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Fatal Flaws in Food Safety Assessment: Critique of the Joint FAO/WHO Biotechnology & Food Safety Report

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Third World Network

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Summary

The Biotechnology and Food Safety Report issued jointly by the Food and Agriculture Organization (FAO) and World Health Organization (WHO), is the result of an Expert Consultation held in Rome, in October 1996. The Consultation was the latest, possibly the most significant, attempt to reach international agreement on the safety of genetically engineered (GE) food. If accepted, it will set international safety standards to be adopted by WHO's Codex Alimentarius Commission, which will determine, not only GE food safety, but also world trade of GE foods. It will be illegal for any country to ban GE food imports, so long as the Codex considers them safe.

The FAO/WHO Report shows up the **glaring inadequacies in safety regulation of GE foods**, designed to expedite product approval with little or no regard for biosafety. It is a case of **'don't need – don't look – don't see'**, effectively giving producers *carte blanche* to do as they please, while serving to diffuse and allay legitimate public fears and opposition.

The 'principle of substantial equivalence', on which all safety assessment is based, is completely unscientific and arbitrary. A GE product assessed to be substantially equivalent (SE) is regarded as safe and fit for human consumption. But the principle is not only vague and ill-defined, it is flexible, malleable and open to interpretation. 'Substantial equivalence' does *not* mean an equivalence of the unengineered plant or animal variety. The GE food could be compared to any and all varieties within the species. It

could have the worst characteristics of all the varieties and still be considered SE. A GE product could even be compared to a product from a totally unrelated species. Worse still, there are no defined tests that products have to go through to establish substantial equivalence. The tests are so undiscriminating that unintended changes, such as toxins and allergens, could easily escape detection. A GE potato, grossly altered, with deformed tubers, was nevertheless tested and passed as SE.

The Consultation explicitly failed to assume responsibility for major areas of GE food safety, such as labelling and monitoring; impacts on biodiversity; and the control of traditional food crops engineered to produce pharmaceuticals and industrial chemicals. The latter will readily cross-pollinate with unmodified food plants and contaminate global food supply for years to come. Also left out are pesticide residues in food crops engineered to be resistant to herbicides, hormone residues and veterinary drugs in BST (or bovine somatotropin) milk from cows fed with this GE bovine growth hormone, which have to be treated for subsequent stress and infections.

Much more serious is **a list of gruesome products that will appear on our dinner table**, if the Report goes unchallenged: a range of 'transgenic wastes' from GE plant residues after engineered industrial chemicals and pharmaceuticals have been extracted, meat from failed GE experimental animals or from animals engineered to produce drugs and human proteins in their milk (e.g. Tracy, the transgenic sheep), meat from pigs engineered with human genes for organ transplants, and crops sprayed with insecticidal GE baculoviruses. The baculovirus is simultaneously engineered by medical geneticists to transfer genes into human liver cells because the virus is particularly good at invading those cells.

The possibility of new viruses being generated and of genes jumping (horizontally) across species barriers, as the result of

GE biotechnology itself, is real, especially in the light of recent scientific findings. The FAO/WHO Report ignores these findings, and sidesteps the whole issue by still maintaining that there is no difference between genetic engineering and conventional breeding methods. The Report is openly partisan to the technology, making unsubstantiated claims for its benefits while omitting to mention the socioeconomic impacts on small farmers, and the viable alternatives to the technology in all forms of sustainable agriculture already practised worldwide.

Recommendations

In view of the gross inadequacies in food safety regulation and the scientific evidence pointing to serious hazards, we recommend a number of measures to safeguard the health of consumers and to protect biodiversity. The precautionary principle also demands that a moratorium on further releases should be imposed until the following measures are implemented.

a. No food crops are to be engineered for producing pharmaceuticals and industrial chemicals, as the engineered crops could be mistaken for food, or cross-pollinate with non-engineered food crops. The onus must be on the producer to prove that any plant genetically engineered is not a food crop.

b. All projects involving genetic manipulation of baculovirus for insecticidal purposes should be discontinued, as this virus is being used in human gene therapy and invades human liver cells readily.

c. Complete characterization of inserted gene sequence(s) of the genetically engineered organism (GEO) must be provided in the application for market approval. This should include any antibiotic-resistance marker gene(s), promoter(s) and enhancer(s) and their effects on the expression of neighbour-

ing genes. The presence of mobile genetic elements and other proviral sequences in the host genome, likely to contribute to secondary mobility of inserts, must also be stated.

d. No GEOs with uncharacterized foreign gene inserts are to be considered for release. No parts of such GEOs, nor of animals from failed experiments in genetic engineering or xenotransplant animals are to be used as human food or animal feed.

e. No GEOs containing antibiotic-resistance genes are to be considered for release or to be used as human food or animal feed.

f. A detailed record of the stability of the GEO over at least five successive generations under field conditions (including drought and heat) is a precondition for market approval. ("Field conditions" does not mean open field conditions.) This must be supported by appropriate data indicating the stability of the insert as well as the level of gene expression under different conditions in successive generations.

g. Data on the frequency of unintended gene transfers, including horizontal gene transfer from the GEO under field conditions, must be included in the application for market approval.

h. Data on the frequency of horizontal gene transfer from the GEO to gut bacteria must be included in applications for market approval.

i. Data on the ability of transgenes and marker genes in the GEO to invade mammalian cells must be included in applications for market approval.

j. A specified set of tests must be carried out to establish "substantial equivalence", which are sufficiently discerning to re-

veal unintended as well as intended effects. The comparator must be the unmodified recipient organism itself, and results of repeated tests must be provided to support the stability of the characteristics over at least five successive generations.

k. Safety assessment must include the GEO's potential to generate pathogens through genetic recombination.

l. Safety assessment must include pesticide residues where they are integral components of the product, as in herbicide-resistant transgenic plants.

m. Product segregation, labelling and postmarket monitoring are non-negotiable conditions for market approval.

Chapter 1

Introduction

The Report on Biotechnology and Food Safety – resulting from the Joint Expert Consultation held by the Food and Agriculture Organization/World Health Organization in Rome from 30 September to 4 October 1996 – is the latest, possibly most significant, attempt to reach international consensus on principles and procedures for the evaluation of safety of food produced by genetic engineering biotechnology. It will set international food safety standards to be adopted by WHO's Codex Alimentarius Commission, which will, in turn, have enormous implications for biosafety and for the world trade of genetically engineered foods. For example, it will be illegal for any country to ban the import of genetically engineered foods that are considered safe by the Codex. Regrettably, the Report reflects and perpetuates the gross inadequacies of current regulatory frameworks:

- It openly favours the technology, making contentious claims for its benefits.
- It fails to assume responsibility for major aspects of food safety, such as environmental impacts, control of traditional food crops being used for pharmaceutical and industrial chemical production, labelling and monitoring.
- It exempts known hazards from safety assessment by restricting the scope of safety considerations.
- It takes as a premise the erroneous claim that genetic engineering does not differ from conventional breeding.
- Its principle of substantial equivalence, on which all safety assessment is to be based, is arbitrary and unscientific.

- It fails to address long-term impacts on health and food security.
- It ignores existing scientific findings pointing to identifiable hazards.

The result is a 'safety assessment' exercise designed to expedite product approval with little or no real regard for safety.

Chapter 2

Biased Partisan Claims for the Technology

The FAO/WHO Report claims that biotechnology accelerates the development of 'better foods', that its benefits are many. These include providing resistance to crop pests, reducing chemical pesticide usage – 'thereby making major improvements in both food quality and nutrition' (p.1). We are told that the use of biotechnological processes, particularly genetic modification, 'is extremely important in devising new ways to increase food production...[and] improve nutrient content' (p.2). Further on, the Report claims that '[r]ecombinant DNA technology has broad application in developing countries and has the potential for very positive impact on their economies...' (p.21). This typical propaganda of the biotech industry has never been backed up by any evidence, and has no place in a safety report.

