

2.4. Non-comparative field studies, with assumptions, projections and reviews without original field data

Here only papers are listed with the full abstracts and the links to the full texts which are based on questionable and theoretical concepts of agricultural ecology, on theoretical extemporations of risk assessment scenarios, stressing possible or future potential negative impacts. It is interesting to realize, that those texts are, with a few exceptions, building on rather negative scenarios and rarely mention possible benefits. This does not necessarily mean, that these are worthless publications, since we need researchers, who do this kind of projection and structural analysis of risk assessment – but we should ask seriously the question of whether this is necessary for the first generation of transgenic crops which are now in the field with such as stupendous success for many years.

Fact is, that we are now in a period, where overwhelmingly positive evidence with the new crops devaluates most of those negative and foremost theoretical scenarios. Many researchers would even say that it is highly questionable, whether we should have gone into such detailed, laborious and costly risk assessment at all.

After all, most of the risk assessment done hitherto is dealing with crops with a single insertion of naturally occurring and widespread genes which are only novel for the crops. In the years after the Asilomar Conference in 1975 precautionary steps in risk assessment have now resulted in the safe use of selected and very well tested transgenic crops of the first generation, they are out in the field in millions of hectares and are bound to stay since they are better for environment, food production in all scales and and for business in agriculture.

Whether this research activity costing billions is really justified to this degree and whether it is justified to call for more and more detailed assessment is highly questionable. The period of cultivating uncritical negative assumptions is closed and also the following up with the direct and unreflected application of the *Precautionary Approach* (often incorrectly and actually unlawfully called *Precautionary Principle*) for those crops of the first generation should be finished now.

Still it has to be said, that in the political situation of Europe, there are many politicians who want to take advantage of such negative wording: After all, one can claim that some of it has been published in peer reviewed journals and if you do not carefully weight the science behind it as defined in this report at the beginning, you could easily take some of the statements for granted.

But after 10 years of widespread and world wide use of the Bt strategy, and after literally hundreds of risk assessment studies (most have been published in peer reviewed literature), we need to seriously change our attitude and risk mindedness.

The times of the Monarch scare and other short lived concerns is now over and the dialogue must start seriously between the people, the researchers and regulators and the author of this review appeals to the politicians to lift import and growing bans which are no more scientifically justified. Those scare scenarios related to GM crops need to be devaluated and are in most cases they are of practical value anymore, and as we know from some economic calculations, they are also financially more and more detrimental.

The Austrian report of (Dolezel et al., 2006) calls for a special mention, see summary below in the list. It could be understood by uninformed people or even by ecologist not familiar with modern agriculture as an alarming report documenting lots of detrimental effects of Bt crops on environment and biodiversity, but the study is seriously flawed: It is (1) a rather poor literature review, by far not complete in its selection of papers, but (2) its most serious flaw is the lack of a comparative perspective as described in the introduction of this report.

Here just one example: the studies of Felke and Langenbruch are cited in the Austrian report in a way which completely neglects the impact of pesticides on those rare butterflies studied (Felke & Langenbruch, 2003; Felke & Langenbruch, 2005; Felke et al., 2002): They were able to demonstrate negative effects on a rare butterfly species in Germany through pollen from a Bt crop, event 176 from Syngenta with its high toxicity in laboratory research under artificial conditions of force feeding. Felke et al. also did not keep to the simple rule of making a comparison to conventional agriculture with its abundant pesticide use, which has long been spotted as the major culprit of biodiversity decline caused through modern agriculture (Longley & Sotherton, 1997; Robinson & Sutherland, 2002). As a consequence, the Austrian study does not meet the scientific rules of a meta study such as the one of (Marvier et al., 2007) and they also do not meet the scientific criteria set up in this review, rules which actually are *undisputable*. This is why below the studies in this category are given without commentary, they cannot be used in a scientific evaluation of risks and benefits of Bt crops on the environment and biodiversity.

Andow, D. and A. Hilbeck (2004). "Science Based Risk Assessment for Nontarget Effects of Transgenic Crops." *Bioscience* **54**(7): 637-649. <http://www.botanischergarten.ch/Bt/Andow-Hilbeck-Biosciences2004.pdf>

Nontarget risk assessment for transgenic crops should be case specific, depending on the plant, the transgene, and the intended release environment. We propose an ecological risk-assessment model that preserves the strengths and avoids the deficiencies of two other commonly used models, the ecotoxicology and nonindigenous-species models. In this model, locally occurring nontarget species are classified into groups according to their ecological function. Within each group, ecological criteria are used to select the species that are most likely to be affected by the transgenic crop. Initial experimental assessments are conducted in the laboratory and consist of two kinds of test: toxicity tests using purified transgene product, and whole-plant tests using intact transgenic plants. For nontarget natural enemy species, it will also be important to evaluate both direct bitrophic impacts and indirect tritrophic impacts. (Andow & Hilbeck, 2004)

Clark, E. A. and H. Lehman (2001). "Assessment of GM crops in commercial agriculture." *Journal of Agricultural & Environmental Ethics* **14**(1): 3-28. <Go to ISI>://000167926400001 AND <http://www.botanischergarten.ch/Bt/Clark-Assessment-commercial-2001.pdf>

The caliber of recent discourse regarding genetically modified organisms (GMOs) has suffered from a lack of consensus on terminology, from the scarcity of evidence upon which to assess risk to health and to the environment, and from value differences between proponents and opponents of GMOs. Towards addressing these issues, we present the thesis that GM should be defined as the forcible insertion of DNA into a host genome, irrespective of the source of the DNA, and exclusive of conventional or mutation breeding. Some defenders of the commercial use of GMOs have referred to the scientific work of GMO critics as "junk science." Such a claim is false and misleading, given that many papers critical of both the utility and safety of GMOs have been published in peer reviewed journals by respected scientists. In contrast, there is a dearth of peer reviewed work to substantiate the frequently heard assertions of either safety or utility in GMOs. The polarity, which now characterizes much of the public discourse on GMOs, reflects not simply scientific disagreement, but also disagreement in underlying value assumptions. Value differences strongly affect the assessment of both benefit and harm from GMOs. The concept of substantial equivalence occupies a pivotal position in the GMO risk assessment process that is used in both Canada and the US. A GMO judged to be substantially equivalent to a conventional product - as have all submissions to date - is presumed to be safe enough for commercialization. The conclusion of safety - from both human health and environmental perspectives - should be based on scientific evidence, corroborated by actual experimentation. However, regulators infer safety largely from assumptions-based reasoning, with little or no experimental validation. The judgement of safety because of substantial equivalence is a dubious argument by analogy. (Clark & Lehman, 2001)

Craig, W., M. Tepfer, G. Degrassi and D. Ripandelli (2008). "An overview of general features of risk assessments of genetically modified crops." *Euphytica* online first, January 11. <http://dx.doi.org/10.1007/s10681-007-9643-8> AND <http://www.botanischergarten.ch/Bt/Craig-Overview-Risk-2008.pdf>

Abstract: The intentional introduction into the environment or market of genetically modified organisms (GMOs) is nearly always governed by a framework of science-based risk assessment and risk management measures. This is usually implemented through the integration of hazard identification and characterisation of all of the elements of risk associated with a new GM crop or derived product. Typical categories of hazards arising from the introduction of transgenic crops include: possible unintended negative health effects in a susceptible subgroup of the consumer (target) population; the evolution of resistance in the targeted pest/pathogen populations when the transgene confers resistance to a pest or pathogen; non-target hazards associated directly or indirectly with the transgenic plant or transgene product outside the plant; and those associated with the integration and subsequent expression of the transgene in a different organism or species following gene flow. The consequences of likely exposure to these and other hazards are considered in this introduction to the main issues raised when evaluating the possible risks arising from the importation or cultivation of genetically modified crops. (Craig et al., 2008)

