Evaluating the effectiveness of isolation distances for field plots of oilseed rape (Brassica napus) using a herbicide-resistance transgene as a selectable marker. Plant Breeding **114**: 317-321.
- 84. Scheffler JA, Parkinson R, Dale PJ (1993) Frequency and distance of pollen dispersal from transgenic oilseed rape (Brassica napus). Transgenic Research **2**: 356-364.

- 85. Scheringer M (1999) *Persistenz und Reichweite von Umweltchemikalien*. Wiley-VCH, Weinheim.
- 86. Schlink S (1998) 10 Jahre Überdauerung von Rapssamen (Brassica napus L.) im Boden.

  Band 11: 221-222, Mitteilungen der Gesellschaft für Pflanzenbauwissenschaften,
  Wissenschaftlicher Fachverlag.
- 87. Schulz R, Jakisch W (2001) *Was leisten neue Winterrapssorten?* Sächsiche Landesanstalt für Landwirtschaft Juli 2001, http://www.landwirtschaft-mv.de/rapsneu.mv.
- 88. SCP (2001) Opinion of the Scientific Committee on Plants concerning the adventitious presence of GM seeds in conventional seeds. (Opinion adopted by the Committee on 7 March 2001). -20pp, SCP/GMO-SEED-CONT/002-FINAL. (http://europa.eu.int/comm/food/fs/sc/scp/index\_en.html) European Commission: Health & Consumer Protection Directorate-General.
- 89. shCherbak VI (2003) *Arithmetic inside the universal genetic code*. Biosystems **70**(3): 187-209.
- 90. Simpson EC, Norris CE, Law JR, Thomas JE, Sweet JB (1999) Gene flow in genetically modified herbicide tolerant oilseed rape (Brassica napus) in the UK. In: Lutman PJW (Hg.) Gene Flow and Agriculture: Relevance for Transgenic Crops. pp. 75-81, British Crop Protection Council.
- 91. Sklar CA (2002) Childhood brain tumors. J Pediatr Endocrinol Metab 15 Suppl 2: 669-673.
- 92. Snow AA, Bente A, Jorgensen R (1999) Costs of transgenic herbicide resistance introgressed from Brassica napus into weedy B. rapa. Molecular Ecology **8**(4): 605-615.
- 93. Spök A, Hofer H, Valenta R, Kienzl-Plochberger K, Lehner P, Gaugitsch H (2002)

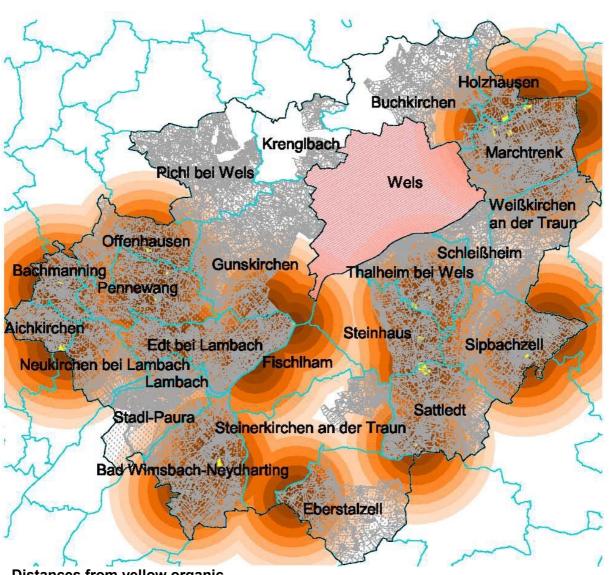
  Toxikologie und Allergologie von GVO-Produkten Empfehlungen zur Standardisierung der Sicherheitsbewertung von gentechnisch veränderten Pflanzen auf Basis der Richtlinie 90/220/EWG (2001/18/EG). UBA-Monographien M-109.
- 94. Squire GR, Crawford JW, Ramsay G, Thompson CE (1999) *Geneflow at the landscape level*. In: Lutman PJW (Hg.) *Gene Flow and Agriculture: Relevance for Transgenic Crops*. pp. 57-64, British Crop Protection Council.
- 95. SRU (1985) *Umweltprobleme der Landwirtschaft. Sondergutachten des Sachverständigenrates für Umweltfragen, Unterrichtung an die Bundesregierung.* Kohlhammer-Verlag. Stuttgart.
- 96. Stokstad E (2002) AGBIOTECH: A Little Pollen Goes a Long Way. Science **296**(5577): 2314.
- 97. Stotzky G (2001) 2000 Progress Report: Toxins of Bacillus thuringiensis in Transgenic Organisms:Persistence and Ecological Effects. National Center for Environmental Research Office of Research and developement US Environmental Protection Agency.
- 98. Sukopp H, Sukopp U (1995) Ökologische Modelle in der Begleitforschung zur Freisetzung transgener Kulturpflanzen. In: Albrecht S and Beusmann V (Hg.) Ökologie transgener Nutzpflanzen. pp. 41-64, Campus Verlag, Franfurt/Main.
- 99. Sustain (1994) Forschungs- und Entwicklungsbedarf für den Übergang zu einer nachhaltigen Wirtschaft in Österreich. Endbericht der Wissenschaftergruppe "Sustain", TU Graz.
- Thompson CE, Squire GR, Mackay GR, Bradshaw JE, Crawford JW, Ramsay G (1999) Regional pattern of gene flow and its consequences for GM oilseed rape. In: Lutman PJW (Hg.) Gene Flow and Agriculture: Relevance for Transgenic Crops. pp. 95-106, British Crop Protection Council.
- 101. Thompson K, Bakker J, Bekker (1997) *The soil seed banks of North West Europe: methodology, density and longevity.* Cambridge University Press.
- Tilman D (1997) Community invasibility, recruitment limitation, and grassland biodiversity. Ecology **78**: 81-92.
- Timmons AM, Charters YM, Crawford JW, Burn D, Scott SE, Dubbels SJ, Wilson NJ, Robertson A, O'Brien ET, Squire GR, Wilkinson MJ (1996a) Risks from transgenic crops [letter]. Nature 380(6574): 487.
- Timmons AM, Charters YM, Crawford JW, Burn D, Scott SE, Dubbels SJ, Wilson NJ, Robertson A, O'Brien ET, Squire GR, Wilkinson MJ (1996b) *Risks from transgenic crops [letter]*. Nature **380**(6574): 487.