Herbicide tolerance and insect resistance are the two most common genetically engineered traits, currently accounting for 54% and 37% respectively of the global area planted with transgenic crops, while viral resistance occupies 14% and quality traits less than 1% (James, 1997).

None of the traits constitute 'major improvements in food quality and nutrition'. On the contrary, each major category carries its own risks for health and biodiversity, some more than others. Herbicide-resistant transgenic crops are used with a companion herbicide; for example, Monsanto has engineered a wide range of crop plants resistant to its top-selling herbicide, the glyphosate-based 'Roundup', which is toxic to animals and human beings as well as

plants (Cox, 1995). Natural insecticides, while safe at the low concentrations found in nature, may be harmful at the high concentrations produced in resistant transgenic plants (Cummins, 1996), while viral-resistant transgenic plants are found to regenerate infectious viruses at high frequencies (Allison, 1997). We shall go into the hazards in greater detail later on.

The claims of benefits to the environment are particularly questionable as the Report specifically excludes environmental considerations from its remit, thereby avoiding any challenge to the claims (see below). These claims are downright misleading when no mention is made of already viable alternatives to increase food production by sustainable agricultural systems all over the world (Reganold *et al.*, 1990; Ho, 1997a, Chapters 2 and 9) nor of the serious socioeconomic impacts of the technology on small peasant farmers worldwide under the monopolistic regime of corporate intellectual property rights and the new world trade laws (Simms, 1997).

Chapter 3

Failure to take Responsibility for Major Aspects of Food Safety

The Report has disclaimed responsibility for several major aspects of food safety, which we shall deal with in turn.

3.1 Environmental impacts

'The Consultation further did not consider environmental safety issues related to the release of food organisms, foods or food components produced using biotechnology, into the environment as these were outside its defined scope' (p.3). On the previous page we are told that in the opinion of Professor Giuliano D'Agnolo, whose Institute hosted the Consultation, 'the environmental issues related to biotechnology have been well defined...' (p.2).

Contrary to Professor D'Agnolo's assertion, the environmental issues related to biotechnology have not been well defined. They have simply been ignored, and continue to be side-stepped by the Report.

The hazards of transgenic plants are by now well recognized (see Ho, 1996; Steinbrecher, 1996). Plants engineered to be resistant to broad-range herbicides will result in indiscriminate killing of a whole range of other plants over vast areas which are important constituents of natural ecosystems, besides the poisoning of human beings and animals, when the herbicides are applied. Similarly, the toxin from soil bacterium, *Bacillus thuringiensis* (Bt), is engineered into plants in a less selective form that is harmful, not just

to pests but also to non-target beneficial insects such as bees (Crabb, 1997). Recent reports indicate that beneficial insects which control pests, such as lacewings and ladybirds, may also be killed or harmed on ingesting pests that have eaten Bt-transgenic crop-plants (Bigler and Keller, 1997; Hawkes, 1997). Furthermore, herbicide-resistant weeds and insecticide-resistant pests are known to evolve rapidly in the field. And superinfectious viruses can be generated from a range of plants engineered to be viral-resistant (see Section 2).

Another environmental hazard from genetically engineered crops is the unintended spread of genes by cross-pollination and by horizontal gene transfer (see Section 7.2).

3.2 Production of pharmaceuticals and industrial chemicals in food

'The Consultation agreed that the safety assessment of pharmaceuticals and industrial chemicals, as such, was outside its remit... the Consultation recognized that the genetic modification of food organisms to produce pharmaceuticals or industrial chemicals may raise ethical and control issues that were outside its remit because the issues were unrelated to food safety' (p.20).

On the contrary, the use of traditional food crops and animals for producing pharmaceuticals and industrial chemicals is a serious food safety issue that ought to be addressed by the Report. Genes producing the pharmaceuticals or chemicals could easily spread by cross-pollination to ordinary food crops and lead to widespread contamination of the world food supply for years to come. Because these products are cryptic, neither farmers nor consumers will be able to tell the difference without the appropriate tests. Additional hazards will come from the spread of genes by horizontal gene transfer through aphids and other insects which feed on the crops, through bacteria in the soil, and above ground (see Sections 6.7. 6.8 and 7.5). The hazards from 'pharm' animals used for pharmaceutical production will be addressed in Section 6.3.

3.3 Labelling and monitoring

'The Consultation also did not consider any issues regarding the labelling of such foods or food ingredients...' (p.3). Monitoring, though not explicitly excluded, is not mentioned at all in the Report. These omissions betray a blatant disregard for safety in view of the many identified hazards already excluded from the remit of the Consultation. Genetic engineering biotechnology is still largely untried and inadequately researched. It has been prematurely rushed to market against the wishes of the vast majority of consumers. In the absence of labelling and postmarket monitoring, it will be almost impossible to identify the sources of hazards, to protect consumers accordingly, or to take appropriate remedial action.

Chapter 4

Restriction of Scope Exempts Known Hazards from Safety Assessment

The FAO/WHO Report further excludes from consideration, 'incidental residues in food resulting from the use of processing aids, or derived from the use of chemicals such as pesticides and veterinary drugs during food production' (p.3).

As a large proportion of current transgenic crops are herbicide-resistant, with a companion herbicide being sold and used as part and parcel of the package, it is not legitimate to exclude from consideration residues derived from the use of the herbicide. But that was precisely what took place in the safety assessment of Monsanto's Roundup-resistant soya bean. It was assessed *without* herbicide application (Tappeser and von Weizsäcker, 1996). Soya bean is known for producing phytoestrogen and to date, no tests have been performed to assess the level of estrogen in genetically engineered soya subject to the kind of repeated applications of Roundup that would take place when grown in the field.[1] Apart from the inherent toxicity of the herbicide, previous research has shown that herbicide spraying can increase the concentration of phytoestrogens (Sandermann and Wellmann, 1988).

Coincidentally, milk produced from cows fed with Monsanto's genetically engineered bovine somatotropin (BST milk) was also not assessed for hormone residues other than BST, nor for antibiotics which were fed to the cows to overcome mastitis and other infections arising from the use of BST. It is significant that the Codex Alimentarius Commission failed to pass a vote to permit the use of BST in June 1997, although it has been marketed since 1994.[2]

Even more serious is the exclusion of 'food-borne pathogens' (p.3). It has now been well documented that new, superinfective viruses can be generated in the many transgenic plants which have been made 'viral resistant' by incorporating the coat-protein and other viral genetic material (e.g. satellite RNA or ribonucleic acid). This is due to recombination between viral transgene(s) and co-infecting viruses (Anderson *et al.*, 1992; Green and Allison, 1994; Palukaitis and Roossinck, 1996; Allison, 1997). The US Department of Agriculture is deliberating restrictions on this category of transgenic plants (Kleiner, 1997). Recombination by similar mechanisms is predicted for another viral sequence, the cauliflower mosaic virus promoter, which is routinely used to boost the expression of transgenes (i.e., the foreign gene) in transgenic plants, although experiments have not yet been carried out to investigate this possibility (Cummins, 1994). The exclusion clause effectively exempts these products from safety assessment for food-borne pathogens that arise from the transgenic technology itself.

Also exempt are a range of genetically engineered baculoviruses, which have been developed for controlling insect pests (Jehle, 1997). This case needs urgent attention, as baculoviral vectors are concurrently being developed for human gene replacement therapy because *these vectors appear to be particularly good at invading human liver cells* (Heitmann and Lopes-Pila, 1993; Hofmann *et al.*, 1995; Sandig *et al.*, 1996). Yet, there is no mention of baculovirus in the entire Report.

