

Annex 2: Online Survey Environment

BEETLE

Biological and Ecological Evaluation
towards Long-Term Effects



Long-term effects of genetically modified (GM) crops on health, biodiversity and the environment: prioritisation of potential risks and delimitation of uncertainties

Reference: ENV.B.3/ETU/2007/0007

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1. Introduction

The main objective of the project was the prioritisation of potential long-term effects of GM plants on the environment including biodiversity as well as the identification of areas of greatest scientific uncertainty. To achieve this goal, the BEETLE approach included consulting an extended expert panel and conducting a stepwise assessment. The conclusions of this allowed a ranking of potential long-term effects to be established, aimed at supporting the Commission in the ongoing task of improving GMO risk assessment and management. In Step 1 of the BEETLE study a broad Literature Review of published peer-reviewed data and internal documents was performed to collect and collate information on established and potential long-term effects (see Annex 1 of the BEETLE main report). Based on this initial collection, the BEETLE team identified in Step 2 about 26 general mechanisms ('processes') and assigned them to seven categories (A-G). Among the 'processes' about 63 more detailed scenarios for specific crop/trait combinations were elaborated that potentially could lead to long-term environmental effects (see Table 6 of the BEETLE main report). The processes and scenarios selected were cross-checked with the Peer-Review Committee. These results were taken as the basis for Step 3, the development of questionnaires sent to an extended expert panel (EEP) via online surveys. Representatives of four stakeholder groups: (i) scientists, (ii), regulators, (iii) representatives of companies, and (iv) representatives of NGOs were invited to participate in the Online Survey. In any case, main selection criterion was the individual scientific expertise. The extended expert panel had the following tasks:

- (i) proof the completeness of the listed processes,
- (ii) approve or correct the preliminary ranking, and
- (iii) identify areas of uncertainty.

The experts specifically assessed whether the listed processes are relevant for the crop/trait combinations cultivated today or in the near future in the EU (Bt maize, HT oilseed rape, HT sugar beet, HT soybean, SM potato). It is important to emphasize that this evaluation did not lead to an absolute or quantitative but a relative ranking and prioritisation among the processes. The expert contributions in the Online Survey helped to identify

- a) processes with a high potential to cause adverse long-term effects, and on which there was good agreement among members of the panel;
- b) areas of uncertainty highlighted by an ambiguous response or a high proportion of the answer 'don't know, insufficient data'.

2. Material & Methods

Based on the literature review, 26 processes (without the questions looking at stacked events) with the potential to cause adverse long-term effects were identified. In the first part of the survey, the experts were asked whether the list for each category was complete. In an open response option they had the opportunity to add possibly unrecognized processes or to comment on the general process list. In the second, more detailed, part of the survey, the experts were asked to evaluate the relevance of the potential long-term effects in relation to crop/trait examples (scenarios). Several potential long-term scenarios were proposed, and the experts were asked to indicate for each of them one of the following likelihoods: 'negligible', 'low' or 'high'. If the experts did not feel competent or had the impression that the data basis was insufficient, they had the opportunity to answer 'don't know, no expert' or 'don't know, insufficient data' respectively. For certain crop/trait combinations, the experts were asked to select among the assessment options 'low', 'medium'¹ or 'high'. Furthermore, the experts were asked whether (i) the mentioned processes should be considered differently with respect to geographically different EU regions, and (ii) whether they would change their assessment if they were assessing stacked events, both intended and unintended. Finally, the experts were asked which field of research regarding cultivation of GMOs in the EU should have the highest priority for financial support.

The structure of the Online Survey Environment allowed a relative assessment based on the experts' knowledge and their personal interpretation of the probability of occurrence of adverse long-term effects. The BEETLE team analysed the expert assessments in detail. An overview of how conclusions were drawn is given in Table 1, 2, and 3. The BEETLE team defined the decision criteria for the overall assessment of the responses (Table 3).

¹ The category 'medium' was used as intermediate category if 'negligible' could already be excluded (see Annex 2 Tables 1, 2, and 3). However, prioritization in the BEETLE study led finally to classification of the potential adverse effects into the categories 'most likely', 'likely', and 'not likely' (see 'BEETLE' Main Report).

Table 1: Assessment options in the Online Survey “Environment” and corresponding BEETLE team interpretation regarding processes

Assessment options:	BEETLE team interpretation
negligible	The likelihood of occurrence of the process causing adverse long-term effects is negligible (can be excluded with a high confidence). Therefore, no need for additional measures is given, remaining risks are covered by general surveillance (monitoring).
low	The occurrence of the process causing adverse long-term effects is possible to a low extent, on a case by case basis additional research or additional measures should be taken into account.
high	The occurrence of the process causing adverse long term effects is possible to a relatively high extent, risk management measures are necessary to protect the environment.
don't know, insuff. data	An assessment of the process is not possible due to insufficient data. A high percentage of this answer to the survey question highlights an area of higher uncertainty; therefore more research is needed to close knowledge gaps.
don't know, no expert	An assessment of the process is not possible due to a lack of personal expertise; this answer was inevitable given the wide field of environmental disciplines addressed.

Table 2: Assessment options in the Online Survey “Environment” and corresponding BEETLE team interpretations regarding specific crop/trait combinations

Assessment options ²	Conclusions
low	The specific process for specific crop/trait combinations may generally occur, but the potential of occurrence is low. No need for additional measures is given, remaining risks are covered by e.g. general surveillance (monitoring).
medium	The specific process for specific crop/trait combinations may generally occur, therefore on a case-by-case analysis additional research or specific measures are necessary.
high	The likelihood of occurrence of the process for specific crop/trait combinations potentially causing adverse long-term effects is relatively high; risk management measures are necessary to protect the environment.

² Used for crop/trait combination if the likelihood of a process causing adverse long-term effects cannot be assessed as ‘negligible’ (see footnote 1)

Table 3: Decision criteria for the overall assessment of processes

Interpretation	Decision criteria	Assessment options	Relative response of the experts
'negligible'	a) The majority (more than 50%) of the experts decided for the response option 'negligible' ¹	negligible low high don't know, insufficient data don't know, no expert	67% 17% 2% 6% 8%
'low'	a) The majority of the experts (more than 50%) responded with the option 'low' ²	negligible low high don't know, insufficient data don't know, no expert	21% 52% 25% 0% 2%
	b) No option received more than 50% of the experts' votes, but a clear tendency was recognizable. More responses were found for the options 'negligible' and 'low' together than for the options 'low' and 'high' together ³	negligible low high don't know, insufficient data don't know, no expert	27% 35% 15% 8% 15%
'high'	a) The majority of experts (more than 50%) responded with the option 'high'	no example found in the online survey	
	b) No option received more than 50% of the experts' votes, but a clear tendency was recognizable. More responses were found for the options 'low' and 'high' together than for the options 'low' and 'negligible' together ⁴	negligible low high don't know, insufficient data don't know, no expert	20% 28% 34% 2% 16%
'area of uncertainty'	a) No clear tendency recognizable for one of the assessment options 'negligible', 'low' or 'high' ⁵	negligible low high don't know, insufficient data don't know, no expert	33% 16% 28% 9% 14%
	b) A disproportionate percentage of the experts responded 'don't know, insufficient data' ⁶	negligible low high don't know, insufficient data don't know, no expert	40% 18% 10% 22% 10%

¹Example taken from process A.1.1

²Example taken from process C.1.1

³Example taken from process A.1.5

⁴Example taken from process F.1.2

⁵Example taken from process A.3.1

⁶Example taken from process F.5.1

Altogether 167 experts were invited to participate in the Online Survey Environment. The majority of the experts were scientists from research institutions and universities. They came from 16 different European and 5 different overseas countries. The participants were selected based on three major criteria: (a) known expertise substantiated by relevant scientific publications cited in the ICGEB database, (b) added value for the requested fields in the BEETLE project (due to the area of specific competence) and (c) known representativeness for important stakeholder groups (see Table 4.). In particular experts from countries with a long experience of GM cultivation were invited to participate from outside the EU.

The ICGEB database³ is a scientific bibliographic collection of studies on 'Biosafety and Risk Assessment in Biotechnology'. On 11 December 2007 the database held a total count of 6,166 records and 11,828 authors. The database is updated monthly and contains scientific articles (full reference + abstract), published in international scientific journals or conference proceedings from 1990 onwards, selected and classified by ICGEB scientists for the main topics of concern for the environmental release of GMO. All the records have been extracted from the internationally renowned applied life sciences database CAB ABSTRACTS [TM], and AgBiotechNet, the online service for Agricultural Biotechnologists from CABI Publishing. The CABI choice is based on the concept of avoiding 'any unnecessary duplication' but collecting very broadly available scientific information. CABI holds the main collection of data on biosafety which are not focused only on human health (main topic of PubMed, free accessible database of scientific bibliographic information, developed by NCBI primarily from MEDLINE and PreMEDLINE).

In addition, some experts were selected based on single publications in a field that was not sufficiently covered by the ICGEB database. Membership in an European Commission Monitoring Working Group, or participation in EU funded research projects highly relevant to BEETLE or national Biosafety Commissions were other selection criteria.

For completeness, representatives of three important stakeholder groups were also invited to participate in the Online Survey: Companies developing the GM plant applications at the EU level, non-governmental organisations (NGO) contributing scientifically to the GMO debate and regulators working in governmental bodies. Known experts were chosen representing the major companies. Members of NGO were selected based on recommendations of the Peer Review Committee, and invited in a similar number as the representatives of the companies for balancing reasons.

³ (see <http://www.icgeb.org/~bsafesrv/bsfdata1.htm>)

3. Results

3.1. General aspects

A total number of 167 experts were invited to participate in the Online Survey. At least 100 experts registered for the Online Survey (see Table 4). However some of them rejected the Online Survey in principle and did not answer the questions or answered only some questions. On average 53 experts (29.7%) responded to each question (range 43-62). The tendency was observed that the number of answers decreased to the end of the Online Survey. From the stakeholder 'NGO' group only one expert answered all questions. Three NGO experts registered but did not answer any questions. The reasons for this reaction remain unclear.

Table 4: Participants of the Online Survey Environment. Presented are number of invited and participating experts for each stakeholder group and the relative proportion.

Stakeholder	Number of participants		Percentage	
	invited	participating	invited	participating
Research institution	114	59	68.3%	59 %
Regulation	24	16	14.4 %	16 %
Industry	15	16	9.0 %	16 %
NGO	14	4	8.4 %	4 %
Other*	-	5	-	5%
Sum	167	100	100 %	100%

*Differences between the 'invited' and 'participating' stakeholder affiliation are caused by the fact that experts relocated themselves to other stakeholder groups after registration.

3.2. Category A: Persistence and invasiveness

In this category potential long-term effects were addressed related to changed fitness effects of crops or wild relatives due to the genetic modification or gene flow respectively. A potential consequence might be an increase of feral crop populations or problems with volunteers or herbicide resistant weeds. In particular, the following three separate processes

- increased fitness,
- effects to out breeding depression and
- increased invasiveness of hybrids

had to be assessed distinctively in accordance to the selected crop/trait combinations.

Potential adverse effects due to an increasing number of volunteers or the occurrence of ferals were only relevant for HT crops according to the experts. The general question of

whether HT plants have the potential to affect the environment adversely was ambiguously answered by the experts (negl. 35%, low 27% high 22%, insuff. data 8%). In-depth analysis revealed a clear picture. The highest probability of potential long-term effects was expected for oilseed rape and sugar beet due to their biology and the existence of cross-compatible wild relatives in Europe. For the other crop/trait combinations the potential was assessed as 'negligible' or 'low'. Interestingly, 10% of the experts were of the opinion that the amount of data is still insufficient for starch-modified potato.

For most of the experts, adverse long-term effects caused by outbreeding depression are 'negligible' in general and 'low' for the listed crop/trait combinations. However 14% of the experts were of the opinion that the data basis is still insufficient.

The potential adverse effects due to hybrids persisting outside of fields were assessed ambiguously by the experts. The answers were characterized by an equally distributed response and two questions drew 'insufficient data' as the response from 11% and 13% respectively of the experts who answered.

3.3. *Category B: Altered gene transfer*

Category B focused on potential long-term effects caused by altered gene transfer. 60% of the experts were of the opinion that phenotypic effects influencing gene flow (like flowering and fecundity) were addressed sufficiently in the environmental risk assessment. Just some experts were more sceptical (13% and 9% respectively) stating that the data basis was insufficient for a reliable risk assessment. The question of whether horizontal gene transfer (HGT) from crops to microorganisms should be assessed during the ERA was answered by 51% of the experts with 'no', whereas only 28% of the experts would recommend taking HGT into consideration. A similar opinion was expressed in response to the question of whether such effects should be assessed for interspecific hybrids and wild relatives in the environmental risk assessment. However, it should be taken into consideration that the listed processes in category B were of general importance and not specific to the selected crop/trait combinations.

3.4. *Category C: Interactions between the GM plant and target organisms*

GM traits may lead to resistance development in target organisms (pests or pathogens). This process applies only for Bt-maize in the EU as the other considered crop/trait combinations (HT and SM crops) do not have any distinct target organism. Potential indirect effects caused by the use of non-selective herbicides on weeds (corresponding to the target organisms in HT crops) were addressed in category F.

Most of the experts were of the opinion that this issue is of 'low' (52%) to 'high' relevance (26%) whereas 20% of the experts stated a 'negligible' potential.

3.5. Category D: Interactions of the GM plant with non-target organisms

Altogether 6 processes were identified potentially causing adverse long-term effects of GM crops to NTOs. These were assigned to

- sublethal toxicity to chronic exposure,
- changes in nutritional composition or value,
- accumulation of toxic compounds, or
- changed interaction with symbionts.

HT crops were not taken into consideration because the recombinant proteins expressed in these crops are ubiquitous and known as non-toxic. Therefore only Bt-crops expressing lepidopteran (ECB; European Corn Borer) or coleopteran (WCR, Western Corn Rootworm) - specific proteins and starch-modified potatoes were assessed.

In most cases the majority of the experts considered that the likelihood of the listed processes causing potential adverse long-term effects was negligible. Exceptions were potential long-term effects on ECB or WCR-resistant Bt-crops to NTO with a close relationship to the target organisms since a similar mode of action of the Bt protein in these NTO's can be expected. However, in several cases more than 10% of the experts held that the available data basis is insufficient, in particular for the following issues: interaction between Bt-proteins and microorganisms (19%), rhizosphere organisms (17%) or mycorrhiza (17%) and between herbivores and starch-modified potatoes (19%).