- Timmons AM, O' Brien ET, Charters YM, Dubbels SJ, Wilkinson MJ (1995) Assessing the risks of wind pollination from fields of genetically modified Brassica napus ssp. oleifera. Euphytica-. **85**: 417-423.
- Tolstrup K, Andersen S, Boelt B, Buus M, Gylling M, Holm P, Kjellsson G, Pedersen S, Ostergard H, Mikkelsen S (2003) Report from the Working Group on: The co-existence of genetically modified crops with conventional and organic crops. Conclusion and Summary. Ministry of Food, Agriculture and Fisheries, http://www.fvm.dk/file/Summary.pdf, January 2003.
- 107. Treu R, Emberlin J (2000) Pollen dispersal in the crops Maize (Zea mays), Oil seed rape (Brassica napus ssp oleifera), Potatoes (Solanum tuberosum), Sugar beet (Beta vulgaris ssp. vulgaris) and Wheat (Triticum aestivum). A report for the Soil Association from the National Pollen Research Unit, www.soilassociation.org.
- 108. Urbanska KM (1992) *Populationsbiologie der Pflanzen*. UTB-für Wissenscchaft 1631 Gustav Fischer Verlag.
- 109. Wagner DB, Allard RW (1991) *Pollen migration in predominantly self-fertilizing plants:* barley. J Hered. **82**(4): 302-304.
- 110. Wahl J (1991) Bewertung der Keimfähigkeit und Triebkraft sowie Untersuchung der Beziehung zwischen Feldaufgang und Labormethoden von 8 Saatgutpartien der Sojabohne Glycine max. L. Merr. Diplomarbeit am Inst. F. Pflanzenbau u. Pflanzenzüchtung, BOKU Wien.
- 111. Wenk N, Stebler D, Bickel R (2001) Warenflusstrennung von GVO in Lebensmitteln.

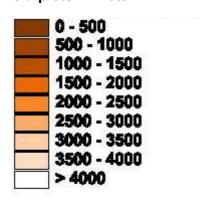
  Prognos Europäisches Zentrum für Wirtschafts- und Strategieberatung. Untersuchung im Auftrrag des Amtes für Gesundheit Schweiz.
- 112. Westbrook JK, Isard SA (1999) *Atmospheric scales of biotic dispersal*. Agricultural and Forest Meteorology **97**: 263-274.
- 113. Williamson M (1993) . Experientia 49(219): 224.
- 114. Williamson M (1996) Biological invasions. Chapman & Hall, London.

# MAP 1 District of WELS LAND

# Protection area needed for organic crops in the district of Wels Land



Distances from yellow organic field plots in meter



# Data courtesy of

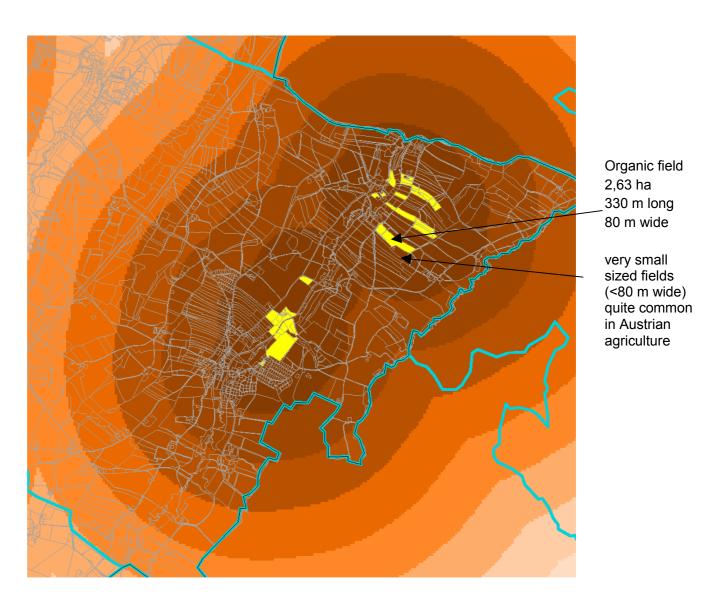
- Federal Ministry of Agriculture, Forestry, Environment and Water Management
- Federal Weights and Measures Office
- Province of Upper Austria (DORIS)

# **Calculation and Interpretation:**

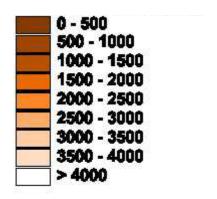
Andreas Bartel, University of Agricultural Sciences, Vienna

Werner Müller, Eco-Risk, Vienna

MAP 2
Small Scale Structure of Austrian Agricultural Fields



Distances from yellow organic field plots in meter



#### **DATA COURTESY OF**

- Federal Ministry of Agriculture, Forestry, Environment and Water Management
- Federal Weights and Measures Office
- Province of Upper Austria (DORIS)

# **Calculation and Interpretation:**

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