Chapter 5

Erroneous Claim that Genetic Engineering is the Same as Conventional Breeding

'Food safety considerations regarding organisms produced by techniques that change the heritable traits of an organism, such as rDNA technology, are basically of the same nature as those that might arise from other ways of altering the genome of an organism, such as conventional breeding' (p.3).

The blurring of the distinction between genetic engineering and conventional breeding – a position adopted by the producers and regulators alike – is the single most important reason for the persistent failure of regulatory systems to protect consumers and biodiversity. *Genetic engineering carries its own inherent hazards which are unique to it, and which must be taken into proper account if we are to really protect health and biodiversity.*

5.1 New hazards are inherent in genetic engineering biotechnology

The unique hazards of genetic engineering have been dealt with in more detail elsewhere (Ho, 1995, 1997a; Antoniou *et al.*, 1997; Ho and Tappeser, 1997). We reiterate them and extend the discussion below.

a. The technology transfers exotic genes to organisms – genes for which no equivalents (alleles) may exist in the genome of the recipient organism – and are, therefore, more likely to have unexpected physiological and metabolic effects.

b. The method of gene transfer involves random insertions of the gene(s) into the genome (Walden *et al.*, 1991), causing correspondingly random genetic effects. In transformation with *Agrobacterium* T-DNA (the T is for Tumour, from the Tumour-inducing plasmid), the most widely used system for plants, the complete vector may be inserted, or a truncated or rearranged form, in single copies or tandem repeats at one or more sites; and insertion mutagenesis (due to insertion within other genes) is relatively common (see Conner, 1995).

c. Special promoter sequences and sometimes enhancers (often from disease-causing viruses) are included with the introduced gene(s) to boost constitutive (continuous) expression, and to effectively place the gene(s) outside regulation by the host cell. These promoters and enhancers are very strong, and are likely to affect the expression of neighbouring genes in the host genome.

On account of (a), (b) and (c), many unintended metabolic and genetic changes can result from the gene transfer, and grossly abnormal transgenic plants and animals have been generated, as well as toxins and allergens (see below).

d. The technology depends on artificially constructed invasive vectors for carrying genes, which are mosaics of different genetic parasites with the ability to invade cells of different species, multiply in them, or insert themselves into the genome. These vectors are designed to deliver genes *into* cells and to overcome cellular mechanisms that destroy or inactivate foreign DNA (deoxyribonucleic acid). They are, therefore, expected to be particularly good at transferring genes horizontally between *unrelated* species, and will do so whether intended or not. Although their mobility function has been removed, they can be moved by 'helper-functions' supplied by other parasitic genetic elements that are present in all genomes. There is already direct evidence of secondary (horizontal) gene

transfers from transgenic plants to a bacterial pathogen (Schluter *et al.*, 1995)[3] and to soil fungi (Hoffman *et al.*, 1994). And these are the only experiments that we know of, which have been carried out specifically to investigate horizontal gene transfer.

e. Many gene-transfer vectors are derived from viruses that cause diseases or bacterial plasmids or transposons (mobile genetic elements) that carry antibiotic-resistance and virulence genes, with their virulence functions removed. However, these gene-transfer vectors may recombine with viruses and plasmids in the host cells to generate new pathogens (Allison, 1997). As mentioned earlier, new superinfective viruses are generated by recombination between viral transgenes and infecting viruses. There is also evidence that while recombination between unmodified viruses may be negligible, modified, manipulated viral genomes are much more prone to undergo further recombination (Allison, 1997; Ho, 1997a; Ho *et al.*, 1997). This raises questions on the safety of gene-transfer vectors which are practically all modified hybrid genomes of viruses, plasmids and mobile genetic elements. This topic alone requires thorough investigations which have yet to be carried out.

f. Most of the gene-transfer vectors carry antibiotic-resistance markers to enable transformed cells to be selected, and these marker genes are routinely left in the transgenic organisms constructed.

The special characteristics inherent to genetic engineering biotechnology, (d), (e) and (f), have to be seen in the context of the current crisis in public health identified by WHO's own 1996 Report – the emergence of old and new infectious diseases which are resistant to treatment by drugs and antibiotics. Furthermore, there is now abundant evidence that horizontal gene transfer and recombination have been responsible for the rapid spread of both

virulence and antibiotic resistances, as we shall examine in more detail in Section 7.5.

In contrast to genetic engineering, conventional cross-breeding usually involves related species, often recombining different forms of the same genes (alleles). These species may have different numbers of chromosomes that differ in gene sequences. Redundant chromosomes or chromosomes that do not have a homologous partner are either lost during cell division or become inactivated by normal cellular mechanisms. Partially homologous chromosomes cause problems in the formation of germ cells and are most likely to lead to sterility of the hybrid. In such cases, induced polyploidy (causing the entire complement of chromosomes to double) is the usual method for ensuring reproductive success of the hybrid, as this restores normal chromosome pairing during germ-cell formation. Polyploidy generally results in an overall increase in size. It may result in changes in metabolism, and the polyploid hybrid should also be assessed for safety with sufficient rigour. The main differences, however, are that *there are no introduced invasive vectors that can insert at random into chromosomes which can potentially undergo secondary movements, nor antibiotic-resistance marker genes, nor strong promoter or enhancer sequences that continuously switch on gene expression, placing the genes outside cellular control.*

Thus, there are clear differences between genetic engineering and conventional breeding. Furthermore, there are already identifiable, if not quantifiable, hazards inherent in current practices of genetic engineering which make adherence to the precautionary principle in food safety paramount. Instead, the Consultation fails to address those hazards, and worse, effectively excludes them from consideration by the seemingly innocuous sentence, 'The presence in foods of new or introduced genes *per se* was not considered by the Consultation to present a unique food safety risk since all DNA is composed of the same elements' (p.4). This statement is nonsensical, as it is the sequence of the DNA which makes all the differ-

ence, especially between a pathogen and a non-pathogen. In addition, it is the special combination of sequences, the form of the DNA, as an invasive vector capable of secondary mobilization and recombination – with its aggressive promoter and enhancer sequences and antibiotic-resistance marker genes – that makes all the difference between food obtained from conventional breeding and genetically engineered foods.

Chapter 6

The Principle of Substantial Equivalence is Unscientific and Arbitrary

The most serious shortcomings of the FAO/WHO Report stem from the principle of 'substantial equivalence' on which all safety assessment is based.

6.1 The principle is intentionally vague and ill-defined so as to be as flexible, malleable and open to interpretation as possible.

'Substantial equivalence embodies the concept that if a new food or food component is found to be substantially equivalent to an existing food or food component, it can be treated in the same manner with respect to safety (i.e., the food or food component can be concluded to be as safe as the conventional food or food component)' (p.4).

This principle is unscientific and arbitrary, encapsulating a dangerously permissive attitude towards producers. At the same time it offers less than minimalist protection for consumers and biodiversity, because it is designed to be as flexible, malleable and open to interpretation as possible.

'Establishment of substantial equivalence is not a safety assessment in itself, but a dynamic, analytical exercise in the assessment of the safety of a new food relative to an existing food.... The comparison may be a simple task or be very lengthy depending upon the amount of available knowledge and the nature of the food or food component under consideration. The reference characteristics for

substantial equivalence comparisons need to be flexible and will change over time in accordance with the changing needs of processors and consumers and with experience' (pp. 4-5).

In other words, one can choose to compare whatever is the most convenient at a particular time, and for a particular purpose. And if on one set of criteria, the product is not substantially equivalent, a different set of criteria could be used, always to the advantage of the producers.

