FINAL RISK ANALYSIS REPORT

APPLICATION A362

Food derived from glyphosate-tolerant corn line GA21

Note:

This report is the "Inquiry" as referred to in Section 17 of the *Australia New Zealand Food Authority Act (1991)* and sets out the reasons for making a recommendation to the Australia New Zealand Food Standards Council under Section 18 of the Act.

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EXECUTIVE SUMMARY

Background

ANZFA received an application from Monsanto Australia Ltd on 10 September 1998, for the approval of food derived from glyphosate-tolerant corn line GA21 under Standard A18 – Foods Produced Using Gene Technology. The corn plants have been genetically modified to be tolerant to glyphosate, the active ingredient in the herbicide known commercially as Roundup®. This report describes the scientific assessment of the application.

Issues addressed during assessment

(i) Safety Evaluation

Food derived from glyphosate-tolerant corn line GA21 has been evaluated according to ANZFA's safety assessment guidelines. This involves an extensive analysis of the nature of the genetic modification together with a consideration of general safety issues, toxicological issues and nutritional issues associated with the new genetically modified (GM) food. This approach has been used to establish whether the food produced from the glyphosate-tolerant corn is as safe and nutritious as foods produced from non-GM varieties of corn.

The sources of the new genetic elements present in corn line GA21 are edible plant species such as rice, sunflower and corn itself, or non-pathogenic soil bacteria such as *Agrobacterium.* The detailed information available on the genetic modification shows that one new protein is expressed in corn line GA21 and that the new genetic material is stably inserted and maintained in corn plants over several generations and in different environments.

The results of extensive compositional data do not indicate that there are any biologically significant differences between glyphosate-tolerant corn line GA21, either untreated or following treatment with glyphosate, and the non-transgenic control in any of the parameters measured. Small but statistically significant differences were observed in the levels of five amino acids (arginine, isoleucine, lysine, valine and serine) of the treated GA21 grain in comparison with the control. These differences were not considered significant in terms of safety or nutrition. Similarly, a small but statistically significant difference was observed in the fatty acid profile (specifically in the level of stearic acid) in the treated GA21 line compared to the control. This difference was not considered to be biologically significant as the value was within the known reported range for commercial corn varieties and is not of concern in terms of safety or nutrition.

Evaluations of the potential allergenicity and toxicity did not reveal any changes to these properties in the modified corn when compared to the conventional counterpart.

The safety of the newly expressed protein has been established by consideration of the results obtained from the biochemical and genetic analyses that demonstrate its

similarity to the conventional form of the enzyme in terms of its functional properties and characteristics.

In assessing all of the above data, ANZFA concluded that glyphosate-tolerant corn line GA21 does not raise any public health and safety concerns.

(ii) Labelling

On the basis of the scientific data considered in the safety evaluation, food derived from glyphosate-tolerant corn line GA21 is considered to be substantially equivalent to food derived from non-GM corn. Therefore, under the current standard, mandatory labelling is not required. However, under proposed changes to the labelling provisions within the standard, some foods derived from glyphosate-tolerant corn may be captured by broader labelling requirements.

(iii) Public Submissions

The Authority has conducted two rounds of public consultation for this application. During the first round, a total of fifty-eight submissions were received in response to the combined Preliminary Assessment report. A total of 25 submissions were received in response to the second round of public consultation over a period of ten weeks.

The majority of submissions did not support the approval of this food, based on concerns about the safety of genetically modified food. The food safety concerns raised in submissions have been addressed in the safety assessment report and in this report.

Conclusion

On the basis of available evidence, ANZFA considers that food derived from glyphosate-tolerant corn line GA21 is as safe for human consumption as food from conventionally produced corn varieties and therefore recommends that the Australian *Food Standards Code* be amended to give approval to the use of this food in Australia and New Zealand.

In accordance with the current Standard A18, ANZFA proposes that, as corn line GA21 is equivalent in terms of its nutritional and compositional properties to its conventional counterpart, labelling of this food is not required. Food derived from glyphosate-tolerant corn will, however, be required to comply with the proposed new labelling provisions once the standard is revised.

INTRODUCTION

The Australia New Zealand Food Authority (ANZFA) is a bi-national statutory body responsible for making recommendations on food standards which, when approved by the Australia New Zealand Food Standards Council (ANZFSC), are adopted by reference and without amendment into food law. ANZFA is currently working to establish a joint *Australia New Zealand Food Standards Code* that will apply in both countries. In the interim, a system of dual standards operates for the majority of the food standards. In the case of Standard A18 – Food Produced Using Gene Technology, this Standard has been accepted by New Zealand, and currently applies in both countries.

Standard A18 was adopted by ANZFSC as a joint Australia/New Zealand standard in July 1998 and came into force on 13 May 1999. Under this Standard, the sale of food produced using gene technology is prohibited unless the food is included in the Table to clause 2 of the Standard. The Standard requires that a pre-market safety assessment be conducted on all foods produced using gene technology. However, the Standard provides an exemption for those foods currently on the market provided that an application was accepted by ANZFA on or before 30 April 1999, that the food is lawfully permitted in a country other than Australia or New Zealand, and that ANZFSC has not become aware of evidence that the food poses a significant risk to public health and safety.

BACKGROUND TO THE APPLICATION

ANZFA received an application from Monsanto Australia Ltd on 10 September 1998, to amend the *Food Standards Code* to include food derived from glyphosate tolerant corn in the Table to clause 2 of Standard A18 – Foods Produced Using Gene Technology.

The principle food products are breakfast cereals, baking products, extruded confectionery and corn chips. Corn starch is used extensively by the food industry for the manufacture of dessert mixes and canned foods. A large proportion of corn starch is converted to a variety of sweetener and fermentation products including high fructose corn syrup and ethanol. In addition, corn oil is commercially processed from the germ for domestic vegetable oil markets. Each of these materials is a component of many foods including dairy products, beverages, confectionery and meat products.

Glyphosate is the active ingredient in the herbicide Roundup® which is used widely as a non-selective agent for weed control in primary crops. The mode of action of glyphosate is to specifically bind to and block the acitivity of 5-enolpyruvylshikimate-3-phosphate synthase (denoted as EPSPS), an essential enzyme involved in the biosynthesis of aromatic amino acids in all plants, bacteria and fungi. The genetic modification in corn line GA21 involves specific changes to the corn EPSPS enzyme to produce a modified version (denoted as mEPSPS) which is significantly less sensitive to glyphosate compared to the unmodified form.

The mEPSPS gene was produced by cloning the wildtype EPSPS gene from corn (*Zea mays*) and introducing two changes using *in vitro* techniques. The changes to the gene

result in the production of the mEPSPS protein with a lower binding affinity for glyphosate, thus allowing sufficient enzyme activity for the plants to function normally in the presence of the herbicide.

The applicant claims that the modification provides potential agronomic benefits to primary producers of corn by allowing a more flexible weed control regime leading to improved crop management, more sustainable agricultural practices and reduced production costs. In addition, the applicant anticipates indirect benefits to flow to processors and consumers through the potential lower production costs.