Cruywagen, G. C., P. Kareiva, M. A. Lewis and J. D. Murray (1996). "Competition in a spatially heterogeneous environment: Modelling the risk of spread of a genetically engineered population." *Theoretical Population Biology* 49(1): 1-38. <Go to ISI>://A1996UB46400001

In recent years regulations have been developed to address the risks of releasing genetically engineered organisms into the natural environment. These risks are generally considered to be proportional to the exposure multiplied by the hazard. Exposure is, in part, determined by the spatial spread of the organisms, a component of risk suited to mathematical analysis. In this paper we examine a mathematical model describing the spread of organisms introduced into a heterogeneous environment, focusing on the risk of spread and plausibility of containment strategies. Two competing populations are assumed, one the natural species and the other an engineered species or strain, both of which move randomly in a spatially heterogeneous environment consisting of alternating favourable and unfavourable patches. The classical Lotka-Volterra competition model with diffusion is used. Analyses of the possible spread and invasion of engineered organisms are thus reduced to finding periodic travelling wave solutions to the model equations. We focus on whether a very small number of engineered organisms can spatially invade a natural population. Initially we investigate the problem for spatially periodic diffusion coefficients and demonstrate that, under the right circumstances and a large enough unfavourable patch, invasion does not succeed. However, if spatially periodic carrying capacities are assumed along with

spatially varying diffusion rates, the situation is far more complex. In this case containment of the engineered species is no longer only a simple function of the unfavourable patch length. BS using perturbation solutions to the nonuniform steady states. approximate invasion conditions are obtained. (C) 1996 Academic Press, Inc. (Cruywagen et al., 1996)

Daniell, H. (1999). "Environmentally friendly approaches to genetic engineering." *In Vitro Cellular & Developmental Biology-Plant* **35**(5): 361-368.<Go to ISI>://000083672000002

Several environmental problems related to plant genetic engineering may prohibit advancement of this technology and prevent realization of its full potential. One such common concern is the demonstrated escape of foreign genes through other crops or toxicity of transgenic pollen to nontarget insects. The high rates of gene flow from crops to wild relatives (as high as 38% in sunflower and 50% in strawberries) are certainly a serious concern. Maternal inheritance of the herbicide resistance gene via chloroplast genetic engineering has been shown to be a practical solution to these problems. Another common concern is the suboptimal production of *Bacillus thuringiensis* (Bt) insecticidal protein or reliance on a single (or similar) B.t. protein in commercial transgenic crops, resulting in B.t. resistance among target pests. Clearly, different insecticidal proteins should be produced in lethal quantities to decrease the development of resistance. Such hyperexpression of a novel B.t. protein in chloroplasts has resulted in 100% mortality of insects that are up to 40 000- fold resistant to other B.t. proteins. Yet another concern is the presence of antibiotic resistance genes in transgenic plants that could inactivate oral doses of the antibiotic or be transferred to pathogenic microbes in the GI tract or in soil, rendering them resistant to treatment with such antibiotics. Cotransformation and elimination of antibiotic resistant genes from transgenic plants using transposable elements via blebbing are promising new approaches. Genetic engineering efforts have also addressed yet another concern, i.e., the accumulation and persistence of plastics in our environment by production of biodegradable plastics. Recent approaches and accomplishments in addressing these environmental concerns via chloroplast genetic engineering are discussed in this review. (Daniell, 1999)

Dolezel, M., A. Heissenberger and H. Gaugitsch (2006). Ecological effects of genetically modified maize with insect resistance and/or herbicide tolerance. Literature research report on behalf of the Bundesministerium für Gesundheit und Frauen, Wien, Austria. Wien, Austria, Bundesministerium für Gesundheit und Frauen,; pp 70 pages.
http://www.bmgfj.gv.at/cms/site/attachments/5/6/2/CH0255/CMS1134457515326/literaturstudie_mais_endbericht.pdf

This review evaluates scientific studies published in peer-review journals during the last 3-4 years that considered ecological effects of insect resistant (Bt) and herbicide tolerant maize. The majority of the scientific studies deal with ecological effects of maize containing the Cry1Ab toxin. In contrast, very few studies are available that considered other Cry toxins used in Bt maize such as Cry3Bb1 or Cry1F. Studies dealing with the Cry1Ab toxin relate to effects on non-target Lepidoptera which were the first non-target effects confirmed for Bt176 maize. New studies confirm these adverse effects of Bt176 pollen to non-target Lepidoptera but show that also MON810 and Bt11 maize pollen or anthers may adversely affect lepidopteran larvae especially under prolonged exposure.

Additive effects can be expected when larvae are exposed to a combination of Bt pollen and anthers containing the Cry1Ab toxin. Generally, effects on lepidopteran larvae are shown to be species and age-specific. Exposure of non-target lepidopteran larvae to Bt maize pollen under field conditions can be highly variable and is still unknown for the majority of European butterfly species. Published studies on ecological effects of Bt maize containing the Cry1Ab toxin also deal with impacts on other non-target organisms than Lepidoptera, such as herbivorous and predatory arthropods. A large range of herbivorous or predatory species have been shown to contain the Cry1Ab toxin when exposed to Bt maize in the field and adverse impacts on some species were confirmed mostly in laboratory studies. There is a definite need for standardization of laboratory feeding assays or tritrophic experiments with non-target herbivores and predators in order to enable the comparability of these studies. It is unclear if these adverse effects which were observed in the laboratory can be also translated to field conditions. Results on nontarget arthropod abundance in Bt and non-Bt maize fields are inconsistent and adverse effects are mostly restricted to single years or locations or certain species. Methodological flaws in the experimental design

and few replications make it unlikely to detect small abundance effects of these non-target organisms in most field studies. According to the studies currently available major effects on non-target species abundance due to Bt maize cultivation seem to be rather unlikely. Therefore the emphasis of further studies should be on the detection of subtle and long-term effects to non-target organisms. Adverse effects of Bt maize containing the Cry1Ab toxin on parasitoids and hyperparasitoids have been shown and are most likely due to indirect and host-mediated effects. Reports on soil persistence and insecticidal activity of Cry1Ab toxins are still controversial although differences in the experimental design or methods used explain to some extent the different results obtained. Some adverse effects of Bt maize on different soil organisms are indicated but confirmation of these indications is still needed. Nevertheless decomposition is most likely different between Bt and non-Bt maize containing the Cry1Ab toxin and was confirmed even for different Bt plant species which is probably the result of differences in lignification patterns between Bt and non-Bt maize. In contrast to non-target studies of Bt maize containing the Cry1Ab toxin only few studies have so far evaluated in depth non-target effects of the Cry3Bb1 containing maize. These studies give only few indications for consistent or major effects of this toxin on non-target organisms. Neither laboratory studies nor field experiments considering effects of the Cry1F toxin or the insecticidal toxins Cry34Ab1 and Cry35Ab1 on non-target herbivores, predators or soil organisms are so far available. Currently no evidence is available confirming negative impacts of Cry1Ab, Cry3Bb1 and Cry1F toxins on pollinators such as honey bees.

