GM ON TRIAL

Scientific evidence presented in the defence of 28 Greenpeace volunteers on trial for their non-violent removal of a GM maize crop

Published by Greenpeace Canonbury Villas London N1 2PN Edited by Michelle Allsopp and Doug Parr Designed by bwa Printed on recycled paper

CONTENTS

INTRODUCTION,	
Dr Douglas Parr, Chief Scientific Adviser, Greenpeace UK	
WIND POLLINATION	5
Professor Jean Emberlin	
BEE POLLINATION	13
Geoffrey Hopkinson	
RISKS TO ORGANIC FARMING	19
Richard Young	
THE WEAKNESS OF THE CONCEPT SUBSTANTIAL EQUIVALENCE	31
Dr. Erik Millstone	
HAZARDS TO FOOD AND ANIMAL FEED	37
Dr. C. V. Howard MB. ChB. PhD. FRCPath.	
HORIZONTAL GENE TRANSFER	45
Professor Terje Traavik	
IMPACTS ON THE SOIL	63
Dr. Max A. Turner and Dr. A. Neil Macgregor	
IMPLICATIONS FOR PESTICIDE USE	69
Peter Beaumont	
FARM SCALE TRIALS AND ENVIRONMENTAL SAFETY	77
Dr. Susan Mayer	
SOURCES	85

INTRODUCTION

Dr Douglas Parr, Chief Scientific Adviser Greenpeace UK

On 26 July 1999, 28 Greenpeace volunteers were arrested at Lyng, Norfolk, for the partial cutting down and removal of a field of genetically modified maize (GM) owned by AgrEvo. Their subsequent arrest and trial for criminal damage and theft is the context for this book. The Greenpeace volunteers were charged with criminal damage and the case came to court in Norwich, beginning on 3 April 2000.

The trigger for the peaceful direct action that those people took was the imminent flowering of the crop and the dispersal of the maize pollen. Shortly before their action it had become clear that the Swiss authorities had banned the release of the same genetically modified maize (known as T25 maize) because of concerns over cross-contamination and the spread of the GM construct.

The GM maize crop was part of the programme of trials of genetically modified herbicide tolerant crops organised by the Department of Environment, Transport and the Regions in the UK. The trials were designed to give limited answers to a range of questions about the impact on wildlife resulting from the changes in agricultural management that herbicide tolerance may bring. Greenpeace has opposed these trials because of their limited nature - a whole range of political, scientific and ethical issues remain unresolved about the development of GM technology. The farm scale trials programme can, at best, address a very small part of them.



Other questions that remain, and which in part drove many people to reject GM food at the supermarkets include (ESRC, 1999):

- Is this necessary?
- Are there better ways to achieve the same ends?
- · Have we taken everything relevant into account?
- Are we being sufficiently cautious about the uncertainties?
- Can we withdraw from it if it doesn't work out as we hope?

Greenpeace criticisms of the Farm scale trial programme specifically are concerned with what it will not consider, including:

- whether the GM crops are safe for humans and farm animals to eat
- the environmental impact of GM crops in comparison with sustainable farming systems such as organic
- the range of species that the herbicides Liberty and RoundUp are known to be toxic to
- the effect of growing one GM crop such as maize in the same field year after year
- the effect of growing a series of different GM crops in rotation
- the environmental impacts of the interaction of these GM crops with any other GM traits
- · the social and economic impact of GM agriculture

Further, there remains a widespread unease about the nature of genetic modification which it would be foolish to dismiss as being the natural conservatism that people exhibit when confronted with novel innovations and technologies. The technique of genetic modification involves re-evaluating our relationship with nature in a more fundamental way than any previous innovation. To dismiss these value-based concerns that are widely held is patronising and unhelpful.

The direct action by the Greenpeace volunteers and their defence of 'lawful excuse' – that they were entitled to take the action that they did because they believed that serious harm to the environment was imminent – meant that expert scientific judgement needed to be assembled to show that the defendants held reasonable beliefs in acting in the way they did. It is not the purpose of this book to rehearse the legal arguments on which the case turns. But the expert scientific statements that were gathered in the course of preparing the case, and that were submitted to the court during the preparation of the defence, seemed to me to be of genuine wider interest, and so Greenpeace has decided to reproduce them in this book. Early in the progress of the trial it became clear that the prosecution felt either unwilling or unable to contest the reasonable nature of the defendants beliefs – that the harm resulting from the dispersal of the GM maize pollen and plant material could be serious and demanded immediate intervention. So in the event none of this material was presented in court.

These statements are not the definitive analysis of scientific certainties, because such statements cannot, neither at this time nor for the foreseeable future, exist. Nor is this in any way a comprehensive review of the scientific case 'against GM crops'. For legal reasons, these statements focus on what was generally known and in the public domain in July 1999. More has come into the public domain since then. Further, there are a whole range of issues that have not been addressed by these expert statements because of their focus on the T25 maize crop that is at the centre of the legal case.

Other crops throw up different issues. In the UK, the use of sugar or fodder beet, or of oil seed rape would raise the question of gene movement into the wild plant community, of agricultural volunteers and feral plant populations, of the ethics of 'genetic pollution' of natural plant species and the ability of ecology to have predictive power. Applications other than herbicide tolerance (e.g. pest resistance, virus resistance) raise other issues about the nature of our agricultural systems and other potential risks.

These scientific statements barely touch on the considerable capacity that scientific experimentation has for unexpected outcomes in the course of experimentation – which has a whole different connotation when experiments are being done in the wider environment and not in the laboratory. And of course none of them look at the deeply political issues of sustainable world food security, consolidation in the global agricultural supply chain, the limit on free flow of information because of intellectual property rights regimes, and the aims and orientation of both private and publicly funded research agendas; which scientific research is actually being carried out (UNESCO 1999)?

The definitive treatise on all the issues that genetic modification raises remains, if ever, to be written. However, the statements gathered together here for this book do represent a body of scientific statements demonstrating that the debate in media terms is often miscast as one of 'science' versus 'emotion'. This is nonsense, as the remainder of this book demonstrates.

The statements contained here are as follows:

Jean Emberlin reports on the dispersal of maize pollen by wind, and how far such pollen is known to travel, including exceptional events which may take the pollen over 100 kilometres whilst still viable.

Geoff Hopkinson describes how bees in particular can act as a carrier for maize pollen over several kilometres and on the worry for beekeepers of contamination of pollen and pollencontaining products (including honey).

Richard Young explains the concerns about cross-contamination of organic crops from pollen movement and a variety of other routes, and why the Soil Association chose to have a standard of zero contamination for GM material in organic crops.



Erik Millstone reports on the weaknesses in the concept of 'substantial equivalence' which underpins much of the food safety assessment process in regulatory regimes around the world, and how the concept may actually be inhibiting research into the real effects of genetic modification on food.

Vyvyan Howard identifies some of the hazards that arise from genetically modified food such as chronic low-dose toxicity and the difficulty of doing an assessment of risk compared to the identification of the hazard.

Terje Traavik reports both on the unpredictability of genetic modification; and the frequency of and potential hazards from the phenomenon of horizontal gene transfer, where organisms, particularly microbes, are able to pick up and utilise DNA from their surroundings or from other organisms.

Max Turner and Neil McGregor identify the risks to soil functioning that genetic modification brings through genetic alteration, potential effects on soil nutrient cycling and productivity, and changed usage of agrochemicals.

Peter Beaumont reports on the potential for changes in herbicide use as a result of genetic modification to produce herbicide tolerant crops, and points out weaknesses in the regulatory framework related to GM herbicide tolerant crops.

Sue Mayer points out the limitations of the farm-scale trials programme; what it will be able to achieve and the questions that will remain unanswered however successful the proposed research agenda, and the capacity of scientific experimentation for unexpected outcomes.

A further statement was submitted to the court by Peter Lund but because he felt it needed more work before dissemination it has been omitted – the area of unpredictability that was covered in that statement is partially addressed by Terje Traavik.

I would like to thank all these people for their willingness to give of their time in preparation of the Greenpeace case. In preparation of the book I must also thank Michelle Allsopp for her editing skills and John Sauven for dealing with the production.

In conclusion, these statements represent a body of information about the hazards of genetic modification which show that those who promote these crops and foods have a case to answer. Those who have only heard the soundbite debate over the media should be aware that there is a substantial scientific case that that lies behind those who are critical of these 'advances'. In the light of the naïve way that the advantages (if any) of GM crops are propounded, surely one should question whether they should even go forward at all.

WIND POLLINATION

Professor Jean Emberlin

PERSONAL EXPERTISE

Lam director of the National Pollen Research Unit, President of the British Aerobiology Federation, a director of European Pollen Information and a council member of the International Association for Aerobiology. My experience includes over 12 years work in airborne pollen dispersal and forecasting pollen concentrations in relation to weather factors. I have produced a report on maize pollen dispersal for the Soil Association (Jan. 1999) which is directly relevant to this case and have also monitored dispersal of oil seed rape pollen from a GM trial site. My doctorate thesis was on the long range dispersal of air pollutants over complex topography. This background provides useful knowledge about the dispersal of pollen grains in the atmosphere.

I have published numerous research papers on aspects of Aerobiology and present my work frequently at International conferences in the fields of Aerobiology and Allergy.

QUESTION POSED TO WITNESS

I have been asked to advise on what was in the public domain and available in July 1999 on the subject of the distances that pollen from GM crops could travel by wind and hence potentially cause contamination.



SUMMARY

This reports examines the potential risk of contamination by pollen from a genetically modified maize crop (2.8 hectares) at Walnut Tree Farm, Lyng, Norfolk. The crop was expected to start flowering about 7-10 days after the time of the action.

- > Potential aspects of contamination from the GM pollen include cross-pollination with other maize, collection by bees and subsequent presence in honey, and deposition on surfaces, such as tree leaves where insects may forage. The crop is not inter-fertile with any UK wild or crop relatives. The percentage of cross breeding with other maize crops in the near vicinity (e.g. up to 800m) will depend on factors such as separation distance, local barriers to pollen movement, such as woods and hedges, and for crops at moderate to long distances (e.g. > 800m) downwind it will depend more on weather and topography.
- > Organic farms are situated 7.2, 10.4 and 11.2 km from the crop and nine or ten conventional farms with maize crops are within 3 km. It is not possible to quantify the risk of contamination to sites at distances of between 7 to 11 km from the GM crop because insufficient research has been done on medium to long range dispersal to predict with reasonable confidence the actual concentrations likely to be found at downwind distances over 500m. Also the dispersal patterns depend a lot on the weather conditions prevailing at the time of pollen release. However it is feasible to provide a general indication of potential maize pollen spread from the location.
- > Evidence from previous work can be used as a basis for generalised estimates of percentages of maize pollen concentrations remaining airborne downwind in low to moderate wind speeds compared with concentrations at 1m from the source. These are approximately 2% at 60m, 1.1% at 200m and between 0.75% and 0.5% at 500m. The implications of these figures for potential cross-pollination are considered in this report but it is emphasised that they should be used as rough guidelines only. In addition it should be noted that dispersal gradients would be altered by factors such as climatic conditions and local topography. Transport on the airflow over longer distances is likely to occur under a range of weather situations including uplift and horizontal movement in convection cells, and uplift and transport in frontal storms. The maize pollen grains remain viable for about 24 hours in normal weather conditions so pollination could occur at sites remote from the source (e.g. 180 km). Dispersal away from the vicinity of the crop also takes place by carriage on bees. Maize pollen is collected by bees in notable amounts. In this way the pollen is transported several miles from the crop plot in suitable weather conditions.
- > It is pertinent to note that the problems are not limited to maize pollen. Airborne oil seed

Potential aspects of contamination from the GM pollen include cross-pollination with other maize, collection by bees ... and deposition on surfaces such as tree leaves where insects may forage

rape pollen sampled downwind from a GM oil seed rape plot was found at distances ranging from 35m to 475m from the edge of the crop (*National Pollen Research Unit*, 1999).

STATEMENT OF THE WITNESS

Maize pollen features

Maize is a highly variable, naturally cross-pollinated crop, in which all forms hybridise freely (*Purseglove, 1972*). It is generally pollinated by wind and gravity and is also visited by bees (*e.g. Percival, 1950*). Maize pollen is amongst the largest of that of the grass (*Gramineae*) family with dimensions in the region of 90 to 125 x 85 microns (*Erdtman, 1952; Smith, 1990*) a volume of about 700 x 10⁻⁹cm³ and a weight of about 247 x 10⁻⁹g (*Goss, 1968*). Published data for the length of time that maize pollen remains viable under natural conditions differs from about 24 hours through to several days. Pollen is shed over a period of 2-14 days, with most plants typically taking 5-8 days, with maximum shedding about the third day (*Purseglove, 1972*). There is usually an overlap of pollen shedding and silk emergence on the same plant but under normal field conditions at least 95% of the ovules are fertilised by pollen from other plants. Maize pollen is produced in enormous quantities. Estimates for the numbers of pollen grains produced by an average-sized plant range from 14 million to about 50 million to fertilise approximately 1000 kernels per plant (*Evans, 1975*) so that there are 20,000 to 30,000 pollen grains for each silk (*Purseglove, 1972*). Maize is typically cultivated at c.20,000 plants per acre giving a pollen output of approximately 154 pounds/70kg per acre.

Dispersal downwind of pollen from maize crop

There is general agreement that the typical downwind dispersal pattern of maize pollen by the airflow in low to moderate wind speeds results in a relatively steep deposition gradient due to the large size of the pollen and its rapid settling rate. Published results of measurements of the deposition gradients differ considerably. One point of comparison is the measure of the half distance, that is, the distance over which the concentration decreases by half as the distance from the source increases by a constant increment (the half distance). Some studies report short half distances, for example, 8.25m, whereas others, e.g. Jones and Newell (1948) report data showing a half distance of 47.47m indicating a potential for cross-pollination over several



hundred metres. Using data from Jones and Newell (1948) the pollen concentration at 500m is estimated to be 0.05% of that at 1m from the crop. McCartney (1994) also uses the maize pollen half distance of 47m in a negative exponential relationship with similar results.

Most estimates of maize pollen dispersal have been derived from deposition rates yet these do not always reflect airborne concentrations due to distortion resulting from dilution and diffusion within the airflow. Factors such as frictional turbulence and thermal convection, which can cause steep deposition gradients, can transport large numbers of pollen grains to great heights where they can disperse over long distances. Raynor, Ogden and Hayes (1972) found that at 60m from the source the total amount of maize pollen remaining airborne was about 5% of that at 1m from the source, and that the deposition per unit area at this distance downwind was only 0.2% of that near the source. In these trials, attenuation of airborne maize pollen grains between the source and 60m was 50:1 whereas attenuation of deposition was 2500:1. This much heavier deposition near to the source corresponds to the grains that never became effectively airborne. This point is relevant when considering data based on deposition readings. Taking concentrations at 1m as 100% the mean results of 15 samples indicated concentrations at 54.9m to be 1.3% (SD 3.30) and at 59.5m to be 1.1% (SD 2.1).

In contrast, the results from Jones and Newell (1948) show approx. 1% concentrations remaining at 427m and those from Jones and Brooks (1950) approximately 0.75% at 503m. It is likely that these shallower slopes of the deposition curves result from greater source areas and the greater wind speed occurring during their work. Faster winds would result in dispersal over larger distances but they would also cause more depletion by impaction enhanced (McCartney, 1990). In most of the data on maize pollen dispersal from empirical studies, the loss in air concentrations compared with deposition are uniformly high indicating an over estimate of deposition or an underestimate of air concentrations (Monteith, 1975).

More reliable information on potential spread of pollen comes from observations of outcrossing. For example Jones and Brooks (1950) researched the effectiveness of distance and border rows in preventing outcrossing in corn. In one of three years the outcrossing exceeded the maximum mixture permitted by international standards at distances of 300m isolation. At a distance of 400m from the contaminating field the mean percent outcrossing was 0.42, and at 500m the mean percent outcrossing was 0.32. Other cited levels of cross breeding between maize at various distances include Jones and Newell (1948) 7.2% at 250m, Jones and Brooks (1950) 2.47% at 200m and Salamov (1940) 0.21% at 805m. It is interesting to note that at 500m in the study by Jones and Brookes, and at 800m in the study by Salnov, give a broadly similar level of outcrossing is given despite the latter study not benefiting from the prevailing wind. This corresponds to the typical leptokurtic distribution and may indicate that a low 'background' level of pollen may occur around a field throughout the time of pollen release. Jones and Newell

(1948) state that these relatively low percentages of total pollen caught at 300m represent considerable numbers of pollen grains and must be considered omnipresent sources of contamination in field production.

Based on consideration of the limited evidence available the generalised estimate figures for maize pollen concentrations from the donor plot at downwind distances under dry weather conditions with low to moderate wind speed are as follows, (expressed as percentages of concentrations at 1m from the source, Qd):

- 60m from the crop edge would be approximately 2%
- 200m downwind from the source would be approximately 1.1%
- 500m from the source would be approximately 0.75% to 0.5%

These relatively small percentages could still result in considerable concentrations in the receptor plot due to the large amounts of pollen released from maize. If it assumed that there was no competing pollen being released from within the receptor plot, these amounts could result in high rates of cross-pollination. For example, a conservative estimate of pollen production per plant is in the order of 25 million grains. This could result in the following approximate amounts of pollen per plant in the donor plot being available for pollination in the receptor plot over the duration of pollen release. Obviously the amounts would not be consistent for all plants in the plot as there would be some deposition and filtering within the stand. In order to arrive at a realistic estimate of the amount of cross-pollination likely to occur with these percentages and amounts, various aspects need to be considered such as synchronisation of maturation of the flowers (both male and female parts), relative concentration strengths of the pollen produced by the donor plot and the receptor plot at the point of pollination, (there may be an overlap in pollen production periods in the two stands so there may be competition for pollination between pollen from the two sources) and the amount of self or cross sterility in the variety.

If it is assumed that there is an overlap in pollen release between the two plots of maize there will be competition for pollination. The donor plot source strength (Qd) will need to be considered in relation to receptor plot source strength (Qr). If flowering is synchronous and both stands produce equal amounts of pollen then the relative concentrations of pollen from the donor plot and the receptor plot can be considered, for example, at 60m this will be $Qd \times 2\%$: Qr giving a qualification to the probability of pollination. For instance, taking the figures given in the bullet points above, the Qd component in the prevailing pollen concentration at the edge of a receptor plot at 60m would be 2%, at 200m would be 1.08% and 500m would be 0.74 to 0.49%. At these concentration ratios the rates of cross-pollination at 500m would be in the order of 1 kernel per 135 to 204. These estimates should be considered as rough



guidelines only as they would be altered by factors such as climatic conditions which effect the transport of pollen, and the numbers of bees and other insects around, which would be likely to increase the amounts of pollen transfer.

Longer range transport is poorly understood but the evidence presented in the following section indicates that viable pollen from the GM site could travel to at least 12 km distant under suitable weather conditions and could pollinate maize grown on organic and conventional farms in the area.

Potential long range dispersal

In certain weather conditions, particles including pollen grains, can travel long distances on the airflow. Long-distance dispersal of maize pollen needs to be considered within the constraint of its viability time (in the region of 24 hours under normal weather conditions, Purseglove, 1972).

Vertical transport of pollens takes place by several mechanisms. On warm days with low wind speeds convective currents driven by heating of the ground by the sun lead to mixing through the boundary layer. This activity has a marked diurnal influence with particles being dispersed laterally through convection cells during the day, and descending when the convection subsides (*Oke, 1978*).

Most anemophilous pollen will be liberated during day time in dry, warm weather. Days like this usually have thermals rising turbulently that will have a positive effect in bringing pollen grains up into the higher strata. The upper limit for convective ascent is marked by the thermal inversion, often shown by the presence of cumulus clouds. Convective cells are typically 1-3 km in diameter, reaching some 1-2,000m in height and last about 20 to 30 minutes each, during which time they can move downwind. The individual cells may form composite cells 5-10 km across and last for several hours. Upward velocity of cell tops reaches 0.5-1.5m/s and horizontal expansion of 0.5-1.0m/s (Hardy and Ottersten, 1968). Some pollen grains will have reached the inversion layer when the bubble collapses. It may then be transported horizontally considerable distances depending on weather conditions. During the evening and night time, convection will cease and the pollen will tend to fall towards the ground but this may be impeded by low level inversions. The usual length of time available for pollen to travel as it is kept aloft by convection is a maximum of one day. This would be equivalent to a distance of about 50-180 km, although it is well known that much longer transports do occasionally take place (Faegri & Iversen, 1989) when suitable meteorological conditions occur.

On days with less solar heating and higher wind speeds, pollen can be dispersed vertically by turbulence generated either by instability in the lapse rate or by rough surfaces such as uneven topography. Biological particles introduced into the boundary layer have been observed in detectable concentrations to distances of several hundred kilometres downwind

(e.g. Hjelmroos, 1991). Penetrative transport to great heights can also take place through updraughts generated by deep intense convective storms. In such storms large masses of air, originally lying near the surface, are transported in a few tens of minutes to heights typically of the order of 8 to 12 km. At such heights, in the middle latitudes, winds are often very strong, in the range 25 to 50m/s, so that pollen can travel great distances in a matter of hours (Mandrioli et al., 1984).

Hirst et al. (1967) sampled air for pollen and spores over the North Sea. Their results include a case in which a pollen cloud generated over Britain could later be found as a pollen concentration cloud over the North Sea. Pollen released during one day was found the following day 300-400 km off the coast. Transport took place over the sea where dispersal conditions could be different from those over land depending on the weather. For example, pollen transport over land could be enhanced by increased convection but conversely the concentrations could be depleted by more deposition due to turbulence. Tyldesley (1973) found appreciable quantities of arboreal pollen (up to 30 per m³) in the air in the treeless Shetlands, 250-380 km away from the nearest forests, in connection with favourable meteorological conditions, i.e. cyclonic storms.

A frontal storm can lift air masses several kilometres up in the air in a very short time and thus place pollen grains far above the day and night cycle (Faegri & Iversen, 1989). Once pollen has arrived in the upper atmosphere it can travel for many hundreds of kilometres on the airflow until finally being deposited or it may be captured by water drops and return to the surface in precipitation (Mandrioli, 1984). In general long range transport occurs most efficiently in dry conditions with limited mixing depth and moderate to high wind speeds.

With mean horizontal wind speeds of 2m/s, that can occur on summer days, and with convection currents that could keep the pollen grains aloft, maize pollen could travel 1 km in 4.16 minutes, or 7.2 km in an hour (potentially 172.8 km in a day). In wind speeds of 10m/s some pollen grains would travel greater downwind distances before deposition than in slower wind speeds. Winds of 10m/s would give rise to turbulent conditions in the boundary layer keeping some pollen airborne for longer than in non turbulent air flows. If the pollen remained airborne it could travel 36 km in an hour and nearly 864 km in 24 hours.