3.6. Category E: Processes related to effects on ecological functions'

Three ecological functions (services) were addressed in detail, which might be potentially altered by GM plants over the long-term: (i) soil fertility, (ii) biological control and (iii) pollination. None of the experts asked for additional processes or ecological functions to be considered. Most of the experts assessed the likelihood of potential adverse long-term effects on ecological functions as 'negligible' for the listed crop/trait combinations. In two cases - potential effects of increased lignin content in Bt-maize and sub-lethal toxic effects to natural enemies - 12% and 14% respectively of the experts were of the opinion that the data basis is insufficient.

3.7. *Category F: Impacts on specific cultivation, management and harvesting techniques*

This category integrated different aspects of potential indirect effects of GM plants on agriculture. Major processes included the use of agrochemicals including fertilizers, changes in susceptibility against pathogens, in agrobiodiversity and landscape structure. The answers were characterized by a high number of answers in the assessment option 'insufficient data' or 'no expert' in general. One reason could be that the data basis for the assessment is deficient. At the same time a high number of the consulted 'generalists' among the experts may have felt lacking in personal expertise. High uncertainty was expressed in particular for cases regarding the use of GM plants with complementary non-selective herbicides.

Many experts believed that weed communities might be affected on a low level through changes by tillage, herbicide drift or development of secondary pests (in the context of Bt-maize). Questions regarding increasing number of volunteers, increasing development of tolerant weeds or consequences of changes in weed communities causing changes in ecological functions were answered ambiguously. In contrast, all other aspects were assessed by the majority of those experts who did not answer 'don't know' as 'negligible'.

3.8. *Category G: Potential interactions with the abiotic environment*

Potential impact of GMOs on the abiotic environment was addressed in Category G. The processes included increased production of greenhouse gases, increased mineral nutrient erosion, fertilizer leaching into water bodies affecting water quality and altered soil particle exchange sites, e.g. due to increased release of Bt-protein or of herbicides and their metabolites into the soil. In all cases the majority of the experts assessed the likelihood of adverse potential long-term effects on the abiotic environment as 'negligible'. However, a noteworthy number of experts (30% in two cases) felt that they did not have the expertise to answer the questions in this category.

3.9. *Regional aspects*

For each group of processes mentioned in categories A-G, the experts were asked whether the assessment needs differentiation concerning geographical regions in Europe. In many cases the majority of experts answered 'yes' or the responses were ambiguous. This relates to the conclusion that there seems to be a need for more regional approaches within the risk assessment; since many experts felt uncertain. Regional aspects played a relatively minor role in the assessment of nutritional composition, toxic compounds, interactions between GM crops and mycorrhiza and bacteria, fitness change due to root exudation, effects on pollination, effects due to fertilizer use and all aspects taken into account in category G (Potential interactions with the abiotic environment).

3.10. Stacked events

For each category mentioned the experts were asked for their view on stacked events and a potential differentiation with regard to intended or unintended stacks. For all processes, the majority of the experts did not consider it necessary to differentially assess intended and unintended stacks. About 10% of the experts were of the opinion that the data basis is insufficient for the following processes: interactions with NTO, effects on ecological functions and effects on cultivation and management.

3.11. Open Comments

The experts were additionally asked whether the list of processes was complete within each category. In general, most of the experts were of the opinion that the listed processes in the different categories were complete (ranging from 70% to 82%).

Some experts missed a specific process in one category, but this process was listed later – assigned by the BEETLE team – in another category. No principally new processes were added by those experts who had the feeling that the categories were incomplete. Instead, the majority of experts who were critical expressed general doubt that the strategy used would be suitable to assess potential long-term effects.

Single comments were made stating that

- the questions in the survey were inherently biased, or
- all questions would be answered in the environmental risk assessment or
- such a list could never be complete because it depends on the specific genetic modification in terms of a 'case-by-case approach'.

Three issues were addressed by various experts with respect to different categories and repeated several times:

- (i) What is or will be the baseline for comparisons as some of the potential long-term effects could also be true for classic breeding and conventional agriculture?
- (ii) The need to balance benefits and adverse long-term effects by using e.g. a risk benefit analysis; and
- (iii) The need for case-specific approaches; definitions of general processes would not be helpful.

3.12. Final Question

At the end of the survey the experts were asked to which field of research regarding cultivation of GMOs in EU the highest priority for financial support should be given. The answers were expected to provide additional hints regarding the most important areas of uncertainty. The relative majority of experts recommended to invest money into the area Cultivation and Management, followed by the issues Non-Target Organisms, Gene Flow (persistence and invasiveness) and Ecological Functions.

4. Summary

The Online Survey Environment provided additional information for the prioritization process and helped to identify areas of uncertainty. However, it has to be considered that the results reflect a frequency distribution of the personal opinions and assessments of the participating experts since they were selected for their personal expertise but not randomly. Thus it is not allowed to deduce a probability of incidence of potential long-term effects.

Interestingly, the experts came to relatively clear assessments for most processes and cases. Potential long-term effects were considered likely (assessment options 'low' or 'high') in relation to

1. Cultivation and Management in particular of HT crops,
2. increased tendency of Persistence and Feralization of HT oilseed rape and HT sugar beet, including their potential hybrids with wild relatives,
3. Ecological Functions - specifically soil functions,
4. ECB or WCR-resistant Bt-crops and NTOs closely related to the target organisms, and
5. Potential Resistance Development of target organisms.

Areas of greatest uncertainty were indicated by ambiguous responses of the experts or a high percentage of the response 'don't know, insufficient data'. These were single processes and cases from the categories

- Cultivation and management,
- interaction with NTOs and
- Persistence and Invasiveness.

Over all categories, interactions between GM crops and soil organisms and effects in relation to starch-modified potatoes were conspicuously classified in this 'uncertainty' way.

A majority of experts agreed that potential adverse effects caused by (i) 'invasiveness, persistence or hybridization' of GM crops and/or hybrids with their wild relatives or by (ii) changes in 'cultivation and management' should be considered differently with respect to geographically different EU-regions. In contrast no geographical differentiation in EU regions was demanded in relation to effects on soil, ecological functions and the abiotic environment.

The overall level of selected competence was strong given the fact that most experts did not make use of the 'don't know, no expert' response. Only for some cases in the categories cultivation and management (F) and abiotic environment (G), a relatively high number of experts felt lacking in competence and thus the conclusions drawn from the survey in these categories should be interpreted carefully.

The structure and content of the Online Survey Environment (OSE) was based on the literature review begun and the preliminary prioritisation begun by the BEETLE team and PRC (Steps 1 and 3). In the OSE (step 3) the participating experts confirmed (i) that the potential processes causing potential long-term effects were complete and (ii) that the same was true for the preliminary prioritisation by the BEETLE team in most cases.

The most important areas of uncertainty were

- impact of cultivation and management,
- impact on soil organisms,
- regional aspects in the assessment, and
- the evaluation of stacked events.

Therefore, these issues were selected for further discussion in the fifth step of expert consultation, the Creative Space Workshop (see Fig. 5 of the BEELTE main report).

Appendix 1: Documentation Online Survey ‘Environment’

I. Introduction

The questionnaire is differentiated into 7 category assessments for potential long-term effects.

- **Category A:** Persistence and invasiveness
- **Category B:** Altered gene transfer
- **Category C:** Interactions between GM plant and target organisms
- **Category D:** Interactions of the GM plant with non-target organisms
- **Category E:** Effects on ecological function
- **Category F:** Impacts of the specific cultivation, management and harvesting techniques
- **Category G:** Potential interactions with the abiotic environment
- **The final question**

In the OSE each category assessment was divided into two steps:

- **Step 1:** Collection of important biological *processes* caused by the intended phenotype of the genetically modified (GM) plant that may lead to long-term effects in general. Our first two questions here are:
 - “Do you agree with the processes listed for potential long-term-effects?” and
 - “Are there - according to your expertise - important processes of potential long-term effects to add?”
- **Step 2:** Prioritization of the probabilities of biological processes causing long-term effects based on selected crop/trait combinations. The user of the questionnaire is asked to assess a selected process (presented in *thesis* format) according to the expected probability: *negligible*, *low*, *high* and *don't know*. We selected according to available literature crop/trait combinations with highest likelihood for detailed expert assessment.

Remark: The expert assessment should primarily focus on long-term effects caused by the intended phenotype of the GM plant.

II. Assessment of processes and cases (crop/trait combinations)

A. Category A: Persistence and invasiveness

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

A.1	The new GM trait causes increased fitness of the GM cultivar. The GM cultivar persists inside and outside fields, becomes invasive over time and/or changing environmental conditions, and finally affects other plant species. The process is favored by (i) increased stress tolerance (e.g. towards temperature, water, salinity), (ii) increased number of progeny, (iii) decreased pathogen susceptibility, (iv) increased pest tolerance/resistance, (v) increased tolerance against herbicides.			
A.2	Outbreeding depression of the GM trait in wild relatives causes reduced GM hybrid fitness. With continuous gene swamping the recipient wild population has less and less genetic barrier, more GM hybrids are released, the wild species becomes less fit in natural or semi-natural habitats and the size of populations decreases. The process is favored by (i) decreased stress tolerance (e.g. towards temperature, water, salinity), (ii) decreased number of progeny, (iii) increased pathogen susceptibility, (iv) decreased pest tolerance/resistance.			
A.3	GM hybrids persist in and outside fields. The new GM trait causes an increased fitness after gene flow and introgression into wild relatives (hybrids). The outside fields persisting GM hybrids become invasive over time or changing environmental conditions, and finally affects other plant species. The process is favored by (i) increased stress tolerance (e.g. towards temperature, water, salinity), (ii) increased number of progeny, (iii) decreased pathogen susceptibility, (iv) increased pest tolerance/resistance			
A.4	A combination of two or more GM traits causes increased fitness. The GM hybrids persist inside and outside fields, invasive over time and/or changing environmental conditions, and finally affects other plant species. The process is favored by (i) increased stress tolerance (e.g. temperature, water, salinity), (ii) increased number of progeny, (iii) decreased pathogen susceptibility, (iv) increased pest tolerance/resistance, (v) multiple herbicide tolerance, (vi) molecular genetic interactions between GM traits			
	Is the list for this category complete?	⁴ Σ	59	100%
		Yes	41	69%
		No	18	31%
	If not, other important processes to assess? Please explain your answer. ⁵			

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⁴ The table presents response options (column 1, row 2ff), the total number of participants (column 2, row 1), the distribution to the different response option (column 2, row 2ff) and the percentage of the different response options (column 3).

⁵ Answers to the open field questions are listed under III Open field answers.

Step 2: Prioritization of long-term effects

A1 Increased fitness of the GM cultivar				
Trait: Insect pest resistance				
<p>Scenario: Bt maize cultivars are protected against insect pests either to <i>Ostrinia nubilalis</i>, <i>Sesamia nonagrioides</i> or <i>Helicoverpa armigera</i> (shortened as ECB) or to <i>Diabrotica virgifera virgifera</i> (shortened as WCR). These cultivars will primarily be cultivated in pest infestation areas. The pest protection trait results in an increased persistence of GM Bt-maize inside and outside of fields.</p> <p>Remarks: In the EU, GM insect resistance is only developed in Bt maize (lepidopteran or/and coleopteran resistance) until now.</p>				
Case (theses): Adverse long term effects will arise due to...				
A.1.1	increasing numbers of volunteers occur in GM Bt maize cropping system	Σ	64	100%
		Negligible	43	67%
		Low	11	17%
		High	1	2%
		insuff. data	4	6%
		no expert	5	8%
A.1.2	the occurrence of feral Bt maize plants at sites outside of fields (e.g. by seed spillage) establishing viable populations and consecutively increased invasiveness of Bt maize; possibly affecting nearby non-cultivated habitats	Σ	64	100%
		Negligible	49	77%
		Low	8	13%
		High	0	0%
		insuff. data	2	3%
		no expert	5	8%
A.1.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	64	100%
		Yes	26	42%
		No	24	39%
		insuff. data	5	8%
		no expert	7	11%

Trait: Herbicide tolerance				
<p>Scenario: Herbicide tolerant crops express proteins conferring tolerance against non-selective herbicides like glyphosate or glufosinate. HT crops will have selective advantage at sites where the non-selective herbicides are applied.</p> <p>Remarks: Herbicide tolerant GM crops to be considered are HT maize, HT oilseed rape, HT soybean, HT sugar beet.</p>				
Case (theses): Adverse long term effects will arise due to..				
A.1.4.	increasing number of volunteers occurring in GM HT cropping systems	Σ	62	100%
		Negligible	21	35%
		Low	17	27%
		High	13	22%
		insuff. data	2	3%
		no expert	8	13%
	If the case would apply, for which HT crop would you expect <i>high</i> , <i>medium</i> or <i>low</i> tendency for altering persistence?			
	HT-maize	Σ	53	100%
		Low	43	81%
		Medium	9	17%
		High	1	2%
	HT-oilseed rape	Σ	51	100%
		Low	8	16%
		Medium	20	39%
		High	23	45%
	HT-soybean	Σ	47	100%
		Low	43	91%
		Medium	4	9%
		High	0	0%
	HT-sugar beet	Σ	49	100%
		Low	21	43%
		Medium	20	41%
		High	8	16%

A.1.5	the occurrence of feral HT plants at sites outside of fields (e.g. by seed spillage) establishing viable populations if the herbicide is applied (at sites like railway tracks or roadsides)	Σ	62	100%
		Negligible	17	27%
		Low	22	35%
		High	9	15%
		insuff. data	5	8%
		no expert	9	15%
	If the case would apply, for which HT crop would you expect <i>high</i> , <i>medium</i> or <i>low</i> tendency for feralization?			
	HT-maize	Σ	52	100%
		Low	48	92%
		Medium	3	6%
		High	1	2%
	HT-oilseed rape	Σ	51	100%
		Low	9	18%
		Medium	23	45%
		High	19	37%
	HT-soybean	Σ	46	100%
		Low	43	93%
		Medium	3	7%
		High	0	0%
	HT- sugar beet	Σ	49	100%
		Low	24	49%
		Medium	20	41%
		High	5	10%
A.1.7	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	60	100%
		Yes	34	57%
		No	14	23%
		insuff. data	2	3%
		no expert	10	17%

Trait: Starch modification				
Scenario: GM crops with altered starch composition show an increased tolerance towards certain environmental stressors like frost, heat, salinity etc., providing a fitness advantage.				
Remarks: The GM crop to be considered here is potato expressing amylose- or amylopectin-enriched starch				
Case (theses): Adverse long term effects will arise due to...				
A.1.8.	increasing number of potato volunteers occurring in GM cropping systems	Σ	61	100%
		Negligible	27	44%
		Low	11	18%
		High	4	7%
		insuff. data	7	11%
		no expert	12	20%
A.1.9	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	60	100%
		Yes	27	45%
		No	18	30%
		insuff. data	5	8%
		no expert	10	17%