PUBLIC CONSULTATION

ANZFA received the first six applications for foods produced using gene technology from Monsanto Australia Ltd. Due to commonalities in these applications, a combined Notice of Application (formally referred to as the Preliminary Assessment Report) was advertised on 28 October 1998, which called for public comment on the applications. A total of 58 submissions were received in response to the combined Notice of Application, of which 53 relate to this application. The submissions were primarily from individuals, consumer organisations and special interest groups from both New Zealand and Australia. The submissions are summarised in Attachment 5.

ANZFA subsequently conducted an assessment of the application, including a safety evaluation of the food, and prepared a Draft Risk Analysis Report, taking into account the comments received. Following release of this report, the Authority invited a second round of public comment for a period of 10 weeks from 19 June 2000. A total of 25 submissions were received and these are also summarised in Attachment 5.

NOTIFICATION OF THE WORLD TRADE ORGANISATION

During the ANZFA assessment process, comments were also sought internationally from other Members of the World Trade Organization (WTO). As Members of the WTO, Australia and New Zealand are signatories to the agreements on the Application of Sanitary and Phytosanitary Measures (SPS Agreement) and on Technological Barriers to Trade (TBT Agreements) (for further details on WTO, see Attachment 4). In some circumstances, Australia and New Zealand have an obligation to notify the WTO of changes to food standards to enable other member countries of the WTO to make comment.

As there is significant international interest in the safety of these foods, the proposed changes to Standard A18 are considered to raise potential Technical Barrier to Trade or Sanitary/Phytosanitary matters and were therefore notified to the WTO.

ISSUES ADDRESSED DURING ASSESSMENT

1. Safety assessment

The safety assessment was performed according to the safety assessment guidelines prepared by ANZFA[1] and considered the following issues: (1) the nature of the genetic modification; (2) general safety issues such as novel protein expression and the potential for transfer of antibiotic resistance genes to gut microorganisms; (3) toxicological issues; and (4) nutritional issues.

Nature of the genetic modification

Glyphosate-tolerant corn line GA21 was generated using a particle acceleration transformation system. The trait has been introduced into corn plants by the addition of a modified version of the corn gene encoding the EPSPS protein (denoted as mEPSPS), plus controlling elements essential for protein expression in plants. Detailed information on the specific amino acid changes incorporated into the modified enzyme were provided by the applicant and have been classified as confidential commercial information as defined by Section 3 of the *ANZFA Act* (1991) as amended.

As well as the mEPSPS gene, other DNA elements transferred into the corn include the actin 1 promoter and first intron from rice, which has been shown to direct constitutive protein expression in corn, together with an optimised chloroplast transit peptide sequence to direct translocation of the mEPSPS protein to chloroplasts, where the protein is functionally active. The transit peptide sequence is derived from plant sequences obtained from corn and sunflower ribulose 1,5-bisphosphate carboxylase oxygenase (RuBisCo), an enzyme present in all photosynthetic plants.

The 3' untranslated region of the nopaline synthase gene (NOS 3') is present, also for regulatory purposes, providing the appropriate eukaryotic polyadenylation signal. This bacterial sequence is obtained from the Ti plasmid of *Agrobacterium.* Because a purified fragment of DNA was used in the transformation, no extraneous bacterial genes, including laboratory marker genes, were transferred.

Genetic analysis of successive generations of corn plants from this line indicates that the additional gene has been stably integrated into the corn genome.

General safety issues

Corn has a long history of safe use as a food for both humans and other animals. The sources of the new genetic elements present in corn line GA21 are edible plant species such as rice, sunflower and corn itself, or non-pathogenic soil bacteria such as *Agrobacterium.* There are no antibiotic resistance genes present in the modified corn.

One new protein is expressed in corn line GA21, namely the modified corn EPSPS, which carries some specified amino acid changes compared to the wildtype enzyme. Under the regulation of the constitutive plant promoter, the mEPSPS protein is expected to occur throughout the whole plant. Although several methods of analysis showed that the mEPSPS is expressed in the edible grain from the plant at levels approximately ten times higher than endogenous EPSPS expression levels, the safety

[1] ANZFA (1999) Guidelines for the safety assessment of foods to be included in Standard A18 – food produced using gene technology.

assessment concluded that the higher levels did not raise any health concerns. The high prevalence of this family of plant and microbial proteins in the human diet supported this conclusion.

Toxicological issues

Almost three-quarters of the corn kernal is composed of starch with smaller amounts of protein, oil and other nutritionally valuable substances. There are no known naturally occurring toxins in corn and it is not regarded as an allergenic food. Allergenicity testing confirmed that the amino acid substitutions in the modified corn enzyme do not alter the food with respect to its potential allergenicity. Furthermore, an acute oral toxicity study in mice did not indicate any differences in gross pathology between control animals and those administered a single variable dose of the mEPSPS protein. The conclusion from this study is that there is no evidence of toxicity in young, developing animals following administration of the protein at a dose up to 500 times higher than the likely human daily exposure.

The expression levels of the protein were measured and the results indicate that the protein is present in relatively low abundance in the grain. A study on the stability of the new protein in conditions that mimic human digestion, indicate that it is chemically labile and therefore provides strong evidence that the protein is readily digested in the mammalian digestive tract.

Nutritional issues

Comprehensive data from a range of compositional analyses conducted on grain from both untreated and treated corn line GA21 and the non-transgenic control were presented for assessment. The compositional components measured included proximates (protein, fat, ash, carbohydrates, moisture, acid detergent fibre and neutral detergent fibre), amino acid composition, fatty acids profile, calcium and phosphorus. In addition, data were provided on proximates, calcium and phosphorus in the forage, but as this portion of the plant is not for human consumption, the data were not considered in the assessment process.

The results of the compositional data do not indicate that there are any biologically significant differences between glyphosate-tolerant corn line GA21, either untreated or following treatment with glyphosate, and the non-transgenic control in any of the parameters measured. Small but statistically significant differences were observed in the levels of five amino acids in the treated GA21 grain, in comparison with the control. The differences were observed for arginine, isoleucine, lysine, valine and serine, but these were not considered to be significant in terms of safety or nutrition.

Similarly, a small but statistically significant difference was observed in the fatty acid profile (specifically in the level of stearic acid) in the treated GA21 line compared to the control. This difference was not considered to be biologically significant as the value was within the known reported range for commercial corn varieties and is not of concern in terms of safety or nutrition.

Conclusion

From the scientific literature, it is well established that EPSPS enzymes from various plant and microbial food sources are already part of the human diet, have a history of use as a food and feed over thousands of years, and are not associated with any health concerns. The detailed analyses that have been carried out on glyphosate-tolerant corn line GA21 indicate that it may be regarded as equivalent to its conventional counterpart in terms of its composition, safety and end use.

2. Labelling of food derived from corn line GA21

Clause 3 of Standard A18 prescribes mandatory labelling of a food produced using gene technology when it contains new or altered genetic material *and* where it is not substantially equivalent in any characteristic or property of the food. However, under the current standard, there is no requirement for labelling of foods that do not differ substantially from the conventional counterpart in any characteristic or property other than by the presence of a new expressed genetic trait. Consequently, there is no requirement to currently label food derived from corn line GA21.

It should be noted that on 28 July 2000 the Australia New Zealand Food Standards Council agreed to a revised standard which requires labelling of foods produced using gene technology where novel DNA and/or protein is present in the final food and also where the food has altered characteristics (irrespective of whether it contains novel DNA and/or protein). The revised Standard A18 will come into effect 12 months after the date of gazettal. Once the new standard comes into effect, foods derived from glyphosate-tolerant corn line GA21 will have to be labelled according to the new provisions.