The transfer of maize pollen by bees and other insects

Maize pollen is mostly dispersed on the wind but it is also collected by bees (*Percival*, 1947, 1955; *Nowakowski and Morse*, 1982; *Vaissiere and Vinson*, 1994) and may be transported by flies. Maize pollen is not uncommon in honey but would not be the major pollen type. There is often 90% of one main type and a 10% mix of many different species, which does contain some wind-pollinated types such as grass and maize (*Hodges*, 1984). Estimates on the distance bees will travel to find pollen or nectar differ. Hooper gives a distance of about 1.5 miles (1976).



Morse (1972) agrees with this for the majority of pollen collection but says that a bee might travel up to 8 or 9 miles if necessary. More recently it has been widely accepted that bees will regularly travel about 3 miles from the hive but will not go this far if a good source of pollen is available closer. However, the observations of bees collecting pollen from the tassels of maize are probably inconsequential in terms of gene flow as maize is diclinous. With the tassels being entirely separate from the non-nectariferous female flowers, bees and other insects are likely to have little motivation for visiting them. As Bateman (1947a) points out "bees often visit the tassels to collect pollen but they do not visit the silks". Therefore the potential for cross-fertilisation via insects is probably highly limited when compared to crop species with bisexual (monoclinous) flowers. However, the pollen carried by bees can become a constituent of honey, which may present difficulties to organic beekeepers whose bees are foraging on GM crops. In addition to transport by bees it is likely that occasional random encounters with other insects could lead to the transfer of maize pollen to neighbouring fields.

Conclusion

It is clear that maize pollen spreads far beyond the 200m cited in several reports as being an acceptable separation distance to prevent cross-pollination. In conditions of low to moderate wind speed the low background concentrations of maize pollen emanating from a crop could spread to a distance of 12 km or more. The impact of this on pollination would depend on the source strengths and relative states of flower maturation. In certain weather conditions, especially those resulting in updraughts, a small percentage of the maize pollen would travel much further downwind. It is not possible to quantify the amounts involved with any reasonable accuracy. The GM maize is likely to be collected by some of the bees within a radius of several km and become a constituent of their honey.

The problems of pollen escape from GM crops are not limited to maize. In trials monitoring oil seed rape pollen between 35m and 475m downwind from a GM crop (National Pollen research Unit, 1999) the maximum concentration recorded was 146.77 grains per m³. This was at 425m from the crop edge, obviously well beyond the exclusion barrier distance. DNA analysis was positive for GM components for pollen sampled at 465m and 475m from the crop edge. In addition, all six of the samples of pollen collected by bees in the vicinity of the crop were positive for GM components. Overall, the results illustrated the fact that oil seed rape pollen can travel on the airflow to at least 425m from the crop edge in notable concentrations and that it is also carried by bees.

It is clear that maize pollen spreads far beyond the 200m cited ... as being an acceptable separation distance

BEE POLLINATION

Geoffrey Hopkinson

PERSONAL EXPERTISE

I have kept bees since 1949 in a total of 5 counties. In addition, I have long standing experience in the study of changing countryside flora and the effect of these changes on beekeeping. I hold the Senior Beekeeping certificate of the British Beekeepers Association and also the National Diploma in Beekeeping, serving that Examination Board as its Chairman and Moderator. I am an executive member of the Council for Environmental Education and a member of their Biodiversity Education Working Party. I am a writer and lecturer on beekeeping topics,

with a specialisation in countryside relationships and pollination and flora. I edited the "Learning From The Land" pack by the Food and Farming Education Service.

QUESTION POSED TO WITNESS

I have been asked to advise on what was in the public domain and available on the subject of maize pollen collection by honey bees as of July 1999.



SUMMARY

This report examines the inter-relationship of *Zea mays* (maize) and *Apis mellifera* (the honey bee).

- > Honey bees collect maize pollen at that time in the season when colony numbers are at their peak. This natural action on the part of honeybees has become more evident to the beekeeper as maize acreages have increased in recent years. Honeybees are capable of carrying this pollen several miles with the possibility of some pollen being carried from the hive on outward foraging flights by other bees. Maize pollen can be found in honey, depending on the area, and is also found in trapped pollen produced for the Health Food trade.
- > If GM crops of maize are grown within three miles of an apiary site or feral bee colonies, it would not be possible to discount the possibility of some pollen being transferred to non GM maize within that area.
- > By the same token, pollen from GM maize would most certainly find its way into the eventual honey crop, although in varying concentrations. It would also be a constituent of harvested pollen sold within the various 'health food' outlets.
- > The effect on the small-scale producer within the present climate of GM opposition would be nothing but harmful, but at a degree yet to be evaluated.

STATEMENT OF THE WITNESS

Introduction

Zea mays is invariably listed as a wind pollinated member of the grass family (Gramineae) in which male flowers are produced at the top of the plant in panicles, referred to as 'tassels', while the female inflorescences, or ears, are produced lower down and are enclosed in sheaths, from which emerge long drooping stigmas called 'silks'. Flowering generally extends over about two weeks and the 'silks' receive pollen, which has been scattered by the wind (Procter and Yeo, 1979).

Maize as a pollen source for bees

There is conclusive evidence that honeybees will collect pollen. With regard to maize, in 1945, F. N. Howes noted that "the male heads on tassels furnish an abundance of pollen which is sometimes collected by bees" (Howes, 1947). In 1982, Nowakowski reported, "It has been calculated that many sweetcorn varieties in the USA produce over 170 kg/ha. The dry pollen is released in the morning and the bees collect it then: they do most easily if it has been wetted by rain" (Nowakowski, 1982). There are also Romanian reports of maize pollen being harvested by a detasselating device and yield of 50 kg/ha being stored for feeding back to bees in times

of pollen scarcity. In 1998, Clive de Bruyn (de Bruyn, 1998), confirmed the observations of F. N. Howes and includes maize in his list of 47 major forage plants for bees. It is however given a score of 3 within a 2 to 9 scale of importance. Rex Walker, in 'Pollen Identification for Beekeepers', a standard text within British beekeeping circles, lists maize pollen as being very large $(90\mu m)$ or 0.09mm as opposed to the Ericaceae $(30-50\mu m)$ and Malus $(35\mu m)$.

Maize is an important staple crop in Africa. J. Amoako (1997), working on Ghanaian beekeeping, places *Zea mays* as the most important pollen source. Atallah, Aly and Eshbah (1988) parallels this observation based on their work in Egypt.

Maize in the UK

Although maize is grown in lowland areas south of Ayr and in areas around Newcastle, the bulk is found south of the Mersey to Humber line and below 400ft above sea level. That is not to discount Lancashire and Yorkshire where site selection becomes much more important. This geographical distribution places the total acreage of maize grown within the flight range of several thousand hives, each containing a nominal 50,000 bees at the height of the season and coincidental with flowering times. The advances in maize 'breeding' are such that with improved hardiness and earlier ripening, there has been a gradual increase in the acreage devoted to silage maize and, in consequence, more beekeepers are experiencing contact with this farm crop.

Flight radius of bees and pollen collection

The flying distances of the foraging worker honeybee will vary according to seasonal weather conditions linked with forage availability. Their search is for nectar (carbohydrate), pollen (protein), water and propolis (collected as resin from trees and used to seal the hive).

The distances bees will fly in search of the above are variously reported and Eckert, an American researcher, recorded colony gains from crops growing seven miles away in the abnormal situation of irrigated crops in an arid area of Wyoming (Eckert, 1933). Thomas Seeley, another American researcher, recorded the bulk of pollen collection taking place within a radius of 1 km but with appreciable quantities being gathered up to 4 km (Seeley, 1985). The general rule of thumb amongst the British beekeepers is to consider a flight range of 3 miles as the effective outside limit with increased returns as the distance between hive and food source decreases. Crops within a half mile radius would be considered as highly likely to yield surplus honey.

The presence of apiary sites in the proximity of GM crop trials is only part of the problem. In the whole of the British Isles, where bees are kept for profit, there exists a concentration of 'feral' bees; these being absconded swarms lodging in hollow trees, false roofs or rock cavities. Some are of short-lived tenure while others may survive for several years. In many instances these only become obvious in the height of the season or at swarming time but they do represent



a contribution to the total foraging force in any area. A swarm issuing from the hive in May or June, and taking up residence in a hollow tree or false roof, would be at it's peak, coincident with maize flowering times.

There is another consideration when attempting to assess reliable parameters for pollen collection and it's subsequent utilisation. Some pollen grains will adhere to the body hairs of the bee and it may well be foraging in the opposite direction to the source of that particular pollen. In other words, the effective range for the transfer of pollen, albeit in small quantities could be doubled.

Pollen

The pollen provides the essential vegetable protein source within the colony metabolism, in addition to supplying lipids (fats) and certain carbohydrates. A colony would require in the region of 20kg of pollen over a season, representing some 2,000,00 pollen loads.

The protein content of pollen can vary between 7% and 30% and, although maize is at the lower end of this spectrum, bees will gather pollen from all manner of sources in times of dearth. Pollen shortage is most likely to occur in intensively farmed arable areas where the absence of indigenous weeds, trees and hedges often results in deleterious effects on hives left on isolated sites after working oil-seed rape and/or field bean crops. The southern Lincolnshire fens provide many examples of this condition.

Pollen in honey and health products

The transmission of the pollen to the hive is followed by a transfer of the pollen loads for storage in the vicinity of the brood cluster. Although this is destined as brood food the small size of even the largest pollen grains is such that a certain percentage finds its way into stored honey. Extraction of honey by centrifugal force also results in some pollen being transferred to the honey crop. This fact is important in that the isolation of pollen grains from honey samples is an essential analytical tool in identifying the floral source of the honey or at least narrowing down the country of origin.

The pollen in any bulk supply can be removed by pressure filtration, an expensive procedure resulting in a cleaner, brighter product and therefore attractive to the major packers of imported honeys. Such a process is quite beyond the scope of the small-scale producers who are the backbone of British beekeeping. For 'farm gate' sales, the fact that the locally produced honey contains pollen grains is very often an attractive selling point with the consumer.

Pollen in any bulk supply can be removed by pressure filtration ... such a process is quite beyond the scope of the small-scale producers

The quantity of pollen in any sample will vary according to the floral source and the method of extraction. "Some honey samples may contain only a few thousand grains per 10gms, while the figure in others may be as large as 160,000." and, "A normal honey should contain between 2,500 to 6,000 pollen grains per 10gms" (Seeley,1985).

Although there is an optimum quantity of pollen required for effective colony metabolism, and although pollen shortage can occur, in the majority of areas bees will gather and store pollen surplus to the colony requirement. This gives rise to the condition known as 'pollen clogged combs' in which this old, unused pollen has dried out and is discarded by the bees.

There exists a market for pollen within the health food arena. There is a recognised technique for installing a proprietary pollen trap at the entrance to the hive. The bees are obliged to pass though a mesh of such size as to cause some of the pollen loads to be scraped from the "pollen baskets" or "corbicula" on the hind legs of the worker bee. Pollen is subsequently harvested from a collection tray for either eventual sale within the whole-food organisation or as a stimulative feed for bees the following spring. In some small-scale beekeeping enterprises, the added value of pollen sales is the key to financial viability. It is reported that one homeopathic dealer in the North of England purchases 12 tonnes of pollen each year for sale in the health food market.

This dried, clean pollen is used as a food supplement. Quantities varying from a teaspoonful to a tablespoon of pollen may for instance be added to the consumer's breakfast cereal each morning. Another market for harvested pollen is in the preparation of propolis lozenges, where it can be used as a coating agency. (Propolis is the resin collected by bees to seal the hive and is sought within the homeopathic trade for it's antiseptic properties).



RISKS TO ORGANIC FARMING

Richard Young

PERSONAL EXPERTISE

I am currently Policy Advisor to the Soil Association and since 1974 I have produced both crops and livestock on approximately 400 acres land according to the organic principles of the Soil Association. My further qualifications are that: from 1985 until the present time I have been a member of the Soil Association's Agricultural Standards Committee: from 1987 until 1991 I took ultimate responsibility for the development of the Soil Association's organic inspection and certification scheme as honorary chairman of its over-arching standards committee, the 'Symbol Committee'; during the same period I also held the following honorary positions - co-opted member of the Soil Association Council, chairman of the Livestock Standards' Committee, member of the Producers' Advisory Committee to the UKROFS Board, and the following paid positions: Deputy Director British Organic Farmers, Editor "New Farmer and Grower". I have researched and written on a range of agricultural and food-related issues and have been the author, co-author and/or editor of a number of Soil Association reports.

OUESTION POSED TO WITNESS

I have been asked to advise on the risks to organic farming from genetically modified food crops based on what was in the public domain and available on the subject in July 1999.



SUMMARY

- > Organic farming has been developed over many decades as a systems-based alternative to the use synthetic fertilizers and pesticides, many of which have damaging environmental effects and all of which require large quantities of energy in their extraction, manufacture, transport and application.
- > Organic farming in the UK is currently expanding faster than ever before at 25% per annum. However, demand is growing by 40% per annum. This is causing strong price premiums in favour of organic food. With price depression in much of the conventional farming industry, organic production, while still small at 1.3% of the farmed area, is seen to have a bright future. Government has also recently allocated £140 million for the Organic Aid Scheme which makes payments to farmers while converting their land.
- > Organic farming organisations in the UK, EU and internationally are united in their opposition to the use of genetic engineering in the development of food crops and have prohibited the growing of genetically-engineered crops on organically-farmed land and prohibited the use of genetically-engineered ingredients in processed organic foods.
- > This opposition is based on the imprecise and unpredictable nature of genetic engineering technologies and on the fact that they:
 - · pose a potentially unacceptable threat to human health
 - involve practices incompatible with the principles of sustainable agriculture, as defined by organic farming organisations
 - make likely irreversible damage to the environment
 - · make likely the loss of species biodiversity
 - involve the release of organisms of an unrecallable nature
 - · pollute other farms with genetically modified organisms
 - · remove the right of choice, for farmers and consumers
 - · violate farmers' fundamental property rights and threaten their economic independence
- > The growing of genetically engineered crops on conventional farms especially in a small country like Britain, poses serious risks of contamination of organic crops by a range of means. Organic farming bodies have sought to address this problem as far as is possible by changes to organic production standards legally binding rules which govern the production, processing and marketing of organic produce. However, since organic and conventional production often takes place on adjacent land, and since some conventionally-produced inputs (principally seed, feed and manure) are permitted in organic farming, the Soil

Organic farming organisations in the UK, EU and internationally are united in their opposition to the use of genetic engineering in the development of food crops ... based on the imprecise and unpredictable nature of genetic engineering technologies

Association, the UK's foremost organic farming body, is also campaigning to prevent the continued growing of genetically-engineered crops in the UK, even for trial purposes, because of the uncontrollable and unpredictable nature of their spread. Contamination of organic crops will lead to loss of organic status and involve substantial financial penalties.

STATEMENT OF THE WITNESS

I have been involved with the specific development of Soil Association policy on genetic engineering since March 1998. The views set out in this report are mine, but they are based closely on and essentially reflect those of the Soil Association.

Background – genetically modified food crops – the Soil Association's position Until about seven years ago the Soil Association knew very little about genetic modification and decided at council level to assess individual cases on their merits in relation to their acceptability within organic farming systems. Over the following two years, however, the organisation received representations from a number of individual members, producers, consumers and scientists about the potential dangers posed by genetic modification technology. As a result of this, the organisation's interest in the subject increased and it soon became apparent that serious questions about the long-term safety of genetically modified food crops remained unanswered.

Far from being a benign technology, as was generally claimed, the Soil Association realized the new and sometimes imprecise nature of genetic engineering technologies and the speed with which they have been developed and used meant there was a distinct possibility that the introduction of genetically modified crops into agriculture could:

- · cause serious ecological imbalance and further speed the decline in species biodiversity
- · lead to unforeseen and even unpredictable effects for human health

Furthermore, the Soil Association felt that it was also not outside the realms of probability that the use of genetically modified bacteria and viruses as vectors in recombinant DNA technology could lead to new and potentially untreatable bacterial infections and viral diseases.

The organisation's initial reaction to the introduction of genetic engineering technologies had been that it should await further research, but as the range of potential problems began to emerge,



the Soil Association realized that it had a duty to its members and to consumers of organic food to adopt the precautionary principle and prohibit their use in organic food production.

It also gradually became apparent to the Soil Association that while genetic modification was being promoted as a way to produce a 'sustainable' agriculture, the concept of 'sustainability' that was being used bore little resemblance to the organisation's own view of what 'sustainability' in food production actually meant.

The Soil Association's view on this was that in a world of diminishing resources, the only durable way of producing a 'sustainable' agriculture is through what is known as a 'systems' approach, in which the long term health of the soil is maintained by the avoidance of toxic chemicals, by balancing exploitative cropping with fertility building phases and where agriculture as a whole generates and recycles as great a part of its resources as possible. This strongly held view has underpinned the development of organic farming, not just in the UK, but throughout the world for more than fifty years. What the Soil Association has come to see as fundamentally unacceptable about the genetic modification of food crops is that in almost every respect, this will make farmers more, not less, dependent upon bought-in and finite resources and that this will increase the trend towards the 'industrialization' of food production and the degradation of soils; trends which the association has been working to reverse since it establishment in 1946.

Organic standards and genetic modification - an overview

Organic farming standards are legally binding rules and regulations governing the production, storage, processing and marketing of all food displayed and marketed as being 'organic'. Throughout the European Union organic farming is governed by Council Regulation EU 2092/91. Member states are free to set higher standards where they wish, as are designated 'organic sector bodies' such as the Soil Association. Individual member states and/or sector bodies may promote unique features of their own standards and distinguish these by their own certification logos, but they cannot prohibit the sale of produce bearing the description 'organic' if it complies with the EU regulation.

As a result of its considerations about genetic modification, the Soil Association council took a decision that it was opposed, in principle, to the genetic modification of crops and that these crops would be prohibited from use in organic farming under the detailed production standards which the organisation sets and polices.

Since only about 70% of organic producers in the UK operate to Soil Association standards, it was felt necessary to present the case for opposition to the use of genetically modified crops to the United Kingdom Register of Organic Food Standards (UKROFS), the EU-designated national body for the maintenance of national minimum organic food and farming standards

and for the implementation of the EU regulation on organic farming. Members of the UKROFS Board and Certification Committee are appointed by Ministers to represent the broad cross-section of interests in organic food production and processing in the UK. After a series of meetings and over a significant period of time, when individual board and certification committee members were able to discuss the issues with the organisations they represented, the UKROFS Board took an identical position to that of the Soil Association and this was reflected in agreed detailed and published changes to its own standards.

The argument was then presented by UKROFS and the Soil Association at an EU level where it complemented similar concerns throughout the European organic farming movement and led, over a period of about two years, to agreed amendments to the EU Regulation on organic food production which similarly exclude the use of genetically modified crops in organic food production throughout the Community. These were adopted as part of Council Regulation (EC No 1804/1999) on 19 July 1999 which came into force on 24 August 1999.

At an international level there is also unanimous opposition within organic farming bodies to genetic engineering of food crops. This position was ratified in the declaration of 22 November 1998 when delegates from more than 60 countries representing the world's leading organic farming organisations at the 12th scientific conference of the International Federation of Organic Agricultural Movements (IFOAM) in Argentina issued the following statement:

"IFOAM is calling for governments and regulatory agencies throughout the world to immediately ban the use of genetic modification in agriculture and food production since it involves:

- Unacceptable threat to human health
- · Negative and irreversible environmental impacts
- · Release of organisms of an unrecallable nature
- · Removal of the right of choice, both for farmers and consumers
- · Violation of farmers' fundamental property rights and endangerment of their economic independence
- Practices which are incompatible with the principles of sustainable agriculture as defined by IFOAM".

Soil Association campaign against genetically modified food

The Soil Association is a registered charity and its council is democratically elected by the organisation's paid up members. Its organic inspection and certification programme is run by a wholly-owned separate limited company, SA Cert, but standards are set by Council for the benefit of the associations' consumer members and to ensure fair trade among its producer members.

The Soil Association's stance on genetic modification has arisen through a groundswell of opinion among its members and details of the organisation's developing position on genetically modified food have been communicated to members through its two membership journals, Living Earth and Organic Farming, and range of newsletters including Organic Focus and



Certification News. Support for the position it has adopted continues to be reflected in a substantial post bag on this issue.

The Soil Association has given a commitment to its members and all consumers of organic food certified under its logo, that it will allow no genetically modified ingredients in organic food production. In order to honour this pledge it has developed a strategy which includes:

- changes to its published standards and inspection procedures
- the introduction of monitoring for contamination, where deemed appropriate
- · the withdrawal of organic status, where necessary
- the maintenance of a continuing campaign against the growing of any genetically modified crops in the UK, in order to reduce and if possible eliminate the risk of contamination of organic crops and land and those conventionally-produced inputs currently permitted under organic standards

Standards

Standards have been amended several times over the last few years as the scale of concerns relating to genetically modified crops has become apparent. The process of standards revision is an ongoing one. The Soil Association has a number of standards committees responsible for different areas, which pass their concerns and recommendations to Council for considerations or ratification. The UKROFS Board is also keeping standards on genetic engineering under review and has established a working group:

"To ensure that the eventual UKROFS Standards adequately reflect the requirements in EC Regulation 1804/1999 that genetically modified organisms or their derivatives shall not be used in the production of organic food, and to present to the Board of UKROFS any amendments or additions they consider necessary" (UKROFS, 1999).

Two key Soil Association standards state:

- 2.4.4 "Organic products must be free of contamination from GMOs and their derivatives. Accordingly, operators must take all necessary measures to prevent any such contamination of organic products during production, processing, storage and transport."
- 2.4.5 "Organic certification may be withdrawn from land, crops or products where, following an evaluation and, where appropriate, analysis, the Certification Committee considers that there is contamination or a specific risk of contamination from GMOs or their derivatives. Withdrawal periods for contaminated production units will be decided on a case by case basis" (*Soil Association, June 1999*).

These standards are augmented by detailed standards relating to each area where risks are perceived to exist.