A2 Outbreeding depression after hybridization of the GM crop with wild relatives				
Trait: Herbicide tolerance				
<p>Scenario: Hybridization will primarily occur for GM oilseed rape or GM sugar beet, because cross compatible wild relatives are present for these crops in Europe. Hybrids may potentially be less viable in comparison to the original wild type.</p> <p>Remarks: IR traits are not taken into consideration because there is presently no Bt crop cultivated having wild relatives in the EU (only Bt-maize).</p>				
Case (theses): Long-term adverse effects of HT crops...				
A.2.1	due to outbreeding depression long-term gene introgression will decrease population sizes of wild relatives of GM HT crops (up to extinction)	Σ	60	100%
		Negligible	28	47%
		Low	11	18%
		High	2	3%
		insuff. data	8	13%
		no expert	11	18%
	If the case would apply, for which crop/wild relative complex would you expect <i>high</i> , <i>medium</i> or <i>low</i> tendency for population decrease?			
	HT oilseed rape / wild relative complex	Σ	45	100%
		Low	27	60%
		Medium	14	31%
		High	4	9%
	HT sugar beet / wild relative complex	Σ	45	100%
		Low	31	69%
		Medium	10	22%
		High	4	9%
A.2.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	60	100%
		Yes	31	52%
		No	17	28%
		insuff. data	3	5%
		no expert	9	15%

A3 GM hybrids persist in and outside fields, and finally become invasive				
Trait: Herbicide tolerance				
Scenario: GM hybrids with herbicide tolerance traits will have selective advantage at sites where the non-selective herbicides are applied				
Remarks: Hybridization with wild relatives is not relevant for (Bt) maize or soybean in Europe. Oilseed rape wild relatives are widespread and may grow in and outside of fields; Sugar beet wild relatives are found mainly in costal regions and as weedy crop/wild complexes in beet rotation.				
Case (theses): Adverse long-term effects will arise due to...				
A.3.1	out-competing natural genotypes of related (cross compatible) weedy relatives by HT-hybrids in GM HT cropping systems.	Σ	57	100%
		Negligible	19	33%
		Low	9	16%
		High	16	28%
		insuff. data	5	9%
		no expert	8	14%
A.3.2	replacing wild relatives of GM HT crops by gene swamping or by competition specifically in habitats where herbicides are occasionally used (like railway tracks or roadsides)	Σ	58	100%
		Negligible	14	24%
		Low	17	29%
		High	11	19%
		insuff. data	6	10%
		no expert	10	17%
A.3.3.	replacing other plant species in plant communities where herbicides are occasionally used (like railway tracks or roadsides)	Σ	58	100%
		Negligible	14	24%
		Low	18	31%
		High	7	12%
		insuff. data	8	14%
		no expert	11	19%
A.3.4	According to cases A.3.1 to A.3.3: If these cases would apply, for which crop/wild relative complex would you expect <i>high</i> , <i>medium</i> or <i>low</i> tendency for altering fitness and competitive ability?			
	HT oilseed rape / wild relative complex	Σ	47	100%
		Low	17	36%
		Medium	16	34%
		High	14	30%
	HT sugar beet / wild relative complex	Σ	47	100%
		Low	26	55%
		Medium	16	34%
		High	5	11%
A.3.5	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	57	100%
		Yes	32	56%
		No	12	21%
		insuff. data	3	5%
		no expert	10	18%

A4 Stacked events				
Case (theses):				
A.4.1	Concerning the processes mentioned in A1 to A 3: Would you change your assessments if you look at GMO with stacked traits (for maize: combination of IR/HR; for oilseed rape and sugar beet: combination of different HT traits); in comparison to GMO with single traits?	Σ	60	100%
		Yes	13	22%
		No	41	68%
		insuff. data	2	3%
		no expert	4	7%
A.4.2	Would you differentiate your assessment between intended (stacks as result of breeding) and unintended stacks (stacks as a result of unintended or unavoidable gene flow during cultivation)?	Σ	59	100%
		Yes	19	32%
		No	33	56%
		insuff. data	3	5%
		no expert	4	7%

B. Category B: Altered gene transfer

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

B.1	GM trait reduces pollination, e.g. due to a decreased attractiveness for pollinators (altered color, altered scent)			
B.2	GM trait causes altered flower phenology, which leads after gene introgression and over time to genetic isolation of wild relatives			
B.3	GM trait causes an altered compatibility between GM crops and conventional varieties or between GM crops and their wild relatives, e.g. reducing or favoring outcrossing			
B.4	GM trait alters fecundity resulting from altered number of seeds produced, which may cause increased seed (gene) flow from GM crops to wild plant populations			
B.5	GM trait increases <i>frequency</i> of horizontal gene transfer from plant to microbial populations introducing new traits into microbial communities			
	Is the list for this category complete?	Σ	52	100%
		Yes	37	71%
		No	15	29%
	If not, other important processes to assess? Please explain your answer.			

Step 2: Prioritization of long-term effects

B.1 General questions				
Trait: General questions				
Remarks: All processes mentioned in B.1-B.5 are not relevant for the selected crop/trait combinations. Consequently, just general questions will be asked on effects on altered gene transfer.				
Case (theses): Do you think...				
B.1.1	...that phenotypic effects such as altered flower or fecundation biology is sufficiently assessed during the GM approval procedure and variety registration currently applied in the EU?	Σ	55	100%
		Yes	34	62%
		No	7	13%
		insuff. data	5	9%
		no expert	9	16%
B.1.2	...that frequency of gene transfer from GM plants to microorganisms should be assessed during the approval process	Σ	55	100%
		Yes	16	29%
		No	28	51%
		insuff. data	5	9%
		no expert	6	11%
B.1.3	...that such effects have to be assessed in interspecific hybrids and wild relatives?	Σ	54	100%
		Yes	20	37%
		No	22	41%
		insuff. data	6	11%
		no expert	6	11%

C. Category C: Interactions between GM plant and target organisms

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

C.1	GM traits lead over time to development of resistance in target organisms (pests or pathogens), which results in a loss of environmentally desired plant protection tools Remark: According to our categorization potential effects of the use of non-selective herbicides to weeds are addressed in category F			
C.2	Stacked events: GM crops with stacked traits need a different assessment of the above mentioned long term effects and processes			
	Is the list for this category complete?	Σ	48	100%
		Yes	34	71%
		No	14	29%
	If not, other important processes to assess? Please explain your answer.			

Step 2: Prioritization of long-term effects

C.1 GM traits lead over time to development of resistance in target organisms (pests or pathogens)				
Trait: Insect resistance				
Scenario: Continuous large-scale cultivation of Bt maize cultivars in pest-infested landscapes may result in development of resistance of the target insects (Lepidoptera or Coleoptera) to the Bt-proteins.				
Remarks: GM crop to be taken into account is GM Bt maize.				
Case (theses):				
C.1.1	Adverse effects affecting Bt maize crops will arise due to resistance development of target insects	Σ	52	100%
		Negligible	11	21%
		Low	27	52%
		High	13	25%
		insuff. data	0	0%
		no expert	1	2%
C.1.2	If this case would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	52	100%
		Yes	35	67%
		No	15	29%
		insuff. data	0	0%
		no expert	2	4%

C2 Stacked events				
Case (theses):				
C.2.1	Concerning the processes mentioned in C1 to C2: Would you change your assessments if you look at GMO with stacked traits (for maize: combination of IR/HR; for oilseed rape and sugar beet: combination of different HT traits); in comparison to GMO with single traits?	Σ	52	100%
		Yes	17	33%
		No	30	58%
		insuff. data	4	8%
		no expert	1	2%
C.2.2	Would you differentiate your assessment between intended (stacks as result of breeding) and unintended stacks (stacks as a result of unintended or unavoidable gene flow during cultivation)?	Σ	52	100%
		Yes	17	33%
		No	30	58%
		insuff. data	3	6%
		no expert	2	4%

D. Category D: Interactions of the GM plant with non-target organisms

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

D.1	GM traits cause adverse effects on plant- associated non-target organisms due to sublethal toxicity (chronic exposure) by consumption of pollen and plant tissue (e.g. on herbivores, pollinators, soil organisms, predators, parasitoids)			
D.2	GM traits alter nutritional composition of plants, leading to reduced fitness in plant-associated (non-pest) herbivores or decomposers			
D.3	GM traits cause altered nutritional value of host or prey organisms in trophic interactions, which affects trophic interactions in higher trophic levels. Adverse effects become apparent in non-target-organism communities (e.g. predators, parasitoids)			
D.4	GM traits cause accumulation of toxic compounds in various environmental compartments (e.g. accumulation in soil), which decreases abundance of (e.g. beneficial) NTO			
D.5	GM traits cause adverse effects on rhizosphere (plant-associated) bacteria and mycorrhizal fungi, e.g. due to altered root exudations impacting soil communities (populations size and community structure)			
D.6	GM trait specific root exudations could lead to fitness changes in NTO-involved symbiotic organisms e.g. involved in nitrogen fixing activities, mycorrhizal fungi			
D.7	Stacked events: GM crops with stacked traits need a different assessment of the above mentioned long term effects and processes.			
	Is the list for this category complete?	Σ	50	100%
		Yes	35	70%
		No	15	30%
	If not, other important processes to assess? Please explain your answer.			

Step 2: Prioritization of long-term effects

D.1 GM traits cause adverse effects on plant-associated non-target organisms				
Trait: Insect pest resistance				
<p>Scenario: Bt proteins act as toxins protecting crops against insect pests. However, Bt proteins are known to be highly specific to the target species groups. Due to expression of <i>lepidopteran</i> or <i>coleopteran</i> specific proteins in all parts of GM Bt plants (including pollen and roots) during the whole vegetation period not just when the larvae of target species are exposed, e.g. European Corn Borer and other lepidopteran target organisms (ECB) or Western Corn Rootworm (WCR) but also the larvae of non-target butterflies or beetles.</p> <p>Remarks: In the EU, lepidopteran or coleopteran specific Bt proteins are only used in maize until now.</p>				
Case (theses): Adverse long-term effects expected for...				
D.1.1	...ECB resistant maize on lepidopteran species	Σ	51	100%
		Negligible	21	41%
		Low	18	35%
		High	6	12%
		insuff. data	3	6%
		no expert	3	6%
D.1.2	...WCR resistant maize on coleopteran species	Σ	51	100%
		Negligible	20	39%
		Low	18	35%
		High	4	8%
		insuff. data	5	10%
		no expert	4	8%
D.1.3	...both traits (ECB resistance or WCR resistance) on bees	Σ	51	100%
		Negligible	38	75%
		Low	2	4%
		High	2	4%
		insuff. data	6	12%
		no expert	3	6%
D.1.4	...both traits (ECB resistance or WCR resistance, including higher trophic levels e.g. predators like Chrysopidae) on other insects	Σ	51	100%
		Negligible	28	55%
		Low	9	18%
		High	1	2%
		insuff. data	7	14%
		no expert	6	12%
D.1.5	...both traits (ECB resistance or WCR resistance) on microorganisms	Σ	51	100%
		Negligible	27	53%
		Low	7	14%
		High	1	2%
		insuff. data	10	20%
		no expert	6	12%
D.1.6	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	26	51%
		No	20	39%
		insuff. data	2	4%
		no expert	3	6%

D.2 GM traits alter nutritional composition of plants				
Trait: Insect resistance/Starch modification				
Scenario:				
<ol style="list-style-type: none"> Nutritional composition of GM crops is altered by newly expressed proteins (e.g. Bt). Altered starch composition caused by the genetic modification (amylose or amylopectine content) of potato tubers could alter fitness of plant associated (non-pest) herbivores or composers. Only NTOs feeding on tubers should be taken into consideration because altered starch composition occurs only in the tubers due to tuber specific promoter and knock down of granule bound starch synthase. 				
Case (theses):				
D.2.1	Long-term adverse effects are expected on herbivore or decomposer populations feeding on Bt protein expressing crops due to altered nutritional composition (Scenario 1).	Σ	51	100%
		Negligible	31	61%
		Low	9	18%
		High	0	0%
		insuff. data	6	12%
		no expert	5	10%
D.2.2	Long-term effects are expected on herbivore populations feeding on altered starch expressing potato tubers (Scenario 2)	Σ	51	100%
		Negligible	24	47%
		Low	8	16%
		High	1	2%
		insuff. data	11	22%
		no expert	7	14%
D.2.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	19	37%
		No	25	49%
		insuff. data	3	6%
		no expert	4	8%

D.3. GM traits cause altered nutritional value of host or prey organisms in trophic interactions				
Trait: Insect resistance				
Scenario: Bt-susceptible herbivores (2nd trophic level organism) feeding on Bt-toxin expressing host plants (1st trophic level organism) show a reduced nutritional value leading to a reduced fitness of predators or parasitoid coleopterans.				
Case (theses):				
D.3.1	Long-term adverse effects are expected on populations or diversity of predators or parasitoids feeding on larvae with altered nutritional value.	Σ	51	100%
		Negligible	22	43%
		Low	14	27%
		High	4	8%
		insuff. data	7	14%
		no expert	4	8%
D.3.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	19	37%
		No	23	45%
		insuff. data	3	6%
		no expert	6	12%

D.4 GM traits cause accumulation of toxic compounds in various environmental compartments				
Trait: Insect resistance				
Scenario: Bt-proteins will either accumulate in decomposed substrate from Bt protein expressing crops or will be bound and accumulate in soil particles from decomposed Bt plant residues				
Case (theses): Long-term adverse effects are expected...				
D.4.1	...on decomposing populations feeding on plant residues from Bt expressing crops or feeding as saprophytes on dead organic substrate in soils.	Σ	51	100%
		Negligible	28	55%
		Low	10	20%
		High	3	6%
		insuff. data	5	10%
		no expert	5	10%
D.4.2	on aquatic organisms due to accumulation of Bt proteins into water bodies being leached from soils or from Bt maize pollen or detritus being transported into waters	Σ	51	100%
		Negligible	28	55%
		Low	10	20%
		High	3	6%
		insuff. data	6	12%
		no expert	4	8%
D.4.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	18	35%
		No	27	53%
		insuff. data	3	6%
		no expert	3	6%

D.5 GM traits cause adverse effects on rhizosphere (plant-associated) bacteria and mycorrhizal				
Trait: Insect resistance				
Scenario: Exudates containing Bt protein may affect population size and activity of the rhizosphere organisms. There are no studies available showing the presence of transgene products in exudates of HT crops. Therefore only Bt-maize is taken into consideration				
Case (theses): Adverse long-term effects on abundance and diversity are expected on...				
D.5.1	...rhizosphere microorganisms (except Rhizobia) of Bt-crops expressing lepidopteran specific or coleopteran specific proteins	Σ	51	100%
		Negligible	29	27%
		Low	3	6%
		High	0	0%
		insuff. data	9	18%
		no expert	10	20%
D.5.2	saprophytic or pathogenic fungi of the rhizosphere expressing lepidopteran specific or coleopteran specific proteins	Σ	51	100%
		Negligible	30	59%
		Low	3	6%
		High	1	2%
		insuff. data	8	16%
		no expert	9	18%
D.5.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	11	22%
		No	26	51%
		insuff. data	4	8%
		no expert	10	20%