6.2 Comparisons are designed to conceal significant changes resulting from genetic modifications

In practice, the principle allows comparison of the transgenic line to any variety within the species, and even to an abstract entity made up of the composite of selected characteristics from all varieties. This is exemplified in the safety evaluation reported by the company Calgene on several of their products (Redenbaugh *et al.*, 1995). By a judicious use of additional varieties any changes from the control recipient variety could be bracketed. In theory, a genetically engineered line could have the worst features of every variety and still be substantially equivalent. Such comparisons actually *conceal* significant changes resulting from the genetic modification *per se*, which should alert conscientious researchers to a more careful characterization of the genetically modified organism.

Bernard Shaw was reputed to have been propositioned by a beautiful though not-too-bright lady who wanted to have his child so that it would have his brains and her looks. But Shaw was said to have discouraged her by pointing out that the child could end up having *her* brains and *his* looks instead. So, it is the particular *combination* of characteristics that makes all the difference. But under the present safety assessment regime, both combinations would be deemed 'substantially equivalent'. The danger is that particular combinations of nutrients or metabolites might fall within the

'equivalent' range determined in this fashion, and yet be anti-nutritional or outright lethal or toxic.

And if that were not enough, producers are assured that, even when products are not substantially equivalent, they can be shown to be substantially equivalent except for defined differences, and 'further safety assessment should focus only on those defined differences' (p.8). Lest one is in any doubt, it is stated on p.11 that, '[u]p to the present time, and probably for the near future, there have been few, if any, examples of foods or food components produced using genetic modification which could be considered to be not substantially equivalent to existing foods or food components.'

Calgene's genetically engineered Laurate canola oil should, by no stretch of the imagination, be considered substantially equivalent to ordinary canola oil. But, 'other fatty acids components are GRAS [Generally Recognized as Safe] when evaluated individually because they are present at similar levels in other commonly consumed oils.' Similarly, 'substitution of Laurate canola for coconut and palm kernel oils does not raise any safety concerns for intended uses, in part because the major components, the fatty acids laurate and myristate, are identical' (Redenbaugh *et al.*, 1995, p.43).

In other words, it is already a foregone conclusion that most, if not all, products now and for the foreseeable future will be assessed as 'substantially equivalent' and if not, then considered GRAS by a judicious choice of a comparator.

It is significant that the Dutch courts recently ruled Monsanto's genetically engineered soya beans *not* equivalent in quality to natural soya beans, as was claimed in the advertisement of Albert Heijn, the biggest supermarket chain in the Netherlands. Albert Heijn is itself part of the Dutch multinational, Ahold, which owns supermarket chains in many countries around the world. The complaint was filed by the Dutch Natural Law Party (Storms, 1997).

6.3 The principle is weak and misleading even when it does not apply, effectively giving producers *carte blanche*

Given that 'substantial equivalence' can be interpreted in the widest possible sense – and if not, by a judicious choice of comparator – in order that a product can be considered as GRAS, it is difficult to imagine which remaining products cannot pass muster.

The Report recognized that 'products could be developed which could be considered to have no conventional counterpart and for which substantial equivalence could not be applied' (p.11). For example, 'products derived from organisms in which there has been transfer of genomic regions which have perhaps been only partly characterized' (p.11). This gives the impression that such are hypothetical cases that might arise in future.

But that is not so. The Report failed to point out that at least one such transgenic organism already exists: Tracy, the sheep engineered with a large segment of the human genome – most of which contains unknown sequences with unknown functions – to produce huge quantities of alpha-antitrypsin in her milk (Colman, 1996). Tracy and her clones may be walking incubators for cross-species viruses to arise by recombination between human and sheep viral sequences. All genomes contain endogenous proviral sequences, and recombination between endogenous and exogenous viral sequences is already implicated in several kinds of animal cancers (see Ho, 1997a, Chapter 13). One might think that the Report would treat such cases with extra caution. Not so.

We are assured that even if a food or food component is considered to be not substantially equivalent, producers need not despair, for 'it does not necessarily mean it is unsafe and not all such products will necessarily require extensive testing' (p.12). The Report is clearly preparing the grounds for slipping those products through a regulatory framework that is already worse than toothless.

Further on, in Section 6.6 on 'Food organisms expressing pharmaceuticals or industrial chemicals' (p.19), there is the telling statement, 'The Consultation recognised that, generally, the genetically modified organism would not be used as food without prior removal of the pharmaceutical or industrial chemical' (p.19). This is a prelude to serving up the rest of Tracy and the 'elite herd' cloned from her, or more likely, superannuated 'pharm' animals and any failed transgenic experiment, whatever, as meat for our dinner tables. Transgenic technology is very inefficient and generates a lot of transgenic wastes – given the large numbers of failed experiments. Such 'foods' from transgenic wastes may be sources of exotic, cross-species food-borne viruses, as mentioned earlier. Furthermore, they will be exempt from safety assessment if the Report is to be taken seriously. A similar category of transgenic waste could be the left-over carcasses of pigs engineered for xenotransplantation.

All the signs are that the producers are handed *carte blanche* to do as they please for maximum profitability, with the regulatory body acting to allay legitimate public fears and opposition.

6.4 Insufficiency of background information for assessing substantial equivalence

The procedure for establishing substantial equivalence, described in less than three pages in the 27-page Report (pp.6-8), comes under two headings: background information on the characterization of the modified organism and actual determination of substantial equivalence, or characterization of the food product itself.

One glaring omission in the background information is the propensity of the transgenic organism for generating pathogenic viruses by recombination (and whether experiments have been carried out to investigate this propensity). This information is highly relevant for assessing impacts on biodiversity as well as food safety, in view of our current knowledge that superinfecting viruses may

be generated from many transgenic plants at high frequencies and that insecticidal recombinant viruses may attack human liver cells. There is also disturbing new evidence that viral DNA can survive digestion in the gastrointestinal tract of mice, with large fragments getting into the bloodstream and into many kinds of cells (Schubbert *et al.*, 1994).[4]

Likewise, information on the stability of transgenes, and potential for mobility of introduced genes, which are mentioned on p.6 of the Report, ought to be based on data collected over a number of generations, documenting the stability of the insert as well as expression of the transgenes and the transgenic line in successive generations. This is so that both consumers and farmers can have confidence in quality control. In a paper presented at a WHO workshop, the author states, 'The main difficulty associated with the biosafety assessment of transgenic crops is the unpredictable nature of transformation. This unpredictability raises the concern that transgenic plants will behave in an inconsistent manner when grown commercially' (Conner, 1995, p.27).

In general, the inheritance of genetically engineered traits are non-Mendelian in subsequent generations (Schuh *et al.*, 1993) necessitating clonal propagation. In early 1997, 60,000 bags of genetically engineered canola seeds, enough for planting 600,000 acres, had to be recalled after they were sold in western Canada, because an unexpected gene, not yet approved for market, turned up in the seeds. The seeds were bred and sold by Limagrain, under licence from Monsanto.[5] If the transgenic plants had been monitored for genetic stability of both the transgenes and the transgenic line in successive generations – *as they should have been* – and careful records kept, those seeds would never have reached the market. This incident also indicates the necessity for product segregation, clear labelling and postmarket monitoring as part of the condition for market approval.

Under background information, it is also crucial to include the upstream and downstream effects of transgenic promoter and enhancer sequences, as well as the presence of genetic elements in the host that might compromise the stability of the transgenes.

A further serious omission in the background information is the explicit requirement to disclose the presence of marker genes, especially antibiotic-resistance marker genes which are considered in Section 6.7.