Preventing the contamination of organically-managed land and organically-produced crops

1) Contamination from the spread of pollen from genetically modified crops:

Research commissioned by the Soil Association from the National Pollen Research Unit and published in January 1999 (*Emberlin et al., 1999*) suggested that pollen from maize can travel many miles under certain conditions. This view has been reinforced by a further report (*Treu and Emberlin, 2000*), which cites evidence that cross-pollination can also occur with other major agricultural crops over significant distances. The growing of genetically modified crops on UK farmland, whether as small-scale trials, field-scale trials or commercial production, therefore presents a threat to the organic status of similar crops on organic farms where cross-pollination may take place with a related food species crop or a weed species on, or within future pollinating distance of, an organic farm. Distances over which cross-pollination risks arise vary according to species from relatively short distances in the case of cereal varieties to very considerable distances in the case of the brassica family.

2) Spread of recombinant DNA by other means:

There is also a possible longer term threat to organic crops in future years if it proves possible for genetically modified DNA to transfer by other means to subsequent crops on organically farmed land. Transfer on agricultural machinery shared between farms is an obvious example and standards have been amended to address this aspect. However, there are other less well understood ways in which such transfer of genetic material could take place. The Soil Association recognizes the incomplete nature of the science in this area and its own less that total understanding even of the existing evidence, but it is nevertheless extremely anxious to protect organic farms from any possible pollution which might arise in this way. This is quite simply because it is not yet known what the long term implications of the possible transfer of DNA from GM crops might be for the future organic status of crops, land and whole farms.

It is not, for example, yet clear to the Soil Association whether DNA from pollen or other plant material from genetically engineered crops may be able enter the soil ecosystem where pollination does not occur. It is also not clear whether this DNA can transfer from one field or one farm to another via soil biological or microbiological organisms.

Hoffman et al. (1994) showed that genes from genetically engineered plants have transferred to soil fungi and Schluter et al. (1995) observed a similar effect in the laboratory with soil bacteria. It is well known that soil fungi such as mycorrhiza, play a fundamental role in the uptake of nutrients by plants, but it is not known by the Soil Association, or documented in the scientific literature as far as we are aware, whether this might provide a mechanism for the transfer of genetically modified material from soil microorganisms to future crops. If such



transfer is possible, it could in future threaten the organic status of crops on land anywhere such transfer takes place and as such would have considerable financial implications.

Since the observations of Watanabe in 1959 (Watanabe, 1963), the ability of bacteria to transfer antibiotic resistance via extra-chromosomal genes has been well studied and documented. Much has been written on the possible threat posed by the use of antibiotic-resistant marker genes in some genetically engineered crops. In the UK such comment appears to have led to a greater use of varieties where such genes have been 'inactivated'. However, the Soil Association remains unconvinced at the present time that this process will necessarily remove the risk of all such antibiotic resistance genes passing to soil bacteria and from them eventually to farm animals. Antibiotic resistance genes used appear often to be members of the aminoglycoside family. Many members of this important group of antibiotics are known to be cross-resistant, for example, gentamycin and apramycin. This is significant because early members of this family of antibiotics, such a streptomycin, are again having to be used in the treatment of otherwise antibiotic-resistant strains of tuberculosis (Young et al., 1999). Resistance to aminoglycosides can also become attached to multiple-drug resistance plasmids and where this occurs can confer resistance to as many as eight different types of antibiotics including glycopeptides drugs of last resort for many antibiotic-resistant infections. Antibiotics are permitted for the treatment of infection in organically managed farm animals and any further pressure which increases the rate of development of resistance in the aminioglycosides could have serious repercussions for animal health and welfare.

The Soil Association is also concerned that the 'horizontal transfer' of genetic material could proceed rapidly if DNA from genetically modified crops is taken up by soil organisms. While it is assumed that migration via microorganisms is likely to be relatively slow, it is feared that if genetic material enters invertebrate life, and subsequently then other creatures, such as insects, birds and mammals, this could increase the rate and range of spread very substantially. The rapid spread of the New Zealand flatworm in parts of Scotland and Northern Ireland (Alston, 1991), after their accidental introduction, demonstrates how rapidly new organisms can colonize large areas of land once released into the environment.

Preventing the contamination of conventionally-produced inputs permitted in organic farming

Several types of conventionally-produced inputs are currently permitted in organic farming. This is because the relatively modest area of land under organic production has not yet reached the 'critical mass' required to provide the full range of needs of all organic farms, especially since some of these are limited in what is available to them by geographical and/or climatic considerations. As such the potential contamination of conventionally-managed crops and land

Inputs containing genetically engineered contaminants are prohibited in principle because the technology of genetic engineering is seen as the antithesis of the organic approach to food production. There are also specific risks associated with each input

is of serious concern to the Soil Association and to individual organic farmers. This is particularly so since the safeguards provided by organic standards do not apply to most (or possibly any) conventionally-managed farms and the contamination of crops on such farms could rapidly erode the availability of conventionally-produced inputs for use on organic holdings.

Inputs containing genetically engineered contaminants are prohibited in principle because the technology of genetic engineering is seen as the antithesis of the organic approach to food production. There are also specific risks associated with each input as the following examples illustrate:

1) Seeds

The use of conventionally-grown seed is gradually being phased out in organic production. However organic grades of many species and varieties are not currently available or not available in sufficient quantities to meet demand. Where this is the case, conventionally-grown seeds are permitted by way of derogation. The contamination of conventionally-grown (or for that matter organically-grown) seed with an admixture of genetically modified seeds would clearly not be acceptable in organic farming since it would provide a direct and rapid route for the introduction of genetically modified DNA into organic systems and would be likely to produce a crop which was contaminated with genetically-modified strains. The spread of pollen from genetically modified crops to neighbouring conventional farms could therefore have serious consequences for organic producers who may in future find it difficult to obtain suitably pure seed or lose the organic status of resulting crops where contamination of seed is not detected before planting.

2) Bought-in feed

Organic livestock producers are permitted to buy in certain proportions of conventionally produced livestock feed. The percentage is calculated on a daily dry matter basis and has recently been reduced in each category. Currently the following proportions of conventional feed are permitted:

Beef and Sheep 10% Dairy 15% Pigs and Poultry 20%



While the range of genetically-modified crops so far cultivated in the UK is relatively limited, it may be relevant to note that organic dairy farmers, for example, would be permitted to buy in silage made from forage maize on conventional holdings, a crop which would be susceptible to possible cross-pollination from genetically modified maize under certain conditions.

In addition, crop residues such as straw are permitted for livestock bedding. Any feed or bedding material contaminated with genetically modified ingredients is prohibited as an input into organic farming systems. The contamination of a significant number of conventionally-produced crops in the UK would therefore pose a serious threat to the future viability of organic livestock production, because it would reduce the overall quantity of conventionally-produced livestock feed available to the organic sector and could cause acute shortages in some localities.

The Soil Association is specifically concerned to prevent the use of feed containing genetically modified ingredients because it believes that we do not yet have sufficient scientific evidence to be sure that recombinant DNA is entirely broken down in the gut. Indeed research at the Nutrition and Food Research Institute in Zeist in the Netherlands suggests that it may survive intact long enough to transfer genes to normal gut micro-organisms (*MacKenzie*, 1999). Animal manures could therefore also become contaminated with genetically modified bacteria of animal origin in addition to genetically modified crop material.

3) Conventionally-produced livestock manures

Organic producers are also permitted to buy in manure from some conventionally-managed livestock systems. Suitable systems are defined in current organic standards, but in general can be described as those which are more extensive. Standard 2.4.9 states:

"With effect from 1 July 1999, fertilizers, composts, manures and other nutrient inputs containing GMOs or their derivatives are prohibited, with the exception of manures from livestock that have consumed feeds containing GMOs or their derivatives. With effect from 1 January 2000, use of manures from animals which have been fed these materials within three months is also prohibited".

The specific concern with manures from stock fed on crops or ingredients containing genetically modified organisms is the risk of genetically modified material passing through an animal's digestive system in tact, bypassing the animal as spilt or soiled feed, or passing recombinant DNA through the animal via other organisms such as bacteria. Each of these cases could subsequently result in contamination of the soil organisms on an organic holding.

4) Conventionally-managed land

Conventionally-managed land may acquire organic status after a conversion period lasting three years. However, where genetically-engineered crops have been grown this period has been extended to five years and could be extended further if evidence emerges suggesting contamination

is persisting longer than this. A five year conversion period, however, makes the economics of converting land to organic production considerably less attractive and the Soil Association is therefore also anxious to prevent the growing of genetically modified crops on conventional land for the additional reason that this will effectively restrict the future availability of land for conversion to organic production.

The current state of organic farming in the UK

After many decades of being ignored, and even ridiculed, organic farming has recently experienced a dramatic change of fortune. Demand is growing by 40% per annum while supply is increasing only by 25% per annum. As a result, organic food commands substantial price premiums. In addition, producers can qualify for conversion grants under the Organic Aid Scheme and benefit from the generally lower production costs associated with the non-use of synthetic fertilizers and pesticides. Contrasted with the generally depressed prices in much of the conventional farming industry, organic production is increasingly seen as having a bright economic future.

The number of applications from conventional farmers to convert land to organic methods has doubled each year for the last five years. The result of this is that by April last year there were 240,000 hectares of UK land in organic production or conversion to organic (1.3% of the total farmed area). The recent rapid rate of expansion can be gauged from the fact that of this, a total of three times as much land was in conversion as in full organic production.

Conclusion

The Soil Association recognizes the necessity for organic methods to be economically viable and has played a pivotal role in developing the various components of this which are now largely in place.

However, its prime motivation in encouraging and facilitating a change from conventional to organic methods is the long-held belief that many aspects of production systems which rely heavily on the input of synthetic and often toxic chemicals and the exploitative cropping patterns and farming methods which these make possible, are causing serious and in some instances, irreversible degradation of the environment, biodiversity, the biosphere, human health and society as a whole. As such, the Soil Association wants to see as many farms as possible change to organic methods. This will, however, be seriously threatened where genetically modified crops have been grown or contamination from 'genetic pollution' has occurred.

It is outside the scope of this paper to address each specific concern, or the detailed arguments relating to the actual and potential risks associated with the various aspects of genetic engineering technologies that have been applied to crop production. However, it may be concluded that the Soil Association perceives the genetic engineering of food crops as a



dangerous, ill-conceived and fundamentally-flawed departure from tradition breeding technologies which not only threatens the future viability of organic farming but poses a threat of immeasurable proportions to the future survival of many species on this planet including, ultimately the human race itself.

This may explain why the Soil Association has become institutionally committed to the campaign against the introduction of genetically engineered crops into UK agriculture, why it is demanding a ban on all trials of genetically engineered crops in the UK and why it has called for a minimum six mile exclusion zone round all organic farms, while this demand remains unheeded.

The Soil Association perceives the genetic engineering of food crops as a dangerous, ill-conceived and fundamentally-flawed departure from traditional breeding technologies which not only threatens the future viability of organic farming but poses a threat of immeasurable proportions to the future survival of many species on this planet including, ultimately the human race itself

THE WEAKNESS OF THE CONCEPT SUBSTANTIAL EQUIVALENCE

Dr. Frik Millstone

PERSONAL EXPERTISE

I am a senior lecturer in Science Policy at Sussex University. Since 1974 I have been conducting research into the public and environmental health and safety aspects of technological change in the food and agricultural sector. I am one of the UK's leading academic commentators in my chosen field. I have a first degree in Physics, and three postgraduate degrees in Philosophy, including a doctorate.

OUESTION POSED TO WITNESS

I have been asked to advise on what was in the public domain and available on the subject of the supposed 'substantial equivalence' of genetically modified foods to their non-genetically modified relatives in July 1999.



SUMMARY

- > Substantial equivalence (the concept by reference to which all genetically modified (GM) foods have so far been approved) is a pseudo-scientific concept because it is a commercial and political judgement masquerading as if it were scientific.
- > It is, moreover, inherently anti-scientific because it provides an excuse for not requiring biochemical or toxicological tests. It therefore serves to discourage and inhibit potentially informative scientific research.
- > It has, moreover, been misapplied, even on its own terms, within the regulatory process.
- > If policymakers are to provide consumers with adequate protection, then the concept of substantial equivalence will need to be replaced with a practical approach which would actively investigate the safety and toxicity of GM foods rather than merely taking them for granted, and which could give due consideration to public health principles as well as to industrial interests.

STATEMENT OF THE WITNESS

Introduction

Whenever official approval for the introduction of genetically modified (GM) foods has been given in Europe or the United States, regulatory committees have invoked the concept of 'substantial equivalence'. This means that if a GM food can be characterized as substantially equivalent to its 'natural' antecedent, it can be assumed to pose no new health risks and hence to be acceptable for commercial use. At first sight, the approach might seem plausible and attractively simple, but it is misguided, and should be abandoned in favour of one that includes biological, toxicological and immunological tests rather than merely chemical ones.

The concept of substantial equivalence has never been properly defined; the degree of difference between a natural food and its GM alternative before its 'substance' ceases to be acceptably 'equivalent' is not defined anywhere, nor has an exact definition been agreed by legislators. It is exactly this vagueness which makes the concept useful to industry but unacceptable to the consumer. Moreover, the reliance by policymakers on the concept of substantial equivalence acts as a barrier to further research into the possible risks of eating GM foods.

Acceptable daily intake

The concept of substantial equivalence emerged in response to the challenge confronting regulatory authorities in the early 1990s. Biotechnology companies had developed several GM foods and, to reassure their customers, wanted official approval for their introduction. But government statutes did not cover GM foods, nor provide the authority to regulate these

innovations. Legislation could be amended, but that would not address the core problem of how to assess the risks. One obvious solution at that time would have been for legislators to have treated GM foods in the same way as novel chemical compounds, such as pharmaceuticals, pesticides and food additives, and to have required companies to conduct a range of toxicological tests, the evidence from which could be used to set 'acceptable daily intakes' (ADIs). Regulations could then have been introduced to ensure that ADIs are never, or rarely, exceeded.

From the point of view of the biotechnology industry, this approach would have had two main drawbacks. First, companies did not want to have to conduct toxicological experiments, which would delay access to the marketplace by at least five years, and would add approximately \$25 million per product to R&D costs. Second, by definition, using ADIs would have restricted the use of GM foods to a marginal role in the diet. An ADI is usually defined as one-hundredth of the highest dose shown to be harmless for laboratory animals. Thus, even if laboratory animals show no adverse effects on a diet consisting exclusively of a test material, human intake would still be restricted to 1% of the human diet. The biotechnology companies want to market GM staples, such as grains, beans and potatoes, which individually might account for as much as 10% of the human diet, and collectively might provide more than half of a person's food intake.

The adoption of the concept of substantial equivalence by the governments of the industrialized countries signaled to the GM food industry that as long as companies did not try to market GM foods that had a grossly different chemical composition from those of foods already on the market, their new GM products would be permitted without any safety or toxicology tests. The substantial-equivalence concept was also intended to reassure consumers, but it is not clear that it has served, or can serve, that purpose. Although toxicological and biochemical tests, and their interpretation, are notoriously problematic and contested, and are slow and expensive, they can provide information vital to consumer protection (European Commission, 1980).

Trying to have it both ways

The challenge of how to deal with the issue of risk from consuming GM foods was first confronted in 1990 at an international meeting, consisting of officials and industrialists but no consumer representatives, of the UN Food and Agriculture Organisation (FAO) and the World Health Organisation (WHO), (WHO, 1991). The FAO/WHO panel report makes intriguing reading, because what it fails to mention is as important as what is discussed. It does not use the term 'substantial equivalence' or mention ADIs. It implies that GM foods are in some important respects novel, but it then argues that they are not really novel at all – just marginal extensions of traditional techniques. These inconsistencies are inevitable, given that the industry wanted to



argue both that GM foods were sufficiently novel to require new legislation and a major overhaul of the rules governing intellectual property rights to allow them to be patented, yet not so novel that they could introduce new risks to public or environmental health.

The biotechnology companies wanted government regulators to help persuade consumers that their products were safe, yet they also wanted the regulatory hurdles to be set as low as possible. Governments wanted an approach to the regulation of GM foods that could be agreed internationally, and that would not inhibit the development of their domestic biotechnology companies. The FAO/WHO committee recommended, therefore, that GM foods should be treated by analogy with their non-GM antecedents, and evaluated primarily by comparing their compositional data with those from their natural antecedents, so that they could be presumed to be similarly acceptable. Only if there were glaring and important compositional differences might it be appropriate to require further tests, to be decided on a case-by-case basis.

Unfortunately, scientists are not yet able to reliably predict the biochemical or toxicological effects of a GM food from knowledge of its chemical composition. For example, recent work on the genetics of commercial grape varieties shows that, despite detailed knowledge going back for centuries of the chemistry and flavour of grapes and wines, the relationship between the genetics of grapes and their flavour is not yet understood (*Bowers et al., 1999*). Similarly, the relationship between genetics, chemical composition and toxicological risk remains unknown. Relying on the concept of 'substantial equivalence' is therefore just wishful thinking: it is tantamount to pretending to have adequate grounds to judge whether or not products are safe.

The results of Arpad Pusztai's experiments with GM potatoes and their interpretation remain a matter of controversy (see Nature, 1999), but his starting hypothesis was that GM potatoes would be substantially equivalent to non-GM potatoes. Pusztai interpreted his results as indicating that the GM potatoes exerted adverse biochemical and immunological effects, which could not have been predicted from what was known of their chemical composition (Ewan and Pusztai, 1999). The kinds of experiments which he conducted are not legally required and are therefore not routinely conducted before GM foods are introduced into the food chain.

Failure to define 'Substantial Equivalence'

The concept of 'substantial equivalence' was first introduced in 1993 by the OECD (OECD, 1993), and was subsequently endorsed in 1996 by the FAO and WHO. Given the weight that the concept has been required to carry, it is remarkable how ill-defined it remains, and how little attention has been devoted to it.

Substantial equivalence is a pseudo-scientific concept because it is a commercial and political judgement masquerading as if it were scientific

The OECD document states:

"For foods and food components from organisms developed by the application of modern biotechnology, the most practical approach to the determination is to consider whether they are substantially equivalent to analogous food product(s) if such exist ... The concept of substantial equivalence embodies the idea that existing organisms used as foods, or as a source of food, can be used as the basis for comparison when assessing the safety of human consumption of a food or food component that has been modified or is new."

That is the closest there has been to an official definition of substantial equivalence, but the definition is too vague to serve as a benchmark for public health policy.

GM glyphosate-tolerant soya beans (GTSBs) illustrate how the concept has been used in practice. The chemical composition of GTSBs is, of course, different from all antecedent varieties, otherwise they would not be patentable, and would not withstand the application of glyphosate. It is quite straightforward to distinguish, in a laboratory, the particular biochemical characteristics which make them different. GTSBs have, nonetheless, been deemed to be substantially equivalent to non-GM soya beans by assuming that the known genetic and biochemical differences are toxicologically insignificant, and by focusing instead on a restricted set of compositional variables, such as the amounts of protein, carbohydrate, vitamins and minerals, amino acids, fatty acids, fibre, ash, isoflavones and lecithins. GTSBs have been deemed to be substantially equivalent because sufficient similarities appear for those selected variables.

But this judgement is unreliable. Although we have known for about 10 years that the application of glyphosate to soya beans significantly changes their chemical composition (for example the level of phenolic compounds such as isoflavones), (Lydon and Duke, 1989), the GTSBs on which the compositional tests were conducted were grown without the application of glyphosate (Padgette et al., 1996). This is despite the fact that commercial GTSB crops would always be treated with glyphosate to destroy surrounding weeds. The beans which were tested were, therefore, of a type which would never be consumed, while those which are being consumed were not evaluated. If the GTSBs had been treated with glyphosate before their composition was analysed it would have been harder to sustain their claim to substantial equivalence. There is a debate in the research community on whether such changes to the chemical composition are desirable or undesirable, but it is an issue which remains unresolved, and which has been neglected by those who have deemed GTSBs and non-GM soyabeans to be substantially equivalent.



Acknowledged limitations

Only one official organization has recognized some of the limitations of the concept of substantial equivalence. A Dutch government team has acknowledged that "compositional analysis ... as a screening method for unintended effects ... of the genetic modification has its limitations ... in particular regarding unknown anti-nutrients and natural toxins", and it has given a lead by trying to explore some alternatives (*Kuiper et al., 1998*). The Dutch team accepts that comparisons of relative crude compositional data provide a very weak screen against the introduction of novel genetic, biochemical, immunological or toxicological hazards, and they have suggested a finer-grained screen to test for differences in some of the relevant biological variables, such as DNA analysis, messenger-RNA fingerprinting, protein fingerprinting, secondary metabolite profiling and in vitro toxicity testing. If the use of such a finer screen revealed that a GM food contained a relevant novelty then the case for further studies would be far stronger, and those studies might benefit from having some clues as to which end-points might be most worthy of investigation.

Conclusions

Substantial equivalence is a pseudo-scientific concept because it is a commercial and political judgement masquerading as if it were scientific. It is, moreover, inherently anti-scientific because it was created primarily to provide an excuse for not requiring biochemical or toxicological tests. It therefore serves to discourage and inhibit potentially informative scientific research. The case of GTSBs shows, moreover, that the concept of substantial equivalence is being misapplied, even on its own terms, within the regulatory process. If policymakers are therefore to provide consumers with adequate protection, and genuinely to reassure them, then the concept of substantial equivalence will need to be abandoned, rather than merely supplemented. It should be replaced with a practical approach which would actively investigate the safety and toxicity of GM foods rather than merely taking them for granted, and which could give due consideration to public health principles as well as to industrial interests.

HAZARDS TO FOOD AND ANIMAL FEED

Dr. C. V. Howard MB. ChB. PhD. FRCPath.

PERSONAL EXPERTISE

I am a toxico-pathologist who specializes in the effects of low-dose subacute toxicity in the fetus and the newborn. For the past decade my co-workers and I have been studying the effects of intra-uterine growth retardation on the fetus and demonstrated that this can have a permanent effect on the development of the kidneys (Hinchliff et al., 1992), lungs (Beech, 1997) and nervous system (Howard et al., 1995).

OUESTION POSED TO WITNESS

I have been asked to advise on what was in the public domain and available on the subject of GM foods in relation to toxicology and human health in July 1999.



SUMMARY

- > Although certain areas of possible hazard associated with GM technology can be identified in a qualitative manner, it is not possible to accurately quantitate them. For example, the likelihood of the emergence of strains of pathogenic organisms resistant to antibiotics as a result of GM food technology is unknown. Under the circumstances where hazard assessment is not quantifiable, risk assessment becomes 'opinion'. The current testing of GM foods by comparing their broad chemical analysis across whole groups of cultivars, 'substantial equivalence', will not, in my opinion, pick up subtle changes to the immune, endocrine and nervous systems to which the fetus and infant are susceptible during periods of imprinting.
- > There is very little evidence available for the general public to examine, to establish the degree of testing of novel GM crops for the hazards outlined above. Although some data is available, it is difficult to obtain and does not contain enough detail to allow for complete interpretation, as illustrated in the section on chronic low dose effects. Such evidence as is available suggests that a number of possible hazards from the use of genetically modified crops in food and animal feed have not been addressed.
- > Measures which would improve public views of GM foods and crops would include thorough feeding and toxicity/allergy assessment trials undertaken by the regulatory licensing authorities, with the inclusion of volunteer human feeding studies.