Only few studies are currently available that consider ecological effects of herbicide tolerant maize. The results of the British Farm Scale Evaluations were re-analysed recently. Exclusion of pre-emergence atrazine treatments from the analyses resulted in fewer positive effects of the herbicide tolerant maize fields compared to the non transgenic maize fields than reported previously both for weeds and arthropods. Other studies have, however, shown that the continuous use of glyphosate can change weed communities. Other studies considering herbicide tolerant maize predict the increase of infections by root pathogens due to the delay of the herbicide application. The occurrence of herbicide tolerant weeds has been observed since the large-scale introduction of herbicide tolerant crops and a further increase in abundance and frequency of herbicide resistant weeds is expected to occur. (Dolezel et al., 2006)

Donkin, S. S., J. C. Velez, A. K. Totten, E. P. Stanisiewski and G. F. Hartnell (2003). "Effects of feeding silage and grain from glyphosate-tolerant or insect-protected corn hybrids on feed intake, ruminal digestion, and milk production in dairy cattle." *Journal of Dairy Science* **86**(5): 1780-1788.<Go to ISI>://000182985600026

Lactating dairy cows were used to determine effects of feeding glyphosate-tolerant or insect-protected corn hybrids on feed intake, milk production, milk composition, and ruminal digestibility. Corn resistant to European corn borer (*Ostrinia nubilalis*) infestation (Bt-MON810), or its nontransgenic control (Bt-CON), were planted in alternating fields, during two successive years. One-half of each strip was harvested for whole plant corn silage and the remainder was allowed to mature and harvested as grain. Effects of feeding diets containing either Bt-MON810 or Bt-CON grain and silage were determined in two experiments (1 and 2) conducted during successive years. In experiment 3, glyphosate-tolerant Roundup Ready corn (RR-GA21) or its nontransgenic control (RR-CON) corn were grown in alternating fields during one cropping season. Diets contained 42 to 60% corn silage and 20 to 34% corn grain from Bt-MON810, RR-GA21, or the appropriate nontransgenic counterpart; treatments were applied using a switchback design. Cows were fed ad libitum and milked twice daily. There were no differences for nutrient composition between silage sources or between grain sources within an experiment. Data for experiments 1 and 2 indicated similar dry matter intake (DMI), 4% fat-corrected milk (FCM) production, and milk composition between Bt-MON810 and Bt-CON diets. There were no differences for DMI, 4% FCM production, and milk composition between RR-GA21 and RR-CON diets. There was no difference in ruminal degradability, determined separately for corn silage and corn grain, for RR-GA21 or Bt-MON810-hybrids compared with their respective controls. These data demonstrate equivalence of nutritional value and production efficiency for corn containing Bt-MON810 compared with its control and for RR-GA21 corn compared with its control. (Donkin et al., 2003)

Driehuis, F. and S. Elferink (2000). "The impact of the quality of silage on animal health and food safety: A review." *Veterinary Quarterly* **22**(4): 212-216.<Go to ISI>://000165306200009

This paper reviews the microbiological aspects of forage preserved by ensilage, The main principles of preservation by ensilage are a rapid achievement of a low pH by lactic acid fermentation and the maintenance of anaerobic conditions. The silage microflora consists of beneficial micro-organisms, i.e. the lactic acid bacteria responsible for the silage fermentation process, and a number of harmful micro-organisms that are involved in anaerobic or aerobic spoilage processes. Micro-organisms that can cause anaerobic spoilage are enterobacteria and clostridia, *Clostridium tyrobutyricum* is of particular importance because of its ability to use lactic acid as a substrate. Silage-derived spores of *C. tyrobutyricum* can cause problems in cheese making. Aerobic spoilage of silage is associated with penetration of oxygen into the silage during storage or feeding. Lactate-oxidizing yeasts are generally responsible for the initiation of aerobic spoilage, The secondary aerobic spoilage flora consists of moulds, bacilli, listeria, and enterobacteria, Mycotoxin-producing moulds, *Bacillus cereus*, and *Listeria monocytogenes* in aerobically deteriorated silage form a serious risk to the quality and safety of human and to animal health. (Driehuis & Elferink, 2000)

Giovannetti, M. (2003). "The ecological risks of transgenic plants." Rivista Di Biologia-Biology Forum 96(2): 207-223.<Go to ISI>://000186191700003

Biotechnologies have been utilized "ante litteram" for thousands of years to produce food and drink and genetic engineering techniques have been widely applied to produce many compounds for human use, from insulin to other medicines. The debate on genetically modified (GM) organisms broke out all over the world only when GM crops were released into the field. Plant ecologists, microbiologists and population geneticists carried out experiments aimed at evaluating the environmental impact of GM crops. The most significant findings concern: the spread of transgenes through GM pollen diffusion and its environmental impact after hybridisation with closely related wild species or subspecies; horizontal gene transfer from transgenic plants to soil microbes; the impact of insecticide proteins released into the soil by transformed plants on non-target microbial soil communities. Recent developments in genetic engineering produced a technology, dubbed "Terminator", which protects patented genes introduced in transgenic plants by killing the seeds in the second generation. This genetic construct, which interferes so heavily with fundamental life processes, is considered dangerous and should be ex-ante evaluated taking into account the data on "unexpected events", as here discussed, instead of relying on the "safe until proven otherwise" claim. Awareness that scientists, biotechnologists and genetic engineers cannot answer the fundamental question "how likely is that transgenes will be transferred from cultivated plants into the natural environment?" should foster long-term studies on the ecological risks and benefits of transgenic crops. (Giovannetti, 2003)

Greenpeace (2002). Environmental Dangers of Insect Resistant Bt Crops. Greenpeace Briefing Pack December 2002, Greenpeace: pp 4.

Of the 52 million hectares of genetically engineered (GE) crops grown throughout the world in 2001, 23% (12 million hectares) were varieties developed to be resistant to insects. Most such crops are created by inserting a synthetic version of a gene from the naturally occurring soil bacterium, *Bacillus thuringiensis* (Bt), so that the plants produce their own Bt toxins to destroy pests. Insect resistant Bt maize, cotton and potatoes have already been grown extensively on a commercial scale, particularly in the USA, and many other Bt crops are under development (e.g. oilseed rape, rice and tomatoes). However, there is strong evidence to indicate that the rush to commercialise Bt crops will have serious environmental consequences. Impact on non-target beneficial organisms In its natural form, Bt has been used by farmers practising organic and other sustainable growing methods since the 1950s as a spray to kill pests without damaging nontargeted insects or other wildlife. The Bt toxins produced by insect resistant crops such as Novartis' and Monsanto's GE maize, e.g. MON810 however, are significantly different and have been shown to be harmful to beneficial predator insects.

Natural Bt sprays have no effect on non-target organisms because the bacterial "pro-toxin" is in an inactivated state and only becomes toxic when processed in the gut of certain (targeted) species of insect larvae. In contrast, many insect resistant plants contain an artificial, truncated Bt gene and less processing is required to generate the toxin. It is therefore less selective, and may harm non-target insects that do not have the enzymes to process the pro-toxin, as well as the pests for which it is intended.

Research has suggested that transgenic Bt plants could also be harmful to non-target organisms that feed on pests exposed to their toxins. The impact of Bt corn pollen on monarch butterflies is the most well-

known example of this phenomenon. Other organisms that have been shown to be affected by Bt crops are lacewings – beneficial insects that play an important role in the natural control of crop pests. Both earthworms and collembola (other small soil-dwelling invertebrates) have been shown to be affected by Bt crops.