3. Issues arising from public submissions

3.1 General issues

It was noted that very few of the comments received during the first round of public consultation relating to the 6 Monsanto applications which were advertised together, specifically addressed the details of the individual applications. Furthermore, many of the submissions received in response to both the first and second rounds of public consultation raised issues of a general nature relating to gene technology or issues that had already been addressed in the safety assessment report (see Attachment 2). A discussion of the general issues relating to gene technology that were raised in public submissions can be found in Attachment 6.

3.2 Specific issues

Issues raised in first round of public comment that are specific to the assessment of this application (see Attachment 5 for summary)

3.2.1 Toxicity of glyphosate

Many submissions express the concern that growing a crop with a herbicide-resistant trait inevitably results in higher levels of usage of the herbicide, with concomitant concerns in relation to the potential toxicity of the herbicide in food and its effects on human health.

Evaluation

This is an issue that is raised frequently in relation to genetic modifications that confer resistance to a herbicide, in this instance, glyphosate. However, as glyphosate is used commercially on conventional crops, this is not an issue that is peculiar to the transgenic corn in this application. In Australia, the National Registration Authority for Agricultural and Veterinary Chemicals (NRA) is responsible for assessing the toxicity of agricultural chemicals prior to their incorporation into farming practices, especially in the production of food crops. This is a rigorous process that entails investigation into the human and animal toxicity of the chemical, its effects on the environment and the potential effects of occupational exposure to the chemical. Consequently, a wide range of scientific data and technical information is taken into consideration when determining the maximum permissible amount of glyphosate residues in food, referred to as the Maximum Residue Limit (MRL).

The toxicity of glyphosate has been extensively studied in animal testing of a range of different species including rats, dogs, mice, rabbits, guinea pigs and monkeys. The testing of the toxicity of glyphosate also included long term studies in which animals were exposed to varying levels of the herbicide over periods of time in excess of 2 years. An assessment of this toxicological data has been undertaken by the Commonwealth Department of Health and Aged Care to support the establishment of acceptable daily intake levels. The results of the animal studies indicate that glyphosate exhibits a very low degree of toxicity.

Furthermore, in the agricultural environment, when applied to emerged weeds, glyphosate shows no residual activity. This is because it binds strongly to soil particles and is readily broken down by soil microorganisms. Because of the rapid transportation from the leaves of treated plants to the roots, it is effective in destroying perennial weeds that can survive other herbicides that only affect the above-ground parts of the weed plant.

Toxicity of other chemicals in Roundup® herbicide

Polyoxyethyleneamine (POEA) is used as a surfactant in agricultural chemicals, including glyphosate formulations, and in other preparations for human use, for example, shampoo. In June 1996, the NRA issued a Community Brief advising the public that it had undertaken a review of the use of glyphosate formulations in and around aquatic areas with particular reference to the toxicity of surfactants to aquatic organisms. This was in response to a report which showed the surfactant in certain glyphosate formulations to be more harmful to frogs than the active ingredient, glyphosate. Following this review, the NRA made specific proposals relating to the inclusion of a warning statement on all agricultural glyphosate product labels precluding use on or adjacent to waterways.

3.2.2 Transfer of genes from different species

Several submitters including Noeline Gannaway and Colin Kell expressed opposition to genetic modification of food crops because the process often involves the introduction of genes from other species, unrelated to the particular plant.

Evaluation

In this application, the gene introduced into corn line GA21 is a corn gene, carrying two minor modifications that were introduced in the laboratory. The modifications were selected on the basis of previously reported detailed scientific information on the enzymatic properties of the protein product. Therefore, in this case, there has been no transfer of foreign genes from a different species.

3.2.3 Commercial in confidence status of application

The National Council of Women of Australia expressed an objection to the Commercial-in-Confidence status on a section of the application concerning specific molecular changes to the corn EPSPS enzyme. The Council asserts that the consumer is being asked to accept the product with blind confidence that the food will not result in adverse health effects.

Evaluation

Section 3 of the *Australia New Zealand Food Authority Act 1991* as amended, defines the essential criteria for confidential commercial information relating to food. This section provides legal protection, under specified circumstances, to information that may have been provided to the Authority by any member of the community in relation to a particular food. Any request for confidentiality is assessed case-by-case on the basis of a written submission to justify the claim in terms of the necessary criteria. Once approved as a valid claim, Section 39 of the ANZFA Act prohibits the disclosure of confidential commercial information by way of public register files.

In relation to glyphosate tolerant corn line GA21, the applicant claimed that specific molecular information was commercial-in-confidence and therefore should not appear on public register files. Accordingly, the nature of the information was assessed as to its commercial value and the claim justified for the following reasons.

On the basis of published data on the glyphosate binding affinity of naturally occurring EPSPS enzymes, the applicant has intentionally altered two of 445 amino acids comprising the whole enzyme. The specific alteration has decreased the sensitivity of the corn EPSPS to glyphosate and conferred resistance to the herbicide at the whole plant level. This discovery has a commercial value and, if revealed, would allow competitors in the biotechnology field to readily reproduce the product. The changes, per se, are of no significance to the normal functioning of the plant at either the DNA or protein level. In relation to the food, the amino acid substitutions are conservative in the sense that they represent amino acids common to all protein of biological origin consumed as part of normal human diets. Nevertheless, as an integral part of the safety assessment of corn line GA21, the specific changes have been considered in terms of their impact on particular characteristics of the protein in the

plant, and the potential of the modified protein to cause adverse effects when consumed as food.

In summary, the confidential status of the specific changes incorporated into mEPSPS do not compromise the rigour of the safety assessment process, as all relevant details have been provided to ANZFA. The applicant's request for confidentiality has been assessed as valid in this case as the circumstances are consistent with the criteria for confidential commercial information, as set out in Section 3 of the *ANZFA Act*.

Issues raised in second round of public comment that are specific to the assessment of this application (see Attachment 5 for summary)

(i) *Toxicity of POEA*

One submitter, G. C. Morgan from New Zealand, commented on the review of the toxicity of glyphosate preparations containing the surfactant, polyoxyethyleneamine (POEA) by the National Registration Authority for Agricultural and Veterinary Chemicals (NRA) in 1996. The Draft Risk Analysis Report for this application stated that the outcomes of the NRA review required inclusion of a warning statement on commercial glyphosate preparations and precluded the use of the formulation on or adjacent to waterways.

Monsanto Australia Ltd. submitted that ANZFA's discussion on the use of POEA in glyphosate preparations was not complete, and that the report included no information about the variety of different formulations of Roundup® herbicide.

Response

In the Draft Risk Analysis Report to this application there is a discussion on the POEA surfactant in commercial preparations of Roundup® (see above in Issues raised in the first round of public comment).

In response to this discussion, the applicant advised ANZFA that Roundup® has many formulations worldwide and not all of these formulations contain POEA. The company further advised that following the 1996 review, although many agricultural glyphosate products required changes, this did not include all formulations. For example, Roundup Biactive® is actually registered for use in and on waterways because the surfactant in this particular formulation is approved on the basis of a rigorous toxicological assessment and profile.