STATEMENT OF THE WITNESS

Introduction

In this report I will address the state of knowledge about GM foods in the public domain as of July 1999. Firstly I will consider the difference between risk and hazard assessment, which is in my opinion absolutely central to this case. I will then address each of the areas of risk with respect to the level of knowledge of the associated hazards available in the public domain.

My interest in the field of GMO crops is the potential risk associated with subtle changes to the composition of the food chain over long periods of time, for example increases in levels of anti-nutritional factors. For staples in the human diet, this could involve whole of life exposure, including intrauterine life.

In view of the novelty of the technology of genetic modification, these potential risks should be set in the context of the precautionary stance taken with the rigorous testing of new pharmaceutical agents. It is estimated that it costs in the order of US\$400,000,000 to produce a new drug, because of the initial laboratory toxicological testing which is then followed by three phases of human volunteer clinical trails. This is for substances which will be taken on a voluntary basis, indicated by the need for treatment of a medical condition, usually for a short

period of time and in fractions of a gram. On the other hand, we have no choice over whether we eat or not. In addition, food is consumed in kilograms per day, amounting to many tonnes over a lifetime. There are strong arguments with such a new and, as yet, incompletely understood technology, for an approach which is at least as precautionary as the testing of new pharmaceutical agents, as recently discussed in the journal Nature (Howard and Saunders, 1999). At present this is not the case.

Risk versus hazard assessment

The concepts of risk and hazard assessment hold true for the management of the introduction of GMO crops. In essence:

- Hazard assessment requires the direct measurement of some parameter of relevance. That requires good hazard identification, because risk assessment cannot address hazards that have not been assessed. This means that it requires a scientific investigation, which there fore needs the investment of resource for labour and materials. An example with respect to GMO crops would be the effect of a novel food in feeding studies on experimental animals and/or humans. In each case, the studies should address the most sensitive indices of harm that can be identified. For example, to investigate the effects of antinutritive factors it would, in general, be more meaningful to use animals that were in a developmental stage of life, rather than fully grown, the former being more susceptible.
- Risk assessment results in a statement of the likelihood of the occurrence of a hazard. Risk assessment is therefore an interpretation of a hazard assessment and can be based on a spectrum of input data ranging from 100% experimental evidence on one hand to 100% speculation (model assumptions) at the other. Experimental evidence can be of more or less relevance to the risk under consideration, depending upon the experimental design. Risk assessments based on assumptions rather than measurements cost very little. They can also mean very little if the assumptions made are unrealistic or the experiments performed are not relevant (*Johnston et al., 1998*). The outcome of risk assessment can be qualitative (e.g. "It is considered unlikely that ...") or quantitative (e.g. "The likelihood of a lifetime cancer risk is estimated to be < 1 in a million").

Should novel GM cultivars be treated in the same way as traditionally produced cultivars?

There was considerable debate over this point in the case of The Alliance for Bio-Integrity vs Donna Shalala in the District if Columbia, USA. In the documents submitted by plaintiffs in support of reply to defendants' opposition to plaintiffs motion for summary judgement (Docket No. 98.1330(CKK)), a number of Administrative Records were exhibited from US



Government Officials to Dr James Maryanski of the US FDA, commenting on the "Statement of Policy: foods from Genetically Modified Plants". These highlight the level of disagreement between scientists working on the regulation of these novel foods:

Dr. Linda Kahl, PhD, of the US FDA Office of Compliance stated:

"The processes of genetic engineering and traditional breeding are different, and according to the technical experts in the agency, they lead to different risks. There is no data that addresses the relative magnitude in the risks – for all we know, the risks may be lower for genetically engineered foods than for foods produced by traditional breeding. But the acknowledgement that the risks are different is lost in the attempt to hold to the doctrine that the product and not the process is regulated".

Dr. Louis J Pribyl, PhD, FDA Office of Microbiology stated:

"The document is inconsistent in that it says (implies) that there are no differences between traditional breeding and recombinant, yet consultations and premarket approvals are being bartered around, when they have not been used for foods before. In fact the FDA is making a distinction, so why pretend otherwise.

"The unintended effects cannot be written off so easily by just implying that they too occur in traditional breeding. There is a profound difference between the types of unexpected effects from traditional breeding and genetic engineering, which is just glanced over in this document. This is not to say they are more dangerous, just guite different, and this difference should be and is not addressed".

Dr. Mitchell J Smith, PhD, of the FDA Office of Microbiology stated:

"My general conclusion is that this issue turns the conventional connotation of food additives on its head. It also conveys the impression that the public need not know when it is being exposed to 'new food additives', for lack of a better descriptor.

"The statement 'organisms modified by modern molecular and cellular methods are governed by the same physical and biological laws as are organisms produced by classical methods' is somewhat erroneous because in the former, natural barriers of breeding have been broached".

Dr. Gerald B. Guest, DVM, Director of the FDA Centre for Veterinary Medicine stated:

"As you state in the Notice, the new methods of genetic modification permit the introduction of genes from a wider range of sources than possible by traditional breeding. The FDA will be confronted with

Although certain areas of possible hazard associated with GM technologies can be identified in a qualitative manner it is not possible to accurately quantitate them

Under the circumstances where hazard assessment is not quantifiable, risk assessment becomes "opinion"

new plant constituents that could be of a toxicological or environmental concern. The Notice further describes unintended or pleiotropic effects that pose unknown safety concerns. It has always been our position that the sponsor needs to generate the appropriate scientific information to demonstrate product safety to humans, animals and the environment."

Dr. Guest goes on:

"Unlike the human diet, a single plant product may constitute a significant proportion of the animal diet. For instance 50-75 percent of the diet of most domestic animals consists of field corn. Therefore, a change in nutrient or toxicant composition that is considered insignificant for human consumption may be a very significant change in the animal diet."

and

"Residues of plant constituents or toxicants in meat and milk products may pose human food safety problems. For example, increased levels of glucosinolates or erusic acid in rapeseed may produce a residue problem in edible products".

The comments above came from senior scientists experienced in the regulation of foods and medicines. When such a level of disquiet and range of opinion exists among scientists, it further emphasises the need for extreme caution and rigorous toxicology in introducing this technology.

This is important because there is a substantial body of scientific opinion which contests that the risks that GM foods pose are qualitatively different than from foods produced by conventional breeding, thus potentially representing new hazards, or known hazards for which quantitative information is not available. An example is in the use of antibiotic resistance 'marker' genes in GM crops. The hazard of transfer of these genes to pathogenic bacteria is identified, but the probabilities of occurrence are virtually unknown from experimental data, making risk assessment more a matter of opinion than rigorous assessment.

Potential sources of hazard from genetically modified crops, with particular reference to glufosinate tolerant corn T25.

Acute toxicity

This is generally accepted as being a low order risk and, in general, toxicity testing by animal feeding studies does address the problem. The 14 day feeding trial performed for AgrEvo and reported in the document 'Glufosinate tolerant corn T25-EC90/200' ($part\ C,\ p46-47$) appears, from the limited information provided, to adequately address this aspect.



Chronic low dose toxicity

The question assessed in this section is "What are the likely health effects of widespread and irreversible subtle compositional changes to the human food chain?" The members of society most at risk to damage from chronic low dose toxicity from GMO foods, are in my opinion, likely be those who are still undergoing development, that is the foetus and the infant.

There is no evidence that any studies have been performed to assess developmental toxicity, either generically or specifically, for the AgrEvo maize. Specifically, the study in the document 'Glufosinate tolerant corn T25-EC90/200' (part C, p46-47) mentioned above, does not give information about the age and weight of the animals used. In addition it had a duration of only 14 days. It is unlikely that immature animals were used because it would have been highlighted. Furthermore, the results indicate that 'body weights were unaffected by treatments', which suggests that adult animals were being treated. Thus it is my opinion that this study does not consider developmental toxicity and cannot be regarded as having any relevance for long-term low-dose toxicity.

To date, the only research in the public domain to have addressed developmental toxicity in an animal GM feeding study was performed by the group of Dr. Arpad Pusztai PhD at the Rowett Institute in Aberdeen, which was successful in winning a Europe-wide competitive tendering process against 26 other applicants, for £1.6 million from the Scottish Office. This money was earmarked specifically for the development of sound protocols for the hazard assessment of novel GM foods by animal toxicity testing. As of July last year, this work had not been published in a peer-reviewed journal but had received considerable public attention. It was understood that aspects of it, addressing unexpected histological changes in the gastro-intestinal tract, had been submitted for peer review. That paper has, in fact, been subsequently published in the Lancet (Ewan and Pusztai, 1999).

That group examined the effect of potatoes that had the gene for the expression of the snowdrop (GNA) lectin introduced to increase their resistance to nematode attack. Dr. Pusztai, who is recognised as a leading expert on lectin protein chemistry, had studied the GNA lectin for over seven years and it had been selected because of its known low mammalian toxicity. The group found that there was organ weight loss and reduced lymphocyte responsiveness in the group of rats fed the transgenic potato but that there were no significant differences in control animals given parent stock potatoes with a dietary 'spike' of GNA lectin. These effects were unexpected and the cause is not known and needs to be the subject of additional research. However, it does emphasise the fact that it is irresponsible not to look for such effects. The only other information in the public domain on feeding studies with GMO foods used mature rats which had stopped laying down muscle and which were on very high protein diets.

Allergy

There is no effective biochemical, *in vitro* or *in vivo* animal model, for allergy prediction in humans. Before widespread use of GM crops in the food chain is allowed, actual testing for allergy in humans is, in my opinion, necessary to try to minimise the chance of adverse effects on general release, which is likely to be irreversible. AgrEvo have addressed this aspect of the toxicology (in the document 'Glufosinate tolerant corn T25-EC90/200' (part C, p46)) by looking for glycosylation motifs in the phosphinothricin acetyltransferase (PAT) protein sequence. They did not find any and concluded that 'since some allergens are glycosylated … [and] … it is very unlikely that the PAT protein will be glycosylated in plants'. Thus no direct allergy testing appears to have been performed.

Animal Feed

The hazards of genetic modification of crops with respect to animal feed are poorly understood. To the best of my knowledge, no specific hazard with respect to human health as a result of farm animals being fed GM crops has been identified, with the exception of the use of antibiotic resistance marker genes. The T25 corn in question contains the ampR Ampicillin resistance gene of pIJC18 from *E. coli*, a gut bacterium which expresses β-lactamase in bacteria. The risks of horizontal gene transfer from this part of the construct (*Ho et al., 1998*), are not addressed in the documentation that I have seen and will be addressed by another witness. There are other reasons for being wary of the use of GM crops for such purposes, not least for animal welfare and human health reasons, as reported in the statement of Dr. Guest of the US FDA above. Single plant sources can be a much greater component of an animal diet than for a typical human one. Animals may consume plant parts and plant by-products that are not consumed by humans.



HORIZONTAL GENE TRANSFER

Professor Terje Traavik

PERSONAL EXPERTISE

I am Professor and Head, Department of Virology, School of Medicine, University of Tromsø, Norway, and Scientific Director at the Norwegian Institute of Gene Ecology, Tromsø. I have been leading and executing research within the fields viral ecology (arboviruses), viral carcinogenesis and gene ecology for the last 27 years. I have used recombinant DNA/genetic engineering methods in my research for the past 17 years. Through these

years I have contributed approx. 170 international research articles and book chapters.

QUESTION POSED TO WITNESS I have been asked to advise on what was in the public domain and available on the subject of horizontal gene transfer in July 1999.



SUMMARY

- > Horizontal gene transfer from GMOs is a real option. Such events may result in extensive and unpredictable health, environmental and socio-economic problems. Under some circumstances the consequences may be catastrophic.
- > Our present level of knowledge about horizontal gene transfer is inadequate for reliable risk assessments. This applies to GMOs in general as well as to any particular GMO.
- > Currently available methods to detect and monitor horizontal gene transfer from GMOs are inadequate.
- > There are no ways to interfere once a horizontal gene transfer cascade has been initiated. It may take a long time even to become aware that it has been initiated.

STATEMENT OF THE WITNESS

My views are based on scientific details given in the following account – horizontal gene transfer under natural conditions: unpredictability and lack of knowledge calls for applying the precautionary principle to deliberate release of GMOs.

Risk factors and hazards

In order to carry out a genetic modification, a recombinant, genetic vector has to be constructed. The vector is intended to carry the cloned gene safely into the chosen organism and order the gene to become expressed, i.e. produce the protein which it codes for.

A vector will, in addition to the chosen gene, be composed of a number of other DNA elements. Typically, it needs a control element (promoter/enhancer) necessary for expression of the gene, and an extra gene coding for resistance to an antibiotic or another cytotoxic substance. An alternative to the highly controversial antibiotic resistance genes for GM plants, is a gene (csr-1) providing resistance to the herbicide chlorsulfon (*Bergelson et al., 1998*).

Vector DNA molecules are transferred across cell membranes aided by so-called biolistics ("gene-canon"), chemical treatment or by exposing cells to an electric field. Thereafter vector molecules are transported to the cell nucleus to become inserted (integrated) into the chromosome(s) of the recipient cell. Integration takes place at unpredictable locations in the chromosomes.

Genetic modification is different from traditional cultivation and breeding

It is often argued that genetic modification represents a more precise, but not fundamentally different, kind of breeding or cultivation. This must be rejected, because:

- Unnatural recombinations are created. Genetic material is recombined between species for which there is no, or very low, probability of natural progeny.
- · New, exotic genes or DNA sequences are introduced into unpredictable chromosomal

locations. Conventional breeding is shuffling around aberrant versions (alleles) of the same genes, while these are fixed in the chromosomal locations they have been given by evolution. Gene technology introduces new, exotic genes. Their location within the recipient cell DNA is unpredictable and with no possibility of targeting. This may result in unpredictable effects on the metabolism, physiology and biochemistry of the recipient, transgenic organism – effects not to be detected with traditional methods of control. Therefore, in the context of precision, it is even questionable whether the techniques for genetic modification of cells and organisms deserve the label "technology", since this is a word associated with predictability, control and reproducibility.

- Vectors used are efficient genetic parasites. They are gene shuttles developed to move and express genes across species borders and ecological barriers.
- Vectors are mosaics of genetic elements and sequences derived from most efficient genetic
 parasites (viruses/plasmids/mobile elements). Many of them are able to invade and insert
 their DNA into the chromosomes of any kind of cell, with possible genetic or metabolic
 harm in consequence.
- Vectors are specifically constructed to break species barriers. In transit they may pick
 up and transfer genes from new host organisms or from the genetic parasites of these.
 Such newly created genetic mosaics may then become transferred to new species, or
 recombine between them to result in pathogenic viruses with potential to infect earlier
 refractory hosts etc. During such relays, genetic rearrangements and mutations may arise
 at any given time, with unpredictable results.
- Vectors carry resistance genes which in themselves may represent new, or enhance existing, public health and environmental problems (i.e. antibiotic resistance in pathogenic bacteria or herbicide-tolerant "super-weeds").

Changes in GMOs or their products

The most serious scientifically based arguments against large scale, commercial use of the first generation GMOs are based on the unpredictability with regard to where in the recipient cell chromosomes insertion of vector DNA takes place. The consequences of insertion may vary considerably according to the precise insertional location (*Doerfler et al., 1997*). Some of the most prominent uncertainties are related to the fact that the recipient organism has received a new promoter/enhancer. These elements are governing the gene expression levels of their attached transgenes, but after insertion they may also change the gene expression and methylation patterns in the recipient chromosome(s) over long distances up- and downstream from the insertion



site. Promoter/enhancers function in response to signals received from the internal or external environment of the organism. For a GMO this results in unpredictability with regard to:

- The expression level of the inserted foreign gene(s).
- Expression of a vast number of the organism's own genes.
- Influence of geographical, climatic, chemical (i.e. xenobiotics) and ecological changes in the environment.
- Transfer of vector sequences within the chromosomes of the organism, and vertical and/or horizontal gene transfer to other organisms.

Changes in ecosystems and the environment in a broad sense

Genetic pollution from GMOs is a real option. This can be exerted by cross-pollination, unplanned breeding and horizontal gene transfer. Such events may result in extensive and unpredictable health-, environmental and socioeconomic problems.

Risk and assessment of risk

The term "risk" is very often confused with "probability", and hence used erroneously. Risk is defined as the probability that a certain event will take place multiplied by the consequences arising if it takes place. The atomic bomb makes a good basis for conceiving the contents of the term. With regard to development and commercialization of GMOs we often are neither able to define probability of unintended events nor the consequences of them. Hence, the present state of ignorance makes scientifically based risk assessments impossible. This calls for invoking the "precautionary principle".

Documented hazards and risks

During the short time period that GMOs (mostly plants) have been employed, a number of warning signals have already emerged.

1) Changes in the GMO or its products:

For a long while, genetically engineered bovine growth hormone (BGH) that was injected
into cows in order to increase milk production, was identical with its natural counterpart.
However, later on it was demonstrated by independent research that epsilon-N-acethyllysine
was substituted for lysine in the engineered hormone (Violand et al., 1994). Such amino
acid substitutions may have unpredictable consequences for the conformation and functions

Genetic pollution from GMOs is a real option. This can be exerted by cross-pollination, unplanned breeding and horizontal gene transfer. Such events may result in extensive and unpredictable health-, environmental and socioeconomic problems During the short period of time that GMO's (mostly plants) have been employed, a number of warning signals have already emerged

of proteins. Recently, indications have been published that milk from BGH treated cows may contribute to increased mammary cancer risk by enhancing the IGF-1 concentration in milk (Outwater et al., 1997; Hawkinson et al., 1998; Gebauer et al., 1998).

- Tobacco plants were genetically engineered to produce gamma-linolenic acid. Instead the
 plants mainly produced the toxic product octadecatetraenic acid. Unmodified tobacco
 plants do not contain this substance (Reddy and Thomas, 1996).
- When yeast was genetically modified to obtain increased fermentation, it was unexpectedly discovered that the metabolite methyl-glyoxal accumulated in toxic and mutagenic concentrations (*Inose and Murata*, 1995).
- When a gene from Brazil nut was inserted in soybean plants, unexpected, strong allergic reactions were recorded in nut-allergic persons who had never had any problems with soybean products. The inserted gene did not code for any known allergen (Nordlee et al., 1996).
- A bacterium (Bacillus amyloliquefaciens) was genetically engineered to produce increased levels of the amino acid L-tryptophan, which has wide spread application in tablets as a nutritional supplement. In the tablets, small amounts of a toxic, tryptophan related molecule were identified (Sidransky et al., 1994). Whether this was the cause of EMS (easinophilia-myalgia syndrome) which resulted in 37 deaths and 1500 cases of chronic neurologic and autoimmune symptoms have never been clarified, mainly because the genetically modified stock of bacteria were not available for investigations (Australian Gen-Ethics Network, 1994).

2) Environmental effects

Recently, it was demonstrated that self-pollinating GM plants may have a forced, augmented capability to cross-pollinate other plants, with a resulting transfer of inserted transgenes (Bergelson et al., 1998). The unpredictability was demonstrated by the fact that inbred, identical plants, genetically modified in separate experiments, had differing abilities to cross-pollinate other plants. Although the experiments were carried out on a single plant species, Arabidopsis thaliana, these results have general interest, also because the inserted gene (csr-1) has been introduced in various plant species as an alternative selection marker to replace antibiotic resistance genes.



GM cotton plants with inserted herbicide tolerance genes have demonstrated two types of malfunctions. In some cases the plants dropped their cotton bolls, in others, the tolerance genes were not properly expressed, so that the GM plants were killed by herbicide (*Fox, 1997*). The manufacturers blamed extreme climatic conditions, indirectly assessing opponents claims of unpredictability.

Persistence of naked nucleic acids in Nature

Free DNA in the environment

This is a key problem in connection with pre-assessments of damaging effects and includes not only the power of resistance of nucleic acids generally, but also the length of fragments which can persist for how long under what conditions (for recent review, see Nielsen et al., 1998).

The new branches of science, molecular palaeontology and molecular archaeology, show quite clearly that relatively long chains of chromosomal DNA can survive for a long time under certain conditions. There is proof of survival over thousands of years (*Pääbo et al., 1988*). Controlled biochemical studies concerning the breakdown of DNA in solution under "normal" conditions imply that DNA generally will be severely degraded, if not totally broken down, after 40-50,000 years (*reference in Morell, 1993*).

When it comes to survival of DNA under natural environmental conditions, on the whole little research has been done in this important field. Moreover, most of the reported trials have used pure, homogeneous clay and sand as the DNA recipient, and these have completely different properties from the far more heterogeneous and complex, naturally occurring soils. This is illustrated by a recently published study (*Ogram et al., 1994*) which shows the dramatically differing extents in which varying lengths of DNA fragments (from about 2 to 23 kilo base pairs, kbp) can be adsorbed by various types of soil. This work also very convincingly demonstrates how adsorption to solid surfaces can have major consequences for DNA survival, because different types of soil particles give varying degrees of protection from DNase attack. Even the largest fragments, under favourable circumstances, could be recovered intact after several weeks. Romanowski et al. (1992, 1993a and b) also showed that the type of soil is important and demonstrated the continued existence of transformable plasmids 60 days after release. Widmer et al. (1997) found that tobacco and potato transgenic plant marker gene nptII persisted in soil for 77-137 days.

Free DNA has been found in all the ecosystems (sea water, fresh water, sediments) so far investigated (*Lorenz & Wackernagel*, 1994), even though DNases are widely distributed.

Many extraction methods exist to analyse for DNA in the environment (see, for example, Torsvik & Goksøyr, 1978; Steffan et al., 1988). All told, larger amounts of DNA are extracted directly from the soil than can be achieved by extraction from the cells in the soil (Steffan et al., 1988), thus serving as direct evidence of the occurrence of free DNA. Investigations also exist which

show that naked DNA molecules in soil originate from micro-organisms which are no longer present in the habitat (*Spring et al., 1992*), yet another indication that phenotypically and genetically dead are two quite different things in an ecological context.

Protection of naked DNA in Nature

Particles found in soil and sediment, such as quartz, feldspar and clay minerals, as well as those suspended in naturally occurring water, have the ability to bind both organic and inorganic material. When DNA is bound to some of these types of particles, it is protected from being broken down and must therefore be looked upon as a source for the transfer of genetic information (Lorenz & Wackernagel, 1994, Nielsen et al., 1998).