D.6 GM trait specific root exudations could lead to fitness changes				
Trait: Insect resistance				
Scenario: Bt protein exudations into the rhizosphere may affect abundance, diversity and activity of symbiotic organisms. Studies on altered root exudates are not available for HT plants. Only Bt-maize is taken into considerations since it is the only Bt crop cultivated in EU. For maize N-fixation processes as in nodules of rhizobia are not relevant, too. Consequently, only potential interactions between Bt-maize and mycorrhizal fungi are taken into consideration.				
Case (theses):				
D.6.1	Adverse long-term effects are expected on symbiotic activity of mycorrhizal fungi in Bt-maize plants if Bt-maize were continuously cultivated on one field over several years expressing lepidopteran specific or coleopteran specific proteins	Σ	50	100%
		Negligible	27	54%
		Low	5	10%
		High	0	0%
		insuff. data	9	18%
		no expert	9	18%
D.6.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	50	100%
		Yes	9	18%
		No	25	50%
		insuff. data	6	12%
		no expert	10	20%

D.7. Stacked events				
Case (theses):				
D.7.1	Concerning the processes mentioned in D1 to D 6: Would you change your assessments if you look at GMO with stacked traits (for maize: combination of IR/HR; for oilseed rape and sugar beet: combination of different HT traits); in comparison to GMO with single traits?	Σ	50	100%
		Yes	9	18%
		No	32	64%
		insuff. data	5	10%
		no expert	4	8%
D.7.2	Would you differentiate your assessment between intended (stacks as result of breeding) and unintended stacks (stacks as a result of unintended or unavoidable gene flow during cultivation)?	Σ	50	100%
		Yes	9	18%
		No	31	62%
		insuff. data	5	10%
		no expert	5	10%

E. Category E: Effects on ecological function

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

E.1	GM traits cause changes in soil fertility (e.g. nutrient cycling, organic matter decomposition, biological N-fixation) due to (i) additive, synergistic or delayed effects on non-target organisms including symbionts and (ii) altered nutritional composition of the plant and impact on decomposition			
E.2	GM traits cause changes in biological control due to (i) additive, synergistic or delayed changes in diversity and abundance of natural enemies and (ii) additive, synergistic or delayed changes in tritrophic interactions			
E.3	GM traits cause changes in pollination due to (i) additive, synergistic or delayed sublethal effects -> changes in diversity and abundance of the pollinator community and (ii) additive, synergistic or delayed altered attractiveness of flowers			
E.4	Stacked events: GM crops with stacked traits cause a different assessment of the above mentioned long term effects and processes.			
	Is the list for this category complete?	Σ	48	100%
		Yes	37	77%
		No	11	23%
	If not, other important processes to assess? Please explain your answer.			

Step 2: Prioritization of long-term effects

E.1 GM traits cause changes in soil fertility				
Trait: Insect resistance				
Scenario: Specific GM traits cause interference for ecosystem functions. Adverse effects result specifically via (i) the GM proteins accumulating in ecosystem sink compartments (e.g. soils) and (ii) retardations of decomposition of GM Bt maize necromass due to increased content of lignin in Bt maize residues				
Case (theses):				
E.1.1	Bt toxins produced by Bt maize will partially be accumulated in necromass of maize residues and will be incorporated into soil organic matter. Additionally, the Bt toxins will be adsorbed at soil minerals like clay. Bt residues in soil will have adverse long term effects on decomposition of soil organic matter where Bt maize is cultivated on the same fields during subsequent years	Σ	52	100%
		Negligible	28	54%
		Low	8	15%
		High	3	6%
		insuff. data	4	8%
		no expert	9	17%
E.1.2	In Bt maize a significant increase of lignin content in tissues was reported which could result in retardations of Bt residue decomposition. Disturbances in decomposition processes as a result of increased lignin content in Bt maize residues will occur in the long term	Σ	52	100%
		Negligible	26	50%
		Low	9	17%
		High	2	4%
		insuff. data	6	12%
		no expert	9	17%
E.1.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	18	35%
		No	21	41%
		insuff. data	3	6%
		no expert	9	18%

E.2 GM traits cause changes in biological control				
Trait: Insect resistance				
Scenario: Biological Control is the reduction of pest populations by natural enemies (predators, parasitoids etc.). Cultivation of BT crops may affect the diversity and abundance of natural enemies				
Case (theses): Adverse long-term effects are expected for Bt maize, specifically on...				
E.2.1	...natural enemies due to sublethal toxic effects and in consequence for a successful biological control of pests, too	Σ	52	100%
		Negligible	32	62%
		Low	7	13%
		High	1	2%
		insuff. data	8	15%
		no expert	4	8%
E.2.2	natural enemies due to a decreasing number of prey/hosts and in consequence for a successful biological control of pests, too	Σ	52	100%
		Negligible	28	54%
		Low	12	23%
		High	4	8%
		insuff. data	4	8%
		no expert	4	8%
E.2.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	51	100%
		Yes	24	47%
		No	21	41%
		insuff. data	3	6%
		no expert	3	6%

E.3 GM traits cause changes in pollination				
Trait: Insect resistance				
Scenario: Only Bt maize expresses GM IR proteins which are potentially toxic to NTOs; thus only Bt maize is considered as realistic crop/trait example.				
Case (theses): Bt toxins...				
E.3.1	...as produced by Bt maize are expressed in pollen. Long term adverse effects on pollinators such as honey bees will affect pollination of crops	Σ	52	100%
		Negligible	36	69%
		Low	6	12%
		High	1	2%
		insuff. data	5	10%
		no expert	4	8%
E.3.2	...will cause an overall diminished attractiveness of plants for pollinators	Σ	52	100%
		Negligible	38	73%
		Low	6	12%
		High	0	0%
		insuff. data	4	8%
		no expert	4	8%
E.3.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	50	100%
		Yes	18	36%
		No	27	54%
		insuff. data	1	2%
		no expert	4	8%

E.4 Stacked events				
Case (theses):				
E.4.1	Would you differentiate your assessment between intended (stacks as result of breeding) and unintended stacks (stacks as a result of unintended or unavoidable gene flow during cultivation)?	Σ	51	100%
		Yes	13	25%
		No	30	59%
		insuff. data	5	10%
		no expert	3	6%

F. Category F: Impacts of the specific cultivation, management and harvesting techniques

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

F.1	GM plant [management] causes increased/alterd use of agrochemicals (e.g. pesticides) controlling herbicide tolerant weeds, persistent GM crops (volunteers) with adverse effects on NTO and/or ecological functions.			
F.2	GM plant [management] causes indirect changes in susceptibility of crops against plant pathogens with adverse effects on NTO due to increased use of other pesticides			
F.3	GM plant [management] causes indirect changes and adverse effects on agro-biodiversity due to knock-on-effect of additive, synergistic or delayed effects cropping systems in agricultural landscape			
F.4	GM plant [management] causes indirect changes in fertilizer use with adverse effects on NTO and/or ecological functions			
F.5	GM plant [management] causes indirect changes in landscape structure due to coexistence measures (e.g. larger fields, larger distances between specific crops), resulting in loss of connectivity and reduced local biodiversity			
F.6	GM crops with stacked traits cause a different assessment of the above mentioned long term effects and processes			
	Is the list for this category complete?	Σ	48	100%
		Yes	38	79%
		No	10	21%
	If not, other important processes to assess? Please explain your answer.			

Step 2: Prioritization of long-term effects

F.1 GM plant [management] causes increased/alterd use of agrochemicals				
Trait: Herbicide tolerance				
<p>Scenario: Use of GM HT crops favours continuous applications of same complementary herbicides on fields cultivated with GM HT crops in successive years, either if the same HT crop is cultivated continuously or if several HT crops are cultivated in rotation. Changes are the result of: (i) in-season application of non-selective herbicides, (ii) relative flexibility with respect to timing of application, (iii) more efficient control of weeds.</p> <p>Remarks: GM crops to be taken into account are HT Oilseed rape, HT sugar beet, HT maize and HT soybean</p>				
Case (theses): In comparison to conventional herbicide application and management long term effects are expected due to...				
F.1.1	...increasing number of GM crop volunteers occurring in fields	Σ	50	100%
		Negligible	17	34%
		Low	13	26%
		High	11	22%
		insuff. data	2	4%
		no expert	7	14%
F.1.2	...increased number of weeds being tolerant to non-specific herbicides	Σ	50	100%
		Negligible	10	20%
		Low	14	28%
		High	17	34%
		insuff. data	1	2%
		no expert	8	16%
F.1.3	...a shift in weed communities causing changes in ecological functions (e.g. refuge for predators important for biocontrol)	Σ	50	100%
		Negligible	8	16%
		Low	17	34%
		High	13	26%
		insuff. data	5	10%
		no expert	7	14%
F.1.4	...increasing use of minimal tillage system favoring changes of weed communities which leads to a decrease in ecological functions (e.g. refuge for predators important for biocontrol)	Σ	49	100%
		Negligible	16	33%
		Low	11	22%
		High	7	14%
		insuff. data	7	14%
		no expert	8	16%
F.1.5	...decreasing non-crop plant biodiversity and thus feed resources for herbivores and associated taxa like birds	Σ	48	100%
		Negligible	21	44%
		Low	7	15%
		High	9	19%
		insuff. data	4	8%
		no expert	7	15%
F.1.6	...later application date of herbicides (in higher crop and weed stands) leads to increasing drift of non-selective herbicides into field margins or adjacent fields resulting in changes of plant communities	Σ	50	100%
		Negligible	19	38%
		Low	13	26%
		High	6	12%
		insuff. data	3	6%
		no expert	9	18%

F.1.7	...the use of additional chemical additives in herbicide formulations (e.g. due to late-season herbicide management), which cause additional environmental problems!	Σ	50	100%
		Negligible	22	44%
		Low	3	6%
		High	6	12%
		insuff. data	4	8%
		no expert	15	30%
F.1.8	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	48	100%
		Yes	23	48%
		No	19	40%
		insuff. data	2	4%
		no expert	4	8%

Trait: Trait: Insect resistance

Scenario: Cultivation of IR maize alters crop management if secondary pests evolve

Case (theses): In view of Bt maize cultivation long term effects are expected due to...

F.1.9	...the development of secondary pests leading to new insecticide application (e.g. with adverse effects on NTO)	Σ	50	100%
		Negligible	18	36%
		Low	17	34%
		High	6	12%
		insuff. data	7	14%
		no expert	2	4%
F.1.10	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	49	100%
		Yes	27	55%
		No	15	31%
		insuff. data	4	8%
		no expert	3	6%

F.2 GM plant [management] causes indirect changes in susceptibility of crops against plant pathogens

Trait: Herbicide tolerance

Case (theses): In view of non-selective herbicide application long term effects are expected due to...

F.2.1	...altered farming practices e.g. low-till farming, altered weed management etc. This effect will cause increased susceptibility of the crops to plant pathogens with adverse effects on NTOs due to increased use of other pesticides.	Σ	49	100%
		Negligible	24	49%
		Low	4	8%
		High	4	8%
		insuff. data	9	18%
		no expert	8	16%
F.2.2	...reduction of weed populations which will lower the attractiveness of insect pests for natural predators. This effect could enhance insect infestations with adverse effects on NTOs due to increased use of other pesticides.	Σ	49	100%
		Negligible	21	43%
		Low	9	18%
		High	5	10%
		insuff. data	8	16%
		no expert	6	12%
F.2.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	48	100%
		Yes	25	52%
		No	16	33%
		insuff. data	2	4%
		no expert	5	10%

F.3 GM plant [management] causes indirect changes and adverse effects on agro-biodiversity				
Trait: Herbicide tolerance / Insect resistance / Starch modification				
Scenario: GM crop cultivation will change cropping systems towards fewer crop rotations and an abundance of various crop species causing long term effects on agricultural landscape.				
Case (theses): In comparison to conventional management practices long term effects are expected due to...				
F.3.1	...decrease in biodiversity via loss of habitat niches	Σ	50	100%
		Negligible	20	40%
		Low	12	24%
		High	9	18%
		insuff. data	6	12%
		no expert	3	6%
F.3.2	... increased fertilizer use in GM cropping systems affecting NTO abundance or community structure	Σ	50	100%
		Negligible	21	42%
		Low	8	16%
		High	4	8%
		insuff. data	12	24%
		no expert	5	10%
F.3.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	49	100%
		Yes	22	45%
		No	17	35%
		insuff. data	7	14%
		no expert	3	6%

F.4 GM plant [management] causes indirect changes in fertilizer use				
Trait: Herbicide tolerance				
Scenario: GM crop cultivation will lead to an altered herbicide regime which indirectly affects fertilizer use due to long-term effects on the availability of inorganic nutrients				
Remarks: This case applies mainly for soybean				
Case (theses): Long term adverse effects are expected by...				
F.4.1	...toxic effects of non-selective herbicides on rhizosphere organisms affecting mineralization or N-fixation, leading to adverse effects on ecological functions and increased fertilizer use (with impact on eutrophication of soils and water bodies)	Σ	50	100%
		Negligible	21	42%
		Low	1	2%
		High	2	4%
		insuff. data	7	14%
		no expert	19	38%
F.4.2	...increased phosphorus application in GM cropping systems leading to adverse effects on mycorrhiza fungi and ecological functions	Σ	50	100%
		Negligible	17	34%
		Low	2	4%
		High	3	6%
		insuff. data	7	14%
		no expert	21	42%
F.4.3	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	45	100%
		Yes	9	20%
		No	15	33%
		insuff. data	6	13%
		no expert	15	33%

F.5 GM plant [management] causes indirect changes in landscape structure				
Trait: Herbicide tolerance				
Scenario: GM crop cultivation will lead to an altered cultivation practice due to the coexistence requirements (e.g. following minimum distance needs).				
Case (theses): Long term adverse effects are expected by...				
F.5.1	...enlargement of fields connected with increasing habitat fragmentation and increasing isolation of plant and animal populations.	Σ	50	100%
		Negligible	20	40%
		Low	9	18%
		High	5	10%
		insuff. data	11	22%
		no expert	5	10%
F.5.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	47	100%
		Yes	25	53%
		No	15	32%
		insuff. data	4	9%
		no expert	3	6%

F6. Stacked events				
Case (theses): .				
F.6.1.	Concerning the processes mentioned in F1 to F 5: Would you change your assessments if you look at GMO with stacked traits (for maize: combination of IR/HR; for oilseed rape and sugar beet: combination of different HT traits); in comparison to GMO with single traits?	Σ	50	100%
		Yes	18	36%
		No	28	56%
		insuff. data	2	4%
		no expert	2	4%
F.6.2	Would you differentiate your assessment between intended (stacks as result of breeding) and unintended stacks (stacks as a result of unintended or unavoidable gene flow during cultivation)?	Σ	49	100%
		Yes	12	24%
		No	30	61%
		insuff. data	5	10%
		no expert	2	4%

G. Potential interactions with the abiotic environment

Step 1: Collection of important biological processes caused by the intended phenotype of the GM plant that may lead to long-term effects

G.1	Use of GM plant causes adverse effect on the abiotic environment due to increased production of green house gases by GM plants [climate change]			
G.2	Use of GM plant causes adverse effect on the abiotic environment due to increased mineral nutrient erosion and fertilizer leaching into water bodies affecting water quality			
G.3	Use of HT or IR GM plant causes adverse effects on the abiotic environment by covering soil particle exchange sites e.g. due to more release of Bt protein or of herbicide molecules including their metabolites into soils			
G.4	GM crops with stacked traits cause a different assessment of the above mentioned long term effects and processes.			
	Is the list for this category complete?	Σ	45	100%
		Yes	37	82%
		No	8	18%
	If not, other important processes to assess? Please explain your answer.			