Other POEA based formulations of glyphosate that were registered for use in waterways were the subject of more restrictive labelling requirements. In addition, still other POEA based formulations of glyphosate that were not registered for use in waterways, were required to incorporate changes to the precautionary statements already on the label.

It should be noted that the toxicity of these substances is evaluated as part of the comprehensive toxicological assessment of the herbicide conducted by the NRA and these considerations are critical to the determination of a Maximum Residue Limit for the herbicide in a food. These matters are therefore already an integral component of

the safety assessment processes, and apply to conventional crops as much as to genetically modified crops since the herbicide is also used at particular stages of development on traditional unmodified crops (for example, at the pre-emergent stage and immediately before harvest, depending on the crop in question).

This issue therefore is not a primary consideration for food safety in relation to the genetic modification in glyphosate-tolerant corn. It is considered during the course of assessments to determine a Maximum Residue Limit for the use of the herbicide on a food crop, including crops that are conventionally produced.

(ii) Effects of the gene insertion

The submission from Canberra Consumers Incorporated included comment on several technical points relating to the genetic modification, and stated that although the inserted gene is a corn-derived gene, there are examples of plants where insertion of additional (same-species) genes (for example, in Application A387 – High Oleic Soybeans) has had a major effect on plant composition.

Response

As stated in the Safety Assessment (Attachment 2), the mEPSPS gene is a modified corn gene that has been introduced into corn plants via standard plant transformation techniques. The introduced gene is under the control of a plant derived promoter sequence from rice and provides for constitutive expression of the modified protein. Although the precise location of the insertion site is not defined, the insertion event itself is well characterised. The molecular data provides information on the nature of the genetic elements incorporated into the plant DNA (that is, whether whole or partial insertions occurred) and the number of insertion events. The functional properties of the newly introduced gene, are also well defined in terms of data on the expression of the new protein in corn kernels and other plant tissues.

From the initial transformation event, the characteristics of the modified line have been assessed and monitored in the laboratory, the glasshouse, field trials and finally to commercialisation. The glyphosate-tolerance trait can be easily observed by application of the herbicide to the plant, and this served as the means of selection throughout the developmental stages, obviating the requirement for the inclusion of additional marker genes (for example, antibiotic resistance genes) to monitor transgene expression.

Moreover, the compositional data confirm that the transformed line exhibits the expected levels of the major nutritional components of corn. Taken together, these data contribute to the overall conclusion that the modified corn is equivalent to the unmodified form in terms of food safety.

The precise chromosomal location of the endogenous (natural) EPSPS gene is not known and therefore it is not possible to determine if the newly introduced mEPSPS gene is located on the identical chromosome. However, the location of either of the genes is not a necessary factor for studying their functional properties and is certainly not an essential feature of the assessment in relation to food safety. The transformation process has not resulted in any manipulation of the endogenous gene

and it remains in the plant. The enzyme activity of the new mEPSPS protein substitutes for the lack of the natural EPSPS activity only in the presence of glyphosate, when the herbicide inactivates the natural enzyme. The modified plants then rely on the introduced mEPSPS activity (via expression of the transgene), for continuing cellular function.

This type of genetic modification should not be confused with the phenomenon of gene silencing which occurs in Application A387 – High Oleic Soybeans. In this application, the insertion of a second identical copy of the *GmFad 2-1* gene into soybean plants has resulted in the genetic silencing of the endogenous *GmFad 2-1* gene. The silencing of the two genes has predictably resulted in substantial compositional changes to the fatty acid profile of the soybeans and those changes were the focus of the safety assessment for that application.

Although gene silencing is a relatively recently described phenomenon, it has been subject to intense research and investigation over the past decade and a significant body of literature exists on the biochemical mechanisms that underlie the phenomenon. Research is still ongoing but it is becoming increasingly apparent that gene silencing is a normal mechanism of gene control in plants.

Homology-dependent gene silencing in transgenic plants may operate either at the transcriptional level or the post-transcriptional level. Silencing at the transcriptional level is usually associated with methylation of the promoter and the suppression of RNA transcription whereas silencing at the post-transcriptional level is usually associated with a failure to accumulate messenger RNA, probably through targeted degradation.

Gene silencing is one possible outcome of plant transformation when using particular genetic constructs. It has been estimated that many thousands of transformed cells are screened in the laboratory for the desired phenotype, and only those cells which respond in the intended manner, are retained for further analysis and progression to later stages of development.

In summary, the safety assessment considers data on the nature of the genetic modification, general safety issues including toxicity and allergenicity and nutritional issues including the levels of key components of the food, together with the levels of anti-nutritional factors or natural toxicants. This approach ensures that both the intended and unintended effects of the modification are considered in determining the nutritional properties and compositional characteristics of genetically modified foods.

(iii) Comparison of data

Several submitters commented that where small but statistically significant differences were measured in some of the compositional analyses between the genetically modified corn and the control, the assessment allowed for a broader comparison of values corresponding to a literature or reference range for that parameter.

Response

If a comparison of the compositional data reveals a statistically significant difference between the modified plant and the control in any of the parameters tested, that difference becomes the subject of further evaluation. The evaluation allows for a broader comparison with values in a published literature range to account for the considerable natural variation in plant composition due to environmental factors, even when plants are grown in the same location and harvested at the same time.

The focus of the evaluation is to consider whether the difference represents a food safety concern rather than to demonstrate absolute identity with the comparator. This approach is consistent with internationally accepted assessment methods and is widely adopted by food regulatory agencies in Europe and the United Kingdom[2].

4. Risk management

Under Standard A18 a GM food must undergo a safety assessment in accordance with ANZFA's safety assessment guidelines. The requirement for the food to be labelled must also be assessed in accordance with the labelling criteria specified in Clause 3 of the standard.

On the basis of the conclusions of the safety assessment report, together with a consideration of the public submissions, it is proposed that Table 1 to clause 2 of Standard A18 be amended to include food derived from glyphosate-tolerant corn line GA21. The proposed amendment is provided in Attachment 1.

In relation to labelling of the food, the safety assessment report found that food derived from glyphosate-tolerant corn line GA21 is considered to be equivalent to food derived from conventional corn in terms of the general properties and characteristics which define it as a food. Therefore, under the current standard, mandatory labelling is not required. However, under proposed changes to the labelling provisions within the standard, some foods derived from glyphosate-tolerant corn will be captured by broader labelling requirements.

In relation to the concerns raised in the public submissions with regard to gene technology and GM food, ANZFA has prepared a public discussion paper on the safety assessment process for GM food[3]. This is widely available and may assist in addressing some of the concerns raised by the public. Other government and industry bodies are also addressing the broader concerns in relation to gene technology.

5. Regulatory Impact Assessment

The benefits and costs associated with the proposed amendment to Standard A18 have been analysed in a draft Regulatory Impact Statement (Attachment 3). The benefits of the proposed Standard A18 amendment to approve food derived from glyphosate-

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[2] Joint FAO / WHO Expert Consultation on Foods Derived from Biotechnology, Switzerland, 29 May – 2 June 2000.

[3] ANZFA (2000) GM foods and the consumer: ANZFA's safety assessment process for genetically modified foods. ANZFA Occasional Paper Series No. 1.

tolerant corn primarily accrue to the food industry and government, with potentially a small benefit to the consumer.