Uptake of nucleic acids in the mammalian organism

In many biological systems, it has been demonstrated that mammalian cells can take up foreign DNA in a manner that permits biological activity. This is, of course, precisely the basis for transfections in cell cultures, genetic modifications of plants and animals, for gene therapy and for DNA vaccination. However, are the epithelial surfaces in the gastrointestinal or respiratory tracts of the mammal impervious barriers to the uptake of introduced foreign DNA, or can such DNA penetrate into the organism from the extensive epithelial surfaces in the body?

These questions have recently been evaluated (*Schubbert et al., 1994*). Mice were pipette-fed with circular or linear double-stranded M13 bacteriophage DNA, or this was added to the feed pellets. Sensitive hybridisation methods and PCR were then used to identify M13 sequences in the faeces and blood.

The results showed that 2-4% of the introduced M13 DNA could be identified in the faeces and 0.01-0.1% in the blood, where the DNA was found in both the serum and the cell fraction. Separate fragments measuring up to 1692 bp out of the 7250 bp total size of the M13 genome were found up to 7 hours after uptake. No difference was found between circular and linear DNA.

In more recent work, the same research group demonstrated that ingested DNA under some circumstances may be taken up from the intestines of mice, inserted into chromosomes and vertically transmitted to offspring (*Doerfler et al., 1997; Schubbert et al., 1997; Doerfler et al., 1998*).

The authors assume that other types of DNA would behave in the same manner, but they add that this must be investigated experimentally. These observations raise several challenging questions:

To what extent can DNA which is taken up from the intestines be internalised by cells in various organ systems? Can foreign DNA in the blood stream of a pregnant female pass across the placenta and enter the foetus (*Doerfler et al., 1998*)? Can foreign DNA which is taken up from the intestine contribute to mutagenesis and oncogenesis?



The genomes of the polyoma viruses (SV-40, BK virus, mouse polyoma, etc.) are small (ca. 5 kbp), circular, double-stranded DNA molecules which are able to function as expression vectors in mammalian cell cultures. Transfection of cell cultures with naked, genomic polyoma virus DNA results in infection with production of virus particles. In a series of viral infection trials carried out at the Department of Virology in the University of Tromsø, one of the controls was naked genomic virus DNA injected intravenously into rabbits and mice. Based on what was known from the literature, and so-called conventional wisdom, it was assumed that DNA under such circumstances would be rapidly broken down by nucleases and, in practice, be devoid of biological activity. It was therefore most surprising, as well as being a lesson to us, that both viral genetic expression and full, productive viral infection were indeed initiated in the animals (Rekvig et al., 1992; Fredriksen, 1993; Fredriksen et al., 1994).

That nucleic acids are taken up and have biological activity is obviously not a general phenomenon. Throughout the history of evolution, animals and people have been receiving foreign DNA from other animals and plants through uptake of nutrients and breathing of air. The problem is just, yet again, that we know that in the case of a few, perhaps rare, combinations of nucleic acids and circumstances, nucleic acids will be able to be taken up from the mucous membranes. However, we have no knowledge of the sequences, structures or environmental factors which can contribute to such stability. Nor can we therefore, at the present time, predict what type of DNA will avoid rapid breakdown in the organism and which environmental factors may contribute to this.

Horizontal transfer of nucleic acids and genetic information

For any given gene construct or GMO which is released, or escape, to the environment, the state of our present knowledge neither allows pre-assessment of probability nor consequences of horizontal gene transfer. Hence, according to the definition of risk, risk assessments become impossible at the moment. Only extensive research on the mechanisms of horizontal gene transfer and on ecosystem interconnections can change this situation.

Horizontal (lateral) gene transfer is defined as non-sexual transfer of genetic information between genomes (*Kidwell*, 1993). The expression is generally used about transfer between core genomes in different species, but it can also be applied to genetic transfer between different organs in the same or different species. Transfer with the aid of parasitic species or symbionts to host species can also be included (*Timmis & Scott*, 1984).

Horizontal transfer is thus distinct from the ordinary form of gene transfer which takes place vertically from parent to offspring. There is now good evidence that horizontal transfer takes place for both genomic (usually non-mobile) sequences and sequences derived from transposable genetic elements or mobile introns.

Horizontal transfer of genes is now an indisputable fact, and the most important question that remains is whether such transfer takes place at a speed that significantly affects evolution

Horizontal transfer of genes is now an indisputable fact, and the most important question that remains is whether such transfer takes place at a speed that significantly affects evolution.

The debate has mainly concerned horizontal transfer of entire genes, but for *E. coli*, *Streptococcus* and *Neisseria* species it has been shown that far shorter elements are stably transferred and can give rise to mosaic genes *(review in Lorenz & Wackernagel, 1994)*. There are strong indications that this takes place in eucaryotic organisms, too, exemplified by cytochrome c in plants and betaglobines in mammals *(Syvänen, 1994)*. Theoretically, shorter DNA sequences will, beyond our knowledge, be able to contain control elements for expression of genes (e.g. promoters or enhancers) which can change the amounts of some gene products in the recipient, perhaps with substantial biological consequences.

Barriers to horizontal transfer

For horizontal transfer to take place, genetic material has to overcome at least two types of hypothetical barrier (*Heinemann, 1991*), an introduction barrier and an establishment barrier. These barriers ought to make contact between genetic donors and recipients difficult, degrade genetic material, exclude foreign material from replication and/or segregation processes, and prevent the expression of genes which are required for inheriting transferred molecules. It is clear that the introduction barriers are often broken and that a network for genetic exchange between organisms exists.

In addition, we know that establishment barriers, too, may be broken, but we do not know the mechanism and can therefore not guard against such highly undesirable occurrences.

General mechanisms for horizontal gene transfer

The occurrence, and mechanisms for, horizontal (lateral) transfer of genes have been remarkably little studied, especially in eucaryotic cells and organisms. However, there are some brief and useful reviews of the topic (Heinemann, 1991; Landman, 1991; Bogosian & Kane, 1991; Powers et al., 1991; Thakur et al., 1991; Kidwell, 1993; Lambowitz & Belfort, 1993; Dreiseikelmann, 1994; Capy et al., 1994; Lorenz & Wackernagel, 1994; Harding, 1996; Wöstemayer et al., 1997; Nielsen et al., 1998).

It is usual to distinguish between the following general mechanisms:

• Transduction: Foreign genetic material that has been included in a virus genome can be transferred to a new cell which is infected by the virus, a bacteriophage in bacteria (review in Dreiseikelmann, 1994) or, for example, a retrovirus in mammalian cells.



- Conjugation: Genetic material on bacterial plasmids which also steer the transfer process itself. Requires close physical contact between two bacteria cells. Both *E. coli* and the plant pathogen Agrobacterium tumefaciens can transfer DNA to eucaryotic species (*review in Heinemann, 1991*). The conjugation phenomenon is partly responsible for the development of antibiotic resistance in bacteria.
- Transformation: Often called transfection when it concerns mammalian cells.
 Free DNA is taken up in cells.
- Transpositioning: Genetic information is moved to other parts of the genome, to plasmids or to other genomes with the help of what are called transposable or mobile elements. Various forms of these elements have been discovered in practically all the species that have been investigated in the three kingdoms (archebacteria, procaryotes and eucaryotes).

Horizontal transfer of genes in procaryotic micro-organisms

The horizontal transfer of genes between several distantly related procaryotes is very convincingly documented (Levy & Miller, 1989; Sprague, 1991; Mazodier & Davies, 1991; Maynard Smith et al., 1991; Dreiseikelmann, 1994; Syvänen, 1994; Lorenz & Wackernagel, 1994; Nielsen et al., 1998).

1) Relative importance of DNA uptake mechanisms

Horizontal transfer of accessory DNA elements is assumed to be one of the most important factors in the adaptation to the environment of the microbial community and for the evolution of new metabolic pathways within the community. Plasmids are elements which are assumed to constitute important parts of the horizontal gene stream. Plasmid genes often code for products that give selective advantages in stressed or hostile environments (*Trevors et al., 1987*), and plasmids are able to undertake autonomous replication and promote their own transfer. With the help of simple conjugation, many trials have produced bacteria which have new biodegradable properties (see, for example, Latorre et al., 1984, Smets et al., 1993).

Transduction is another potentially important mechanism for gene dispersal in the environment. The possibility of transferring both chromosomal and plasmid-localised genes between bacterial populations on plant surfaces and in aquatic environments using this mechanism has been demonstrated in many studies (*Ogunseitan et al., 1992; Zhou et al., 1993; Dreisekelmann, 1994*), but has not been attributed any importance in a risk context.

Biotechnological solutions to problems in agriculture often entail the release and dispersal of GMO's to surfaces on crops and other plants. Recent studies have shown not only genetic exchange by transduction between bacterial populations on the same plant, but also, transfer to other plants when this is made possible by closely-spaced planting (*Kidambi et al., 1994*).

The efficiency of horizontal gene transfer between microbes is dramatically illustrated by the occurrence of antibiotic-resistant genes, even in apparently non-contaminated biotopes and ecosystems (see Davies, 1994; Kruse, 1994; Anderson & Sandaa, 1994; Kruse & Sørum, 1994; Kruse and Jansson, 1997). Recently, Nikolich et al. (1994) have demonstrated naturally occurring transfer of a tetracycline-resistant gene between intestinal bacteria (Bacteriodes spp. and Prevotella spp.) from domestic livestock (pigs, cattle, sheep) and man.

Transformation means that free DNA is directly taken up by bacteria, and the term natural genetic transformation is used to differentiate uptake occurring under natural conditions from artificial laboratory procedures. Natural genetic transformation can be looked upon as the most widespread transfer mechanism, since the other known mechanisms are steered by genes located on plasmids or transposons (conjugation), or on bacteriophages (transduction).

It has been shown that extracellular DNA is present in natural environments and that such DNA can be taken up in bacteria. Simulation trials have demonstrated that many bacteria can achieve competence for DNA uptake under conditions which can arise in natural habitats. (Competence is defined as the ability to take up free DNA from the ambient medium). These observations are compatible with bacterial gene transfer performed by free, naked DNA taking place on a significant scale (*Lorenz & Wackernagel*, 1994).

2) Genetic transformation under natural conditions

Competent bacteria cells must exist in the environment (the habitat), and a number of specific conditions must also be satisfied: i) the bacteria must come into contact with DNA; ii) the DNA must have a minimum length; iii) specific cations must be present in optimal amounts; iv) the DNA must follow an internalisation route which gives biological meaning.

These rules apply for chromosomal DNA; investigations of free plasmid DNA have not been reported. Model studies, however, show that plasmid DNA is liberated from bacteria together with chromosomal DNA (*Lorenz et al., 1991*). Such liberated plasmids maintain their transforming activity in soil (*Romanowski et al., 1992, 1993*).

· Transformation in a microcosm

A number of experimental studies have been performed to investigate genetic transformation in a microcosmos set-up using samples taken from natural habitats such as soil and water. Most of the studies concern the ability for transformation in bacterial strains within a microcosm. The obvious, main conclusion to be drawn from these studies, which have taken place over many years, is that in no instances are there any specific model organisms which are representative for other naturally transformable bacteria in a given habitat (*Lorenz & Wackernagel, 1994*). Naturally occurring genetic transformation has been proved in all kinds of habitat, but otherwise the results are often highly contradictory. This probably only reflects that it is



impossible to generalise results achieved with an ecotype of a given species of bacteria (Leff et al., 1992).

· Transformation on solid surfaces

It has been shown that many bacteria are transformed by DNA bound to sand and clay particles (review in Lorenz & Wackernagel, 1994). For some species, the transformation is more effective than with corresponding DNA concentration in solution, and for others it is less effective.

· Complexity of ecosystems.

Most microcosmos studies have been concerned with variations in bulk soil (*Nielsen et al.*, 1998). But real ecosystems are very complex, varying in soil composition, chemical pollution and organisms. Horizontal gene transfer is also likely to take place within the digestive systems of protozoa, nematodes, insect larvae, earthworms and other soil-inhabiting macro-organisms (*Adamo and Gealt, 1996; Daane et al., 1996; Schlimme et al., 1997*).

- Transfer of chromosomal DNA across species boundaries

 A number of studies prove that chromosomal genes can be transferred by transformation
 between species and even across higher taxonomic boundaries (Lorenz & Wackernagel, 1994).
- Plasmid transformation across species boundaries

The transfer of plasmids between cells from different species requires that the plasmid can initiate replication in many different hosts. The plasmid is taken to the nucleus of the recipient as single-stranded fragments. Reconstitution of the circular, double-stranded plasmid DNA molecules does not require sequence homology to the recipient's DNA, but, on the contrary, recombination and repairing enzymes which are compatible.

Even though special conditions are stipulated and varying restriction enzyme systems are present in different species, plasmids and transposons can be transferred between all the different parts of the procaryotic world (*Lorenz & Wackernagel*, 1994). For recombinant shuttle vectors, which are constructed to function in both procaryotic and eucaryotic hosts, the picture will obviously be both more complicated and unpredictable.

• Naturally occurring transformable bacteria

The very first attempt at transformation that has been described (*Griffith, 1928*) still illustrates ecologically important aspects. Dead, pathogenic (S-shaped) bacteria could transfer their pathogenic properties to non-pathogenic (R-shaped) bacteria following injection into mice.

Naturally occurring genetic transformation has been proved in all kinds of habitat but otherwise the results are often highly contradictory. This probably only reflects that it is impossible to generalise results achieved This illustrates the difference between phenotypic and genetic death, and that genetic life may have dramatic ecological consequences in the event of transformation or other means of genetic transfer.

Many habitats exist which have a high potential for gene exchange by transformation. One example is tubers which contain large quantities of rhizobacteria. These are able to develop natural competence and transport free DNA into their cytoplasm. Other examples of localities with high concentrations of bacteria which may favour transformation of free DNA, or gene transfer by cell contact, are the intestines of both vertebrates and invertebrates, protozoans and surficial, mesophyllic and intracellular space in plants (*Lorenz & Wackernagel*, 1994).

3) Estimates of transformation frequencies in the environment

Comparisons of the transformation frequencies in microcosm experiments, which simulate natural habitats, and those attained under optimised laboratory conditions reveal many minor and major differences. The differences between transformation frequencies recorded in microcosm experiments and those truly occurring in Nature may be at least as great. The reason is that the variations in both environmental parameters such as temperature, pH, supply of nutrients, minerals, particle composition, contamination, total number of micro-organisms and the composition of populations, as well as the barriers to transformation cannot be simulated in the microcosm. Nonetheless, the many data that have been amassed in recent years strongly imply that there may be high transformation frequencies in some natural habitats (Lorenz & Wackernagel, 1994; Nielsen et al., 1998).

4) Barriers to transformation

Physiological, genetic and microcosmic studies have shown that transformation frequencies vary both within and between species (*Lorenz & Wackernagel, 1994; Nielsen et al., 1998*). Some varying factors and mechanisms exist within the cells and the environment which may limit the efficiency of transformation in natural bacterial ecosystems, and thus constitute barriers to transformation. These include the effects of restriction enzymes in bacteria, the occurrence and level of competence that occurs in nature and various environmental factors. However, a number of important questions remain completely unanswered.

- Which factors decide the transformation frequencies in a given habitat?
- What decides the actual concentration of transforming DNA in a given habitat?
- What is the difference between transforming and non-transforming DNA, and is the difference general or related to one or more specific recipients?
- · Is access to nutrients decisive?



5) Importance and extent of horizontal gene transfer between micro-organisms

The horizontal transfer of genes plays a positive role by giving genetic flexibility and adaptability to a shifting environment and also providing the basis for species formation by sexual isolation. Horizontal transfer between species offers possibilities for spreading environmentally adapted genes to more species within the same environment.

The extent of horizontal gene transfer may be far greater than the classical studies have succeeded in revealing (*Lorenz & Wackernagel*, 1994; Nielsen et al., 1998), because such studies have concentrated on demonstrating known differences in phenotypical traits over short time intervals as evidence that transfer has taken place. This only reveals the changes that are being sought, and not changes from possible horizontal gene transfer which are only apparent under specific conditions. However, phenotypical changes from such transfer in single populations may have great ecological effect over time, because the balances in the ecosystems are upset.

Horizontal gene transfer in eucaryotes

The existence of a number of conflicting species relationships has been recognised for many years without any ultimate explanation having been found because the available analytical methods have been too inexact to differentiate between alternative explanations. This situation has now changed with the arrival of modern sequencing methods and PCR. In the years to come, access to sequence data from many different organisms will permit more reliable estimates of the frequency of horizontal transfer of genes (Kidwell, 1993; Nielsen et al., 1998).

1) Genomic sequences

Recently, a number of previously suspected horizontal gene transfers between procaryotes and eucaryotes were re-examined in detail (Smith et al., 1992). Five cases were judged as probable in varying degrees, one was considered impossible, and the remainder were characterised as improbable. The probable ones concerned transfers both from procaryotes to eucaryotes, between eucaryotes and from eucaryotes to procaryotes. Eucaryotic to procaryotic transfers include transfer from plants to bacteria (Smith & Doolittle, 1992; review in Nielsen et al., 1998) and from mammalian virus to E. coli (Doolittle et al., 1989). In one of these cases, there are strong indications of horizontal transfer having taken place through several stages, first from an animal to a bacterium, thence continuing between several species of bacteria. This concerned the Fibronectin type III (Fn3) domain, which is common among animal proteins, but not plants and fungi, and usually not bacteria either (Bork & Doolittle, 1992).

As mentioned above, several cases of horizontal transfer from procaryotes to eucaryotes have been documented (Smith et al., 1992; Nielsen et al., 1998), whereas there are few credible examples of such transfer of genomic, non-mobile sequences between eucaryotes. Those which are reported, and which are accompanied by strong indications, seem to involve viruses,

There are strong indications that horizontal gene transfer takes place between eucaryotes across species, family and kingdom boundaries. It appears to be extremely important to have this relationship investigated because even exceptionally rare cases may have far-reaching, unpredictable and serious ecological consequences

pseudogenes or multigenic families (*Kidwell, 1993*). However, there is at least one well documented case, a quite "recent" transfer of retrovirus sequences from apes to one of the close ancestors of the domestic cat. There also exists a well-documented case of transfer between a gallinaceous bird and rat (*Kidwell, 1993*).

2) Mobile elements and introns

Eucaryotic transposable elements (TE) can be placed in two classes, class I and class II according to their transposing mechanisms (*Kidwell, 1993*):

3) Biological vectors for horizontal gene transfer

In bacteria, gene transfer regularly takes place across species boundaries, based on transformation, conjugation, transduction and transpositioning (Mazodier & Davies, 1991) and compared with procaryotes it is assumed that stronger barriers to gene transfer exist in eucaryotes (Kidwell, 1993). Nevertheless, there are strong indications that horizontal gene transfer takes place between eucaryotes across species, family and kingdom boundaries (Stachel & Zambryski, 1989).

It appears to be extremely important to have this relationship investigated because even exceptionally rare cases may have far-reaching, unpredictable and serious ecological consequences (Syvänen 1984, 1985, 1987a and b, 1994).

In a few situations, such as in a close symbiotic relationship, physical proximity between donor and recipient may permit horizontal transfer (Zambryski et al., 1989). A striking example here is the distribution of related nuclear group I introns among lower eucaryotic organisms with a pronounced symbiotic and phagocytic way of life (Vader et al., 1994; Brul & Stumm, 1994). However, in most situations a genetic vector has to be envisaged in order to achieve a horizontal transfer.

This is, of course, what gene technology is very largely concerned with and dependent upon – genetic shuttles (vectors) to transport DNA sequences between species which, in terms of reproduction, are completely isolated from one another (*Kidwell, 1993*). It surely should not be too surprising if corresponding mechanisms were active under natural conditions.

A list of the possible, naturally occurring vectors would have to include:



- Transposable elements (TE)
- Plasmids
- Extra chromosomal nucleated ribosomal DNA
- Viruses
- Chlamydias
- · Mycoplasmas
- Spiroplasm
- Rickettsias
- Bacteria
- Protozoans
- Nematodes
- Fungi
- · Parasitic arthropods

It is easy to imagine a genetic element that passes like a relay baton from one type of vector to another while in the course of the race being caught by and established in the host organisms of the vectors. Such 'batons' are particularly obvious in connection with transposable elements (TE) (Finnegan, 1989).

An example of a recent TE invasion exists in connection with the P element of *Drosophila melanogaster* (fruit fly). This invasion began in laboratory strains in the USA during the 1950's and has since spread rapidly to banana fly populations throughout the world (*Capy et al., 1994*). Rickettsia-like organisms are assumed to be involved as genetic vectors in this horizontal gene transfer of TE between different *Drosophila* species. Horizontal spreading of transposable elements between such distantly related organisms as banana flies, nematodes and fungi has been proved, but the underlying mechanisms are not known (*Capy et al., 1994*).

Transposable elements of the Tc1/mariner family are found in many species of the animal kingdom, including humans. The widespread distribution of this transposon family seem to be the result of horizontal transfer between a high number of species. The mariner transposons easily cross species and kingdom borders, i.e. from fish to mouse and human cells (*Luo et al., 1998; Ivics et al., 1997*) and from a nematode into human cells (*Schouten et al., 1998*).

Some viruses, for instance, within the ecological grouping arboviruses, easily cross species boundaries. Some arboviruses may even alternate between vertebrates and blood-sucking arthropods as an essential part of their life cycle (Anderson, 1970; Traavik, 1979; Syvänen, 1987). Proof exists that retroviruses are able to function as transfer vectors between different mammals (Duesberg, 1983) and that host-cell mRNA can be carried from one vector to another (Ikawa et al., 1974). Retroviruses may contribute significantly to transduction of genetic material between

The extent of horizontal gene transfer may be far greater than the classical studies have succeeded in revealing, because such studies have concentrated on demonstrating known differences in phenotypical traits over short time intervals ... this only reveals the changes that are being sought and not changes from possible horizontal gene transfer which are only apparent under specific conditions. However, phenotypical changes from such transfer in single populations may have great ecological changes over time, because the balances in the ecosystems are upset

mammalian cells and individuals (Coffin, 1990; Milot et al., 1994; Stevens and Griffith, 1994).

There are good indications that baculoviruses, which are now used in recombinant versions as laboratory tools and for combating insect pests, have spread mobile elements between various insect host species (Miller & Miller, 1982; Friesen & Nissen, 1990; Jehle et al., 1995). Some baculovirus species have proved to contain transposable elements from their host organisms. The transposable elements can, of course, have picked up foreign genes from, for example, a GMO or a plasmid which was located in a particular host species (Capy et al., 1994). In this connection it is also worth noting that baculoviruses, originally claimed to be strictly insect-related, have now been proven to infect human cells.