Step 2: Prioritization of long-term effects

G.1 Use of GM plants and climate change				
Case (theses):.				
G.1.1	Adverse effects on abiotic environment are expected for cultivation of Bt maize, HT oilseed rape, HT sugar beet, HT soybean and starch modified potato due to an intensification of agriculture solely related to the GM crops. Its cultivation will be connected with higher use of fossil energy resources, global deforestation and decline of organic soil fraction resulting in additional release of carbon dioxide	Σ	49	100%
		Negligible	32	65%
		Low	3	6%
		High	2	4%
		insuff. data	6	12%
		no expert	6	12%
G.1.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	47	100%
		Yes	17	36%
		No	21	45%
		insuff. data	1	2%
		no expert	8	17%

G.2 GM plants and mineral nutrient erosion				
Case (theses):				
G.2.1	Adverse effects on abiotic environment are expected for Bt maize, HT oilseed rape, HT sugar beet, HT soybean and starch modified potato, due to an intensification of agriculture with higher use of fertilizers and reduction in natural nitrogen fixation (toxic herbicide effects) resulting in an increase of mineral nutrient erosion and leaching of fertilizer into water bodies affecting water quality	Σ	49	100%
		Negligible	24	49%
		Low	4	8%
		High	4	8%
		insuff. data	3	6%
		no expert	14	29%
G.2.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	47	100%
		Yes	15	32%
		No	19	40%
		insuff. data	4	9%
		no expert	9	19%

G.3 HT or IR GM plants and soil exchange capacity				
Case (theses):				
G.3.1	The use of GM plants causes adverse effects on the abiotic environment by covering soil particle exchange sites e.g. due to enhanced release of ionic protein and enhanced release of molecules of the non-selective herbicides into soils. The reduced soil exchange capacity will negatively affect ecological functions of soil (pH-buffer, mineral nutrient availability).	Σ	49	100%
		Negligible	26	53%
		Low	2	4%
		High	0	0%
		insuff. data	6	12%
		no expert	15	31%
G.3.2	If one or more of these cases would apply, would you recommend the potential effects mentioned should be considered differently with respect to geographically different <i>EU-regions</i> ?	Σ	46	100%
		Yes	13	28%
		No	19	41%
		insuff. data	4	9%
		no expert	10	22%

G.4 Stacked events				
Case (theses):				
G.4.1.	Concerning the processes mentioned in G1 to G3: Would you change your assessments if you look at GMO with stacked traits (for maize: combination of IR/HR; for oilseed rape and sugar beet: combination of different HT traits); in comparison to GMO with single traits?	Σ	48	100%
		Yes	8	17%
		No	29	60%
		insuff. data	4	8%
		no expert	7	15%
G.4.2	Would you differentiate your assessment between intended (stacks as result of breeding) and unintended stacks (stacks as a result of unintended or unavoidable gene flow during cultivation)?	Σ	48	100%
		Yes	8	17%
		No	30	63%
		insuff. data	4	8%
		no expert	6	13%

III. Open field answers

A. Step One Answers for Category A

The above deal with the direct effects of the cultivar and its descendants but do not include nontarget effects.

Why are you only anticipating effects to other plants species? This is a quite narrow view. Effects could also be indirect effects to e.g. animals that due to increased abundance of the GM plant loose their open hunting ground, provides more shade etc. I know that you are dealing with some of my above mentioned aspects later in this questionnaire, but a problem arises in trying to fragment the effects into categories as you do here. By doing that you loose the systems-view, and therefore I think that your approach is highly dangerous, as it simplifies the complicated interactions in nature - your categories are often interlinked. It is necessary that we have broader, more holistic questions, when we discuss long time effects. Also an increased life span of the GM plants, e.g. in the form of perennially (that would really be a risky trait to insert, luckily it is hardly possible yet) or just prolonged growth season might favour the processes as well of the "increased number of offspring" you mention. The questions you present to us in step 2 are difficult to answer, as the answer will depend on a number of factors connected to particular scenarios. E.g. in some cases volunteers numbers might increase - in others decrease. Your questions are much too general to be answered precisely. Remember that the climate in the EU will change in the future, as global warming will entail average temperatures increases of 3-4 C, and changed precipitation patterns (in this century). This is a black horse in future prediction on interactions between GM plants and the environment, and will increase uncertainty of risk assessments. PS for your information: It has been shown now (e.g. our publication; Hauser et al., 2003; Johannessen et al., 2006 a and b) that in quite a number of scenarios crop-wild hybrids are more fit than the wild parent, and they can actually also be more fit than the crop parent. I think that it is general knowledge, that hybrid depression is not to be expected in the case of hybridization in many crop-wild complexes.

Phenotype of GM plant make them unable to participate to the interplay of positive biotic interactions with endophytic or soil microorganisms, depressing their resistance capacity against stress and pest against which these microbes enhance adaptation

Organisms harbouring GM-Traits lack fitness. They have an advantage only under selective conditions, i.e. herbicide treatment! Otherwise they consume more energy for the synthesis of constitutively expressed proteins. Natural populations which are in Hardy-Weinberg equilibrium will very fast minimize the amount of GM-plants if selective pressure does not occur due to natural selection processes. This is simple population genetics.

Recombination of traits/events that originate from different GMHPs by natural processes (secondary stacking) - after gene flow, introgression etc.

The list is extremely valuable for the risk characterisation step in environmental risk assessment, but in practice, the list might be too theoretical (or mainly based on worst-case scenarios).

It is not the question, whether the list is complete, and you do not leave the possibilities to the experts to answer as an introduction more fundamental questions on whether A1 to A4 are appropriate or not. This means that my NO does not just signal an incomplete list of questions, but a fundamental critique of A1 to A1, the critique is so fundamental, that I seriously ask myself whether I should participate in the survey or not - maybe better not, otherwise I make myself the complice of a deeply flawed view of risk assessment of GM crops, where for instance the BENEFITS are not at all considered, in a complete breach with Article 19 of the CBD, the fundament of the Cartagena Protocol. And more: A1 to A4 concentrate on GM crops, which is not scientifically correct. If you do scientific risk assessment, then it should be always with a baseline and also with considering the risks of NOT adopting the novel approach, in this case GM crops. If you do not follow these principles, then the whole approach is deeply biased. Concretely: A1 how many classic breeds cause increased fitness? ANY agricultural specialist will tell you, that the only REAL problem we have is the one with Brassica, being a crop very close to its ancestral wild relatives. But this does INCLUDE the non-transgenic Brassicas, this means: the way you construct question A1 is highly biased and focuses in an unscientific way on GM crops. A2: Outbreeding depression: dramatic cases in classic breeding: loss of fragmentation of spindle, seed dispersal seriously hampered, in Triticae etc. etc. A3: ALL those concerns are much more realistic with classic breeds, a normal situation in agriculture however. A4: Classic breeds can be seen as super-stacked events, with hundreds of new genes crossed in and this in a basically uncontrolled way. Here the scientific basis for a new (old) view of GM crops: it is simply scientifically unfounded to claim absolute novelty for the GM crops (of 1st generation). There are however differences to the classic breeds: Whereas the classic breeder (and natural mutation processes) work more or less at random, the genetic engineer works in a targeted way, and can analyse it at the latest post festum what really happened - and it also has to be clearly stated that in contrast to natural mutation the GM crops are, after an average of 10 years of testing, released to the billions in the field within a short time. Arber, W. (2000) Genetic variation: molecular mechanisms and impact on microbial evolution. Fems Microbiology Reviews, 24, 1, pp 1-7 ://000084915900001 AND <http://www.botanischergarten.ch/Mutations/Arber-Gen-Variation-FEMS-2000.pdf> Arber, W. (2002) Roots, strategies and prospects of functional genomics. Current Science, 83, 7, pp 826-828 ://000178662800019 and <http://www.botanischergarten.ch/Mutations/Arber-Comparison-2002.pdf> Arber, W. (2003) Elements for a theory of molecular evolution. Gene, 317, 1-2, pp 3-11 ://000186667000002 and <http://www.botanischergarten.ch/Mutations/Arber-Gene-317-2003.pdf> Arber, W. (2004) Biological evolution: Lessons to be learned from microbial population biology and genetics. Research in Microbiology, 155, 5, pp 297-300 ://000222736200001 AND <http://www.botanischergarten.ch/Mutations/Arber-Evolution-Lessons-2004.pdf> see the discussion in: Ammann, K. (2007) Reconciling Traditional Knowledge with Modern Agriculture: A Guide for ...

... Building Bridges. In Intellectual Property Management in Health and Agricultural Innovation a handbook of best practices, Chapter 16.7 (eds A. Krattiger, R.T.L. Mahoney, L. Nelsen, G.A. Thompson, A.B. Bennett, K. Satyanarayana, G.D. Graff, C. Fernandez & S.P. Kowalsky), pp. 1539-1559. MIHR, PIPRA, Oxford, U.K. and Davis, USA The general link to the www.ipHandbook.org. (as of September 2007) AND <http://www.botanischergarten.ch/IP/Press-Release-ipHandbook-Online-20071101.pdf>, AND the Flyer: <http://www.botanischergarten.ch/Patents/ipHandbook-Flyer1.pdf> AND chapter 16.7 <http://www.botanischergarten.ch/TraditionalKnowledge/Ammann-Traditional-Biotech-2007.pdf> free of copyrights AND the exported bibliography with the links: <http://www.botanischergarten.ch/TraditionalKnowledge/Exported-Bibliography-links-Ammann-2007.pdf> The biased way of questioning devaluates this survey considerably, and the laudable goal stated by the project leaders to produce a fair comparison between GM and non-GM crops is thus seriously hampered. and more: I fear that in each case we would like to set as experts an IMPOSSIBLE, then this will go with "negligible" into LACUNE OF KNOWLEDGE in the end, and this is NOT acceptable, and the possible answers leave no IMPOSSIBLE in cases where no scientific evidence is present and also the scenario is highly improbable to impossible. You are seriously restricting the freedom of answers of the experts this way. according to a correspondence I have seen with Dr. Piet van der Meer, you have carefully chosen a balanced range of experts, and if I understood it correctly, then you invite equal numbers of so called NGO experts, which are either deeply ignorant and deficient of access to peer reviewed literature or researchers who are companions of NGO units because they have a vested interest in continuing to invest millions in worthless risk assessment research. (risk assessment research has enough work to do with truly novel traits derived from pharming goals and from synthetic biology, its a shame that we still waste money on risk assessment of the GM crops of the first generation, where ALL the experience is truly positive, just see the following examples in the literature: Wilson, T.A., Rice, M.E., Tollefson, J.J., & Pilcher, C.D. (2005) Transgenic corn for control of the European corn borer and corn rootworms: a survey of Midwestern farmers' practices and perceptions. *Journal of Economic Entomology*, 98, 2, pp 237-247 ://000228259000001 AND <http://www.botanischergarten.ch/Bt/Wilson-Survey-Midwestern-2006.pdf> Burkness, E.C., Hutchison, W.D., Weinzierl, R.A., Wedberg, J.L., Wold, S.J., & Shaw, J.T. (2002) Efficacy and risk efficiency of sweet corn hybrids expressing a *Bacillus thuringiensis* toxin for Lepidopteran pest management in the Midwestern US. *Crop Protection*, 21, 2, pp 157-169 ://000174435900009 AND <http://www.botanischergarten.ch/Burkness-Consistent-Benefits-2002.pdf> Ammann, K. (2005) Effects of biotechnology on biodiversity: herbicide-tolerant and insect-resistant GM crops. *Trends in Biotechnology*, 23, 8, pp 388-394 ://000231342700005 and <http://www.sciencedirect.com/science/article/B6TCW-4GG2HJM-2/2/b23d0cc8c6846b9f6625162f3351b0ae> and <http://www.botanischergarten.ch/TIBTECH/Ammann-TIBTECH-Biodiversity-2005.pdf> Conner, A.J., Glare, T.R., & Nap, J.-P. (2003) The release of genetically modified crops into the environment. Part II. Overview of ecological risk assessment. *The Plant Journal*, 33, 1 %R doi:10.1046/j.0960-7412.2002.001607.x, pp 19-46 <http://www.blackwell-synergy.com/doi/abs/10.1046/j.0960-7412.2002.001607.x> AND <http://www.botanischergarten.ch/Bt/Conner-Release-Review-2-2003.pdf> Gray, A.J. (2004) Ecology ...

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You have identified hazards and described factors that MAY affect persistence (but often do not). I wasn't sure whether you were asking whether adverse long term effects are likely to occur if, for instance there is an increase in GMHT maize volunteers/ ferals, or whether increased volunteers and ferals are likely to occur (in the long-term) as a result of cultivating GMHT maize i.e. whether I should assume that increased volunteer numbers translate to an adverse environmental effect. I chose the latter interpretation.

PvT: I am not sure that the following is adequately covered in the above: How does the GM trait affect the intrinsic barrier that may exist between crop and wild relative in producing hybrid offspring? Is this part of 'in/decreased number of progeny' (e.g phenology overlap, endosperm balance). Likewise, how does the trait affect the dispersal characteristics as feral crop or as hybrid/introgressed material? DH: The GM gene and its location in the genome can not be seen as loose effect on top of those of hybridization as such. New linkages will be formed in which different genes, potentially including the GM, could strengthen, reinforce or cancel each other. In effect the phenotype caused by the GM in the crop is not always 1:1 to be translated to a crop-wild relative hybrid phenotype. So there might be not only genetic interactions between stacked GM-traits (as in A.4) but also between non-GM and GM traits.

A combination of two or more GM traits decrease fitness. The wild species becomes less fit in natural or semi-natural habitats and the size of populations decreases.