Changes in populations of both other pests and of natural enemies have been documented in Bt cotton. Data from China show that use of Bt crops can exacerbate populations of other secondary pests, including aphids, lygus bug, whitefly, Carmine spider mite and thrips⁶. Studies there have shown significant reductions in populations of the beneficial parasites *Microplitis* sp. (88.9% reduction) and *Campoletis chloridae* (79.2% reduction) in Bt cotton fields. Data being collected in India indicates higher levels of aphids and jassids in Bt cotton fields. Scientists in the United States have recently demonstrated impacts of Bt corn on field populations of *Coleomegilla maculata*, a beneficial predatory insect commonly found in corn fields.

The disturbing conclusion is that Bt toxins from GE plants can kill non-target species and be passed higher up the food chain, an effect which has never been observed with the Bt toxin in its natural form.

Groot, A. T., Dicke, M. (2002). "Insect-resistant transgenic plants in a multi-trophic context." Plant Journal 31(4): 387-406. <http://www.botanischergarten.ch/Bt/Groot-Multi-trophic-2002.pdf>

So far, genetic engineering of plants in the context of insect pest control has involved insertion of genes that code for toxins, and may be characterized as the incorporation of biopesticides; into classical plant breeding. In the context of pesticide usage in pest control, natural enemies of herbivores have received increasing attention, because carnivorous arthropods are an important component of insect pest control. However, in plant breeding programmes, natural enemies of herbivores have largely been ignored, although there are many examples that show that plant breeding affects the effectiveness of biological control. Negative influences of modified plant characteristics on carnivorous arthropods may induce population growth of new, even more harmful pest species that had no pest status prior to the pesticide treatment. Sustainable pest management will only be possible when negative effects on non-target, beneficial arthropods are minimized. In this review, we summarize the effects of insect-resistant crops and insect-resistant transgenic crops, especially Bt crops, from a food web perspective. As food web components, we distinguish target herbivores, non-target herbivores, pollinators, parasitoids and predators. Below-ground organisms such as *Collembola*, nematodes and earthworms should also be included in risk assessment studies, but have received little attention. The toxins produced in Bt plants retain their toxicity when bound to the soil, so accumulation of these toxins is likely to occur. Earthworms ingest the bound toxins but are not affected by them. However, earthworms may function as intermediaries through which the toxins are passed on to other trophic levels. In studies where effects of insect-resistant (Bt) plants on natural enemies were considered, positive, negative and no effects have been found. So far, most studies have concentrated on natural enemies of target herbivores. However, Bt toxins are structurally rearranged when they bind to midgut receptors, so that they are likely to lose their toxicity inside target herbivores. What happens to the toxins in non-target herbivores, and whether these herbivores may act as intermediaries through which the toxins may be passed on to the natural enemies, remains to be studied. (Groot, 2002)

Hilbeck, A. (2001). "Implications of transgenic, insecticidal plants for insect and plant biodiversity." Perspectives in Plant Ecology Evolution and Systematics 4(1): 43-61. <http://www.botanischergarten.ch/Bt/Hilbeck-Implications-2001.pdf>

Genetically modified plants are widely grown predominantly in North America and to a lesser extent in Australia, Argentina and China but their regions of production are expected to spread soon beyond these limited areas also reaching Europe where great controversy over the application of gene technology in agriculture persists. Currently, several cultivars of eight major crop plants are commercially available including canola, corn, cotton, potato, soybean, sugar beet, tobacco and tomato, but many more plants with new and combined multiple traits are close to registration. While currently agronomic traits (herbicide resistance, insect resistance) dominate, traits conferring "quality" traits (altered oil compositions, protein and starch contents) will begin to dominate within the next years. However, economically the most promising future lies in the development and marketing of crop plants expressing pharmaceutical or

"nutraceuticals" (functional foods), and plants that express a number of different genes. From this it is clear that future agricultural and, ultimately, also natural ecosystems will be challenged by the large-scale introduction of entirely novel genes and gene products in new combinations at high frequencies all of which will have unknown impacts on their associated complex of non-target organisms, i.e. all organisms that are not targeted by the insecticidal protein. In times of severe global decline of biodiversity, pro-active precaution is necessary and careful consideration of the likely expected effects of transgenic plants on biodiversity of plants and insects is mandatory. In this paper possible implications of non-target effects for insect and plant biodiversity are discussed and a case example of such non-target effects is presented. In a multiple year research project, tritrophic and bitrophic effects of transgenic corn, expressing the gene from *Bacillus thuringiensis* (Bt-corn) that codes for the high expression of an insecticidal toxin (Cry1Ab), on the natural enemy species, *Chrysoperla carnea* (the green lacewing), was investigated. In these laboratory trials, we found prey-mediated effects of transgenic Bt-corn causing significantly higher mortality of *C. carnea* larvae. In further laboratory trials, we confirmed that the route of exposure (fed directly or via a herbivorous prey) and the origin of the Bt (from transgenic plants or incorporated into artificial diet) strongly influenced the degree of mortality. In choice feeding trials where *C. carnea* could choose between *Spodoptera littoralis* fed transgenic Bt-corn and *S. littoralis* fed non-transgenic corn, larger instars showed a significant preference for *S. littoralis* fed non-transgenic corn while this was not the case when the choice was between Bt- and isogenic corn fed aphids. Field implications of these findings could be multifold but will be difficult to assess because they interfere in very intricate ways with complex ecosystem processes that we still know only very little about. The future challenge in pest management will be to explore how transgenic plants can be incorporated as safe and effective components of IPM systems and what gene technology can contribute to the needs of a modern sustainable agriculture that avoids or reduces adverse impacts on biodiversity? For mainly economically motivated resistance management purposes, constitutive high expression of Bt-toxins in transgenic plants is promoted seeking to kill almost 100% of all susceptible (and if possible heterozygote resistant) target pest insects. However, for pest management this is usually not necessary. Control at or below an established economic injury level is sufficient for most pests and cropping systems. It is proposed that partially or moderately resistant plants expressing quantitative rather than single gene traits and affecting the target pest sub-lethally may provide a more meaningful contribution of agricultural biotechnology to modern sustainable agriculture. Some examples of such plants produced through conventional breeding are presented. Non-target effects may be less severe allowing for better incorporation of these plants into IPM or biological control programs using multiple control strategies, thereby, also reducing selection pressure for pest resistance development. (Hilbeck, 2001)

Hilbeck, A. and J. E. U. Schmidt (2006). "Another view on Bt Proteins – How Specific are They and What Else Might They Do?" *Biopesticides International* 2(1): 1-50.
<http://www.botanischergarten.ch/Bt/Hilbeck-another-View-2006a.pdf>

The entomopathogenic bacterium *Bacillus thuringiensis* (Bt) and its toxins are extensively used for pest control purposes in agriculture, forestry and public health programmes since the 1930. In addition to spray formulations, transgenic plants containing Bt genes for the expression of the toxins (Bt plants) are commercially available since the mid 1990s and are grown on an increasing percentage of the global agricultural area. A main reason for the importance of Bt as a pesticide is the assumed environmental safety concluded from the high specificity of its endotoxins (Cry proteins) towards a limited number of target organisms, mostly distinct groups of pest insects. While the mode of action of the Cry toxins in these susceptible target insects is well studied, Bt experts claim that several details are still not understood well enough. Although there is considerable experience with the application and the environmental safety of Bt sprays, a number of research papers were published in the past that did report adverse effects on non-target organisms. These and the widespread use of transgenic Bt plants stimulated us to review the published laboratory feeding studies on effects of Bt toxins and transgenic Bt plants on non-target invertebrates. We describe those reports that documented adverse effects in non-target organisms in more detail and focus on one prominent example, the green lacewing, *Chrysoperla carnea*. Discussing our findings in the context of current molecular studies, we argue firstly that the evidence for adverse effects in non-target organisms is compelling enough that it would merit more research. We further conclude from our in-depth analysis that the published reports studying the effects of Bt toxins from Bt pesticides and transgenic Bt plants on green lacewing larvae provide complementary and not contradictory data. And, finally, we find that the key experiments explaining the mode of action not

only in this particular affected non-target species but also in most other affected non-target species are still missing. Considering the steadily increasing global production area of Bt crops, it seems prudent to thoroughly understand how Bt toxins might affect non-target organisms. (Hilbeck & Schmidt, 2006)