6.5 There is no specification of tests for establishing substantial equivalence

Under 'characterization of the food product', we are told it entails 'molecular characterization', 'phenotypic characterization' and 'compositional analysis'. While the latter two categories are elaborated subsequently, 'molecular characterization' has mysteriously disappeared. Nowhere is it specified which methods of molecular characterization are required, nor which molecular information should be established. Such information happens to be crucial for identifying unintended effects. Similarly in a previous document which reports on a WHO Workshop on the principle of substantial equivalence,[6] molecular characterization is left very vague. It refers to 'the inserted DNA'; 'the level and mechanism of expression of the protein', which is considered to be 'more important than knowing the gene copy number'. In other words, the inserted DNA sequence need not be well characterized at all. The WHO's Workshop Report then mentions that 'the level and function of the introduced gene product in the plant may be useful in judging substantial equivalence', again implying that the function of the gene product need not be known as a condition for safety approval. If the gene(s) and gene product(s) transferred are well understood, however, the safety evaluation can then 'focus on the safety of the expression product and/or changes brought about by the expression product'. This is an open endorsement of a totally inadequate,

reductionist safety assessment that ignores effects on the system as a whole, especially in the longer term.

In effect, no molecular characterization of the product whatsoever is required. Not even the level of expression of the introduced transgene(s) or marker gene(s) need to be ascertained, much less the effects of promoters and enhancers on neighbouring genes, as judged by the samples of papers presented in the WHO workshop on substantial equivalence.[7] If one happens to know what has been transferred, then the safety assessment can focus only on the gene product and its effects. So the two main categories of characterization of the food product are, simply, phenotypic characteristics:

- agronomic, morphological and physiological and compositional comparison

- key nutrients and toxicants which are *known* to be inherently present in the species.

6.6 There is no requirement to test for unintended effects, current tests are undiscerning and may even serve to conceal unintended effects

Although the FAO/WHO Report recognizes the possibility of 'indirect consequences' (p.4) and that 'assessment of the safety of genetically modified organisms must address both intentional and unintentional effects that may result as a consequence of the genetic modification of the food source' (p.5), these effects are limited to phenotypic changes that are readily apparent, and alterations in the concentrations of major nutrients or increases in the level of natural (known) toxicants. There is thus no specific requirement to test for unintended effects, *per se*.

Similarly, while it is stated that 'attention must be paid to the impact of growth conditions on the level of nutrients and toxicants ... attention must be paid to the impact of different soils and climatic

conditions' (p.5). These are not elaborated further, and certainly not required for safety assessment recommended in the Report.

The range of tests which are actually carried out, as exemplified by WHO's Workshop Report on applying the principle of substantial equivalence, is not sufficiently discerning to pick out unintended effects. Unless there are gross morphological or phenotypic changes, there is no need to look for them. And even when there are gross abnormalities, the product can still be assessed to be 'substantially equivalent'. One paper presented in the WHO workshop reports, 'Field trials on the transgenic lines used in these studies showed marked deformities in shoot morphology and poor tuber yield involving a low number of small, malformed tubers during field trials .... These changes were attributed to somaclonal variation during the tissue culture phase of transformation .... Despite these marked morphological abnormalities, virtually no changes in tuber quality attributes were detected...' (Conner, 1995, p.30). So much for the discerning power of the tests carried out.

There were no metabolic profiles done by routine techniques such as High Pressure Liquid Chromatography (HPLC), nor two-dimensional gel electrophoresis to scan for unintended expression of genes – again, another routine technique. The compositional analyses reported are limited to uninformative amino-acid profiles, or to known components present at levels greater than 0.1%, or 0.01% at best. And, as mentioned earlier, the arbitrariness of the comparator will already hide any changes due to the transferred gene(s) *per se*, which should alert researchers to unintended effects. Instead, the tests are aimed specifically at intended effects only, and if anything, to conceal secondary, unintended effects as much as possible.

The hazard of unintended effects is already well attested to by the US epidemic of eosinophilia-myalgia syndrome in 1990, resulting in more than 1,500 affected and 37 deaths, which is linked to the consumption of L-tryptophan produced by a genetically modifed

strain of *Bacillus amyloliquefaciens* (Mayeno and Glich, 1994). Several trace contaminants identified on HPLC have been implicated in pathogenesis.

A metabolite, methylglyoxal, was found to accumulate at toxic, mutagenic levels in yeasts engineered with multiple copies of one of several yeast glycolytic enzymes to increase the rate of fermentation (Inose and Murata, 1995). Recently, tobacco plants genetically engineered to produce the gamma-linoleic acid, also unexpectedly produced octodecatetraenoic acid, a substance previously unknown in natural tobacco plants (Reddy and Thomas, 1996). In the absence of a metabolic profile on the product, unintended toxic metabolites might easily have escaped notice in the safety assessment.

It is equally important to check for unintended gene products being produced, which will not be revealed by routine amino-acid analyses of total lysates, as is done by Calgene for canola meal (Redenbaugh *et al.*, 1995). A minimum requirement should be a two-dimensional gel electrophoretogram of the total proteins (Ho, 1996). Even then, minor modifications in a proportion of the proteins may not be detectable, but which may change the properties of the proteins involved. For example, a proportion of the recombinant porcine and bovine somatotropins synthesized in *E. coli* were found to contain the abnormal amino acid ε-N-acetyllysine in place of the normal lysine, only when reversed-phase HPLC analyses were carried out (Voland *et al.*, 1994).

Key questions on the allergenic potential of transgenic foods are raised by the recent identification of a brazil-nut allergen in soya bean genetically engineered with a brazil-nut gene (Nordlee *et al.*, 1996). It is possible to test for *known* allergens, as in the case of the brazil-nut soya bean, but not for allergenicity to proteins completely new to the foods involved, as acknowledged in the FAO/WHO Report (p.14). It is significant that allergenicity in plants is thought to be linked to proteins involved in defence against pests and dis-

eases (Franck and Keller, 1995). Therefore, transgenic plants engineered for resistance to diseases and pests may have a higher allergenic potential than the unmodified plants. One major novel protein is the insecticide produced by the gene from *Bacillus thuringiensis* (Bt), now incorporated into a range of transgenic crop plants, which had never contained it before. Nevertheless, the producers were able to claim substantial equivalence by pointing to its 'comparability' (not identity!) 'to one of the proteins contained in the commercial microbial formulations that have been used commercially since 1988' (Fuchs *et al.*, 1995, p.66).

One important characteristic of an allergen is that it resists digestion in the stomach (gastric digestion). According to a recent publication (Astwood *et al.*, 1996), known allergens were stable for 60 minutes, whereas non-allergens were fully digested within 15 seconds. While one study claimed that the Bt protein was readily digestible (Fuchs *et al.*, 1995), another report showed that it failed to be completely digested under gastric conditions after two hours (Noteborn and Kuiper, 1995). In both cases, we are assured that the protein is safe. In view of the recent discoveries that predators eating pests which have ingested the Bt toxin in transgenic crop-plants are also harmed (Bigler and Keller, 1997; Hawkes, 1997), it is irresponsible to assume that the toxin is safe for human beings.

We accept that no safety assessment system is foolproof. A case in point is the rigorous testing that goes on with pharmacological products. It is estimated that despite such rigorous testing, 3% of the products approved for market turn out to have such harmful effects that they have to be withdrawn, while an additional 10% have sufficiently harmful side-effects that limited use has to be recommended (Suurkula, 1997). This underlines the importance of segregation, clear labelling and postmarket monitoring of the health and other impacts of genetically engineered foods. Labelling is a matter of traceability and should be a scientific requirement, not only a consumer option.