CONCLUSIONS

ANZFA recommends the adoption of the draft variation (Attachment 1) for the following reasons:

- the introduced genetic elements in glyphosate-tolerant corn line GA21 are not considered to produce any increased public health and safety risk;

- based on the data submitted in the present application, glyphosate-tolerant corn line GA21 is not significantly changed with respect to any nutritional property or characteristic and may be considered equivalent to non-GM corn in terms of food safety and nutritional adequacy;

- food derived from glyphosate-tolerant corn line GA21 is not required to be labelled under the current Standard A18, as it is substantially equivalent to food derived from non-genetically modified corn. The proposed amendments to the labelling provisions of Standard A18 could result in changes to the way in which certain food fractions derived from glyphosate-tolerant corn is to be labelled in the future; and

- the benefits to government, consumers and industry associated with the proposed amendment outweigh the costs.

ATTACHMENTS

1. Draft variation to the Australian *Food Standards Code*
2. Final safety assessment report
3. Regulatory impact assessment
4. World Trade Organisation Agreements
5. Summary of public comments
6. General issues raised in public comments

ATTACHMENT 1

DRAFT VARIATION TO THE AUSTRALIAN FOOD STANDARDS CODE

A362 – FOODS DERIVED FROM GLYPHOSATE-TOLERANT CORN

To Commence: on gazettal

Standard A18 *is varied by inserting into Column 1 of the Table to clause 2 -*

Food derived from glyphosate-tolerant corn line GA21.

If Standard 1.5.2 has been adopted by the Ministerial Council at the time this recommendation is considered, the following applies –

To Commence: on gazettal

Standard 1.5.2 *is varied by inserting into Column 1 of the Table to clause 2 -*

Food derived from glyphosate-tolerant corn line GA21.

ATTACHMENT 2

SAFETY ASSESSMENT REPORT

A362 – FOOD DERIVED FROM GLYPHOSATE-TOLERANT CORN LINE GA21

SUMMARY AND CONCLUSIONS

Nature of the genetic modification

Glyphosate-tolerant corn line GA21 has been developed primarily for agricultural purposes to provide growers with a crop that is tolerant to applications of the broad spectrum herbicide, glyphosate. This trait has been introduced into corn plants by the addition of a modified corn gene encoding the EPSPS protein, a key enzyme in the biosynthesis of aromatic amino acids in plants and microbes. The specific amino acid changes incorporated into the modified enzyme (denoted as mEPSPS) were provided by the applicant and have been classified as confidential commercial information at the applicant's request. The modification alters the sensitivity of the enzyme to glyphosate, resulting in sufficient enzyme activity to allow the plant to function in the presence of the herbicide.

As well as the mEPSPS gene, other DNA elements transferred into the corn include the rice actin promoter and intron, which have been shown to direct constitutive protein expression in corn, together with an optimised chloroplast transit peptide sequence to direct translocation of the mEPSPS protein to chloroplasts, where the protein is functionally active. The NOS 3' untranslated region is present, also for regulatory purposes, providing the appropriate eukaryotic polyadenylation signal. Because a purified fragment of DNA was used in the transformation, no extraneous bacterial genes, including laboratory marker genes, were transferred.

General safety issues

There is a comprehensive set of analytical data for the safety assessment of the transgenic corn. There is only one new protein, namely the mEPSPS enzyme, produced by the genetic modification to the corn plants. Despite some amino acid changes, the mEPSPS shows more than 99.3% homology with the conventional corn EPSPS enzyme. The new protein is present in corn grain at levels approximately ten times that of the endogenous corn protein, however the family of EPSPS proteins are ubiquitous in plant and microbial food sources which are already part of human diets.

Toxicological issues

Corn has undergone substantial genetic breeding by conventional methods over many centuries and has been safely consumed as food and feed for thousands of years. As the changes to the corn enzyme involve substitutions with standard amino acids common to all proteins of biological origin, and do not alter the functional properties of the enzyme, the mEPSPS protein is not considered to be inherently toxic. This was supported by the results of an acute toxicity study in mice, where animals given a variable single dose of the purified mEPSPS protein showed no clinical signs of toxicity and continued to grow normally for the duration of the 14 day study.

Similarly, there is no evidence to suggest that the transgenic corn would be more likely to cause allergies than the conventional counterpart. The mEPSPS lacks similarity to known allergens and protein toxins, is rapidly degraded in simulated digestive systems and occurs at low levels in the protein fraction of the grain.

Nutritional issues

The results of extensive compositional analyses on both treated and untreated plants demonstrate that the levels of the important components in corn grain (protein, total fat, carbohydrate, ash, fibre, fatty acids, amino acids and moisture) and the minerals calcium and phosphorus in this transgenic line are comparable to the non-transgenic control and to available published literature ranges. The safety of the mEPSPS protein to humans is therefore established by consideration of the results obtained from the biochemical and genetic analyses that demonstrate its similarity to the conventional form of the enzyme in terms of its functional properties.

Conclusion

From the scientific literature, it is well established that EPSPS enzymes from various plant and microbial food sources are already part of the human diet, have a history of use as a food and feed over thousands of years, and are not associated with any health concerns. Based on the data submitted in the present application, the conclusion from this assessment is that glyphosate-tolerant corn line GA21 is compositionally equivalent to unmodified corn varieties, and is therefore suitable for human food use with respect to its safety, nutritional properties and wholesomeness.

1. BACKGROUND

Monsanto Australia Ltd have submitted an application to ANZFA to vary Standard A18 of the Australian *Food Standards Code* to include food products derived from glyphosate-tolerant corn, known commercially as Roundup Ready® (RR) corn.

Glyphosate is the active ingredient of the herbicide Roundup® which is used widely as a non-selective agent for controlling weeds in primary crops. The mode of action of glyphosate is to specifically bind to and block the activity of 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS), an essential enzyme involved in the biosynthesis of aromatic amino acids in all plants, bacteria and fungi. Biochemical studies on the EPSPS enzyme from a variety of different species have shown that a natural variation in glyphosate binding affinity exists, particularly across bacterial species (Schultz *et al*. 1985). Further studies on bacterial and plant EPSPS enzymes demonstrated that sequence changes at the active site of the enzyme, a highly conserved region across species, could alter substrate and inhibitor binding properties (Padgette *et al.*, 1991). Tolerance to glyphosate in plants can therefore be achieved by introducing a version of the EPSPS gene producing a protein with a reduced binding affinity for glyphosate, thus allowing the plant to function normally in the presence of the herbicide.

The RR corn described in this application is glyphosate-tolerant corn line GA21. In this line, the glyphosate-tolerant trait is generated in the plants through specific changes to the corn (*Zea mays*) gene which results in the production of a modified EPSPS enzyme, the so-called mEPSPS protein. The modification produces an enzyme which is less sensitive to glyphosate, compared with the unmodified corn enzyme, and thus imparts glyphosate tolerance to the whole plant. The mEPSPS protein exhibits more than 99.3% amino acid homology with the conventional corn EPSPS protein.

Corn is used in the manufacture of breakfast cereals, baking products, extruded confectionery and corn chips. Corn starch is used by the food industry for the manufacture of dessert mixes and canned foods.