Very recently, total sequencing of the *Chlamydia trachomatis* genome has brought strong indications that this intracellular parasite has picked up a high number of genetic elements from eucaryotic host organisms (*Stephens et al., 1998*).

Do pollution and other environmental changes affect the horizontal transfer or other negative impacts of naked DNA?

Very little work seems to have been done with regard to how xenobiotics may interfere with horizontal gene transfer under natural conditions or in microcosm and other types of controlled experiments.

Xenobiotics are, literally, compounds that are alien in the biosphere. Xenobiotics can be defined as compounds which people release into Nature in concentrations that create undesirable impacts. Xenobiotics include many pesticides, heavy metals and organic chemicals.

Different xenobiotics have properties and biological activities that enable us to envisage at least two different sets of possible impacts on the fate of naked DNA in an ecosystem:



- Some xenobiotics can act as mutagens (this applies to radioactive substances, polluting
 industrial chemicals and plant protectants). Mutagens can result in naked DNA that escapes or
 is released having its sequence or structure changed. This, in turn, can affect the possibilities
 for DNA uptake in cells and organisms, horizontal transfer and long-term establishment in
 the ecosystems in ways which are totally unpredictable for us. Kipling & Kearsey (1990)
 have reported examples of minor changes in a DNA sequence altering the host spectrum
 of a transferable genetic element.
- Some xenobiotics can affect cell membrane and/or intracellular functions in ways which can very well be thought to influence the ability of cells to take up and horizontally transfer naked DNA. This concerns the structure of cell membranes and the content of both surface receptors and transport canals, and also, for intracellular signal conversion and gene expression. For instance, xenobiotics which mimic hormones or affect the local conditions in the organ systems of mammals (e.g. respiratory passages) may change the possibilities for both uptake and establishment of foreign nucleic acids in animals and people.

IMPACTS ON THE SOIL

Dr. Max A. Turner and Dr. A. Neil Macgregor

PERSONAL EXPERTISE

I am Max A. Turner, a soil scientist, at the Department of Soil and Earth Science, Massey University, Palmerston North, New Zealand. I graduated M.Agr.Sc (1st class) from Massey University (New Zealand) in 1965 and PhD from the University of Minnesota in 1969. I have been employed at Massey University as a lecturer and researcher since 1970. My area of expertise is in soil fertility, soil-plant relationships and fertilizer use in agriculture and horticulture. I consult within New Zealand and offshore on aspects of soil husbandry and plant nutrition.

I am A. Neil Macgregor, a soil microbiologist, at the Department of Soil and Earth Science, Massey University, Palmerston North, New Zealand. I graduated M.Sc. from University of Otago, New Zealand in 1961, and PhD from Cornell University, Ithaca, New York in 1968. My primary teaching and research responsibility at universities in USA and New Zealand (Massey University) have been soil biology, biochemistry and microbial ecology.

OUESTION POSED TO WITNESSES

We have been asked to advise on what was in the public domain and available on the subject of soil implications from the use of Liberty^(TM)-linked technology, specifically the growth of an experimental maize crop at Walnut Tree Farm, Lyng, Easthaugh, Norwich.



SUMMARY

- > Many significant, soil science related issues have not been addressed in the crop trial at Walnut Tree Farm.
- > Omissions in this case of appropriate soil investigations ensures that potential hazards to ecosystem integrity, resulting from "Liberty(TM)-linked crop technology" cannot be identified or acknowledged.
- > We make the case for soil information about the introduction of novel plant genes and about the effects of glufosinate (phosphinothricin) on soil biological processes.
- > A summary of some of the pertinent, existing, literature on GM crops and glufosinate suggest there is real reason to be concerned about side effects of their use on natural soil processes.

STATEMENT OF THE WITNESSES

Introduction

Our evidence is formulated in relation to what may be called the Liberty^(TM)-linked technology. As we understand this technology, a "glufosinate-tolerant" maize crop is grown (in this particular instance at an experimental site, on a commercial farm) so that the bioherbicide phosphinothricin (*The Merck Index 12th edition, entry 7945*) can be used to control the growth of all other undesirable plants (weeds).

We understand that one of the main rationales for conducting this crop trial is to research, not only the growth and yield of the crop, but also to investigate any major environmental impacts and/or side effects that might arise from growing crops in this unorthodox way. In this respect the technology is new and relatively unresearched and lots of potential problems could occur during, and after, the growth of the crop itself.

To be fully sustainable this method of cropping obviously needs to be shown to be capable of producing the same, or better yields of high quality produce, as is done with more conventional methods; use lower inputs of herbicides and pesticides; and have minimal disturbance to other components of the ecosystem (loosely referred to as the "environment" in some of the published literature). It is however our contention that many soil science issues are not addressed in this type of trial and we make a case for why omitting such investigations is potentially hazardous to the integrity of the ecosystem at large.

We see two main threads of concern about the risk of using GE crop technology packages to the wider environment, more particularly the soil resource, and especially the living organisms within the soil. One thread relates to the crop itself and the other to the herbicide used as part of the package. We address both, but try to distinguish the separate issues. We are also cognisant of the irreversibility of any impact of new technology such as GM crops on soil. So if any major effect on soil were to derive from GM technology it would be self-replicating, possibly with amplification and very likely irreversible.

Genetic Material

In this section we present factual and speculative information about the known and unknown effects of growing GM crops, such as maize, on the soil, soil inhabitants and soil processes. Where possible we show existing literature to support our claims.

Soil is NOT inert - it contains living and non-living material

Some of the organic material comes from roots, some from above-ground plant parts, some from the organisms that live, breed and die in the soil to become ultimately part of its "organic matter". Organic matter is what distinguishes soil from other non-soil materials. It is the most complex material known on earth and is difficult to fully characterize, isolate or alter.

Soil contains genetic material

This genetic material (DNA) occurs within and outside living cells. It will be both free and bound with varying strength to soil constituents.

Crop residues (including stalks, roots and unharvested tops) contribute to the soil pool of genetic information

These occur whilst the crop is growing but also after the crop has been harvested as the residues are naturally processed and decomposed by soil microorganisms. The DNA material is not necessarily degraded but may become a permanent, residual and stable component of the soil fabric (*Lorenz, 1998*).

Soil organic matter has varying half-lives from weeks to years and even longer It is thought that the absorption of DNA by soil organic matter permits for a much longer "persistence" or residual effect of genetic material in the soil; much longer than simply the period over which the crop grows (planting to harvest). If bound to soil organic matter this genetic material could have longer persistence than if it remains unbound. The exact half-life would be difficult to predict but could be appreciably longer than "free" material in the same soil matrix (Wolf et al., 1994).

Soil (and soil life) is rarely monitored during GM crop experiments

We have yet to see trial protocols for GM crops that specify the grounds for evaluation of the effect of growing such crops, on soil biota (micro- or macro-organisms) and soil biodiversity.

It is our opinion that any long-term adverse, and unexpected, effects on soil organisms, or their functioning, could prove to be the greatest major hazard for the environment and a major



deterrent to their continued use as viable, economic propositions. If not included in the protocols for GM trials then this represents a potentially serious omission.

Genetic "mutations" resulting from the biological uptake of novel genetic material could affect soil and plant productivity

It is known that factors affecting the normal functioning of soil microorganisms could cause major disturbances to normal nutrient cycling (Seidler, 1994). It is our opinion that specific areas of concern are nitrification, nitrogen fixation, organic matter formation and decomposition, functioning of vesicular arbuscular mycorrhizae (fungi) and myriads of other microorganisms that fulfill essential roles in soil. Even organisms responsible for the detoxification of herbicides may themselves be affected by genetic modification.

Genetic "mutations" resulting from the uptake of novel genetic material could affect the composition of the soil-plant rhizosphere (both quantitatively and qualitatively)

The rhizosphere is the zone of contact between the plant root and the soil. It is a zone of intense microbial activity. It is possible that genetic changes to organisms in the rhizosphere could affect the normal functioning of plants, disease susceptibility and productivity of crops grown on contaminated soil (*Giovani et al., 1999*).

Genetic pollution of soil is almost impossible to rehabilitate

There are no known techniques or procedures for recalling genetic information back from the soil once it is introduced and becomes an integral part of the soil. In our opinion, to attempt to do so would destroy the very matrix of the soil and render the soil virtually sterile.

Glufosinate

In this section we present information and published views about the possible effects of glufosinate on soil organisms and implications to the functioning of soil and soil-plant relationships. We note that there does not appear to be a large volume of literature on the effects of glufosinate on soils in the published scientific literature, at this time. This probably stems from the fact that it was only registered for use in the USA in 1993 and provisional approval for use in the UK in 1991 (Glufosinate Ammonium Fact Sheet, produced by AgrEvo, the manufacturer). Hence, in our view, this would be an even more important reason to include such studies in the present Norfolk experimental site.

Briefly, glufosinate is claimed to be "persistent and mobile" according to the US EPA. It is degraded by soil organisms with a half-life varying from 3-70 days. Shorter half-life occurs in soils with a high clay and organic matter content. However, no evidence about desorption capabilities after adsorption by soil was located in the literature. One trial apparently reported residues of glufosinate in plant tissue 120 days after the herbicide was applied to the soil. Also in sandy soils, longer persistence can occur where the herbicide is not rapidly degraded.

So movement to aquifers, and drinking water, is likely to be a problem on some soils. The factsheet produced by AgrEvo also mentioned toxicity to animals (including humans) of the surfactant (AES) used in the formulations.

Further information on glufosinate is summarized below:

- Reports on effects of phosphinothricin on soil bacteria are conflicting, which probably stems from the disparate methods used in the divergent studies. However, whilst there have been few published effects of glufosinate, to our knowledge, one report stands out. It is a paper by Drs Ahmad and Malloch from the University of Toronto, Ontario, Canada (Ahmad and Malloch, 1995). The experiment they conducted was designed to determine the effects of phosphinothricin (the active ingredient in Liberty) on soil microbes. They did note that knowledge of the effect of phosphinothricin on soil microflora is "extremely limited". They also noted that it is not known how the enzyme inhibitor, isolated originally from soil Streptomyces species, affects other soil microorganisms.
- The research of Ahmad and Malloch (1995) concluded that phosphinothricin was inhibitory to soil fungi, at realistic field rates of application of the chemical. It had an even more striking effect on soil bacteria, especially those from agricultural soils as compared with those from forest soils. For the former situation, phosphinothricin completely suppressed the activity of 40% of soil bacteria and 20% of soil fungi.

The following are quotes taken directly from the paper by Ahmad and Malloch.

"The variations in the sensitivity of soil microbes to the current field level of phosphinothricin suggest that changes in the composition of soil microflora may be one of the inevitable consequences of using phosphinothricin for chemical weed control. Changing the composition of soil microorganisms can lead to a strong inhibition of plant growth under field conditions. Thus the selection of soil microbes by phosphinothricin may not be without serious consequences for the fertility of agricultural soils."

and

"That phosphinothricin can potentially alter the composition of soil ecosytems has important implications for soil fertility with respect to the balance between saprophytic, parasitic and pathogenic soil activities."

and

"Given that the degradation of organic matter in a soil ecosystem involves a multiplicity of interacting microbial activities, the marked variation observed in phosphinothricin resistance of saprophytic molds suggests a disruptive influence of phosphinothricin on microbial nutrient cycling. Furthermore the lower tolerance limit of mycoparasitic Trichoderma and the high resistance of the plant pathogen V.albo-atrum indicate a negative effect of phosphinothricin on the antagonistic potential of soils."



and finally,

"In conclusion, our data show a capacity for selection of soil microbes by phosphinothricin. A large group of soil bacteria and many fungi appear to be sensitive to the current field levels of phosphinothricin application. The outcome of these effects in terms of soil fertility and microbial diversity may be profound but difficult to predict in these in vitro studies. These considerations can become a useful part of current evaluations of phosphinothricin and related bioherbicides."

Dekker and Duke (1995), and Marshall (1998), refer to glufosinate as a "benign" herbicide that does not persist in the environment. However, in the Fact Sheet (Environmental Risks of Herbicide-Tolerant Oilseed Rape: A review of the PGS hybrid oilseed rape), issued by the UK Department of the Environment, Transport and the Regions states that:

"Although the safety and efficacy of glufosinate (indeed of all herbicides) has been fully evaluated (by PSD and MAFF advisory committees) the chief concern is that the increased use of a broad spectrum herbicide (which kills most plants) will have a deleterious impact on farmland diversity".

In sum, it is our contention that on balance, knowledge of the effects of glufosinate on soil microbiota and soil nutrient cycling is extremely fragmentary and therefore far from well understood. Any field trials that are done to evaluate the effect of glufosinate on the "environment" should be at least measuring soil factors, soil microbiological changes and nutrient cycling processes in order to establish the true effect of the weed control package on the system and its continued integrity.

Knowledge of the effects of glufosinate on soil microbiota and soil nutrient cycling is extremely fragmentary and therefore far from well understood. Any field trials that are done to evaluate the effect of glufosinate on the "environment" should be at least measuring soil factors, soil microbiological changes and nutrient cycling processes

IMPLICATIONS FOR PESTICIDE USE

Peter Beaumont

PERSONAL EXPERTISE

I am a qualified solicitor. I was senior partner in a small firm of solicitors and left the partnership in 1987 to help set up Pesticides Trust, now known as Pesticide Action Network UK (PAN UK). I am employed as Development Director of PAN UK, a science-based charity, concerned with the health and environmental implications of pesticides. My responsibilities include UK and European pesticide policy issues. I wrote the book "Pesticides, Policies and People: A Guide to the Issues" (Pesticides Trust, 1992), and have written and spoken widely on pesticide-related topics and contributions at conferences and seminars, and contribute to

PAN UK's international journal Pesticides News. I hold a number of representative positions on behalf of PAN UK, including membership of the UK government Pesticide Residues Committee and the Pesticides Forum.

OUESTION POSED TO WITNESS

I have been asked to advise on what was in the public domain and available on the subject of the implications of GM crops for herbicide use in July 1999.



SUMMARY

- > Genetically modified crops, by comparison with traditional plant breeding, pose completely new problems.
- > There will be adverse impacts on the development of pest resistance and on the environment. There are concerns about gene transfer, the impacts on certain other crops and the denial of consumer choice.
- > Regulatory policy and risk assessment have not been adequately formulated. The issues of liability and monitoring and assessment have not yet been properly addressed. The control of intellectual property and impact on the local gene pool particularly in developing countries, is a concern.
- > The issues appear currently to be forced by the needs of the corporate sector before regulation, policy and the general public are ready.

STATEMENT OF THE WITNESS

The following sections address problems posed by GM crops, including the issues of pest resistance, and impacts on the environment, the problem of gene transfer and other impacts for food production, including residues, consumer choice and cheaper food.

Resistance problems

It is generally regarded as bad weed management practice to use the same herbicide, or one with the same mode of action, continually on a cropped area. Increased usage over a wide area may lead to resistant weed populations. There is concern at the incidence of resistant weed populations and the implications for weed management of herbicide resistant plants. Herbicides with a high risk for inducing weed resistance through the exertion of selection pressure will have:

- · a single target site of action
- activity and efficacy in killing a wide range of weed species (broad spectrum)
- provide long-term soil residual and season-long control of germinating weeds, or applied many times throughout the year
- and be applied frequently and over several growing seasons without rotating, alternating, or combining with other types of herbicides.

Most of the herbicides to which crops have been made tolerant may present most of these features, and glufosinate-ammonium is one of these. It seems likely that if use of herbicide-resistant crops becomes widespread, then after a few seasons resistance will become a problem. Farmers will still be locked into herbicide use to manage new and increased weed problems.

It is generally regarded as bad weed management to use the same herbicide ... continually on a cropped area. Increased usage over a wide area may lead to resistant weed populations

Impacts on the environment

Impacts of broad spectrum pesticides on biodiversity

Most of the herbicides to which crops have been made tolerant seem to be broad spectrum products – they kill a wide range of weeds. If the use of broad spectrum products increases, this will increase the negative impacts – at a time when agrochemical wisdom is counselling the use of more targeted, selective products aimed at minimising adverse effects.

The increased use of broad spectrum products will adversely affect biodiversity in general, and is likely to be a particular problem at field margins, hedges and ditches which provide habitat refuge and food for other fauna. There has been considerable concern expressed in recent reports of the indirect impacts of pesticides on bird populations. Pesticides may deplete the invertebrates that are part of their diet, and/or deplete their habitats and refuges. Many other agricultural practices besides pesticide use have affected birds, but pesticides remain one of the main causes for concern.

General concerns about Genetically Modified Organisms (GMOs)

This is a problem that the growth of herbicide tolerant crops shares with conventional pesticide usage on unmodified plants. It is very difficult to monitor the impacts of pesticides on wider aspects of the environment such as non-target species and biodiversity. However, the widespread introduction of genetically modified crops aggravates the problem. Firstly, genetically modified organisms once released cannot be recalled from the environment; and secondly such organisms are likely to reproduce. English Nature and Friends of the Earth have drawn attention to the necessary implications for research and the number and scale of topics that need to be addressed.

Specific concerns about glufosinate-ammonium

The principal environmental concern about increased use of glufosinate-ammonium would be the increased risk to water. The compound is extremely mobile, and many of the limitations on its use are designed to minimise risks to water. In general, no use is permitted between the end of September and the beginning of March, to reduce the risks of run off to water over bare ground.

The interim results of one particular research project involving cooperation between government and industry (BRIGHT – the Botanical and Rotational Implications of Genetically Modified Herbicide Tolerance) were recently presented. A number of GM and non-GM crops are being grown at different sites in crop rotations. It appeared that on some crops that were



tolerant to glufosinate-ammonium, applications were permitted under special experimental permits for usage in October and November, increasing the risks to water.

Gene transfer

One of the recognised implications of engineering herbicide-resistance into plants is the possibility of transfer of the resistance gene to other wild or weedy relatives. In farming terms this means whether intransigent, herbicide-resistant volunteer plants or 'bolters' will emerge in following crops or in other non-crop areas such as field margins or roadsides. This is a problem in a number of widely grown crops.

The likelihood of these problems depends in part on the farmer's own rotations and when herbicides are used on the crop in order to prevent bolters as far as possible. The possibility also presents itself that different genes may transfer, causing stacking or the presence of more than one gene in a wild population. Again, either of these options will reduce the farmer's options for weed control and will possibly present new weed problems.

Impacts on other crops

Herbicide drift is a concern with all use of herbicides but this concern is likely to increase if the growing of crops tolerant to broad-spectrum herbicides increases. The problems that may be anticipated are drift to neighbouring broad-acre crops sensitive to broad spectrum herbicides; or to high value crops including vegetables or GM-free crops; or of course to organic crops, with the added possibility of depriving the organic grower of organic status.

Impacts on food

It seems likely that the use of herbicide tolerant crops will result in an increase in the use of the particular herbicide or herbicides to which the crop is tolerant. It remains to be demonstrated whether the nature of the residues in the crop that will result are toxicologically equivalent to the un-modified parent herbicide residue, for instance, the residues in the crop may be less or more toxic than the parent compound; and whether the levels of residue themselves will be acceptable.

The other issue in terms of food production is that of consumer choice. The introduction of genetically modified soya was met with enormous consumer resistance, and indeed, anger, in that the consumer's right to choose non-genetically modified product was being denied.

As to the wider issue of whether herbicide tolerant crops will mean more and cheaper food, the issue is too complicated to provide a simple answer. The immediate impacts at a farm level will depend initially on the price of genetically modified seed and comparable herbicide costs, and how well the GM crop itself performs under field conditions. However, seeds and pesticides are only part of a farmer's costs, and the importance of these varies with the economics of the

particular crop grown. More important are pest and seasonal conditions and factors such as support prices and other fixed costs. The European Union has made proposals to reform the Common Agricultural Policy in order to reduce the agricultural budget, to meet the challenge of enlarging membership to the Union to include the major agricultural economies of Eastern Europe, and to reduce subsidies in accordance with the requirements of the World Trade Organisation. In these circumstances, agriculture is about to undergo many changes and the possible impact of herbicide tolerant crops is too difficult to isolate.

Regulatory issues

This section addresses some of the issues for regulators posed by herbicide tolerant crops in particular. There is currently no overarching policy agreement to cover such crops and the specific areas of risk assessment, liability, intellectual property and enforcement are concerns.

The lack of a policy

The regulation of pesticides is effected through policy, legislation, codes of practice, and policy fora. The initial problem for GMOs, whether herbicide tolerant crops or insecticide producing plants, is that there is a policy vacuum. The use of pesticides is governed by the pesticide minimisation policy, which is in turn given effect by the Food and Environment Protection Act 1985 (FEPA) and subsequent legislation, a number of established codes of practice, and established stakeholder fora. This is not yet the case for GMOs.

Although legislation, derived from EU directives, exists, there are wide policy disagreements. A number of EU states have, for different reasons, banned certain GM crops. After initially dismissing talk of a moratorium, even when advised by its own statutory advisor English Nature, the UK government then announced in 1998 a one-year moratorium on the commercial sowing of herbicide tolerant crops. This has subsequently been extended.

The UK government appeared to recognise the lack of policy direction and set up the Ministerial Cabinet Committee on Biotechnology and Genetic Modification to examine the issue. As a result, the Agriculture and Environment Biotechnology Commission (AEBC) has also announced – it will cover the use of biotechnology in agriculture and its environmental effects. Membership will include environmentalists, farmers and lay members, and regular public consultation will be required. The Commission will undertake strategic analysis, advise on guidelines, address broader issues regarding the acceptability of GM activities and identify gaps in the regulatory and advisory frameworks.

The Advisory Committee on Releases to the Environment (ACRE) has also recently been empowered to set up a Wider Biodiversity Issues subgroup.



Risk assessment and the impacts of GMOs

ACRE is a Department of the Environment, Transport and the Regions (DETR) advisory committee established to provide the relevant government departments with advice principally about the risks that might arise from the release of GMOs. For some time there has been concern at the regulatory gap between ACRE and the remainder of the normal pesticide regulatory apparatus, led by the Ministry of Agriculture, Fisheries and Food (MAFF) and headed by the Advisory Committee on Pesticides (ACP). The ACP is an independent committee that advises government ministers on pesticide issues. Overall responsibility for the impacts of GMOs on agriculture or the environment was beyond the remit of ACRE, but MAFF and the ACP declined also to offer guidance, taking the view that the matter was one for the market only and not for government.