Please note that a thorough environmental risk assessment (e.r.a.) for the placing on the EU market of each genetically modified organism is carried out in accordance with Commission Decision 2002/623/EC establishing the guidance notes supplementing Annex II of Directive 2001/18/EC. In particular, detailed information regarding the genetically modified organism is taken into account, including: a) the inserted genetic material (i.e., size, structure, copy number, location); b) developmental stages and parts of the plant where the insert is expressed and characteristics of the novel proteins expressed; c) how the GM plants differ from the non-GM plants; d) genetic and phenotypic stability of the GM plant; e) whether there is any change to the ability of the GM plant to transfer genetic material to other organisms; f) potential toxic, allergenic or other harmful effects of the genetic modification on human health; g) safety of the GM plant to animal health; h) mechanism of interaction between the GM plant and target organisms; i) potential changes in interactions of the GM plant with non-target organisms; j) potential interactions with the abiotic environment; k) detection and identification methods for the GM plant; and, l) information about previous releases of the GM plant. As a result, the e.r.a. takes into consideration all of the mechanisms for potential adverse effects that have been prescribed under Point 4.2.1 of the Annex to Commission Decision 2002/623/EC, namely: a) The spread of the GMO in the environment b) The transfer of the inserted genetic material to other organisms, or the same organism whether GM or not c) Phenotypic and genetic instability d) Interactions with other organisms (other than exchange of genetic material/pollen) We are not aware of other mechanisms (or processes) that may lead to long-term effects.

Outbreeding of traits rendering the seeds of the recipient wild population sterile. The size of the recipient wild population decreases with continuous use of traits rendering the next generation of plants sterile. Stacked events: a combination of two or more GM traits reduces the fitness of GM hybrids (see above)

The cited 'theses' and 'important biological processes' are already captured in the environmental risk assessment prepared by the applicants according to the EFSA guidance document and are intensely reviewed by EFSA and member states expert bodies prior to issuing a positive scientific opinion. To date all data and studies confirm that the GM crops do not lead to any other effects (neither short nor long-term) than conventionally bred and cultivated crops in the agricultural environment. The traits that are developed for GM crop plants and are cited in the survey as 'favouring adverse long term effects' are identical to the breeding goals pursued by plant breeders world wide for centuries (e.g. disease and stress tolerant plants, higher yielding and more nutritious plants, plants reducing the environmental footprint of humans) with a variety of tools, therefore GM crops do not have the potential to exert any other effects on the environment as current crop varieties and agricultural practices do

The 4 main themes are OK I think. However, I do not fully agree with the lists of favoured processes. In my opinion the likelihood whether one of the listed processes will lead to long term effects can not be considered similar. In general if GM crops are altered in only one trait caused by a monogenic modification that also affects only a single gene the likelihood of LTE is (very) low. In cases of fitness, characteristics of spreading and survival or one specific type of resistance, many other characteristics of the plant may be more important to determine overall fitness, invasiveness, etc. A monogenic (or better a mono-phenotypic) change will have very little effect. On the other hand, a general rule of thumb in the ERA is that changes in the composition of a macrocomponent of plants may result in physiological changes of the plant. E.g. increased frost-tolerance in amylose-free potatoes (which is by the way proven not to occur) or changes in levels of oil in seed or polyol accumulation in cells may result in altered characteristics in survival or dissemination of the (GM) plant. In many cases this involves polygenic modifications or transcription factors affecting more than one biosynthetic pathway. These modifications might result in to increased stress tolerance. In short the assessment of LTE largely depends on the specificity of the introduced trait. PS Regarding the answers below, I assume that effects are assessed against the local baseline (how do the unmodified crops behave agronomically and ecologically in the different geographical regions). In that sense the ERA is already differentiated in geographical regions. (so many answers would be yes). However I think that the mentioned effects themselves should not be assessed differentially as they affect the baseline in a similar way independently of the geographical region. So the effects need to be considered always, only the magnitude of the (un)foreseen effects may differ over the different geo-graphical regions. Also in most cases it is not the modification which directs the answer whether or not effects should be assessed differently for the geographical regions but the recipient organism. 'Exotic' crops like cotton, maize, potato or soy behave ecologically more or less similar over the EU-territory since there are no wild relatives. On the other hand for crops like OSR ecological effects are far more likely due the presence of sexual compatible wild relatives. These arguments complicate the answering of the questions in a straightforward fashion.

don't forget soil quality mediated by microbial activity and indirect effects by changing plant species and rotations needs to be elaborated in discussions and workshops
It is unclear to me whether the data collected would always allow the proper computation of fitness as the growth rate of the population over a single generation from one life-stage to a corresponding one in the next generation. I do not give definite examples but I cannot believe that the four listed scenarios cover all those likely to occur. The question seems to me to go against the case-by-case approach and I have no confidence that the list is complete
It is not clear from these categories for GMHPs used in agriculture what the basis for comparison is. While the processes of assessing fitness changes, the importance of the nature of the crop is not clear. For example, traditional breeding has long had increased yield as a desired endpoint and this may have translated into potential increased progeny.
I don't agree with all of the above scenarios. Even though I am an expert in this field, I don't understand why these effects are all listed together here. I don't think I can help with this survey because the scenarios listed below this section also seem very odd. How can maize be feral or weedy? I have never heard of this, other than with volunteers that do not form permanent populations. Why would it be bad for a weed to receive a transgene that causes outbreeding depression (mentioned above)? Most weeds are not in danger of extinction and it would be fine if they became less abundant in the margins around crop plants. Also, I don't think genetic swamping would be more likely with a GM crop than a non-GM crop because many wild individuals (with lots of genetic diversity) would receive the transgene and a few genes that are linked to it. Looking further down your list of questions, I don't have enough knowledge of specific weed problems with weed beets and B. rapa in the EU. I don't think there is much chance of crop-wild hybridization with other weed species that are related to sugar beet or oilseed rape. Further down, you imply that the spread of HT wild relatives in ruderal areas will be a problem - I don't understand why, unless these species are very serious weeds already and they are already treated with glyphosate. Sorry I can't be more helpful.
A1. I don't think that GM is invasive in all cases. It depends on the trait e.g. in the case of herbicide the fitness will depend on the spray of the specific herbicide A2; The fitness of hybrid combination depends on the hybrids concerned and over relative is close to the GM
Invasiveness is also a question of world trade and tourism. Maybe seeds of GMOs or their offspring are more able to enter new habitats than conventional plants?
Each GM trait should be considered individually, case -by-case thus generic statements can never cover all possibilities or be complete
GM cultivars or hybrids with wild relatives do not necessarily have to be more fit, the opposite can be true, depending on the type of parameter we speak about.

B. Step One Answers for Category B

Seems OK; a clever scientist can always come up with more ideas...
GM traits may also increase pollination GM traits may change flowering time and therefore affect outcrossing to wild relatives GM trait may change seed survival and therefore affect spatial distribution and outcrossing with relatives Changes in the crop types grown may change gene transfer. Example: GM oilseed rape would be popular in Denmark, where few and inefficient herbicides can be applied in this crop. A HT genotype may increase the area with oilseed rape, and thus the gene transfer to wild B. rapa. Again trying to group the effects like you do here, will simplify the system, so that some effects may not be foreseen.
Questions again biased, because there is no baseline included, all those phenomena occur also in classic breeding and conventional agriculture
DH: GM trait alters pollen morphology (size, shape, weight) increasing or decreasing its crossing * distance relationship relative to the wild relative. In addition to B.4. in cases increased/decreased seed production is caused by a changed flower production: GM trait alters the pollen quantities released relative to the wild relative
Unintended effects in both the crops and mating species, due to the presence of a transgene are hard to foresee. Moreover the location of the transgene in the genome plays also a major role. Therefore this list cannot, by definition, be complete
GM trait does not impair pollen production and pollination capacity but renders developing seeds sterile
I would like to comment that the above effects are not so much effects affecting the environment but are almost all factors affecting the rate of dissemination of the GM trait. The answers of the questions below are therefore very dependent on the trait itself. Therefore I find it difficult to give specific answers whether or not a specific aspect should be considered in detail or whether there is reason to suspect environmental harm. In many cases gene flow will not result in adverse effects. According to this point of view the questions below can only be regarded when, based on the trait, mitigation measures are considered to be necessary.
The list is too long to write. Theoretically, anything could be altered by a specific transgene or combinations of transgenes and molecular baggage that controls their expression.
GM trait increases weediness and surviving ability of the modified crops in nature.
There is no demonstration for such traits

As mentioned above, a thorough e.r.a. for the placing on the EU market of each genetically modified organism is carried out in accordance with Commission Decision 2002/623/EC establishing the guidance notes supplementing Annex II of Directive 2001/18/EC. In particular, detailed information regarding the GMO is taken into account, including: a) the inserted genetic material (i.e., size, structure, copy number, location); b) developmental stages and parts of the plant where the insert is expressed and characteristics of the novel proteins expressed; c) how the GM plants differ from the non-GM plants; d) genetic and phenotypic stability of the GM plant; e) whether there is any change to the ability of the GM plant to transfer genetic material to other organisms; f) potential toxic, allergenic or other harmful effects of the genetic modification on human health; g) safety of the GM plant to animal health; h) mechanism of interaction between the GM plant and target organisms; i) potential changes in interactions of the GM plant with non-target organisms; j) potential interactions with the abiotic environment; k) detection and identification methods for the GM plant; and, l) information about previous releases of the GM plant. As a result, the e.r.a. takes into consideration all of the mechanisms for potential adverse effects that have been prescribed under Point 4.2.1 of the Annex to Commission Decision 2002/623/EC, namely: a) The spread of the GMO in the environment b) The transfer of the inserted genetic material to other organisms, or the same organism whether GM or not c) Phenotypic and genetic instability d) Interactions with other organisms (other than exchange of genetic material/pollen) Therefore, potential effects arising from gene transfer are thoroughly assessed during the GM approval procedure.

see previous answer - I have no confidence that this list can be complete - this goes against the case-by-case approach completely

may be

GM trait might change the relation of endophytes with consequences for viability or stability

Risk assessments are not generic but case by case, so no list of cases is complete as new ones are always being introduced. e.g. parthenocarpy, autogamy, transplastomics,

C. Step One Answers for Category C

Not sure.

Imagination sets the limits here. New types of transgens may have other long term effects. Secondary pest may invade, if the target organism is efficiently controlled. Example: the Bt-GM cotton case in China. Under EU conditions where farmers will introduce refugia, I find invasion of secondary insects much more likely than development of resistance in the target organism. I know that you are dealing with this aspect later, but these effects can also be treated here.

see remarks under A
C.3. Development of secondary pests leading to the application of additional plant protection products
The GM trait could alter the behaviour of the target pest (e.g. feeding on other host plants)
The mechanism of interaction between novel protein(s) expressed in the GM plant and the corresponding target organisms are described in detail in the notification and evaluated in detail in the e.r.a. carried out in accordance with Commission Decision 2002/623/EC establishing the guidance notes supplementing Annex II of Directive 2001/18/EC. To summarise, the very high specificity of the mode of action of the expressed proteins has ensured that any potential development of resistance will not lead to the loss of any other plant protection tools. In fact and on the basis of the favourable outcome from the numerous evaluations carried out by safety and regulatory authorities worldwide and including EFSA, we can conclude that currently authorised GMOs should be considered as the most 'environmentally desired' plant protection tools.
Assessment of C.1 relative to current conventional means of pest control (i.e. conventional insecticides)
Comment: To avoid resistance development resistance management should be applied, as it is the case in North America. Once the acreage of Bt-maize is more substantial in Europe, resistance management would probably be applied so that this point should not be a problem.
Comment on C1 - Experience shows us that disease resistance bred into crops is likely to breakdown at some-point. As this questionnaire is focusing on the long-term, I have assessed the hazard of resistance occurring as high. However, whether this means that there translates into an adverse environmental consequence is debatable and I do not feel qualified to classify this likelihood. The crop reverts to the wildtype (susceptible) phenotype and the conventional counterpart (comparator) may not be sprayed with a Bt product. The alternative maybe a new generation of (GM) crop containing different (stacked) resistance genes, which would not necessarily have an adverse environmental effect.
see previous answer
GM trait alters behaviour of the target pest (e.g. feeding on other host plants) Resistance development impairs the use of microbial preparations of Bt
Technically this category should include efficacy and comparisons of the efficacy of other control mechanisms. It is not clear from the category whether the base case is being reported and used as a comparative basis.
The durability has to be assessed.

GM traits require a tool to prevent adaptation of target organisms (e.g. high-dose strategy, refuge strategy, and change with alternative measures). The long term effectiveness of such preventive measures has to be assessed.

I do not understand what you are listing - two completely unrelated characters -resistance in target organisms and assessment of stacked traits - How is this a category? There are a whole range of factors influencing interactions which should be studied on a case by case basis.

D. Step One Answers for Category D

Don't know.

You are talking a lot about effects of root exudates. Do not forget that the decomposing of the GM plant (which may take quite some time) may also release the transgenic product - perhaps this is what you are thinking about in D.4? In D.4 you are also talking about toxic compounds that might cause effects and decrease abundance. It is not necessary toxic compounds that affect the environment; compounds with hormonal effect may affect the NTOs, so that NTOs INCREASE in abundance or are affected differently. These effects are much in parallel to what we see today, where human use of medicines and cosmetics are jeopardizing some ecosystems through sewage systems and water run-off - abundance of the NTO may increase or decrease, or totally other effects occur like unisexual organisms becoming hermaphroditic. I also refer to the China-Bt-cotton case, where the clearing of an ecological niche (the target organism is controlled) opens up for increased abundance of other insects.

The nontarget organism questions are basically solved, no need for further research, except for purely scientific reasons. From the point of view of practical agriculture all necessary steps have been made, see the final reports of ECOGEN, which have now been published, there much more citations of a vast scientific literature <http://www.botanischergarten.ch/Bt/Bibliography-ECOGEN-200801016.pdf>

1. Gm traits alter trophic structures that affect biological control functions. I saw later that this topic is mentioned in E section.

D.8. GM trait (e.g. Bt toxin) causes adverse effect on NTOs as it is mediated through the food chain (indirect effect; the NTO may not be associated with the GMO but with the herbivore feeding on the GM plant - toxin transfer to higher trophic levels) D.9. occurrence of secondary pests leads to switch to other plant protection products - thus affecting NTOs

GM traits cause changes in profile of damaging insects (stimulating shift)

GM crops alter ecological balance by removing the target pest.