Approximately 30% of the corn grown in Australia is manufactured into foods for human consumption, with the remainder used as stockfeed or exported. A small proportion (400 tonne in 1995/96) of corn products is imported in the form of high-fructose corn syrup, according to market demand. In New Zealand also, crop planting regimes are variable. Due to the diverse uses of corn products, there is a requirement to import corn products, mainly in the form of high-fructose corn syrup, to meet manufacturing demand. The RR trait has not been introduced into sweet corn or popcorn varieties.

2. DESCRIPTION OF THE MODIFICATION

Monsanto studies submitted:

D.A. Dixon, L.A. Turner, T.A. Dowey, T.C. Lee and J.N. Leach, 1997. Molecular Analyses of Roundup Ready Corn Line GA21. Performing Laboratory: Monsanto Company, Molecular Analysis Centre, Report No. MSL-15205, Study 97-01-46-10.

R.P. Lirette, D.A. Dixon, S-Z. Pang, L. Albee, R. Krieb, C. Hironaka, J. Astwood and R.S. Sidhu,1998. Additional molecular characterisation of Roundup Ready® corn line GA21. Performing Laboratory: Monsanto Life Sciences Company, Report No. MSL-15335, Study 97-01-46-12.

2.1 Methods used in the genetic modification

Corn line GA21 was generated by transformation of corn (*Zea mays*) using a particle acceleration transformation system. This method of transformation allowed for a specific linear fragment of DNA incorporating only the gene of interest together with essential controlling elements to be transferred to the plant. Since the introduced DNA contains a gene encoding for herbicide tolerance (in this case, the mEPSPS gene), the plant cells are grown in the presence of glyphosate and only those cells which carry the DNA modification continue to grow.

2.2 Function and regulation of the novel genes

A specific 3.4 kb DNA fragment from plasmid pDPG434 was purified by agarose gel electrophoresis and subsequently introduced into embryogenic corn cells. The purified fragment, referred to as the gene cassette, contained only the modified corn EPSPS gene fused to an optimised chloroplast transit peptide sequence and controlling DNA elements essential for expression in plant cells (see below). The mEPSPS gene is under the regulation of the rice actin promoter and rice actin intron, and the 3' untranslated region of the nopaline synthase gene (NOS 3') from the Ti plasmid of *Agrobacterium*.

Although plasmid pDPG434 contained other bacterial genes and controlling sequences for selection and replication in the laboratory, these sequences were not contained within the purified fragment used in the transformation and therefore are not present in the plant.

2.2.1 Actin Promoter

To direct expression of the inserted gene, a rice actin promoter was used (McElroy *et al*, 1990.). The promoter region is comprised of a 1.37 kb DNA sequence corresponding to the 5' region of the rice actin 1 gene, containing the promoter site and first intron. This plant derived promoter provides for constitutive expression in all parts of the corn plant.

2.2.2 The mEPSPS gene

The modified EPSPS gene was produced by cloning the wildtype EPSPS gene from corn (*Zea mays*) in plasmid pDPG434 and introducing specific changes to the DNA using standard *in vitro* techniques (see Padgette *et al*., 1991). Although full details of the exact nature of the changes have been provided to ANZFA for assessment, at the request of the applicant, this information has been deemed confidential commercial information. The specific changes to regions of the corn gene result in the production of the mEPSPS protein with enzymatic activity that is glyphosate-insensitive relative to the unmodified form of the enzyme.

2.2.3 The chloroplast transit peptide

Natural EPSPS enzyme is located within chloroplasts, the site of aromatic amino acid biosynthesis in plant cells. As for many proteins with subcellular locations, newly synthesised preproteins are directed to a particular organelle by a transit peptide usually at one end of the mature protein. Following delivery to the organelle, the short transit peptide is cleaved from the mature protein and is rapidly degraded (della-Cioppa *et al*. 1986).

To direct the mEPSPS protein to the chloroplast of plant cells, the mEPSPS gene is fused to chloroplast transit peptide (CTP) sequences, to generate an optimised transit peptide (OTP) (Lebrun et al., 1996). These transit sequences are derived from plant sequences isolated from corn and sunflower ribulose 1,5 –bisphosphate carboxylase oxygenase (RuBisCo). The mEPSPS gene with its OTP sequence is approximately 1.7 kb in size. The entire deduced amino acid sequence of the mEPSPS preprotein including the 125 amino acids of the OTP was provided. The fusion of the mEPSPS with the OTP results in an additional amino acid (methionine) at the amino-terminal end of the mature mEPSPS protein, following cleavage of the transit peptide.

2.2.4 NOS 3' untranslated region

The NOS 3' untranslated sequence is derived from the common soil bacterium *Agrobacterium tumefaciens*. It is a region of DNA copied from the bacterial nopaline synthase gene (*nos*) that operates in plants by providing the polyadenylation signal for stable expression.

2.3 Characterisation of the genes in the plant

2.3.1 Molecular characterisation of corn line GA21

Molecular characterisation of the integrated DNA present in Roundup Ready corn line GA21 was performed using untransformed corn DNA and plasmid pDPG434 as reference material. Using Southern blot analyses, genomic DNA from the transformed line was analysed for the number of sites of insertion of the plasmid DNA into the corn genome and the integrity of the genetic elements contained within the inserted DNA.

The results of experiments using several different probes specific for the purified DNA fragment showed that a single insertion event has resulted in the introduction of a segment of DNA of approximately 18.5 kb. Further analysis demonstrated that the inserted segment contained three complete copies in tandem of the plasmid fragment used in the transformation plus an incomplete copy. The partial copy has been demonstrated to include the rice actin promoter and a truncated mEPSPS gene which lacks the NOS 3'untranslated region.

Further evidence in support of this molecular characterisation was provided by the results of a Western blot analysis performed in order to assess the equivalence of the modified EPSPS protein produced by corn line GA21 to that expressed in bacteria in the laboratory. This experiment showed that only one immunoreactive protein of the

expected apparent molecular weight (approx. 47 kD) is found in crude extracts of the transformed corn tissue.

2.3.2 Characterisation of the 5' and 3' ends of the inserted DNA in GA21 corn

The above characterisation (see 2.3.1) of the modification in line GA21 including the results from Southern blot analyses, allowed predictions of the genetic makeup of the inserted DNA based on size and sensitivity to restriction enzyme digestion. These reports established that the inserted DNA of line GA21 contains, as a single insert, three copies of the mEPSPS gene cassette, plus an incomplete copy consisting of the rice actin promoter, the optimised transit peptide and a truncated mEPSPS sequence without the NOS 3' untranslated region. However, these studies did not provide molecular detail about the ends of the inserted DNA, particularly where these are adjacent to flanking corn DNA. With regard to the direction of transcription (and hence protein synthesis) the starting end of the DNA segment is denoted as the 5' end, with the opposite end denoted as the 3' end.

Testing and experiments in the laboratory allowed for cloned segments of the corn GA21 genomic DNA to be analysed in greater detail by direct DNA sequencing. The DNA sequence data was verified by polymerase chain reaction (PCR) analysis of genomic DNA from the GA21 corn. Sequence analysis showed a truncated rice actin promoter at the 5' end of the GA21 insert. The truncated promoter contained the last 148 base pairs (bp) of the 3' end of the rice actin promoter including the rice actin intron. This partial rice actin promoter is expected to be functional and enable the production of the full length mEPSPS protein, based on the detailed characterisation of the rice actin promoter in the published literature (McElroy et al., 1990).