The consequence of the lack of agreement as to who should be responsible for assessing the indirect and cumulative effects of GMOs on agriculture and the environment meant that it is unclear if, or how these factors are considered in risk assessment. Partly in recognition of these concerns it seems, government has secured an agreement with industry that it will not proceed to commercial plantings of GM crops until farm-scale trials have yielded satisfactory results that there is no significant or lasting damage to the environment.

On a European level, there has not yet been agreement about risk assessment issues and criteria to be included in amended Directives governing release. On the international level, the Convention on Biological Diversity is a UN process intended to protect biodiversity and promote the safe use of genetically modified crops and genetic resources in general is still some way from agreement. A recent meeting of the parties to the Convention (and the US, a major biotechnology producer, is not a party) failed to advance the process.

Liability

Hand in hand with the lack of policy and confusion about risk assessment goes the lack of a liability regime for GMOs. It was only in 1985 that all pesticides were regulated by the FEPA legislation. Many commentators have suggested that the current concerns about GMOs and their impacts on food, the environment and the economy (including organic farmers) require a liability regime to be established. It should perhaps be for those who propound the safety of GMOs to undertake liability in case of fault or mistake. If there is no risk as to safety, there will

[on liability] reassurances as to safety are unconvincing if those who do the reassuring ... have an interest only in the profits of development and require this to be underwritten by the public purse

be no risk as to liability. But at present, reassurances as to safety are unconvincing if those who do the reassuring – usually the biotech companies – have an interest only in the profits of development, and require this to be underwritten by the public purse.

Because GMOs once released cannot be recalled, issues of what constitutes damage, together with remoteness and quantification, need discussion and clarification. The European Commission is in the process of proposing a liability regime for chemicals and GMOs, and this initiative has been supported by the European Parliament in its review of GMO legislation. It is helpful that the government has recently called for consideration of the feasibility of, and possible criteria for, a liability regime or regimes to cover release and marketing of GMOs.

Monitoring and Enforcement

In the area of pesticides, experience has shown that problems in general have only become apparent after chemicals have passed through the regulatory system and have been disclosed by post-market monitoring or other means. It has also become clear that field results often vary greatly from laboratory results. Given the qualitatively different nature of the problems posed by herbicide tolerant crops, there is an urgent need to establish an effective monitoring and crop management regime. This does not currently exist.

For GM crops, self-regulation has been proposed by industry in the form of the Supply Chain Initiative on Modified Agricultural Crops (SCIMAC) Code of Practice. This is intended 'to establish a consistent, industry-wide approach to the supply of information relating to GM crops ... and to promote practical guidelines for the management of specific aspects of GM crops'. Industry good practice and stewardship is always helpful. However, industry management schemes have not been noticeably successful – particularly in terms of resistance management – unless there has been statutory backing and enforcement. SCIMAC presumes that farmers will be educated to carry out best practices. This is unlikely to be the case in the real world. SCIMAC makes no mention of sanctions or enforcement.

SCIMAC's Guidelines for growing newly developed herbicide tolerant crops restate good agricultural practice and exhort growers to follow established legal requirements, but state little else. There is a requirement to notify neighbours and adjoining owners; but not organic farmers or the managers of nearby Sites of Special Scientific Interest or other particularly vulnerable environments or features.

Statutory enforcement of current regulation is the responsibility of the Health and Safety Executive. Currently enforcement of the whole pesticides regime is the responsibility of approximately 80 HSE inspectors. HSE inspectors inspect on the basis of targeted programmes or complaint. This translates to a visit every ten years or so for the average farmer. It is understood that perhaps only two inspectors have been allocated the responsibility for GM



sites. Given the likely area of GM crops in due course, it is unlikely that HSE have the extra resources adequately to enforce current GM trials, recent prosecutions notwithstanding.

Intellectual property

Herbicide tolerant crops are patented. The farmer enters into a strict agreement to pay for the seed and to allow the seed company to enter on his land at any time to ensure that seed is not saved from the crop. The right of farmers to save seed is therefore abrogated. This is likely to be a problem in developing countries, where farmers will become locked into an agriculture that depends increasingly on inputs purchased from an agrochemical or seed company. Local varieties of crops will fall into disuse and the gene pool will be depleted. This would translate into a loss of crop varieties and consequently, a possible increased risk of crop failure in the future. A further issue, again more pertinent for developing countries, is that agrochemical and seed companies patent indigenous genetic resources and then sell these back to local farmers.

FARM SCALE TRIALS AND ENVIRONMENTAL SAFETY

Dr. Susan Mayer

PERSONAL EXPERTISE

I have a first class honours degree in Pharmacology, a degree in Veterinary Science and a PhD in veterinary cell biology/immunology. I am currently Executive Director of GeneWatch UK and an Honorary Research Fellow at the University of Lancaster. I have been studying and undertaking research on the science and regulation of genetically modified (GM) foods and crops for ten years.

OUESTION POSED TO WITNESS

I have been asked to advise on what was in the public domain and available in July 1999 on the subject of the farm-scale trials and the extent to which the trials will be able to answer questions of environmental safety of GM herbicide tolerant crops.



SUMMARY

- > The farm scale trials with GM herbicide tolerant maize are intended to address concerns about the potential impact of growing herbicide resistant crops on the agricultural environment and wildlife. In large part, these trials are to answer the criticisms made by English Nature and others that the approval process of GM crops takes no account of the potential for secondary effects on biodiversity.
- > The glufosinate tolerant maize being tested in the farm scale trial at Lyng, Norfolk, was given approval for cultivation in Europe under the Deliberate Release Directive (90/220/EEC) in April 1998 (EEC, 1998a). The maize is still awaiting plant variety approval before it can be sold to farmers and there is no licence for the use of glufosinate on GM glufosinate tolerant crops.
- > However, the trials have been entered into before a proper scientific review of the information required for a full assessment of the effects of GM crops on natural biodiversity is available and before levels of acceptable cross-contamination of organic and non-GM crops have been agreed.
- > In addition, the design of the trials means that considerable uncertainty will remain about the long term safety even when they are completed.
- > Despite this uncertainty, the trials are being presented to the public by the Government and the scientists involved a misleading manner by making excessive claims for the predictive ability of data provided by the farm scale trials.

STATEMENT OF THE WITNESS

Inadequacies of GM trial designs

Since the maize was approved under the Deliberate Release Directive it has been acknowledged that there are shortcomings in the environmental risk assessment system and the Directive is in the process of being revised (*EEC*, 1998b). Because the maize was assessed under the existing regulations, confidence cannot be placed in its environmental safety assessment. That the trials are taking place at all is a clear admission that not enough is known to ensure their safety.

The Department of the Environment Transport and the Regions has acknowledged that the scope of the environmental risk assessment system used to evaluated GM crops in the past is inadequate in its 1999 document 'The Commercial Use of GM crops into the UK: the potential wider impact on farmland wildlife' published by the DETR's Advisory Committee on Releases to the Environment (ACRE), (ACRE, 1999). In particular, it identifies a lack of attention to the possible effects on biodiversity. ACRE has subsequently established a sub-committee to look

into what kinds of data would be required to give a more thorough risk assessment of crops taking into account secondary effects on biodiversity and agricultural practices. However, the sub-committee has yet to report. Therefore, there is no guarantee that the farm scale trials will meet the requirements identified. A more scientific approach would have been to wait until the ACRE's sub-committee reported and design experiments accordingly.

Further haste in conducting the trials can be seen in the way they have been managed. Even though the first year of the farm scale trials was apparently intended to determine scientific protocols for following years, the sites and initial planting patterns were decided upon before the scientific steering group was appointed and this may influence the quality of data collected in 1999 and in subsequent years.

Therefore, a careful examination of the gaps in currently available data and a systematic assessment of what we need to know has not been undertaken. In addition, other research which should inform the design of farm scale trials has not been completed. In particular, MAFF are funding a research project known as BRIGHT (Botanical and Rotational Implications of Genetically Modified Herbicide Tolerance) which began in April 1999 and is due to be completed in 2003. As well as measuring weed diversity, the intention is to "provide farmers with practical guidance on the appropriate management of herbicide tolerant crops" (MAFF, 1998). This is the kind of information that should be available before moving to farmer controlled experiments.

Potential impacts of GM trials on the environment not considered

The impacts of the trials and possible gene flow and genetic pollution of organic and non-GM crops has not been resolved in advance of the trials beginning. Under organic standards, no contamination of organic crops is allowed but no standards have been set for allowable levels of cross contamination of conventional non-GM crops. (Note: This is currently being discussed as part of the revision of the Deliberate Release Directive). Therefore it is impossible to know at this time what, if any, level of cross contamination will be acceptable. Before this is known, it is impossible to quantify the threat that these trials pose. Organic farmers and organic associations have not been involved in developing the management guidelines.

With maize, pollen flow over large distances is possible (*Emberlin and Tidmarsh*, 1999), and although no wild related species exist in the UK which can be cross-pollinated, the growing of maize is gradually increasing in the UK and genetic contamination of non-GM maize is therefore more likely. Since some of this maize is grown organically and organic standards specifically exclude GM crops, a conflict of interests exists which needs to be resolved before, not after, the farm scale trials.

The inevitability of cross-pollination has been endorsed in a report from the John Innes



Centre (Moyes and Dale 1999). However, the separation distances of 50-200 metres which are being used in the farm scale trials as recommended in SCIMAC guidelines (SCIMAC, 1999), will be ineffective in preventing cross-pollination completely.

Therefore, the farm scale trials have been introduced before a scientific assessment of the data required for a full evaluation of the impacts on biodiversity of GM crops are complete and before levels of acceptable cross-contamination of organic and non-GM crops have been agreed.

There are also shortcomings in the design of the farm scale trials. I have examined the information that is available in the public domain about the farm-scale trials and attended the DETR's briefing (held on 23rd July, 1999, London) in order to understand the protocols involved. Having considered the protocols, the main doubt about the farm scale trials is whether they will provide reliable data about the potential for long-term cumulative damage to biodiversity as a result of growing herbicide resistant crops in the UK. At the end of the research programme there is every chance that there will be as many unanswered questions as there are now.

Limitations of farm scale GM trials

Limitations of farm scale trials include:

- the lack of a realistic rotation being included. The GM crop will only be grown for one year on each field. Therefore, a small change in any parameter may not (given the statistical power of the experiment) be considered statistically significant in one season but could form part of a significant trend over time as a result of the repeated used of herbicide tolerant crops. Since all major arable crops are being engineered to be herbicide resistant, in a very few years from now the typical arable crop rotation could be using herbicide resistant crops in two or even three years out of every four. At worst, the trials could pronounce the crops 'safe' when over time, serious incremental effects on biodiversity may be seen;
- how to detect possibly small but potentially significant differences reliably when management practices vary and are outside the control of the researchers;
- how to extrapolate data to assess impacts on wildlife as a whole if differences in plant and insect life are found. The relationships between numbers of insects and weeds, the species

The farm scale trials have been introduced before a scientific assessment of the data required for a full evaluation of the impacts on biodiversity of GM crops are complete and before levels of acceptable cross-contamination of organic and non-GM crops have been agreed

Despite these limitations to the farmscale trials, they are being wrongly characterised as providing an unequivocal answer to whether the use of GM maize is safe or dangerous

involved and other parts of the food web are not fully understood. Furthermore, the selected measures of biodiversity may prove not to be the relevant ones in the long-term and are partly driven by what is practically feasible rather than by empirical evidence of their importance. As the scientists conducting the study have said;

"It is not practical to record population responses for all species in the arable system. We are using indicators of biodiversity that are <u>likely</u> to be sensitive to the treatments, and reflect processes that may lead to significant ecological shifts that cannot be detected directly given the time and spatial scales available for the study" (*Firbank et al.*, 1999) [emphasis added].

- that some species, such as birds, mammals and soil micro-organisms, are being excluded
 completely from direct study in the farm-scale trials. In the case of birds and mammals, it
 is hoped that the correct measures of resources available to them are being made but this
 is not certain. Despite micro-organisms being important parts of farmland biodiversity, soil
 health and fertility they are inexplicably excluded from the trials altogether;
- the uncertainty which will inevitably remain after the trials are completed as even on this large scale the information produced will be limited;
- that although the farmers conducting the experiments are instructed to follow particular
 rules when growing the GM crops, these are bound to be broken in the practical farming
 situation but no effort is being made to investigate the consequences of such predictable
 variation in human behaviour. Even in other closely controlled experimental trials there
 have been breaches of regulations covering matters such as separation distances;
- that the GM crop system is only being compared to a conventional system rather than
 organic or low input systems. Research I have conducted in collaboration with Dr Andy
 Stirling of the University of Sussex, shows that there is a broad dissatisfaction (including
 among specialists and industry) with conventional systems (Stirling and Mayer, 1999), and
 therefore the comparison being made in the farm-scale trials is unlikely to command
 confidence in terms of environmental protection.

Therefore, although the farm scale trials may appear more representative than small scale trials they cannot be relied upon to predict what the future may hold in terms of even the ecological consequences, let alone the other consequences, of using GM herbicide tolerant maize.



Inappropriate characterisation of GM trial outcomes

Despite these limitations to the farm scale trials, they are being wrongly characterised as providing an unequivocal answer to whether the use of the GM maize is safe or dangerous. For example, the Department of the Environment has said in the 'Farm-scale evaluation Factsheet' that:

"The results of these farm-scale evaluations <u>will</u> ensure that the managed development of GM crops in the UK takes place safely" (*DETR*, 1999a), [emphasis added].

Mr Meacher, the Environment Minister has made a similar statement:

"The farmscale evaluation of GM crops is extremely important research which <u>will</u> ensure that the managed development of GM crops will take place safely" (DETR, 1999b), [emphasis added].

Furthermore, scientists representing the consortium commissioned to conduct the research have said that:

"The research addresses the concern that the changing management of GM herbicide tolerant crops (GMHT) could result in reductions of weed and invertebrate populations on which farmland birds and other wildlife depend" (*Firbank et al., 1999*).

In these presentations there is no indication, either implicit or explicit, that the experiments address only one part of the question or that doubt may remain. I believe this is a very disturbing state of affairs and, together with the haste with which the farm-scale trials have been embarked upon, suggests that some motivation other than science is behind them. At the very worst the commercial use of the GM maize could be sanctioned on the basis of these trials and yet damaging effects could arise.

When considering the seriousness and irreversibility of any impacts that the large scale use of GM crop may bring any rush to experiment with GM crops on a large scale cannot be justified.

Furthermore, the approach that is being taken in relation to the introduction of GM crops and the use of scientific knowledge reflects more general findings made by specialists in the fields of sociology of science and risk assessment. These reveal systematic failings in the regulation of risk in the past which appear to be being repeated again here. For example, published research I have examined by experts, such as Professor Brian Wynne of Lancaster University, shows:

• scientific disciplines typically understate their uncertainties and limitations of their own knowledge (see eg. Freudenberg, 1992). Likewise, in this case the presentation of the outcome has been one of certainty, not that many questions remain unanswered or that the risks may be underestimated;

- when they arise, the consequence of technical interventions come as total surprises to
 existing scientific knowledge, yet this is rarely acknowledged. For example, some chemicals
 have proved to affect reproduction in some invertebrates and fish. In the domain of genetic
 modification, the phenomenon of transgene silencing was unpredicted.
- that democratic deliberation about the legitimacy of social and ecological experimentation
 are not developed. In this case there has been no debate about whether GM herbicide
 tolerance is considered a positive use of the technology and whether the products will
 be acceptable to consumers.



SOURCES

INTRODUCTION

ESRC Global Environmental Change Programme (1999), The Politics of GE Food, Special Briefing No 5, University of Sussex. See also www.gecko.ac.uk

UNESCO World Conference on Science in Budapest, June 1999. A speaker to the conference pithily described this issue by pointing out that "companies will not do research that benefits campaigners".

WIND POLLINATION

Bateman A.J. (1947). Contamination of seed crops. II. Wind pollination. Heredity I: 235-46.

Erdtman G. (1952). Pollen Morphology and Plant Taxonomy. Angiosperms. Almquist and Wiksells, Sweden.

Evans L.T. (1975). Crop Physiology. Cambridge.

Faegri K. and Iversen J. (1989). Textbook of Pollen Analysis (4th Ed.) John Wiley and Sons Ltd.

Goss J.A. (1968). Development, physiology and bio-chemistry of corn and wheat pollen. Bot. Rev. 34: 333-358.

Hardy K.R and Ottersten H. (1968). Two scales of convection in the clear atmosphere. Proc. Int. Conf. Cloud phys. Toronto: 534-538 (Am. Meteorol. Soc.).

Hirst J.M., Stedman O.J. and Hurst G.W. (1967). Long distance spore transport: Vertical Sections of Spore Clouds over the Sea. J. Gen. Microbiology. 48: 357-377.

Hjelmroos M. (1991). Evidence of long-distance transport of Betula pollen. Grana, 30: 215-228.

Hodges D. (1984) The pollen loads of the honey bee. International Bee Research Association, London.

Jones M.D. and Brooks J.S. (1950). Effectiveness of distance and border rows in preventing outcrossing in

corn. Oklahoma Agricultural Experimental Station – Technical bulletin No 38.

Jones M.D and Newell L.C. (1948). Longevity of pollen and stigmas of grasses: Buffalograss, Buchloe dactyloedees (NUTT) Engelm., and corn, Zea mays L. Journal of the American Society of Agronomy, 40 (3): 195 – 204.

Mandrioli P., Negrini M.G., Cesari G. and Morgan G. (1984). Evidence for long range transport of biological and anthropogenic aerosol particles in the atmosphere. Grana. 23: 43-53.

McCartney H.A. (1990). Dispersal Mechanisms through the Air in: Dispersal in Agricultural Habitats (Eds. Bunce and Howard) pp 133-158. Belhaven Press, London.

McCartney H.A. (1994). Dispersal of spores and pollen from crops. Grana, 33: 76 - 80.

Monteith J.L. (1975). Vegetation and the Atmosphere. Academic Press.

Morse R.A. (1972). The Complete Guide to Beekeeping. Pelham Books.

National Pollen Research Unit. (1999).
Pollen concentrations up to 475m downwind from a field of oil seed rape at model farm, near Watlington, Oxfordshire. June/July 1999.
Report for Friends of the Earth.

Nowakowski J. and Morse R. (1982). The behaviour of honey bees in sweet corn fields in New York state. American Bee Journal, January: 13-16.



Oke T.R. (1978). Boundary layer climates. Methuen. London.

Percival M.S. (1947). Pollen collection by Apis mellifera. New Phyto. 46: 142-165.

Percival M.S. (1950). Pollen presentation and pollen collection. New Phyto. 49: 40-63.

Percival M.S. (1955). The presentation of pollen in certain angiosperms and its collection by Apis mellifera. New Phyto. 54: 353-368.

Purseglove J.W. (1972). Tropical Crops. Monocotyledons 1. Longman Group, London.

Raynor G.S., Ogden E.C., and Hayes J.V. (1972). Dispersion and Deposition of Corn Pollen from Experimental Sources Agronomy Journal, 64: 420 - 427.

Salamov A.B. (1940). About isolation in corn. Sel.i.Sem, 3. (Russian translation by Michel Atanasiev in 1949).

Smith EG. (1990). Sampling and identifying allergenic pollens and molds. Blewstone Press, Texas.

Tyldesley J.B. (1973). Long-range transmission of tree pollen to Shetland. I-III. New Phytologist. 72: 175-190, 691-697

Vaissiere B. and Vinson S.B. (1994). Pollen morphology and its effect on pollen collection by honey bees, Apis mellifera L. (Hymenoptera: Apidae), with special reference to upland cotton, Gossypium hirsutum L. (Malvacaece). Grana. 33: 128-138.

BEE POLLINATION

Amoako J. (1997) Apiculture in Ghana: School of Pure and Applied Biology University of Wales.

Atallah M.A., Aly F.K. and Eshbah H.M. (1988). Pollen Gathering Activity of worker honey bees. Tropical Apiculture Conference. Cairo 1988.

De Bruyn C. (1998). Practical Beekeeping. The Crawood Press.

Howes F.N (1947). Plants & Beekeeping. pub.Faber & Faber.

Proctor M. and Yeo P. (1979) Pollination of Flowers, Collins New Naturalist Series. pub. Collins.

Seeley T. (1985). Honey Bee Ecology. Princeton University Press.

Pollen Identification for Beekeepers. University College, Cardiff Press.

The flight range of the honey bee. Journal of Agricultural Research No. 47.

The behaviour of honey bees in sweetcorn fields. American Bee Journal No122.

RISKS TO ORGANIC FARMING

Alston R. (1991). Living with the New Zealand flatworm, New Farmer and Grower, 33: 22. British Organic Farmers and Organic Growers Society: Bristol.

EEC (1991). Council Regulation (EEC No. 2092/91 of 24 June 1991) on Organic Production of Agricultural Products and Indications referring thereto on Agricultural Products and Foodstuffs.

EC (1999). Council Regulation (EC No. 1804/1999 of 19 July 1999) supplementing Regulation EC No. 2092/91.

Emberlin J., Adams-Groom B. and Tidmarsh J. (1999). The dispersal of maize pollen (Zea mays) – A report based on evidence available from publications and internet sites. National Pollen Research Unit: Worcester, January 1999.

Hoffman T., Golz C., and Schneider O. (1994). Foreign DNA sequences are received by a wild-type strain of Aspergillus niger after co-culture with transgenic higher plants, Current Genetics 27: 70-76.

MacKenzie D. (1999). Gut reaction. New Scientist, 30 January, p.4

MAFF (1998). Code of good agricultural practice for the protection of air (The air code). (MAFF publication no. PB0618).

Moyes, C.L. and Dale, P.J., (1999). Organic Farming and Gene Transfer from Genetically Modified Crops, May 1999. John Innes Centre: Norwich.

Schluter K., Futterer J., Potrykus I. (1995). Horizontal gene-transfer from a transgenic potato line to a bacterial pathogen (Erwinina-chrysanthem) occurs, if at all, at an extremely low frequency. Bio Technology 13: 1094-1098.

Soil Association (1998). Soil Association Standards for Organic Food and Farming, January 1998.

Soil Association (1999). Soil Association Standards for Organic Food and Farming (Genetic Engineering), June 1999.

Soil Association (1999). Genetic Engineering Guidance Notes, Soil Association, September 1999.

Soil Association (1999). The Organic Food and Farming Report, The Soil Association.

Treu R. and Emberlin J. (2000). Pollen dispersal in the crops Maize (Zea mays), Oil seed rape (Brassica napus ssp oleifera), Potatoes (Solanum tuberosum), Sugar beet (Beta vulgaris ssp.vulgaris) and Wheat (Triticum aestivum), National Pollen Research Unit: Worcester, January 2000.