Hilder, V. A. and D. Boulter (1999). "Genetic engineering of crop plants for insect resistance - a critical review." *Crop Protection* **18**(3): 177-191.<http://www.botanischergarten.ch/Bt/Hilder-Insect-Resistance-critical-1999.pdf>

Genetically engineering inherent crop resistance to insect pests offers the potential of a user-friendly, environment-friendly and consumer-friendly method of crop protection to meet the demands of sustainable agriculture in the 21st century. Work to date has concentrated on the introduction of genes for expression of modified *Bacillus thuringiensis* (Bt) toxins. Impressive results on the control of Bt-susceptible pests have been obtained in the laboratory and the field, and the first commercial Bt transgenic crops are now in use. A main alternative approach exploits plant-derived insect control genes. Enhanced resistance to a wide spectrum of pests has been demonstrated in laboratory trials of transgenics expressing various protease inhibitors, lectins, etc, and some promising field trials have been carried out, but the scale of effects produced by plant-derived insect control genes has not been deemed convincing enough to lead to serious attempts at commercialization. Both classes of compounds have limitations: there have been serious failures in resistance to targeted pests in Bt cotton; most plant-derived resistance factors produce chronic rather than acute effects; and many serious pests are simply not susceptible to known resistance factors. We have analysed the characteristics which would be desirable in an ideal transgenic technology: these include being environmentally benign, relatively inexpensive to develop, with a potentially wide spectrum of activity (although targetable at pests and not beneficials), generated by a flexible technology that allows any insect site to be targeted and readily adaptable so that alternatives can be produced as required. We are developing such a technology based on the expression of single-chain antibody genes in crop plants which would be compatible with the likely trends in pesticide discovery using biology-driven target-based methods. The importance of a changed, more socially responsible attitude in this sector is emphasised as is the need for much improved presentation of the benefits and need for responsible deployment of genetically engineered crops. (Hilder & Boulter, 1999)

Hilder, V. A. (2003). "GM Plants and Protection Against Insects - Alternative Strategies Based on Gene Technology" *Acta Agriculturae Scandinavica, B* **53**(Supplement 1): 34-40.<http://www.ingentaconnect.com/content/tandf/sagb/2003/00000053/A00100s1/art00008>
<http://dx.doi.org/10.1080/16519140310015076>

Insect resistant genetically modified (GM) crops may provide a partial substitution technology for chemical pesticides. Those which are available commercially are all based on modified *Bacillus thuringiensis* (Bt) toxins. It is desirable to have a pool of alternative genes that encode insect control proteins (ICPs) having entirely different modes of action. Various classes of compound have been suggested although most have one or more disadvantages compared with Bt toxins. A pool of alternative ICPs would allow pyramiding of resistance genes, which should have a number of advantages: multifactorial resistance is expected to be more durable, to increase the spectrum of pests targeted and to be more effective. Some of these compounds address concerns about durability, spectrum of activity, reliability, environmental impact and public acceptability of GM crops. More speculative alternative approaches which might remove other key constraints on the transgenic approach to crop protection are discussed. The fundamental research necessary for their development is unlikely to be funded from commercial sources (Hilder, 2003)

Howard, J. A. and K. C. Donnelly (2004). "A quantitative safety assessment model for transgenic protein products produced in agricultural crops." *Journal of Agricultural & Environmental Ethics* **17**(6): 545-558.<Go to ISI>://000225838600006 AND <http://www.botanischergarten.ch/Bt/Howard-Quantitative-Protein-2004.pdf>

Transgenic plants are now being used to develop pharmaceutical and industrial products in addition to their use in crop improvement. Using confinement requirements, these transgenic plants are grown and processed under conditions that prevent intermixing with commodity crops. Regulatory agencies in the United States have provided guidance of zero tolerance of these new industrial crops with commodity crops. While this is a worthy goal, it is theoretically unattainable. In spite of the best containment practices, there is a potential risk using any system of production due to unforeseen incidences including natural disasters or exposure to workers. The precautionary principle has been used for numerous regulated articles in addressing the potential risks of new products and technology based on a risk assessment in similar situations. We present here a risk assessment model that could be used as a start to develop an accepted model for the industry. The model is based on current risk models used for other regulated articles, but adapted for these types of products. This could be used to determine action levels in the event of an unintended exposure or to ensure that detection or confinement methods are adequate to avoid risks. As an example, aprotinin, a therapeutic protein now being produced in maize, was evaluated for potential risk to humans using this model. (Howard & Donnelly, 2004)

Jorgensen, S. E., H. C. Luthoft and B. H. Sorensen (1998). "Development of a model for environmental risk assessment of growth promoters." *Ecological Modelling* **107**(1): 63-72.<Go to ISI>://000073406200005

Growth promoters are applied in significant amounts to attain a faster growth of pigs. As a considerable amount of the applied growth promoters is discharged with the manure on agricultural fields, it seems reasonable to assess the environmental risk for the use of growth promoters. It will require development of a model to obtain the predicted environmental concentration (PEC). This paper presents a model which is able to predict the allocation of growth promoters in the seven compartments that are the state variables of the model, namely the amount of growth promoter per square meter in two soil layers on 50 and 200 cm, in the soil water in the same two layers, in surface water, in crops and in ground water. The model could in principle be used in a concrete case of manure applied as natural fertilizer, but as the model is presented it is aimed towards indicating where in the environment the highest concentration of growth promoters should be expected and at what time which can be used to set up an analytical program, and to indicate some 'worst case' concentrations to be able to estimate the risk. The model is applied to simulate concentrations in the environment of two widely applied growth promoters, olaquinox and tylosine. The highest concentrations in crops, fish, surface water and ground water are attained after 20-30 days and the 'worst case' situation may lead to concentrations of several $\mu\text{g/l}$ or $\mu\text{g/kg}$ in fish (about 2 $\mu\text{g/kg}$ for olaquinox and about 6 $\mu\text{g/kg}$ for tylosine), in crops (at harvest about 30 $\mu\text{g/kg}$ for olaquinox and 70 $\mu\text{g/kg}$ for tylosine) and in surface water (a few $\mu\text{g/l}$). The concentrations in ground water were in the order of 30 ng/l for olaquinox and 70 $\mu\text{g/l}$ for tylosine. A sensitivity analysis indicates that it is extremely important to assess the biodegradability. The results clearly indicate that the biodegradability should be estimated if not known from the literature, to obtain a reasonable reliability of model predictions. (Jorgensen et al., 1998)

Lang, A. and E. Vojtech (2006). "The effects of pollen consumption of transgenic Bt maize on the common swallowtail, *Papilio machaon* L. (Lepidoptera, Papilionidae)." *Basic and Applied Ecology* **7**(4): 296-306.<Go to ISI>://WOS:000239199700002 AND <http://www.botanischergarten.ch/Bt/Lang-Effects-Swallowtail-2006.pdf>