Detailed analysis at the 3' end of the GA21 insert established the presence of the full length rice actin promoter, the optimised transit peptide and, as expected, a truncated mEPSPS gene, but lacking the NOS polyadenylation signal. This truncated sequence contained the first 289 bp of the mEPSPS coding sequence, terminating in a translational stop codon. Northern blot analysis showed that, whereas a stable transcript was detected for the complete mEPSPS gene cassette, the truncated mEPSPS sequence does not produce a detectable transcipt. Further evidence that the truncated gene does not express protein was provided by Western blot analysis which showed that only a single band corresponding to the full length mEPSPS protein was expressed in GA21 corn.

The DNA sequence analysis of the 3' end of the GA21 inserted segment also showed the presence of a partial mEPSPS cassette containing the rice actin promoter truncated before the start of the rice actin intron, and fused at the 3' end to corn genomic DNA. Based on DNA sequence data, two putative overlapping open reading frames, ORF-1 (97 amino acids) and ORF-2 (19 amino acids) were identified. However, Northern blot analysis using poly (A+) RNA prepared from leaf tissue of corn line GA21 demonstrated that there was no detectable RNA transcript of this region. This finding is supported by detailed published studies on the rice actin promoter elements which report that in the absence of the intron, protein is not produced (McElroy et al., 1990).

Despite the lack of evidence that the region of proximal corn DNA is transcribed, further investigations were carried out. The putative amino acid sequences

corresponding to ORF-1 and ORF-2 were compared to all known allergens and toxins present in public domain databases. No sequence similarity was found for either ORF-1 or ORF-2 when the comparisons were done according to established criteria for allergen screening (Metcalfe et al., 1996). Similarly, neither ORF-1 nor ORF-2 encoded amino acid sequences meeting established criteria suggesting homology to protein toxins (Doolittle, 1990).

The database searches indicated that the DNA sequences corresponding to ORF-1 and ORF-2 were found to be homologous to highly repetitive DNA regions commonly found in the intergenic regions separating functional genetic loci in corn which represent more than 50% of the corn genome (San Miguel et al., 1996). This is further evidence that the putative ORFs associated with the 3' proximal corn genomic DNA at the integration site in corn line GA21, would not produce any protein.

The results of all of these analyses confirm the characterisation of the inserted DNA. Glyphosate-tolerant corn line GA 21 contains a single inserted segment of DNA which expresses only the full length mEPSPS protein.

2.4 Stability of the genetic changes

Progeny from successive backcrossing of the transgenic line were tested for five generations and the data indicate that the inserted DNA is stably integrated into the corn genome. Analysis of the progeny from one generation of self pollination of the fifth generation of backcrossed plants (BC5F2) also demonstrates the stability of the modification according to Mendelian inheritance. These results are consistent with the genetic and molecular analyses described above.

Conclusion

Based on the detailed molecular characterisation, glyphosate-tolerant corn line GA21 contains, as a single insert, four functional mEPSPS gene cassettes plus a truncated mEPSPS cassette that does not produce a detectable RNA transcript. The only protein expressed from the inserted DNA is the full length mEPSPS protein.

3. GENERAL SAFETY ISSUES

Monsanto studies submitted:

T. C. Lee and M. Bailey, 1996. Assesment of the equivalence of CP4 EPSPS protein produced in *Escherichia coli* and in several insect protected , Roundup Ready® and insect protected/Roundup Ready® corn lines. Performing Laboratory: Monsanto Company, CEREGEN, Report No. MSL-14746, Study 95-01-50-05.

Glyphosate-tolerant corn line GA21 has been assessed according to ANZFA's paper entitled 'Guidelines for the safety assessment of foods to be included in Standard A18 – Food Produced Using Gene Technology' relating to Group D foods. This process is applicable to foods derived from a plant or animal that contains new or altered genetic material (ANZFA 1999).

3.1 History of use

Corn (*Zea mays* L., also called *maize*) has a long history of safe use as a food for both humans and other animals. Being the only important cereal crop indigenous to North America, it has been utilised for thousands of years and was the foundation of the extensive North and South American ancient civilisations. Corn seed was carried to Europe centuries ago, where it became established as an important crop in southern latitudes, moving rapidly to Africa, Asia and other parts of the world.

In countries where corn is an important crop, it is the principal component of livestock feeds, and most of it is fed to farm animals, particularly to ruminants. In only a few countries is corn a major constituent of human diets. In developed countries, corn is consumed mainly as popcorn, sweet corn, corn snack foods and occasionally as corn bread. However, most consumers are not aware that corn is an important source of the sweeteners, starches, oil and alcohol used in many foods, beverages and numerous other products.

In the United States, corn is the largest crop in terms of planted acreage, total production and crop value (National Corn Growers Association, 1997). While corn is generally used as a high energy animal feed, it is also a very suitable raw material for the manufacture of starch which is largely converted to a variety of products for human consumption, such as sweetener and fermentation products including high fructose corn syrup and ethanol. Corn oil is commercially processed from the germ and accounts for approximately nine percent of domestic vegetable oil production. Little whole kernel or processed corn is consumed by humans worldwide when compared to these corn-based food ingredients that are used in the manufacture of many foods including bakery and dairy goods, beverages, confections and meat products.

3.2 Nature of novel protein

As part of the safety assessment of glyphosate-tolerant corn line GA21, the assessment examines the expressed products of the introduced genes and considers the levels of new protein in the grain. In this line, the pre-mEPSPS protein is the only expressed protein product from the inserted gene cassette, the other DNA elements being controlling sequences. Under the regulation of the rice actin promoter, the mEPSPS protein is expected to occur throughout the whole plant, since this promoter has been shown to drive constitutive expression in genetically modified corn.

Since the EPSPS protein is naturally present in plants, bacteria and fungi as part of the basic biochemical makeup of the organism, several scientific studies have compared the amino acid sequences and catalytic properties of the enzyme from a wide variety of different sources (for example, see Schultz *et al*., 1985). Data from these studies shows differences in amino acid sequence of the enzyme from different species, including bacteria and fungi, which directly alters the sensitivity to glyphosate of these naturally occurring forms of the enzyme. Such information promoted further study of the catalytically important amino acid residues of this key metabolic enzyme and provided the scientific background material for development of the mEPSPS in this application. Therefore, with respect to this enzyme in the environment, considerable sequence variation exists across species, and several naturally occurring

versions exhibit a concomitant range of natural tolerance levels to the herbicide glyphosate.

The mEPSPS gene has been completely sequenced and encodes a protein of 47.4 kDaltons consisting of a single polypeptide of 445 amino acids. The amino acid sequence identity between the modified enzyme and the wildtype enzyme from corn is greater than 99.3%.

The new protein was expressed in *E. coli* and purified to allow characterisation of its enzymatic properties. Kinetic and enzyme activity analyses indicate that the mEPSPS enzyme interacts with the normal EPSPS substrates, shikimate-3-phosphate and phosphoenolpyruvate, similarly to the wildtype corn EPSPS enzyme.

3.3 Expression of the novel protein in the plant

Monsanto study:
T.C. Lee *et al*., 1997. Assessment of the Equivalence of Modified Maize 5-Enolpyruvylshikimate-3-phosphate Synthase (mEPSPS) Produced in *Escherichia coli* and in the Roundup Ready® Maize Line GA21.