6.7 The spread of antibiotic-resistance marker genes by horizontal gene transfer is downplayed, by ignoring existing scientific evidence

Antibiotic-resistance marker genes are not mentioned in the FAO/WHO Report until p. 15, under Section 6.2, 'Gene transfer from genetically modified plants', where it is stated that '[t]heir continued use in plants remains critical to the production of genetically modified plants. The Consultation therefore focused on these particular marker genes.' However, all it did was support the conclusions of a previous, 1993 Workshop 'that 'there is no recorded evidence for the transfer of genes from plants to microorganisms in the gut' and that there are no authenticated reports of such bacterial transformation in the environment of the human gastrointestinal tract.'[8] These conclusions are not based on actual experiments that have been done to ascertain if these transfers occur. It is a case of interpreting 'the absence of evidence' as 'evidence of absence'.

We are told that the first conclusion was 'based on the judgement that transfer of antibiotic resistance would be unlikely to occur given the complexity of steps required for gene transfer, expression, and impacts on antibiotic efficacy'. The steps are listed, the first of which is the most crucial, 'the plant DNA would have to be released from the plant tissue/cells and survive in the presence of the hostile environment of the GI tract, including exposure to gastric acid and nucleases' (p.16). But that is untrue.

In the course of digestion, plant DNA *will* be released from the plant cells, and, there is already evidence that large fragments of viral DNA can survive digestion in the gastrointestinal tract of mice (Schubbert *et al.*, 1994). So, it is possible that vector DNA, which carries the antibiotic-resistance marker genes may also resist digestion. The question is whether bacteria in the gut can be transformed by the DNA, and there is an urgent need for experiments to be done to answer this question, in view of the wealth of new

evidence, *since 1993*, on the ease with which transformation occurs in all other environments (see below). Most of the old assumptions supporting the previous judgement that transfer is unlikely may be superseded by the new findings.

Because gene-transfer vectors are already extensively modified – with sequence homologies to a wide range of species, and to resist restriction – they may successfully integrate into many bacterial genomes. It is practically impossible to design vectors that prevent horizontal transfer. Furthermore, as sequence homology is not required for integration into chromosomes or plasmids, homology only makes it more likely to occur. The assumption that antibiotic-resistance marker genes under plant promoters 'would not be expressed in a microorganism' (p.16) is dangerous, as so few bacterial promoters are characterized. While some antibiotic-resistance marker genes are placed under bacterial promoters, as in the Ciba-Ceigy transgenic maize, there are special mobile genetic elements in microorganisms, called integrons, which carry an enzyme catalyzing the integration of antibiotic-resistance genes into specific sites where the integrated genes are then provided with ready-made promoters for expression (Collis *et al.*, 1993). The Report also fails to take account of the ease with which recombination can occur following horizontal gene transfer, whereby any missing promoter for the gene(s) may be regained.

Horizontal gene transfer has been demonstrated between bacteria in the gut of animals as well as human beings *since the 1970s* (Anderson, 1975; Freter, 1986; Doucet-Populaire, 1992). For this reason, gene transfer from genetically modified microorganisms must definitely be considered in the safety assessment of genetically modified microorganisms, as appears to be recommended by Section 6.3 of the Report, 'Gene transfer from genetically modified microorganisms' (pp.17-18). It is stated that '[t]he Consultation affirmed the recommendation from the 1990 FAO/WHO joint consultation ... regarding genetically modified microorganism including: 1) that vectors should be modified so as to minimize the likeli-

hood of transfer to other microbes; and 2) selectable marker genes that encode resistance to clinically useful antibiotics should not be used in microbes intended to be present as living organisms in food' (p.18).

However, as stated above, artificial vectors are already extensively modified; and as modified vectors are often unstable (Old and Primrose, 1996), they may be much more prone to mobilize and to recombine (Allison, 1997; Ho, 1997a; Ho *et al.*, 1997). An additional problem of antibiotic resistance is that of cross-resistance. For example, resistance to kanamycin may be accompanied by resistance to new-generation aminoglycoside antibiotics such as tobramycin and amikacin (Conner, 1995; Smirnov *et al.*, 1994).

The Report states that '[t]he Consultation was not aware of any reports of genes from animal, plant or microbial origin into epithelial cells, except for infectious agents, such as viral DNA.' But there is already evidence that viral DNA can enter the bloodstream and many kinds of cells in mice.[9] Again, vector DNA is modified viral DNA in many cases, and in the absence of results from actual experiments carried out to investigate this possibility, it is not legitimate to conclude that DNA cannot enter epithelial cells, or the bloodstream and from there, gain access to other cells. One major immediate danger in this regard is the genetically engineered baculoviruses developed as insecticides, which are also simultaneously developed as vectors for human somatic gene therapy (see Section 4) which the Report has not even mentioned.

6.8 There is no consideration of unintended gene transfers in the general environment

The Report has avoided any discussion of horizontal gene transfer to microbes and other organisms in the general environment, for which substantial evidence has emerged within the past three to four years. But there is still no explicit requirement to monitor such horizontal gene transfers during field releases. It is a blatant

omission in view of already existing evidence that transgenic plants can transfer transgenes and marker genes horizontally to microbes in the soil (Schluter *et al.*, 1995; Hoffman *et al.*, 1994). Also, reviews of recent findings by many authors (dealt with in detail in Section 7.5) show that *there is essentially no barrier to gene transfer between microorganisms.* The microbes in the environment, in turn, serve as a gene transfer highway and reservoir for multiplication and recombination, from which the genes can spread to practically all other species. Particularly significant are new findings indicating that genetically 'crippled' microorganisms can survive, or go dormant and reappear, after having acquired genes horizontally from some species in the environment to enable them to grow and multiply; that naked DNA can survive for long periods in all environments and retain their ability to transform; that transformation frequencies are high in all environments.

These findings have large implications for the safety of the releases from contained use, which is itself urgently in need of a full reassessment (Ho, 1997b). It is significant that the Norwegian government banned the imports of two rabies vaccines and four transgenic plants containing antibiotic-resistance marker genes in September 1997, *in recognition of the hazards arising from horizontal gene transfer and recombination.*[10]

On account of the possibilities of horizontal gene transfer, it is paramount that no organism containing antibiotic-resistance marker genes, and in particular, unknown, uncharacterized foreign gene sequences, should be considered for release.

Chapter 7

Failure to Take Existing Scientific Evidence into Account

Genetic engineering biotechnology is a rapidly moving area. Many key discoveries have only been made within the past three to four years, as reviewed in detail elsewhere (Ho, 1997a; Ho *et al.*, 1997), which have large implications for the safety of genetically modified foods. The totality of existing scientific findings leads us to conclude that an inadequately researched and inherently dangerous technology has been pushed prematurely to commercialization. We must emphasize that these indications of hazards have emerged from a non-exhaustive search of limited databases, and despite the paucity of specifically targeted research. Relevant data may be missing in some instances simply because the experiments or investigations have not been done. It is unacceptable for the Report to interpret 'the absence of evidence' as 'evidence of absence'. Yet, the Report has registered neither the substantial body of existing scientific findings, nor the hazards indicated by these findings.

7.1 The instability of transgenes and transgenic lines

Transgene instability is now a recognized problem in both farm animals and plants (see Colman, 1996; Lee *et al.*, 1995; Ho, 1996; Steinbrecher, 1997). In transgenic tobacco, 64 to 92% of the first generation of transgenic plants become unstable. Similarly, the frequency of transgene loss in *Arabidopsis* ranges between 50 and 90%. Instability arises both during production of germ cells and in cell division during plant growth. The commonest cause of transgene instability is gene silencing (see Finnegan and McElroy, 1994; Ho,

1996, 1997a, Chapters 8 and 9) – the inability of the introduced gene to become expressed – due to chemical modification (methylation) of the DNA. Other causes are DNA rearrangements and excision of the transgene. The stability of the transgenic line may also be compromised by somaclonal variation – variation arising during plant cell culture after transformation (Cooking, 1989) – which has been known for a long time. Instability may also be due to the tendency of the insert towards secondary mobilization (see Ho, 1997a, Chapter 9).