Effects of exposure to maize pollen of event Bt176 (cultivar "Navares") on the larvae of the European common swallowtail (*Papilio machaon* L.) were studied in the laboratory. First instar larvae were exposed to different pollen densities applied to leaf disks of *Pastinaca sativa* L. for 48 h. Pollen densities applied in this study were in the range recorded from the field. Larvae which were exposed to higher Bt maize pollen densities consumed more pollen and had a lower survival rate. The LD50 with regard to larvae surviving to adulthood was 13.72 pollen grains consumed by first instar larva. Uptake of Bt maize pollen led to a reduced plant consumption, to a lower body weight, and to a longer development time of larvae. Effects on pupat weight and duration of the pupa[period were present but less pronounced and smaller than effects on larvae. Larvae having consumed Bt-maize pollen as first instars had a lower body weight as adult females and smaller forewings as adult males. We conclude that possible effects of Bt maize on European butterflies and moths must be evaluated more rigorously before Bt maize should be cultivated over large areas. (c) 2005 Gesellschaft für Ökologie. (Lang & Vojtech, 2006)

Lethourneau, D. K., G. S. Robinson and R. A. Hagen (2003). "Bt crops: Predicting effects of escaped transgenes on the fitness of wild plants and their herbivores." *Environmental Biosafety Research* 2: 219–246. DOI: 10.1051/ebr:2003014 AND <http://www.botanischergarten.ch/Bt/Lethourneau-Predicting-Escapes-2003.pdf>

One prominent concern about genetically modified crops is the possibility of environmental impacts from the movement of fitness-enhancing traits to wild plant populations. Decisions to deregulate Bt crops in the USA

have relied strongly on arguments that these crops will not interbreed with wild relatives in the permitted growing regions. Limited attention therefore has been directed to analyses of the consequences of gene flow.

To provide a transparent evaluation process for risks associated with insecticidal transgene escape, we crafted a series of questions designed to guide this aspect of the risk assessment. We then explored the current

knowledge base available for answering such risk-related questions for three Bt crops (cotton, rapeseed, and rice). First, we generated a list of wild relatives of these crops. A definitive list of potential transgene recipients is not yet possible for some crops. Sufficient data are not available for some crops to eliminate certain related plant species from consideration of fertile hybrid formation, thus making lists for these crops subject to speculation. Second, we queried the HOSTS database (UK) to obtain a worldwide listing of lepidopteran species that feed on these crops and their wild relatives, and to determine the host range of the larvae. To our knowledge, this list of 502 lepidopteran species is the first such list published for these crops and wild crop relatives. Third, we used a data set maintained by the Canadian Forest Service to assess Bt toxin susceptibility for these lepidopterans. Only 3% of those species have been tested for susceptibility; and the literature suggests that generalizations about susceptibility among taxa are difficult due to the variability within families. Fourth, we consulted the literature to interpret what is known about the ability of lepidopterans to regulate plant fitness or invasiveness. We could not eliminate the possibility of ecological release due to plant resistance against lepidopterans. In fact, there is strong experimental evidence that lepidopteran herbivores do limit the distribution and/or abundances of at least some wild plant species. Neither could we eliminate the possibility that non-target lepidopterans might have important functions in the ecosystem as pollinators or alternate hosts to natural enemies of pest species. This study suggests that crucial data are lacking for the development of a credible scientific basis to confirm or deny environmental risks associated with the escape of Bt transgene constructs to wild relatives. Given the absence of information on the identity, level of susceptibility, and ecological roles of lepidopterans exploiting specific wild relatives of Bt crops, we suggest that new efforts be directed to assessing possible consequences of lepidopteran mortality on resistant wild relatives. (Lethourneau et al., 2003)

Levidow, L. (2001). "Precautionary uncertainty: Regulating GM crops in Europe." *Social Studies of Science* 31(6): 842-874. <Go to ISI>://000173147500003

Through the precautionary principle, governments acknowledge the limits of science as a basis for policy, while seeking to clarify scientific uncertainty. This tension is exemplified by the European risk regulation of genetically modified (GM) crops. The risk debate has been translated into various precautionary approaches, each with its own cognitive framing of the relevant uncertainties. Early safety claims took for granted intensive agricultural models; normative judgements served to downplay uncertainties which were not readily reducible, thus justifying commercial approval of products. In the late 1990s public protest strengthened broader accounts of uncertainty, for example through more stringent environmental norms and more complex causal pathways of potential harm. Fact-finding methods were debated as a value-laden choice for how best to generate more relevant knowledge. As risk-assessment research challenged assumptions in safety claims, critics cited the results as evidence of greater uncertainty. Invoking the precautionary principle, regulatory procedures delayed or restricted commercial use of GM crops. They not only increased the burden of evidence for safety, but also stimulated and requested knowledge about more complex uncertainties. Criteria for relevant evidence were implicitly linked with different framing visions for agriculture. Such value conflicts made scientific uncertainty more important - rather than vice versa. When risk research methods were challenged, fact/value boundaries

were blurred, thus increasing 'uncertainty' - rather than vice versa. In these ways, the risk controversy was constituted by divergent accounts of the relevant scientific uncertainty. Uncertainty was constitutive, not merely contextual. In general, then, precaution offers a means to justify uncertainty - not simply vice versa. (Levidow, 2001)

Levidow, L. (2003). "Precautionary risk assessment of Bt maize: what uncertainties?" Journal of Invertebrate Pathology **83**(2): 113-117.<Go to ISI>://000183491000007 AND <http://www.botanischergarten.ch/Bt/Levidov-Precautionary-Assessment-2003.pdf>

GM crops have become a test case for the conflicting slogans of 'the precautionary principle' versus 'sound science.' The issues can be illustrated by developments in regulatory science for Bt maize in the European Union. As this case study suggests, risk assessment is always framed by some account of the relevant uncertainties. These in turn depend upon how the environment is valued and how scientific questions are posed about cause-effect pathways of potential harm. The slogan of 'sound science' hides such judgements, by representing ignorance or value-judgements as 'science.' By contrast, precaution can challenge such judgements, identify new unknowns, generate different criteria for evidence, open up new scientific questions, and make these judgements more transparent. It is doubtful whether these complexities have been fully acknowledged by specialists, and thus whether the continued risk debate is due solely to a public misunderstanding of science. (Levidow, 2003)

Lorch, A. and C. Then (2007). How much Bt toxin genetically engineered MON810 maize plants actually produce? Bt concentration in field plants from Germany and Spain. Greenpeace: pp 27. <http://www.botanischergarten.ch/Bt/Lorch-Bt-toxin-MON806-2007.pdf>

In the growing season 2006, Greenpeace took leaf samples of commercially cultivated MON810 maize plants in Germany and Spain to determine the Bt toxin (Cry1Ab) concentration. A total of 619 samples from 12 fields were analysed using ELISA tests. MON810 maize is genetically engineered to produce a modified insecticide (Cry1Ab) that naturally occurs in the soil bacterium *Bacillus thuringiensis* (Bt). The production of this toxin is supposed to protect the maize plants from European corn borer larvae (ECB, *Ostrinia nubilalis*). This Greenpeace study shows a surprising pattern of plants that contained only very low Bt toxin levels. However, high levels could be observed in some plants. The variation found on the same field on the same day was considerable, and could differ by a factor of as much as 100. This is in agreement with the results of a new study published in April 2007¹ that concludes that "the monitoring of Cry1Ab expression [of MON810 plants] showed that the Cry1Ab concentrations varied strongly between different plant individuals. In total, the Bt concentrations were much lower than those available from Monsanto for cultivation approval in the US and the EU, with an arithmetic mean of 9.35 μ g Bt/g fresh weight (fw; standard deviation 1.03; range 7.93-10.34 μ g Bt/g fw). Here, our data also corroborate the results of Nguyen & Jehle (2007), who also found lower Bt concentrations (with means between 2.4 and 6.4 μ g Bt/g fw) than those known from the literature. The data recorded by Greenpeace, however, deviate even more from the data published so far. The means ranged from 0.5 to 2.2 μ g Bt/g fw, while Bt concentrations ranged from a minimum of no or 0.1 μ g Bt/g fw to concentrations of about 14.8 μ g Bt/g fw. The results presented here raise far-reaching questions about the safety and the technical quality of the MON810 plants as well as some fundamental methodological questions. (Lorch & Then, 2007)