The cited 'theses' and 'important biological processes' are already captured in the environmental risk assessment prepared by the applicants according to the EFSA guidance document and are intensely reviewed by EFSA and member states expert bodies prior to issuing a positive scientific opinion. To date all data and studies confirm that the GM crops do not lead to any other effects (neither short nor long-term) than conventionally bred and cultivated crops in the agricultural environment. The traits that are developed for GM crop plants and are cited in the survey as 'favouring adverse long term effects' are identical to the breeding goals pursued by plant breeders world wide for centuries (e.g. disease and stress tolerant plants, higher yielding and more nutritious plants, plants reducing the environmental footprint of humans) with a variety of tools, therefore GM crops do not have the potential to exert any other effects on the environment as current crop varieties and agricultural practices do.

Please note that there is no scientific evidence to contradict the conclusions reached by the GMO Panel of the EFSA on the safety of Bt maize cultivation in the EU. On the contrary and in particular, the OECD published in July 2007 a consensus document on safety information on transgenic plants expressing *Bacillus thuringiensis* (Bt) derived insect control proteins. This document has thoroughly reviewed and confirmed the safety and high degree of specificity of the Cry proteins expressed in Bt maize. Additional publications have confirmed the absence of long-term effects on NTOs arising from Bt proteins expressed in authorised GM plants. In particular and in reference to the cases (theses) below it is worth reviewing the following publications: - Marvier et al. (2007) have published a meta-analysis of all available studies carried out with Bt crops that confirms that there is no indication of ecological risk arising from the cultivation of Bt maize. - Rose et al. (2007) conducted laboratory and field studies to investigate the effects of Bt maize pollen on honey bees. These studies show no adverse effects to honey bees, which strongly substantiates the conclusions on the safety of Bt maize cultivation as previously predicted from laboratory studies. Furthermore, this work supports the tiered risk assessment followed by EFSA as being appropriate for the evaluation of the environmental safety of Bt maize cultivation in the EU. - Prafsifka et al. (2007) show that anthers from Bt maize expressing the Cry1Ab protein affect monarch butterfly larvae when sprinkled on milkweed leaves in the laboratory. Larval wandering is a common response to Cry proteins among sensitive Lepidoptera as an avoidance response and serves to reduce the exposure of the insects to the Cry protein. Similar to those from Losey et al. (1999), the laboratory findings are not relevant to field situations (Sears et al., 2001). Behavioral changes are not likely to occur on milkweed plants in the field because the anther density tested is rare and natural feeding behaviors already reduce exposure to Bt anthers (Anderson et al., 2004; EFSA, 2005 and 2006). Thus this study does not present any new evidence of potential environmental risk from Bt maize cultivation. In addition, Hellmich et al. (2001) had confirmed that Cry1F protein expressed in 1507 maize is relatively non-toxic to larvae of the monarch butterfly compared to other Cry proteins. - Farria et al. (2007) report increased numbers of aphids on Bt maize. Plants are well known to produce chemicals that protect them against aphids in response to stress. Well protected Bt maize plants would be expected to produce less of these chemicals due to less damage from caterpillars. One of the benefits of excellent pest control is the reduction in ...

... secondary plant compounds that can be detrimental to both pest and human health (Mattsson 2000, 2006). In fact, the main effect reported by the authors is that more honeydew produced by the aphids may sustain greater populations of beneficial organisms and therefore may be a benefit of the Bt maize. In any case, the magnitude of the difference they measured between the Bt and non-Bt maize was smaller than the magnitude of the difference among different varieties of non-GM maize. - Mulder et al. (2006) report that soil obtained from a field cultivated with Bt maize expressing the Cry1Ab protein shows transient differences in microbial activity compared to soil from a non-GM maize field. One might expect that maize tissue protected from insect pest damage would reduce stress related factors and lessen invasion from pathogens that could lead to differences in utilization by soil microbes. Therefore, a transient difference in soil microbial activity is neither unexpected nor does it represent any environmental risk from Bt maize cultivation. In addition, the reduction in soil compaction and pesticide inputs that would result from shifting to Bt maize could also improve the microbial activity in field soils. - Rosi-Marshall et al. (2007) attempt to extrapolate from minor effects seen in two laboratory tests conducted under unrealistic exposure conditions and speculate that entire aquatic ecosystems might be affected by Bt maize cultivation. However, when these authors looked for effects in actual streams located within maize fields, they found no effects on either individual species or on the aquatic ecosystems. This indicates that the conclusions derived from their extrapolations from their laboratory based studies were incorrect and that the effects did not occur under realistic field situations (see Chambers et al., 2007; and, Pokelsek et al., 2007). In addition, an isogenic control was not included in the studies making it impossible to know what was the effect that the transgene had on caddisflies. (For additional details, please refer to a letter to the editor of the publishing journal by academic experts from several different countries that details some of the serious shortcomings with this work:

http://pubresreg.org/index.php?option=com_smf&Itemid=27&topic=9.msg24). References Anderson, P. L., Hellmich II, R. L., Sears, M. K., Sumerford, D. V. and Lewis, L. C. (2004) Effects of Cry1Ab-expressing corn anthers on monarch butterfly larvae. *Environmental Entomology*, 33:1109-1115
 Andow, D. A. and Zwahlen, C. (2006) Assessing environmental risks of transgenic plants. *Ecology Letters*, 9: pp. 196-214
 Chambers, C. P., Whiles, M.R., Griffiths, N. A., Evans-White, M A., Rosi-Marshall, E. J., Tank, J. L. and Royer, T. V. (2007) Assessing the impacts of transgenic Bt corn detritus on macroinvertebrate communities in agricultural streams. NABS Annual Meeting, Columbia, South Carolina, 2007

<http://www.benthos.org/database/allnabstracts.cfm/db/Columbia2007abstracts/id/373> EFSA (2005) Opinion of the Scientific Panel on Genetically Modified Organisms on a request from the Commission related to the notification (Reference C/ES/01/01) for the placing on the market of insect-tolerant genetically modified maize 1507 for import, feed and industrial processing and cultivation, under Part C of Directive 2001/18/EC from Pioneer Hi-Bred International/Mycogen Seeds. *The EFSA Journal* 181, pp. 1-33

http://www.efsa.europa.eu/EFSA/Scientific_Opinion/op_gm08_ej181_1507_opinion_doc1_2en1,2.pdf
 EFSA (2006) Annex to the Opinions of the Scientific Panel on Genetically Modified Organisms on the insect resistant genetically modified Bt11 and 1507 maize. ...

<p>... http://www.efsa.europa.eu/EFSA/Scientific_Opinion/gmo_opinion_ej181_ej213_annex_en,6.pdf</p> <p>Farria, C.A., Wäckers, F.L., Pritchard, J., Barrett, D.A. and Turlings, T.C. (2007) High susceptibility of bt maize to aphids enhances the performance of parasitoids of lepidopteran pests. PLoS ONE 2(7)</p> <p>Hellmich, R. L., Siegfried, B. D., Sears, M. K., Stanley-Horn, D. E., Daniels, M. J., Mattila, H. R., Spencer, T., Bidne, K. G. and Lewis, L. C. (2001) Monarch larvae sensitivity to <i>Bacillus thuringiensis</i>-purified proteins and pollen. P.N.A.S., 98, 21, pp. 11925-11930</p> <p>Losey, J. E., Rayor, L. S., and Carter, M. E. (1999) Transgenic pollen harms monarch larvae. Nature 399, 214</p> <p>Marvier, M., McCreedy, C., Regetz, J. and Kareiva, P. (2007) A meta-analysis of effects of Bt cotton and maize on nontarget invertebrates. Science 316: 1475-1477</p> <p>Mattsson, J. L. (2000) Do pesticides reduce our total exposure to food borne toxicants? Neurotoxicology, 21, 195-202</p> <p>Mattsson, J. L. (2006) Spray more for safer food! New Zealand Geographic, 77</p> <p>Mulder, C., Wouterse, M., Raubuch, M., Roelofs, W. and Rutgers, M. (2006) Can transgenic maize affect soil microbial communities? PLoS Comput Biol. 2006 Sep 29; 2(9)</p> <p>OECD (2007) Consensus document on safety information on transgenic plants expressing <i>Bacillus thuringiensis</i> – derived insect control proteins. Pokelsek, J. D., Rosi-Marshall, E. J., Chambers, C. P., Griffiths, N.A., Evans-White, M A., Tank, J. L., Whiles, M. R. and Royer, T. V. (2007) Effects of Bt corn pollen on caddisfly growth rates in Midwestern agricultural streams. NABS Annual Meeting, Columbia, South Carolina, 2007</p> <p>http://www.benthos.org/database/allnabstracts.cfm/db/Columbia2007abstracts/id/370</p> <p>Prasifka, P.L., Hellmich, R.L., Prasifka, J.R. and Lewis, L.C. (2007) Effects of Cry1Ab-expressing corn anthers on the movement of monarch butterfly larvae. Environ Entomol 36(1): 228-233</p> <p>Rose, R.I., Dively, G. P. and Pettis, J. (2007) Effects of Bt corn pollen on honey bees: emphasis on protocol development. Apidologie 38 (4): 368-377</p> <p>Rosi-Marshall E. J., Tank, J. L., Royer, T. V., Whiles, M. R., Evans-White, M., Chambers, C., Griffiths, N. A., Pokelsek, J. and M. L. Stephen (2007) Toxins in transgenic crop byproducts may affect headwater stream ecosystems. PNAS, 104, (41): 16204-16208</p> <p>Sears, M. K., Hellmich, R. L., Stanley-Horn, D. E., Oberhauser, K. S., Pleasants, J. M., Mattila, H. R., Siegfried, B. D. and Dively, G. P. (2001) Impact of Bt corn pollen on monarch butterfly populations: A risk assessment. PNAS 98: 11937-11942</p>
Transformation impairs the GM crop`s ability to attract pest predators or parasitoids
You could go on forever with this, but these seem enough.
As previously
Again, the comparative context for assessing these processes is missing. In particular, D.1. has been an irrelevant endpoint in traditional breeding programs, and as such constitutes and unnecessary endpoint for risk assessment of the GM crop. Historically, most arthropods that consume plant tissue are pest and these are not adequately differentiated in the way this process is described.
Not enough assessment to answer

Again, the main list seems complete. However, the way you assess (and answers the questions below) is highly dependent of the trait. HT will most probably NOT result in adverse effects on NTOs. CRY proteins may, likewise NBS-LRR genes (against e.g. Phytophthora) while more broadly acting insecticidal proteins like lectins or broadly acting fungicide resistance most probably will affect a wider range of organisms. Another issue is that the purpose of Bt-crops is to get rid of pests and are therefore not to be protected. That this might also affect predators/parasitoids is considered to be collateral damage, but of low ecological importance. If we would stop cropping maize because of the infestations being too large we would obtain a similar environmental effect.

Again case specific : the NTOs of concern are ALL those exposed to the GMO, especially those of ecological significance and function, that are sensitive to the GM plant and its products. .

E. Step One Answers for Category E

I can't think of anything, but that doesn't mean that the list is complete.

E.2: Why are you only mentioning biological control? Effects can also be expected in chemical control due to the points (i) and (ii), which might in turn affect the ecosystems. You treat that under G - again I am reluctant to describe the effects in categories like you do here. E.3: What if the transgene changes the architecture of the plant, so that it is no longer suitable for nesting place of birds, or do not give hiding to mice etc? Again I think that only imagination sets the limits for effects to ecological function. The effects will totally depend on the transgene.

GM traits do not have other ecological effects than conventionally applied insecticides.

Again, all questions asked here are also valid for conventional agriculture, where those problems are present but do not lead to expensive risk assessment research, but sometimes they are subject of corrections in normal procedures of agricultural policy. one example: instead of asking the absurd question whether bees are harmed through GM crops which may even cause the colony collapse disease, we should care more about the overall stress of bee colonies, which are frequently moved from one place to the other in modern agriculture
http://pubresreg.org/index.php?option=com_smf&Itemid=27&topic=4.0

It is unclear what is meant by "effects on ecological function" (of what?)- or are "ecological services" meant? If the latter is true then point E.5. should be added: E.5. GM traits cause changes in biodiversity (species, population, landscape biodiversity)

Consideration of these same processes in systems using conventional pest control (e.g. conventional insecticides)

Bt maize, such as MON810 and BT176, have been cultivated in Europe since 1998 in a total cumulative area of more than 420,000 hectares, and there are no scientific reports of any confirmed or biologically significant adverse effects to non-target organisms, biological functions or biogeochemical cycles in the European environment or to human and animal health. In addition, a total area of more than 102 million hectares have been cultivated with Bt maize globally (ISAAA, 2006). As in the EU, there are no scientific reports of any confirmed or biologically significant adverse environmental effects arising from the cultivation of Bt maize worldwide. Furthermore, Bt maize results in reduced environmental impact of agriculture as demonstrated by the environmental benefits of large scale Bt maize cultivation in Spain, France, and other EU Member States to date. Bt maize significantly reduces impact of pesticide spray drift: By eliminating or significantly reducing the use of insecticides, Bt maize affects only those target insect pests which attack the maize plant, while broad application sprays of insecticides may harm both target and non-target species. Yield is a good indicator of potential environmental effects. In the case of Bt maize, farmers reported increased yield of up to 19% of Bt maize over similar conventional maize cultivated in Spain. With such increased yield, Bt maize demonstrates that it is a tool which can play an important role in increasing agricultural productivity without an equivalent increase in land area. In addition and as mentioned above, Rose et al. (2007) conducted laboratory and field studies to investigate the effects of Bt maize pollen on honey bees. These studies show no adverse effects to honey bees, which strongly substantiates the conclusions on the safety of Bt maize cultivation as previously predicted from laboratory studies. Furthermore, this work supports the tiered risk assessment followed by EFSA as being appropriate for the evaluation of the environmental safety of Bt maize cultivation in the EU. References ISAAA (2006) Global Status of Commercialized Biotech/GM Crops: 2006. ISAAA Briefs 35-2006. <http://www.isaaa.org/> Rose, R.I., Dively, G. P. and Pettis, J. (2007) Effects of Bt corn pollen on honey bees: emphasis on protocol development. *Apidologie* 38 (4): 368-377

see previous answers

Not enough information

case by case. Allelopathy, etc..

F. Step One Answers for Category F

Ad F.5 Landscape structure might change - not only due to co-existence - but also as a consequence of the popularity of some GM crops, increasing the area with certain types of crops. What effects that may have, will depend on the GM types and their cultivation practise.