Instability, such as secondary mobilization, may be triggered by extreme environmental conditions, such as heat and drought. For this reason, transgenic plants must be tested for stability under these conditions before they are approved for market. All these factors compromise the quality of the product. Not only do they have socioeconomic impacts for farmers, but they have large implications for food security and food safety, since they increase the potential for unintended effects as well as secondary gene transfers.

7.2 The prevalence and scope of horizontal gene transfer in all environments, including the gastrointestinal tract

The full scope of horizontal gene transfer is such that any gene released in any species has a finite probability of being transferred to many other species of both eukaryotes and prokaryotes (Stephenson and Warnes, 1995). Direct transfer has been demonstrated from higher plants to bacteria, and to fungi (Schluter *et al.*, 1996; Hoffman *et al.*, 1994), and from bacteria to plants. The Ti (Tumour-inducing) plasmid of the soil bacterium, *Agrobacterium*, widely used in modified versions as vectors for making transgenic crop plants, actually mediates conjugation between *Agrobacterium* and plant cells (Kado, 1993). This is why the secondary mobility of such vectors in transgenic crop plants cannot be ruled out, and should have been rigorously monitored. It is well known that helper-functions supplied by endogenous elements – which are ubiquitous in genomes – can mobilize elements that do not have

their own enzymes for mobility. Indirect evidence also exists of two-way transfers between bacteria and viruses and the animal kingdom. Most of all, the bacteria and viruses in all environments act as a gene transfer highway and reservoir, for multiplying and recombining genes and from which the genes can spread to all species.

Another means of horizontal gene transfer is via insects that visit plants. Aphids, bees and butterflies, for example, will spread infectious viruses that arise in transgenic viral-resistant plants from recombination (see Section 4 above).

All the means available to microorganisms – transformation, transduction and conjugation – are utilized. New discoveries indicate that the frequencies of horizontal gene transfers in all environments are much higher than previously thought. It has been demonstrated in the marine environment (Frischer *et al.*, 1994; Lebaron *et al.*, 1994), in the freshwater environment (Ripp *et al.*, 1994) and in the soil (Neilson *et al.*, 1994). Horizontal gene transfer occurs preferentially in interfaces between air and water and in the sediment, and especially under nutrient depletion conditions (Goodman *et al.*, 1994). Transfer (of multiple antibiotic resistance) has even been demonstrated in wastewater treatment ponds (Mezrioui and Echab, 1995).

Transformation (by uptake of naked DNA) in the environment (Lorenz and Wackernagel, 1994) is extremely widespread. Both chromosomal and plasmid DNA are able to transform bacteria. Cross-species, cross-genera and even cross-order transfers have been observed with chromosomal DNA, while plasmid DNA has effected cross-kingdom transformations. Similarly, transduction may be substantial in aquatic environments (Bergh *et al.*, 1989), while conjugation is essentially promiscuous when it is realized that retro-transfer from recipient to donor can also occur, and that conjugative transposons can jump between plasmids and chromosomes (Clewell, 1993).

As mentioned in Section 6.7 above, horizontal gene transfer between bacteria has been documented in the gastrointestinal tract of animals as well as human beings.

7.3 DNA is not readily degraded in the environment

Recent findings show that DNA can persist for days, weeks and even months in the environment, especially when adsorbed to solid particles in the soil or in aquatic sediments, where they retain their transforming power (Jager and Tappeser, 1995; Lorenz and Wackernagel, 1994). This is contrary to previous assumptions that DNA-degrading enzymes (DNases) in the environment will rapidly break down DNA. Thus, DNA released from dead plant cells or dead microorganisms may retain the ability to transform other organisms. Transgenic plant exudates, and debris ploughed back into the soil, are very likely to release DNA for transforming soil bacteria and other microbes. In the aquatic environment, dead cells from transgenic fish and other organisms will release DNA capable of transforming bacteria and viruses which are abundant in the aquatic environment. Transgenic domesticated animals or 'pharm' animals will pass dead cells in their faeces in the farmyard, which will release DNA for transforming soil microbes.

7.4 Transgenic bacteria, even those that are 'biologically crippled', may survive and multiply in the environment

This topic has been reviewed recently (Jager and Tappeser, 1995). Individual strains of genetically manipulated microorganisms (GMMs) can survive and outcompete wild-type strains. Even when they seem to disappear after release, GMMs can often go dormant and reappear. A laboratory strain of *E. coli* K12, introduced into the sewage, went dormant and undetectable for 12 days before reappearing, having acquired a new plasmid for multidrug resistance that enabled it to compete with naturally occurring bacteria (Tschäpe, 1994).

7.5 Horizontal gene transfer is now known to be responsible for spreading antibiotic resistance and virulence among pathogens

Non-pathogenic GMMs could evolve into pathogenic ones by horizontal gene transfer. *E. coli* 0157 is one such example; its *Shigella*-like toxins have probably been acquired by horizontal gene transfer from *Shigella*.[11] There have been many publications documenting the spread of antibiotic resistance by horizontal gene transfer and recombination (reviewed by Ho and Tappeser, 1997; Ho, 1997a, Chapter 10). The first evidence involves resistance genes acquiring neomycin-kanamycin resistance (Trieu-Cuot *et al.*, 1985). Since then, horizontal transfer of many other antibiotic-resistance genes has been found, including that of tetracycline resistance and even chromosomally encoded penicillin resistance (Ambilecuevas and Chicurel, 1993; Bootsma *et al.*, 1996; Coffey *et al.*, 1995; Kell *et al.*, 1993; Manavathus *et al.*, 1988; Roberts, 1989; Sougakoff *et al.*, 1987; Speer *et al.*, 1992; Spratt, 1988, 1994).

The same mechanisms for horizontal gene transfer have been shown to be responsible for the emergence of virulence among old and new pathogens since the mid-1980s, including *Streptococcus pyogenes* (toxic-shock syndrome) (Kehoe *et al.*, 1996), group A streptococci isolated from a cluster of cases in the 1993 epidemic in Tayside, Scotland (Upton *et al.*, 1996), *Vibrio cholerae* (Bik *et al.*, 1995), *Mycoplasma genitalium* (Reddy *et al.*, 1995). A fuller report on this topic is in preparation (Ho *et al.*, 1997).

The gravity of these findings should be appreciated in the light of the current crisis in public health worldwide, due to the emergence of old and new pathogens which are resistant to multiple antibiotics, as detailed in the 1996 WHO Report. A fuller discussion of this topic has been presented elsewhere (Ho, 1997a, Chapter 10).

7.6 The ability of viral DNA to survive digestion in the gut

This has been demonstrated by feeding viral DNA to mice. Large fragments survived digestion and were passed out with the faeces. At the same time, viral DNA was found in the bloodstream and in many kinds of cells in the body.[12]

7.7 The ability of recombinant vectors to invade mammalian cells

As mentioned in Section 7.6 above, DNA can easily gain access into cells. Studies made since the 1970s have documented the ability of bacterial plasmids carrying a mammalian virus (SV40) to infect cultured mammalian cells, which then proceeded to synthesize the virus. Similarly, bacterial viruses or baculoviruses can also be taken up by mammalian cells (Heitman and Lopes-Pila, 1994). The baculovirus is so effectively taken up by mammalian cells that, as mentioned earlier, it is now being developed as a gene transfer vector for human gene replacement therapy (Hofmann *et al.*, 1995; Sandig *et al.*, 1996), at the same time that it is being engineered for insecticidal purposes (Jehle, 1997).