UKROFS (1999). Terms of reference and rules of procedure for the Genetic Modification Working Group.

UKROFS (1999). UKROFS Standards for Organic Food Production, January 1999.

Watanabe T. (1963). Infective heredity of multiple drug resistance in bacteria. Bacteriological Review 27:87-115.

Young R., Cowe A., Nunan C., Harvey J. and Mason L. (1999). The Use and Misuse of Antibiotics in UK Agriculture, Part 2, Antibiotic Resistance and Human Health, The Soil Association.

THE WEAKNESS OF THE CONCEPT SUBSTANTIAL EQUIVALENCE

Bowers J. et al. (1999). Science 285: 1562-1565.

European Commission (1980). 'The ADI Debate', Appendix I of Food additives and the consumer', European Commission, 1980, pp. 41-3.

Ewan S. W. B and Pusztai A. (1999). Effect of diets containing genetically modified potatoes expressing Galanthus nivalis lectin on rat small intestine. Lancet 354 (9187): 1353-1354.

Kuiper H. A. et al. (1998). Food Safety Evaluation of Genetically Modified Foods as a Basis for Market Introduction (Ministry of Economic Affairs, The Hague, 1998).

Lydon J. and Duke S.O. (1989). Pesticide Science 25: 361-374.

Nature (1999). Food scientist in GMO row defends 'premature' warning. Nature 398: 98.

OECD (1993). Safety Evaluation of Foods Derived by Modern Biotechnology OECD, Paris, 1993.

Padgette S. R. et al. (1996). Journal of Nutrition 126: 702-716.

WHO (1991). Strategies for assessing the safety of foods produced by biotechnology, WHO, Geneva, 1991.

HAZARDS TO FOOD AND ANIMAL FEED

Beech D. (1997). "Study of the effects of Intra-Uterine Growth Retardation on the development of the lungs and kidneys in humans and two animal models. A stereological investigation.", 1997. PhD Thesis. Brunel University.

Ewan S.W.B and Pusztai A. (1999). Effect of diets containing genetically modified potatoes expressing Galanthus nivalis lectin on rat small intestine. The Lancet 354: 1353-1354.

Hinchliffe S.A., Lynch M.R.J., Sargent P.H., Howard C.V., van Velzen D. (1992). The effect of human intrauterine growth retardation on the development of renal nephrons. British Journal of Obstetrics and Gynaecology 99: 296-301.

Ho M.W., Traavik T., Olsvik R., Midtvedt T., Tappeser B., Howard C.V., von Weizsacker C. and McGavin G. (1998). Gene Technology and Gene Ecology of Infectious Diseases. Microbial Ecology in Health and Disease 10: 33-59.

Howard C.V., van Velzen D., Ansari T., Li Y. & Pahal N. (1995). Unbiased stereological measurements using conventional light microscopy, applied to the study of human intra-uterine growth retardation. Zoological Studies 34: 109-110.

Howard C.V. and Saunders P.T. (1999). Sensible precautions make good science. Nature 401: 207.

Johnston P., Stringer R., Santillo D. and Howard C.V. (1998). "Hazard, Exposure and Ecological Risk Assessment" In: Environmental Management in Practice, Vol. 1 (Eds: Nath, Hens, Compton and Devuyst) Routledge, London.

HORIZONTAL GENE TRANSFER

Adamo J.A. and Gealt M.A. (1996). A demonstration of bacterial conjugation within the alimentary canal of Rhabditis nematodes. FEMS Microbiol. Ecol. 20: 15-22.

Andersen S.R. and Sandaa R.A. (1994). Distribution of tetracycline resistance determinants among gram-negative bacteria isolated from polluted and unpolluted marine sediments. Appl. Environ. Microbiol. 60: 908-912.

Anderson N.G. (1970). Evolutionary significance of virus infection. Nature 227: 1346-1347.

Australian Genethics Network (1994). The troubled helix. Vol. 3 (ISBN 0-85802-115-3 08502-095-5).

Bergelson J. et al. (1998). Promiscuity in transgenic plants. Nature 395: 25.

Bogosian G. and Kane J.F. (1991). Fate of recombinant Escherichia coli K-12 strains in the environment. Adv. Appl. Microbiol. 36: 87-131.

Bork P. and Doolittle R.F. (1992). Proposed acquisition of an animal protein domain by bacteria. Proc. Natl. Acad. Sci. USA. 89: 8990-8994.

Brul S. & Stumm C.K. (1994). Symbionts and organelles in anaerobic protozoa and fungi. Trends Ecol. Evol. 9: 319-323.

Capy, P. et al. (1994). The strange phylogenies of transposable elements: are transfers the only explanation? TIG 10: 7-12.

Coffin J. (1990). In: Fields, B. et al. (eds.) Virology. Raven Press. Pp. 1437-1527.

Daane L.L. et al. (1996). Influence of earthworm activity on gene transfer from Pseudo-monas fluorescens to indigenous soil bacteria. Appl. Environ. Microbiol. 62: 515-521.

Davies J. (1994). Inactivation of antibiotics and the dissemination of resistance genes. Science 264: 375-382.

Doerfler W. and Schubbert, R. (1998).

Uptake of foreign DNA from the environment: the gastrointestinal tract and the placenta as portals of entry. Wien Klin. Wochenschr. 110: 40-4.

Doerfler W. et al. (1997). Integration of foreign DNA and its consequences in mammalian systems. TIBTECH 1997; 312: 401-6.

Doolittle R.F. et al. (1989). Origins and evolutionary relationships of retroviruses. Q. Rev. Biol. 64: 1-29.

Dreiseikelmann B. (1994). Translocation of DNA across bacterial membranes. Microbiol. Rev. 58: 293-316.

Duesberg P.H. (1983). Retroviral transforming genes in normal cells. Nature 304: 219-226.

Finnegan D.J. (1989). Eukaryotic transposable elements and genome evolution. Trends Genet. 5: 103-107.

Fox J.L. (1997). Farmers say Monsanto's engineered cotton drops bolls. Nature Biotechnol. 15: 1233.

Fredriksen K. (1993). Anti-native DNA antibodies induced by BK virus and BK virus native DNA. Ph.D. thesis, Department of Virology, Institute of Medical Biology, University of Tromsø, Norway (ISBN 82-7589-030-6).

Fredriksen K. et al. (1994). On the biological origin of anti-double stranded (ds) DNA antibodies: systemic lupus erythematosus-related anti-dsDNA antibodies are induced by polyomavirus BK in lupus-prone (NZB x NZW) F1 hybrids, but not in normal mice. Eur. J. Immunol. 24: 66-70.

Friesen P.D. and Nissen M.S. (1990). Gene organization and transcription of TED, a Lepidoptera retroposon integrated within the baculovirus genome. Mol. Cell Biol. 10: 3067-3077.

Gebauer, G. et al. (1998). mRNA expression of components of the insulin-like growth factor system in breast cancer cell lines, tissues, and metastatic breast cancer cells. Anti-cancer Res. 18:2A 1191-5.

Griffith F. (1928). The significance of pneumococcal types. J. Hyg. 27: 113-159.

Harding K. (1996). The potential for horizontal gene transfer within the environment. Agro Food Ind. Hi-Tech. 7: 31-35.

Hawkinson S.E. et al. (1998). Circulating concentrations of insulin-like growth factor 1 and risk of breast cancer. Lancet 351: 1393-6.

Heinemann J.A. (1991). Genetics of gene transfer between species. Trends Genet. 7: 181-185.

Ikawa Y. et al. (1974). An association between globin messenger RNA and 60S RNA derived from Friend leukemia virus. Proc. Natl. Acad. Sci. USA 71: 1154-1158.

Inose T. and Murata K. (1995). Enhanced accumulation of toxic compound in yeast cells having high glycolytic activity: a case study on the safety of genetically engineered yeast. Int. J. Food Science Tech. 30: 141-146.

Ivics Z. et al. (1997). Molecular reconstruction of Sleeping Beauty, a Tc1-like transposon from fish, and its transposable cells. Cell 91: 501-510.

Jehle J.A. et al. (1995). TC.14.7: a novel transposon found in Cydia pomonella granulosis virus. Virology 207: 369-379.

Kidwell M.G. (1993). Lateral transfer in natural populations of eukaryotes. Annu. Rev. Genet. 27: 235-256.

Kipling D. and Kearsey S.E. (1990). Reversion of autonomously replicating sequence mutations in Saccharomyces cerevisiae: creation of a eucaryotic replication origin with procaryotic vector DNA. Mol. Cell. Biol. 10: 265-272.

Kruse H. (1994). Antimicrobial resistance - epidemiological aspects. Dr. Scient. thesis, Norwegian College of Veterinary Medicine, Oslo (ISBN 82-7725-015-0).

Kruse H. and Sørum, H. (1994). Transfer of multiple drug resistance plasmids between bacteria of diverse origins in natural microenvironments. Appl. Environ. Microbiol. 60: 4015-4021.

Kruse H. and Jansson J. (1997). The use of antibiotic resistance genes as marker genes in genetically modified organisms. Norwegian Pollution Control Authority, Oslo (ISBN 82-7655-052-5).

Lambowitz A.M. and Belfort M. (1993). Introns as mobile genetic elements. Annu. Rev. Biochem. 62: 587-622.

Landman O.E. (1991). The inheritance of acquired characteristics. Annu. Rev. Genet. 25: 1-20.

Latorre J. et al. (1984). Microbial metabolism of chloroanilines: enhanced evolution by natural genetic exchange. Arch. Microbiol. 140: 159-165.

Leff L.G. et al. (1992). Information spiraling: movement of bacteria and their genes in streams. Microb. Ecol. 24: 11-24.

Levy S.B. and Miller R.V. (1989). Gene transfer in the environment. McGraw-Hill, New York.

Lorenz M.G. and Wackernagel W. (1994). Bacterial gene transfer by natural genetic transformation in the environment. Microbiol. Rev. 58: 563-602.

Lorenz M.G. et al. (1991). Release of transforming plasmid and chromosomal DNA from two cultured soil bacteria. Arch. Microbiol. 156: 319-326.

Luo G. et al. (1998). Chromosomal transposition of a Tc1/mariner-like element in mouse embryonic stem cells. Proc. Natl. Acad. Sci. USA 95: 10769-10773.

Maynard Smith J. et al. (1991). Localized sex in bacteria. Nature 349: 29-31

Mazodier P. and Davies, J. (1991). Gene transfer between distantly related bacteria. Annu. Rev. Genet. 25: 147-171.

Miller D.W. and Miller L.K. (1982). A virus with an insertion of a copia-like element. Nature 299: 562-564.

Milot E. et al. (1994). Association of a host DNA structure with retroviral integration sites in chromosomal DNA. Virology 201: 408-412.

Morell V. (1993). Dino DNA: the hunt and the hype. Science 261: 160-162.

Nielsen K.M. et al. (1998). Horizontal gene transfer from transgenic plants to terrestrial bacteria – a rare event? FEMS Microbiological Reviews 22: 79-103.

Nikolich N. et al. (1994). Evidence for natural horizontal transfer of tet Q between bacteria that normally colonize humans and bacteria that normally colonize livestock. Appl. Environ. Microbiol. 60: 3255-3260.

Nordlee J.A. et al. (1996). Identification of a Brazil-nut allergen in transgenic soybeans. New Engl. J. Med. 14: 688-728.

Ogram O.V. et al. (1994). Effects of DNA polymer length on its adsorption to soils. Appl. Environ. Microbiol. 60: 393-396

Ogunseitan O.A. et al. (1992). Application of DNA probes to analysis of bacteriophage distribution patterns in the environment. Appl. Environ. Microbiol. 58: 2046-2052.

Outwater J.L. et al. (1997). Dairy products and breast cancer: the IGF-1, estrogen and bGH hypothesis. Med. Hypotheses 48: 453-61.



Powers D.A. et al. (1991). Genetic mechanisms for adapting to a changing environment. Annu Rev. Genet. 25: 629-659.

Pääbo S. et al. (1988). Mitochondrial DNA sequences from a 7000 year old brain. Nucleic Acids Res. 16: 9777-9787.

Reddy S.A. and Thomas, T.L. (1996). Expression of a cyanobacterial delta 6-desaturase gene results in gammalinolenic acid production in transgenic plants. Nature Biotechnol. 14: 639-42.

Rekvig O.P. et al. (1992). Antibodies to eukaryotic, including autologous, native DNA are produced during BK virus infection, but not after immunization with non-infectious BKV DNA. Scand. J. Immunol. 36: 487-495.

Romanowski G. et al. (1992). Persistence of free plasmid DNA in soil monitored by various methods, including a transformation assay. Appl. Environ. Microbiol. 58: 3012-3019.

Romanowski G. et al. (1993a). Use of a polymerase chain reaction and electroporation of Escherichia coli to monitor the persistence of extracellular plasmid DNA introduced into natural soils. Appl. Environ. Microbiol. 59: 3438-3446.

Romanowski G. et al. (1993b). Plasmid DNA in a groundwater aquifer microcosmadsorption, DNase resistance and natural genetic transformation of Bacillus subtilus. Mol. Ecol. 2: 171-181.

Schlimme W. et al. (1997). Gene transfer between bacteria within digestive vacuoles of protozoa. FEMS Microbiol Ecol 23: 239-247.

Schouten G.J. et al. (1998). Transposon Tc1 of the nematode Caenorhabditis elegans jumps in human cells. Nucleic Acids Res 26: 3013-3017.

Schubbert R. et al. (1994). Ingested foreign (phage M13) DNA survives transiently in the gastrointestinal tract and enters the bloodstream of mice. Mol. Gen. Genet. 242: 495-504.

Schubbert R. et al. (1997). Foreign (m13) DNA ingested by mice reaches peripheral leukocytes spleen and liver via intestinal wall mucosa and can be covalently linked to mouse DNA. Proc. Natl. Acad. Sci. USA 94: 961-966.

Sidransky H. et al. (1994). Studies with 1,1'-ethylidenebis (tryptophan), a contaminant associated with L-tryptophan implicated in the eosinophilia-myalgia syndrome. Toxicol. Appl. Pharmacol. 126: 108-13.

Smets B.F. et al. (1993). The specific growth rate of Pseudomonas putida PAW1 influences the conjugal transfer rate of the TOL plasmid. Appl. Environ. Microbiol. 59: 3430-3437

Smith M.W. and Doolittle R.F. 1992. Anomalous phylogeny involving the enzyme glucose-6-phosphate isomerase. J. Mol. Evol. 34: 544-545

Smith M.W. et al. (1992). Evolution by acquisition: the case for horizontal transfers. Trends Biochem. Sci. 17: 489-493.

Sprague G.F. jr. (1991). Genetic exchange between kingdoms. Curr. Opin. Genet. Dev. 1: 530-533.

Spring S. et al. (1992). Phylogenetic diversity and identification of nonculturable magneto-tactic bacteria. Syst. Appl. Microbiol. 15: 116-122.

Stachel S.E. and Zambryski P.C. (1989). Generic trans-kingdom sex? Nature 340: 190-191.

Steffan R.J. et al. (1988). Recovery of DNA from soils and sediments. Appl. Environ. Microbiol. 54: 2908-2915.

Stephens R.S. et al. (1998). Genome sequence of an obligate intracellular pathogen of humans: Chlamydia trachomatis. Science 282: 754-759,.

Stevens S.W. and Griffith J.D. (1994). Human immunodeficiency virus type 1 may prefer-entially integrate into chromatin occupied by L1Hs repetitive elements. Proc. Natl. Acad. Sci. USA 91: 5557-5561

Syvänen, M. (1984). Conserved regions in mammalian b-globulins: could they arise by cross-species gene exchange. J. Theor. Biol. 107: 685-696.

Syvänen M. (1985). Cross-species gene transfer: implications for a new theory of evolution. J. Theor. Biol. 112: 333-343.

Syvänen M. (1987a). Cross-species gene transfer: a major factor in evolution? Trends Genet. 2: 63-66

Syvänen, M. (1987b). Molecular clocks and evolutionary relationships: possible distortion due to horizontal gene flow? J. Mol. Evol. 26: 16-23.

Syvänen M. (1994). Horizontal gene transfer: evidence and possible consequences. Annu. Rev. Genet. 28: 237-261.

Thakur M.S. et al. (1991). An environmental assessment of biotechnological processes. Adv. Appl. Microbiol. 36: 67-86.

Timmis J.N. and Scott N.S. (1984). Promiscuous DNA: sequence homologies between DNA of separate organelles. Trends Biochem. Sci. 9: 271-273.

Torsvik V.L. and Goksøyr J. (1978). Determination of bacterial DNA in soil. Soil Biol. Biochem. 10: 7-12.

Traavik T. (1979). Arboviruses in Norway. Dr. philos. thesis. University of Tromsø. Norway.

Trevors J.T. et al. (1987). Gene transfer among bacteria in soil and aquatic environments: a review. Can. J. Microbiol. 33: 191-198.

Vader A. et al. (1994). Nuclear introns from Physarum flavicomum contain insertion elements that may explain how mobile group I introns gained their open reading frames. Nucleic Acids Res. 22: 4553-4559.

Violand B.N. et al. (1994). Isolation of Escherichia coli synthesized recombinant eukaryotic proteins that contain epsilon-N-acetyllysine. Protein Sci. 3: 1089-97.

Widmer F. et al. (1997). Quantification of transgenic marker gene persistence in the field. Mol. Ecol. 6: 1-7.

Wöstemayer J. et al. (1997). Horizontal gene transfer in the rhizosphere: a curiosity or a driving force in evolution? Adv. Bot. Res. Incorp. Adv. Plant Pathol. 24: 399-429.

Zambryski, P.C. et al. (1989). Transfer and function of T-DNA genes from Agrarium Ti and Ri plasmids. Cell 56: 193-201.

Zhou, Y. et al. (1993). Transfer of neurotoxigenicity from Clostridium butyricum to a non-toxigenic Clostridium botulinum type E-like strain. Appl. Environ. Microbiol. 59: 3825-3831.

IMPACTS ON THE SOIL

Agro Evo. Glufosinate Ammonium Fact Sheet http://www.qn.apc.org/pesticidestrust/aifacts/qlufosin.htm

Ahmad I. and Malloch D. (1995). Interaction of soil microflora with the bioherbicide phosphinothricin. Agriculture, Ecosystems and Environment 54 pp 165-174. Elsevier Science.

Dekker J. and Duke S.O. (1995). Herbicide-Resistant Field Crops. In Advances of Agronomy 54: pp 69-116, Academic Press.

Giovani G.D. et al. (1999). Comparison of parental and transgenic alfalfa rhizosphere bacterial communities using BiologGN metabolic finger-printing and enterobacterial repetitive intergenic consensus sequence-PCR (EPIC-PCR). Microbial Ecology 37: pp129-139.

Lorenz M.G. (1998). Horizontal gene transfer among bacteria in soil by natural genetic transformation. In "Microbial Interactions in Agriculture and Forestry" pp19-44. Eds N.S. Subba Rao and Y.R. Dommergues, Science Publishers Inc., New Hampshire, USA.

Marshall G. (1998). Herbicide-Tolerant Crops-Real Farmer opportunity or Potential Environemtnal Problem. Pesticide Science, 52: pp394-402.

Seidler R.J. (1994). Evaluation of methods for detecting ecological effects from genetically engineered microorganisms and microbial pest control agents in terrestrial systems. In Environmental Gene Release, Eds M.J.Bazin and J.M.Lynch. OECD/ Chapman Hall, London.

Wolf D.C. et al. (1994). Isotopic methods for the study of soil organic matter dynamics. In: Methods of Soil Analysis, Part 2. pp865-905. Soil Science of America, Madison, Wisconsin, USA.

IMPLICATIONS FOR PESTICIDE USE

English Nature (1998). Position Statement on Genetically Modified Organisms, Peterborough 1998.

Lebaron H., and McFarland J. (1990). Herbicide Resistance in Weeds and Crops. In: Green, M.B., Lebaron, H., & Moberg, W.K., (eds), Managing Resistance to Agrochemicals. American Chemical Society, Washington, 1990.



MAFF (1999). Review of Usage of Pesticides in Agriculture, and Horticulture Throughout Great Britain 1986 – 1996. Pesticide Usage Survey Report 150. MAFF Publications, London 1999.

Pesticides Safety Directorate (1998). Scientific Review of the Impact of Herbicide Use on Genetically Modified Crops. Pesticides Safety Directorate, York. Dec. 1998.

SCIMAC (undated). Guidelines for growing newly developed herbicide tolerant crops. Supply Chain Initiative on Modified Agricultural Crops (SCIMAC).

SCIMAC (1999). Code of Practice on the introduction of genetically modified crops. Supply Chain Initiative on Modified Agricultural Crops (SCIMAC), May 1999 Publications and Reports of the Advisory Committee on Releases to the Environment.

FARM SCALE TRIALS AND ENVIRONMENTAL SAFETY

ACRE (1999). 'The Commercial Use of GM crops into the UK: the potential wider impact on farmland wildlife'. DETR's Advisory Committee on Releases to the Environment, 1999. URL: http://www.environment.detr. qov.uk/acre/wildlife/index.htm

DETR (1999a). Department of the Environment, Transport and the Regions. Farm-scale evaluations Factsheet. 26th May 1999.

DETR (1999b). Press Release 14th June 1999 "New GM Committee will assess 'science facts not fiction'- Meacher".

EEC (1998a). Official Journal of the European Communities 131 5th May 1998, p 31.

EEC (1998b). A proposal for a Directive to amend Directive 90/220/EEC on the deliberate release into the environment of genetically modified organisms (GMOs) (COM (1998) 85 final).

Emberlin J. and Tidmarsh J. (1999) A report on the dispersal of maize pollen. Soil Association: Bristol.

Firbank L.G. et al. (1999) Farm-scale evaluation of GM crops explained. Nature 399: 727-728.

Freudenberg W.R. (1992). Heuristics, biases, and the not-so-general publics: expertise and error in the assessments of risk. In: Social Theories of Risk. S.Krimsky & D. Golding (eds.) Praeger: Westport, Connecticut.

MAFF (1998). BRIGHT Project Press Release 14 October 1998.

Moyes C.L. and Dale P.J. (1999) Organic farming and gene transfer from genetically modified crops. John Innes Centre: Norwich.

SCIMAC (1999). Supply Chain Initiative on Modified Agricultural Crops – Guidelines for growing newly developed herbicide tolerant crops. May 1999.

Stirling, A. & Mayer, S. (1999) Rethinking Risk. A pilot multi-criteria mapping of a genetically modified crop in agricultural systems in the UK. SPRU, University of Sussex.