Increased distances of fields between gm and non-gm will lead to an increase of uncultivated landscape, building habitat for biodiversity

Please note that the conclusions obtained from the Farm Scale Evaluations (Zeki, 2003) regarding herbicide tolerant crops confirmed that any potential impact on organisms depends on the consumption of weeds. Thus, any potential effect on higher trophic levels is directly correlated to the efficacy of the herbicide applied, within common agricultural management practices, to control weeds in cultivated areas. Furthermore, genetically modified T25 maize showed a significant two-fold increase in weed biomass, as well as a significant two-fold greater weed seed return resulting in more nectar resources for pollinators and more weed seed resources for granivorous birds. By nature of the fact that Bt maize is resistant to attack by certain insect pests, it eliminates the need to use chemical insecticides against those insect pests. This has been demonstrated repeatedly around the world. It is estimated that insecticide use in Spain could be reduced by 35,000 - 54,000 kg/year with Bt maize. Such a reduction would correspond to an area of between 59,000 and 98,000 hectares each year that is no longer sprayed with insecticides against certain insect pests (Brookes, 2002; 2007; Brookes and Barfoot, 2006a; 2006b). With the reduction in insecticide use, farmers have a reduced risk of accidents, spillage and exposure from using such chemicals. Fumonisin, a specific group of mycotoxins, are known carcinogens that can disrupt folic acid metabolism in mammals and are associated with neural tube birth defects in humans. Fumonisin also have been shown to be fatal to horses, swine and rabbits. Insects cause many forms of damage when they feed on parts of the maize plant and this feeding can provide an entry point for *Fusarium*. The growth of this fungus leads to the production of mycotoxins such as fumonisins. In 2005, the European Commission lowered the maximum residue limits (MRLs) for maize products. These reduced MRLs have come into effect in 2007 and crops need to meet these tough new requirements. Because Bt maize is resistant to these insect pests, under conditions of insect pressure, Bt maize is less likely to become infected by *Fusarium* than non-Bt maize making Bt maize another tool to meet Europe's stringent food safety standards. In 2004, the Agence Française de Sécurité Sanitaire des Aliments (AFSSA) published a report on GMOs in food production which analysed the safety and benefits of Bt maize. In particular, the report stated that the introduction of varieties of Bt maize makes it possible to decrease the amount of mycotoxin contamination, resulting from insect attack, in the maize grain. The full report can be downloaded at the following address: <http://www.afssa.fr> References AFSSA (2004), "OGM et alimentation : peut-on identifier et évaluer des bénéfices pour la santé?", July 2004. <http://www.afssa.fr> Brookes, G. (2007) The benefits of adopting genetically modified, insect resistant (Bt) maize in the European Union (EU): first results from 1998-2006 plantings. PG Economics. <http://www.pgeconomics.co.uk/publications.htm> Brookes, G. (2002) The farm level impact of using Bt maize in Spain. PG Economics. <http://www.pgeconomics.co.uk/publications.htm> Brookes, G. and Barfoot, P. (2006a) GM crops: the first ten years – global socio-economic and environmental impacts. PG Economics. <http://www.pgeconomics.co.uk/publications.htm> Brookes, G. and Barfoot, P. (2006b) Global impact of biotech crops: Socio-economic and environmental effects in the first ten years of commercial use. *AgBioForum* 9 (3): 139-151 Zeki, S. (2003) Preface: One contribution of 10 to a Theme Issue 'The Farm Scale Evaluations of spring-sown genetically modified crops'. *Phil. Trans. R. Soc. Lond. B*, 358: 1775-1776 <http://www.journals.royalsoc.ac.uk>

The first question is showing the bias of the whole questionnaire beautifully: very hidden and packed into a complicated syntax you can guess that there MAYBE even some benefits, but the editors did not dare to write so. In setting the possible answers you prevent fully that there are clear benefits in herbicide tolerant no-tillage to be marked. Again in the question about 'increased number of resistant weeds', you suggest that in RR strategy this is the bigger problem than in conventional herbicide applications, whereas factually it is exactly the contrary. Again you force the experts to follow your bias. This is why I refuse to answer those questions. if you need to be documented about the benefits of RR herbicide-no tillage agriculture, I can provide numerous peer reviewed papers. Furthermore there is a rich literature derived from the British Farm Scale Experiments which I can provide, the latest state is that only GM oilseed rape remains negative compared to the non GM crops, the rest is positive. But again below: no word about beneficial effects. conclusion: I set all points on negligible, although this is incorrect

F.7. GM plant management causes adverse effects on agro-biodiversity due to changes in crop rotation (less rotation)

see previous answers

F7. GM plant [management] causes direct changes in crop rotations with adverse effects on soil fertility and pesticide use. F8. GM plant [management] requires measures to prevent adaptation of target organisms on the trait. The long term effectiveness of such preventive measure has to be assess.

Some of the questions listed below do not relate to GM cultivation but to agricultural changes in general. Some of these changes might be induced by GM crops (i.e. changes of in field weed populations due to altered herbicide use in HT crops), others are related to further intensification of agriculture in general which is independent of the adoption of GM crops. In those cases I answered the questions with 'insufficient data'.

case-by case: eg tillage effects,

F1 -F6 are assuming no GM-effects but only the adverse (not the positive)assumptions of changes in management practises

G. Step One Answers for Category G

Again imagination sets the limits here. Example as before: a transgene that changes the architecture of the plant may have effects on the abiotic environment, e.g. microclimate may change.

I find it hard to link the adverse effects with GM in particular rather than an intensification of agriculture in general

GM plants do not influence the environment different from the influence coming from conventional crops. GM plants will be optimized concerning the use of nutrients, water and other factors. They will have an beneficial effect on the long term stabilisation of the environment.
same remarks as above, and again the same conclusions for the questions
Please note that increased yield helps relieve the burden on precious agricultural land resources and allows for the use of biofuels to reduce production of green house gases from fossil fuels. In particular, in regions of Spain particularly affected by insect pests, farmers reported increased yields of up to 19% of Bt maize over similar conventional maize crops. With such increased yield, Bt maize, including 1507 maize, demonstrates that it is a tool which can play an important role in increasing agricultural productivity without an equivalent increase in land area. Furthermore, reduction in fossil fuel use from reduction in chemical synthesis of insecticides and tractor or airplane use in applying pesticides on GM crops because of the reduced need for spraying insecticides on Bt maize.
see previous answers
Use of HT or IR GM plant causes adverse effects on the abiotic environment by increasing soil erosion (lost of soil fertility) if proportion of maize in crop rotations has been increased as a result of the trait.
Case by case.
G1:intensive crop production means intensive reduction of greenhouse gases G2:Minimum Tillage in HT Crops reduces erosion and fertilizer leaching G3 ;Soil particle exchange sites are abundant and herbicides are metabolized All theses are not referring to GM , but if ever to indirect effects

H. Answers to the final question

Looking at GMO environmental effects in comparison to effects and damage caused by current agricultural practise.
Accompany commercial growing of approved GM plants in the field under typical gricultural practise. This is the only means to confirm the current scientifically based conclusion that no adverse effects are epected from approved GM plant passed through the dilligent evaluation process by indepentdent scientific bodies in the EU.
Interaction between GM plant and target organism. Aim is to develop specific strategies to protect crops against pathogens without harming other, not involved organisms.
D. Non-target organisms

<p>That would certainly depend on the transgenic plants, we are dealing with. The transgenic trait would give us a hint as to what aspect might be most interesting for future research. If we assume that we carry out a thorough and targeted risk assessment, then a number of potential effects can be foreseen, and coped with in the risk management. What is not considered in the risk assessment is the cultivation practice. We know that farmers' behaviour can rarely be predicted; for example they might spray more being able to use non-selective herbicides, even though the reverse should be the case. So category F might be worth looking at, however, it would be difficult to study farmers' behaviour before the GM plants are part of the every-day-practice; I would prefer to give the funding to long time monitoring in relation to agricultural practise. I WOULD NOT PUT MORE MONEY INTO CO-EXISTENCE! In relation to GM plants there is one other uncertainty that might influence the effects to the environment from GM plants, and that is the coming global change and the increased environmental variability it represents. Today GM plants are rarely tested in environments with changed precipitation patterns and increased temperature, ozone and CO₂. Maybe we will see some unexpected effects when GM plants are grown in more stressful climates. This could be another aspect to focus on.</p>
<p>I think it is important to gain a better understanding about the ecological perspective of weed population shifts and evolved resistant biotypes.</p>
<p>I would invest some of the money in a survey on synthetic biology and its perils, the rest into a campaign to raise the awareness for the absolute necessity in organizing another world conference on the risk assessment in agriculture with novel methods, so to say a new Asilomar Conference, concentrating on the real needs of risk assessment related to all the novel gene altering and producing methods in synthetic biology and nanobiology. Why did you actually neglect PHARMING? there are still lots of unsolved questions there which urgently need solutions, pharming is on its way worldwide and we should really get active, and forget the nostalgic risk assessment research on Bt - and other first generation GM crops, with maybe some notable exceptions, such as really novel transgenes, novel methods of transformation, pharmaceutical transgenes, risk assessment of biofuel production and certainly more to come. cheers, and all the best, I do not envy you in analyzing the answers.... K.A.</p>
<p>E,D,C</p>
<p>Cannot be answered as such in general. Depends on GM crops and traits currently used.</p>
<p>In general I would give highest priority to category E "effects on ecological functions" But only few research can be done in this category with 100,000 euros.</p>
<p>my choice should be on investigation about the potential effects on the soil and biogeochemical cycles.</p>
<p>D. Interactions of the GM plant with non-target organisms</p>

DH: Persistence and invasiveness. I would suggest to combine two issues (i) the interaction between hybridization and the GM (see comment at A) and (ii) the effect of unintended stacked multiple GM constructs. On both levels interactions can be tested by comparing phenotypes containing, no, single and multiple GM constructs. The interaction between the GM constructs themselves gives as well a indication about their interaction in ferals. If a larger grant would be available above 100,000, the location of the GM construct within the genome could be a next level of research (needing a larger quantity of expensive molecular work).
Although it's not possible to completely avoid a certain subjectivity, as I'm personally involved in the research on the gene transfer and its consequences, I think funds allocation should be proportional to the extent of the identified risks. Yet, I'm not an expert in cultivation and management techniques (category F) but it is clear that most of the long term effects listed here, and their extent, depend directly on the impact on agricultural practices of GM crops. Therefore this field of research should probably receive much more attention than it did till now.
I would use the funding to protect field trials, which are the basis for developing the scientific knowledge that allows for the proper evaluation of the safety of GMOs in accordance with EU regulations and international guidelines.
Establish sounds baselines (NTOs, soil microorganisms, effect of management practices)on the existing agricultural systems.
1. 100 000 € will not be enough to deliver reliable data on longterm effects. To explore longterm effects a combination of laboratory based and field release experiments resp. monitoring ver several years will be needed. 2. The most important questions concern ecological functions: impact on pollination and soil function.
Effects on ecological functions derived from new traits (from GMOs or from other "conventional" technologies)linked with specific cultivation and crop management.
I would spend the euros for category F 'Impacts of cultivation and management'. First, because I am an agronomist and interested in the management of plant production. Secondly, because agronomical changes will evolve and significantly alter crop rotations, intensity of production, etc. This will occur with GM-crops as well as with non-GM crops. The task of scientists will be to decide on a case by case basis which changes are worse. Alternatively, formulating it more positively: Which alteration results in a more positive outcome for the society and the agricultural environment?
Look at the value added effects of GM cultivation rather than concentrating solely on adverse indirect effects
Influence of herbicide resistance traits on the introgression of traits of evolutionary significance (high fitness value) in natural habitats from transgenic crops to weedy relatives.

Gene flow from GM crops and its potential ecological consequences.
E. Effects on ecological function.
Instead of financing any exhaustive environmental potential effects, I would put my money into research of new GM crops, including improved IR and HR crops.
A generic answer to this question is not possible because the potential adverse effects will differ among transgenic crops (crop x trait combinations). The premise of the question is that GM crops as a class have certain common hazardous properties; this premise is untenable.
1. Good agricultural practice (GAP) for growing of selected GM crops (maize, beet, rape, potatoes, soybean) in various European cropping systems. 2. Evaluation a prevention of the real agro-ecological and economical risks within agro ecosystems e.g. resistance, shift in communities, cost-benefit analyses in various crops, natural conditions, and cropping systems. 3. Simplifying of complicated EU law and rules for GMOs growing and traceability of GM products which make impossible to use the modern biotechnology and cause both direct and indirect economical losses.
developing biodiversity maps for NTO risk assessments monitoring management effects of novel crops and processes.
Comparison of GM crop cultivation with current agricultural practice.
In relation to the in the survey mentioned GMOs, being currently available GM crops, I would propose an EU-wide long term monitoring of post-market cultivation. Ideally this effort should be coupled with efforts outside the EU, namely USA, Canada, Argentina, etc. In this way we get a better understanding what happens in real practice. PS I would also like to make a general comment. The number of aspects listed in this survey is quite exhaustive and seems to cover most considerations to be made during the risk assessment. Nevertheless in many cases the questions are formulated ambiguously or different types of modifications are lumped together. This complicated the answering and there was little room to clarify my way of thinking. If also the other experts experienced this issue, the returned answers may not be fully comparable.
Category F: Impacts of the specific cultivation, management and harvesting techniques
F: The interaction of GM traits with changes in agricultural practice - with a view to mitigating the adverse impacts and enhancing the beneficial impacts.
Impact of the specific cultivation, management and harvesting techniques
Ecological effects on NTOs of Bt maize
Development of realistic assessment methods

F. Impacts of specific cultivation, management and harvesting techniques. GM crops need to be given a better chance for use in ag systems in the EU. The current approach favors asking questions that could take years to answer while the European environment is being degraded by conventional ag practices and poor environmental policy. This research (like that that was withheld from the public in Italy) might enable to the public to see a different side to some of these technologies that are available.
Interactions between GM plant and target and non-target organisms
Learning lessons from the GM science debate to see what lessons and techniques can be taken forward into other contentious areas of agriculture and land management, e.g. the bio energy debate
Sorry I don't know enough to have helped with this survey. I think it is probably more productive to study each question in a more focused and scholarly manner than to ask people to fill out a survey.
Interactions of the GM plants with target and non-target organisms
F; focused on long term adverse effects of alternative (conventional) systems that are already known. It seems important to investigate whether GMOs will boost these findings or not.
D
D = effects on the non-target organisms examined with two aims: - detect changes in biodiversity in complex field studies - if they occur, investigations on their causes (i.e. other research categories specified in this questionnaire) would identify the primary specific environmental effects of GM crops. - elaboration of simple laboratory tests for 1st tier assessment of transgene products on representative non-target organisms. Support is also needed for improving quantification of transgene products by ELISA backed-up with biological assays. Most investigations on the environmental impact of GM crops are of little use if transgene products are not quantified reliably.
D-interaction between GM plant with non-target organisms
Impacts of the specific cultivation, management and harvesting techniques
Category A. Introgression, invasiveness and fitness in perennial GM plants.
Do some studies in Canada on Category A: persistence and invasiveness and Category F: Impacts of the specific cultivation, management and harvesting techniques wit HT Canola
As the only risk in reality is gene flow from GM crop to related weeds, within the field (not to wild species outside the field) it should be spent on research on how to mitigate this problem, and not on the impossible scenarios developed in this highly biased questionnaire.