Michaud, D. (2005). "Environmental impact of transgenic crops. II. Impact of recombinant traits." Phytoprotection **86**(2): 107-124.<Go to ISI>://WOS:000236225700003 AND <http://www.botanischergarten.ch/Bt/Michaud-Environmental-Impact-2005.pdf> AND <http://www.botanischergarten.ch/Bt/Michaud-Impact-Proteines-2005.pdf>

A scientific communication reporting the deleterious effects on monarch butterfly larvae of a transgenic corn hybrid expressing a *Bacillus thuringiensis* delta-endotoxin has caused, a few years ago, an unprecedented controversy on the environmental impact of recombinant traits introduced into the genome of agricultural crops. This review, complementing a review in this same issue on transgene migration in the environment (Michaud 2005), addresses the impact of these new traits on the development and survival of different non-target living organisms present in the environment. The impact of these new traits

is first considered at the ecosystem level, in relation with the effects of current agricultural practices on field biodiversity. The impact of these traits is then considered in relation with the specific interactions established in the field or under laboratory conditions between the modified plant and a collection of model organisms including secondary herbivorous pests, predatory arthropods and different species of the soil community. (Michaud, 2005)

Pilson, D. and H. R. Prendeville (2004). "Ecological effects of transgenic crops and the escape of transgenes into wild populations." *Annual Review of Ecology Evolution and Systematics* 35: 149-174.<Go to ISI>://000226244100006

Ecological risks associated with the release of transgenic crops include nontarget effects of the crop and the escape of transgenes into wild populations. Nontarget effects can be of two sorts: (a) unintended negative effects on species that do not reduce yield and (b) greater persistence of the crop in feral populations. Conventional agricultural methods, such as herbicide and pesticide application, have large and well-documented nontarget effects. To the extent that transgenes have more specific target effects, transgenic crops may have fewer nontarget effects. The escape of transgenes into wild populations, via hybridization and introgression, could lead to increased weediness or to the invasion of new habitats by the wild population. In addition, native species with which the wild plant interacts (including herbivores, pathogens, and other plant species in the community) could be negatively affected by "transgenic-wild" plants. Conventional crop alleles have facilitated the evolution of increased weediness in several wild populations. Thus, some transgenes that allow plants to tolerate biotic and abiotic stress (e.g., insect resistance, drought tolerance) could have similar effects. (Pilson & Prendeville, 2004)

Shirai, Y. (2007). "Nontarget effect of transgenic insecticidal crops: Overview to date and future challenges, (in Japanese)." *Japanese Journal of Applied Entomology and Zoology* 51(3): 165-186.<Go to ISI>://000249835500001 AND <http://www.botanischergarten.ch/Bt/Shirai-Nontarget-Effect-2007.pdf>

In the mid-1990s, commercial cultivation of transgenic insecticidal crops (Bt corn, cotton and potato) began in North America. In 1998 and 1999, some researchers warned that these Bt crops may have unexpected negative effects on nontarget butterflies, beneficial natural enemy insects, and soil fauna. Since then, many peer-reviewed articles have been published about the nontarget effects of transgenic insecticidal crops. Most subsequent studies revealed that Bt corn pollen has no harmful effect on nontarget butterflies in the field. Negative effects on predatory or parasitic insects shown in laboratory experiments have never been demonstrated in greenhouse or field studies. Although assessing the effect on soil fauna is difficult compared with on nontarget butterflies or above-ground natural enemies, none of the reports have documented deleterious effects on soil biota. The current commercially used Bt crops appear to have little significant adverse effect on nontarget fauna. Before the approval of commercial field cultivation, many ecological risk assessments are imposed on new types of transgenic crops (new trait event) and the nontarget effect is an essential part of the risk assessment for transgenic insecticidal crops. Many further articles on the nontarget effect will be published for transgenic insecticidal crops including the current Bt crops. (Shirai, 2007)

Conclusion: There is not a single paper cited above under 2.1.5 which could come up with a factual basis for an overall negative judgement on Bt crops. But it has to be said in all fairness that there are some papers in the above enumeration which describe general and highly interesting phenomena of Bt interactions and do not pretend to make negative statements. Its time to concentrate on the plethora of positive evidence, gathered in truly comparative field experiments with a solid background of experimental structure and statistics.

Cited Literature

Andow, D. & Hilbeck, A. (2004)

Science Based Risk Assessment for Nontarget Effects of Transgenic Crops. *Bioscience*, 54, 7, pp 637-649
<http://www.botanischergarten.ch/Bt/Andow-Hilbeck-Biosciences2004.pdf>

Clark, E.A. & Lehman, H. (2001)

Assessment of GM crops in commercial agriculture. *Journal of Agricultural & Environmental Ethics*, 14, 1, pp 3-28
 <Go to ISI>://000167926400001 AND <http://www.botanischergarten.ch/Bt/Clark-Assessment-commercial-2001.pdf>

Craig, W., Tepfer, M., Degrassi, G., & Ripandelli, D. (2008)

An overview of general features of risk assessments of genetically modified crops. *Euphytica*, online first, January 11, pp
<http://dx.doi.org/10.1007/s10681-007-9643-8> AND <http://www.botanischergarten.ch/Bt/Craig-Overview-Risk-2008.pdf>

Cruywagen, G.C., Kareiva, P., Lewis, M.A., & Murray, J.D. (1996)

Competition in a spatially heterogeneous environment: Modelling the risk of spread of a genetically engineered population. *Theoretical Population Biology*, 49, 1, pp 1-38
 <Go to ISI>://A1996UB46400001

Daniell, H. (1999)

Environmentally friendly approaches to genetic engineering. *In Vitro Cellular & Developmental Biology-Plant*, 35, 5, pp 361-368
 <Go to ISI>://000083672000002

Dolezel, M., Heissenberger, A., & Gaugitsch, H. (2006)

Ecological effects of genetically modified maize with insect resistance and/or herbicide tolerance. Literature research report on behalf of the Bundesministerium für Gesundheit und Frauen, Wien, Austria., pp. 70 pages. Bundesministerium für Gesundheit und Frauen., Wien, Austria. Forschungsberichte der Sektion IV, Band 6, 2005
http://www.bmgfj.gv.at/cms/site/attachments/5/6/2/CH0255/CMS1134457515326/literaturstudie_mais_endbericht.pdf

Donkin, S.S., Velez, J.C., Totten, A.K., Stanisiewski, E.P., & Hartnell, G.F. (2003)

Effects of feeding silage and grain from glyphosate-tolerant or insect-protected corn hybrids on feed intake, ruminal digestion, and milk production in dairy cattle. *Journal of Dairy Science*, 86, 5, pp 1780-1788
 <Go to ISI>://000182985600026

Driehuis, F. & Elferink, S. (2000)

The impact of the quality of silage on animal health and food safety: A review. *Veterinary Quarterly*, 22, 4, pp 212-216
 <Go to ISI>://000165306200009

Felke, M. & Langenbruch, G.A. (2003)