7.8 Recombination between viral transgenes and viruses generates superinfectious viruses

This ability has been known since 1994. Plants engineered with one of several viral genes or other sequences can acquire resistance to the virus, although the mechanisms are still poorly understood (Sela, 1996). It is now sufficiently well established that viral transgenes can recombine with other viruses to generate new viruses (Anderson *et al.*, 1992; Greene and Allison, 1994; Palukaitis and Roossinck, 1996; Allison, 1997) for the US Department of Agriculture to consider new restrictions on viral-resistant transgenic plants (Kleiner, 1997). Transgenic plants contain the viral gene in all their cells all the time, thus greatly increasing the probability of recombination.

A more general hazard comes from all transgenic organisms, as different viral sequences are incorporated into a range of gene transfer vectors. The cauliflower mosaic virus promoter, for example, is routinely used in vectors for making transgenic plants (Cummins, 1994). These have similar potential to generate new viruses either in the environment or when ingested by human beings and other animals. There have been no experiments carried out to date to investigate this possibility.

7.9 Antibiotics promote horizontal gene transfer

Recent findings show that the presence of antibiotics greatly increases the frequency of horizontal gene transfer, from one to four orders of magnitude (Davies, 1994; Mazodier and Davies, 1991; Sandaa and Enger, 1994; Torres *et al.*, 1991). Antibiotics are already widely present in the environment, from intensive farming and hospital effluents. The potential for spreading antibiotic resistance by horizontal gene transfer may be therefore, much, much higher than previously thought. *There is an urgent need for appropriate monitoring of horizontal gene transfer in limited field releases before further products are approved.*

Chapter 8

A 'Safety Assessment' Designed to Expedite Product Approval with Little or No Real Regard for Safety

Our detailed analysis of the Report leads us to conclude the following:

- it makes biased, partisan claims in favour of genetic engineering biotechnology;
- it wilfully excludes known hazards from safety assessment;
- it ignores existing scientific evidence pointing to the hazards;
- it presents a 'safety assessment' based on an arbitrary and unscientific 'principle of substantial equivalence' that effectively allows the producer to pass any and every product with impunity and with little or no regard for all aspects of safety.

A recent, leaked document (Penman, 1997) indicates that EuropaBio, representing the interests of the industry, is advised by the public relations company Burson Marsteller – whose clients include Babcock and Wilcox during the Three Mile Island nuclear crisis in the US in 1979, and Union Carbide after the Bhopal disaster in India, which killed up to 15,000 people – to 'stay quiet on risks of gene-altered foods' as 'it cannot hope to win the arguments over the risks posed.' We agree.

So why does a Report on food safety produced by such authoritative international agencies like the WHO and FAO not take any of

the risks seriously? The answer is in the same document leaked by Burson Marsteller, which suggests that 'the best way of eliciting a favourable consumer response to new products must be to use regulators and food producers to reassure the public.'

The predominant philosophy behind current regulation can be summarized by the 'No need – don't look – don't see' futile cycle (Fig. 1). Starting from the 'no need, don't look' basis – the assumption that genetic engineering is no different from conventional breeding, so it is not really necessary to provide special oversight – one progresses to the 'don't look, don't see' safety assessment embodied in the principle of 'substantial equivalence', before ending up in a 'don't see, no need' reinforcement of the position with which one began.

As can be seen, this forms a self-reinforcing cycle, a motor for effectively passing with impunity any and every genetically engineered product to the public that the industry so wish. We repeat, the industry have been handed *carte blanche* to do as they please for maximum profitability, with the regulatory body acting to allay legitimate public fears and opposition.

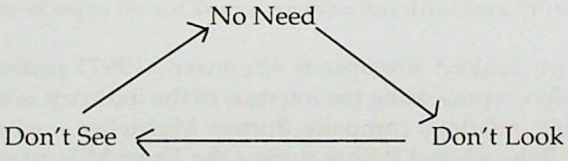

Figure 1. The futile cycle of food safety regulation recommended by the Report

Chapter 9

Recommendations

In view of the gross inadequacies in food safety regulation and the existing scientific evidence pointing to serious hazards, we recommend the following minimum measures to safeguard the health of consumers and to protect biodiversity. A moratorium on further releases must be imposed until these measures are implemented.

a. No food crops are to be engineered for producing pharmaceuticals and industrial chemicals, as the engineered crops could be mistaken for food, or cross-pollinate with non-engineered food crops. The onus must be on the producer to prove that any plant genetically engineered is not a food crop.

b. All projects involving genetic manipulation of the baculovirus for insecticidal purposes should be discontinued, as this virus is being used in human gene therapy and invades human liver cells readily.

c. Complete characterization of inserted gene sequence(s) of the genetically engineered organism (GEO) must be provided in the application for market approval. This should include any antibiotic-resistance marker gene(s), promoter(s) and enhancer(s) and their effects on the expression of neighbouring genes. The presence of mobile genetic elements and other proviral sequences in the host genome, likely to contribute to secondary mobility of inserts, must also be stated.

d. No GEOs with uncharacterized foreign gene inserts are to be considered for release. No parts of such GEOs, nor of animals from failed genetic engineering experiments or xenotransplant animals are to be used as human food or animal feed.

e. No GEOs containing antibiotic-resistance genes are to be considered for release or to be used as human food or animal feed.

f. A detailed record of the stability of the GEO over at least five successive generations under field conditions (including drought and heat) is a precondition for market approval. ('Field conditions' does not mean open field conditions.) This must be supported by appropriate data indicating the stability of the insert as well as the level of gene expression under different conditions in successive generations.

g. Data on the frequency of unintended gene transfers, including horizontal gene transfer from the GEO under field conditions, must be included in the application for market approval.

h. Data on the frequency of horizontal gene transfer from the GEO to gut bacteria must be included in applications for market approval.

i. Data on the ability of transgenes and marker genes in the GEO to invade mammalian cells must be included in applications for market approval.

j. A specified set of tests must be carried out to establish 'substantial equivalence', which are sufficiently discerning to reveal unintended as well as intended effects. The comparator must be the unmodified recipient organism itself, and results of repeated tests must be provided to support the stability of the characteristics over at least five successive generations.

k. Safety assessment must include the GEO's potential to generate pathogens through genetic recombination.

l. Safety assessment must include pesticide residues where they are integral components of the product, as in herbicide-resistant transgenic plants.

m. Product segregation, labelling and postmarket monitoring are non-negotiable conditions for market approval.

Endnotes

1 Personal communication, Pierre Hochuli, Monsanto Europe, April 1997.

2 *Nature Biotechnology* 15, 701, 1997.

3 Despite what is claimed in the title of the paper, the actual observed frequencies were high, from which the authors calculated an extremely low frequency through various unsubstantiated assumptions concerning 'natural' conditions.

4 The work of this group of scientists was reported recently in the *New Scientist*, 4 January 1997, p.24.

5 Reported in *Manitoba Co-Operator* 24/4/97; also *The Ram's Horn*, No. 147, April 1997.

6 *Applications of the Principles of Substantial Equivalence to the Safety Evaluation of Foods or Food Components from Plants Derived by Modern Biotechnology, Report of a WHO Workshop*, SHO/FNU/FOS/95.1, p.7.

7 See note 6.

8 *Health aspects of marker genes in genetically modified plants. Report of a WHO Workshop*, WHO/FNU/FOS.93.6, 1993.

9 See the *New Scientist*, 4 January 1997, p.24.

10 Norwegian decision to prohibit deliberate releases of six genetically modified products approved for marketing in the EU. Third World Network Briefing Paper, circulated in UN Meeting of the *Ad Hoc* Working Group on Biosafety, 13-17 October 1997.

11 Professor Hugh Pennington, BBC Radio 4 Today Programme, February 1997, confirmed by personal communication.

12 See the *New Scientist*, 4 January 1997, p.24.

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