Wirkung von Bt-Mais-Pollen auf Raupen des Tagpfauenauges im Laborversuch. Effect of Bt-maize-pollen on caterpillars of *Inachis Io* in a laboratory assay. *Gesunde Pflanzen*, 55. Jahrgang, 1, pp 1-7
<http://www.botanischergarten.ch/Bt/Felke-Bt-Gesunde-Pfl.pdf>

Felke, M. & Langenbruch, G.A. (2005)

Electronic Source: Auswirkungen des Pollens von transgenem Bt-Mais auf ausgewählte Schmetterlingslarven (ed V.G. Dr. Mathias Otto; Fachgebiet II 2.3 „Bewertung gentechnisch veränderter Organismen), BfN-Skripten
 published by: Bundesamt für Naturschutz, Konstantinstr. 110, 53179 Bonn
www.bfn.de AND <http://www.botanischergarten.ch/Bt/Felke-Langenbruch-Bt-Mais-BfN-2005.pdf>

Felke, M., Lorenz, N., & Langenbruch, G.A. (2002)

Laboratory studies on the effects of pollen from Bt-maize on larvae of some butterfly species. *Journal of Applied Entomology-Zeitschrift Fur Angewandte Entomologie*, 126, 6, pp 320-325
 <Go to ISI>://000177281800010 AND <http://www.botanischergarten.ch/Bt/Felke-Langenbruch-Bt-Mais-Journal-2002.pdf>

Giovannetti, M. (2003)

The ecological risks of transgenic plants. *Rivista Di Biologia-Biology Forum*, 96, 2, pp 207-223
 <Go to ISI>://000186191700003 BEI NEBIS NOCH BESTELLEN

Groot, A.T., Dicke, M. (2002)

Insect-resistant transgenic plants in a multi-trophic context. *Plant Journal*, 31, 4, pp 387-406
<http://www.botanischergarten.ch/Bt/Groot-Multi-trophic-2002.pdf>

Hilbeck, A. (2001)

Implications of transgenic, insecticidal plants for insect and plant biodiversity. *Perspectives in Plant Ecology Evolution and Systematics*, 4, 1, pp 43-61
<http://www.botanischergarten.ch/Bt/Hilbeck-Implications-2001.pdf>

Hilbeck, A. & Schmidt, J.E.U. (2006)

Another view on Bt Proteins – How Specific are They and What Else Might They Do? *Biopesticides International*, 2, 1, pp 1-50
<http://www.botanischergarten.ch/Bt/Hilbeck-another-View-2006a.pdf>

Hilder, V.A. (2003)

GM Plants and Protection Against Insects - Alternative Strategies Based on Gene Technology Acta *Agriculturae Scandinavica*, B, 53, Supplement 1, pp 34-40
<http://www.ingentaconnect.com/content/tandf/sagb/2003/00000053/A00100s1/art00008>
<http://dx.doi.org/10.1080/16519140310015076>

Hilder, V.A. & Boulter, D. (1999)

Genetic engineering of crop plants for insect resistance - a critical review. *Crop Protection*, 18, 3, pp 177-191
<http://www.botanischergarten.ch/Bt/Hilder-Insect-Resistance-critical-1999.pdf>

Howard, J.A. & Donnelly, K.C. (2004)

A quantitative safety assessment model for transgenic protein products produced in agricultural crops. *Journal of Agricultural & Environmental Ethics*, 17, 6, pp 545-558
 <Go to ISI>://000225838600006 AND <http://www.botanischergarten.ch/Bt/Howard-Quantitative-Protein-2004.pdf>

Jorgensen, S.E., Luthoft, H.C., & Sorensen, B.H. (1998)

Development of a model for environmental risk assessment of growth promoters. *Ecological Modelling*, 107, 1, pp 63-72
 <Go to ISI>://000073406200005

Lang, A. & Vojtech, E. (2006)

The effects of pollen consumption of transgenic Bt maize on the common swallowtail, *Papilio machaon* L. (Lepidoptera, Papilionidae). *Basic and Applied Ecology*, 7, 4, pp 296-306
 <Go to ISI>://WOS:000239199700002 AND <http://www.botanischergarten.ch/Bt/Lang-Effects-Swallowtail-2006.pdf>

Lethourneau, D.K., Robinson, G.S., & Hagen, R.A. (2003)

Bt crops: Predicting effects of escaped transgenes on the fitness of wild plants and their herbivores. *Environmental Biosafety Research*, 2, pp 219-246
 DOI: 10.1051/embr:2003014 AND <http://www.botanischergarten.ch/BtLethourneau-Predicting-Escapes-2003.pdf>

Levidow, L. (2001)

Precautionary uncertainty: Regulating GM crops in Europe. *Social Studies of Science*, 31, 6, pp 842-874
 <Go to ISI>://000173147500003

Levidow, L. (2003)

Precautionary risk assessment of Bt maize: what uncertainties? *Journal of Invertebrate Pathology*, 83, 2, pp 113-117

<Go to ISI>://000183491000007 AND <http://www.botanischergarten.ch/Bt/Levidov-Precautionary-Assessment-2003.pdf>

Longley, M. & Sotherton, N.W. (1997)

Factors determining the effects of pesticides upon butterflies inhabiting arable farmland. Agriculture Ecosystems & Environment, 61, 1, pp 1-12

<Go to ISI>://A1997WN51300001 AND <http://www.botanischergarten.ch/Bt/Longley-Factors-Pesticides-Butterflies-1997.pdf>

Lorch, A. & Then, C. (2007)

How much Bt toxin genetically engineered MON810 maize plants actually produce? Bt concentration in field plants from Germany and Spain pp 27 (Report)

<http://www.botanischergarten.ch/Bt/Lorch-Bt-toxin-MON806-2007.pdf>

Marvier, M., McCreedy, C., Regetz, J., & Kareiva, P. (2007)

A Meta-Analysis of Effects of Bt Cotton and Maize on Nontarget Invertebrates. Science %R 10.1126/science.1139208, 316, 5830, pp 1475-1477

<http://www.sciencemag.org/cgi/content/abstract/316/5830/1475> AND

<http://www.botanischergarten.ch/Bt/Marvier-Meta-Analysis-2007.pdf> AND supporting data:

<http://www.botanischergarten.ch/Bt/Marvier-Meta-Analysis-Supporting-2007.pdf>

Michaud, D. (2005)

Environmental impact of transgenic crops. II. Impact of recombinant traits. Phytoprotection, 86, 2, pp 107-124

<Go to ISI>://WOS:000236225700003 AND <http://www.botanischergarten.ch/Bt/Michaud-Environmental-Impact-2005.pdf> AND <http://www.botanischergarten.ch/Bt/Michaud-Impact-Proteines-2005.pdf>

Pilson, D. & Prendeville, H.R. (2004)

Ecological effects of transgenic crops and the escape of transgenes into wild populations. Annual Review of Ecology Evolution and Systematics, 35, pp 149-174

<Go to ISI>://000226244100006

Robinson, R.A. & Sutherland, W.J. (2002)

Post-war changes in arable farming and biodiversity in Great Britain. Journal of Applied Ecology, 39, 1, pp 157-176

<http://www.botanischergarten.ch/Bt/Robinson-Post-war-Changes-Biodiversity-2002.pdf>

Shirai, Y. (2007)

Nontarget effect of transgenic insecticidal crops: Overview to date and future challenges, (in Japanese).

Japanese Journal of Applied Entomology and Zoology, 51, 3, pp 165-186

<Go to ISI>://000249835500001 AND <http://www.botanischergarten.ch/Bt/Shirai-Nontarget-Effect-2007.pdf>