To verify expression, levels of the new protein were evaluated in forage and grain samples collected from five field locations in the U.S. during the 1996 growing season, using an Enzyme Linked Immunosorbent Assay (ELISA). As for these and other tests, corn plants identified by PCR as negative segregants were used as controls.

The polyclonal antibody used in the assay system was also an appropriate reagent for the detection of wildtype corn EPSPS as well as the mEPSPS protein, however the expression of wildtype EPSPS was below detectable levels in the grain of the control samples. It is known that the expression of endogenous EPSPS in plant tissues is very low relative to microorganisms (Mousdale and Coggins, 1984). For treated transgenic GA21 grain, the mean expression of EPSPS protein, representing the sum of the endogenous and modified corn EPSPS expression levels, was 4.6 μg/g fresh weight (range was 1.7-7.4 μg/g fwt). The quantitation limit of the EPSPS ELISA assay was approximately 0.8 μg/g for grain. The results of this assay showed that the expression of mEPSPS protein in corn line GA21 was at least one order of magnitude greater than that of the wildtype EPSPS expressed in the non-transgenic control.

Western blot analysis was used to further assess the expression of the mEPSPS in the modified corn. This technique provides for high specificity and also allows for comparison of the apparent size (molecular weights) of proteins with immunological cross-reactivity present in complex mixtures of crude protein extracts from corn. In particular, this procedure tested for the presence of any related proteins with cross-reactivity with the EPSPS specific antibody, but not of the predicted size, and therefore allowed for the detection of unexpected fusion proteins.

The results from the Western blot procedure indicated the presence, in the GA21 transgenic line, of a single protein band of equivalent molecular weight to the expected mEPSPS as expressed *in vitro* in the laboratory. This procedure also

confirmed that the expression level of mEPSPS in the corn grain was at least ten times greater than that of the endogenous corn EPSPS enzyme.

In summary, the data obtained from both the ELISA and Western blot procedures indicate that the mEPSPS is expressed in corn line GA21 at greater than 10 fold the levels of the endogenous EPSPS in the non-transgenic control, and that the protein produced in the transgenic plant is equivalent to the predicted size according to its characterisation *in vitro*. Furthermore, the absence of any other immunoreactive bands of unexpected size is evidence that the transformation has resulted in the expression of only the mEPSPS protein of the predicted size in the modified plants.

Conclusion

One new protein is expressed in corn line GA21, namely the modified corn EPSPS, which carries some specified amino acid changes compared to the wildtype enzyme. Although several methods of analysis showed that the mEPSPS is expressed in the edible grain from the plant at levels approximately ten times higher than endogenous EPSPS expression levels, this is not considered to be a safety issue due to the prevalence of this family of plant and microbial proteins in the human diet.

3.4 Impact on human health from potential transfer of novel genetic material to cells in the human digestive tract

The human health considerations in this regard depend on the nature of the novel genes and must be assessed on a case-by case basis.

In 1991, the World Health Organization (WHO) issued a report of a Joint FAO[4]/WHO Expert Consultation which looked at strategies for assessing the safety of foods produced by biotechnology (WHO, 1991). It was concluded by that consultation that as DNA from all living organisms is structurally similar, the presence of transferred DNA in food products, in itself, poses no health risk to consumers.

The major concern in relation to the transfer of novel genetic material to cells in the human digestive tract is with antibiotic resistance genes. Antibiotic resistance genes can be present in some transgenic plants as a result of their use as marker genes to select transformed cells. It is generally accepted that there are no safety concerns with regard to the presence in the food of antibiotic resistance gene DNA *per se* (WHO, 1993). There have been concerns expressed, however, that there could be horizontal gene transfer of antibiotic resistance genes from ingested food to microorganisms present in the human digestive tract and that this could compromise the therapeutic use of antibiotics.

In relation to the transfer of novel genetic material from genetically modified food to human cells via the digestive tract, this is extremely unlikely to occur. In considering the potential impact on human health, it is important to note that humans have always consumed large amounts of DNA as a normal component of food and there is no evidence that this consumption has had any adverse effect on human health. Furthermore, current scientific knowledge has not revealed any DNA sequences from

[4] Food and Agriculture Organisation

ingested foods that have been incorporated into human DNA. Novel DNA sequences in genetically modified foods comprise only a minute fraction of the total DNA in the food (generally less than 0.01%) and are therefore unlikely to pose any special additional risks compared with the large amount of DNA naturally present in all foods.

The gene cassette in this modification includes a promoter element from another edible plant, the rice actin promoter, which has been shown to give rise to protein expression in corn plants. This eukaryotic promoter is necessary for expression of the new protein, mEPSPS. However, this element is not functional in prokaryotic gut microorganisms, as critical DNA sequences are not recognised by the protein expression machinery of bacteria, including those normally present in mammalian intestines.

Of significance for this application, the method of transformation allowed for a gel purified specific fragment of plasmid pDPG434 to be introduced into the corn cells, a process which effectively excluded the presence of antibiotic resistance marker genes and other bacterial genetic elements present elsewhere in the whole plasmid. Consequently, no extraneous DNA sequences such as marker genes were ever introduced into this plant line.

Conclusion

There is no biological potential for the transfer of novel genetic material from corn line GA21 to intestinal microorganisms, as a result of the genetic modification. The DNA sequences which give rise to protein expression in the plant are not functional in prokaryotes. Furthermore, microorganisms including bacteria and fungi contain an endogenous EPSPS gene and corresponding protein product not unlike the version produced by the transgenic corn. Due to the variety of naturally occurring EPSPS sequences across species in the environment, there is a natural array of organisms exhibiting a potential for glyphosate tolerance.

4. TOXICOLOGICAL ISSUES

4.1 Levels of naturally occurring toxins

Over 72% of the corn kernel is composed of starch, with smaller amounts of protein, oil and other nutritionally valuable substances. There are no known naturally occurring toxins in corn. While mycotoxins can be detected in corn, these are metabolites produced by fungi that grow on corn kernels as a result of production or storage under adverse conditions. They are not a natural component of sound corn.

4.2 Potential toxicity of novel protein

The detailed protein expression analyses have demonstrated that the only new protein present in corn line GA21 is the mEPSPS enzyme. The mEPSPS gene has been completely sequenced and encodes a 47.4 kDa protein consisting of a single polypeptide of 445 amino acids. The modified corn mEPSPS protein shows high amino acid sequence homology to the wildtype corn EPSPS enzyme (99.3%) as well as to other EPSPS enzymes found in common food crops (for example, soybean and

tomato) that have a long history of safe human consumption, or that are present in fungal and microbial food sources such as Baker's yeast (*Saccharomyces cerevisiae*) or *Bacillus subtilis*. Thus, notwithstanding the minor change of several amino acids, this protein is a member of a family of closely related proteins from plants and microbes that are commonly found in human foods.

The amino acid sequence of the mEPSPS protein was compared to that of known protein toxins listed in the PIR, SwissProt, EMBL and GenBank genetic databases. Based on these computer searches, no evidence for any similarity to known protein toxins was found. The specific amino acid changes in the mEPSPS protein, being standard substitutions with common amino acids that comprise all proteins, are unlikely to result in a protein with toxic properties.

As a further test for potential toxicity, the applicant conducted an acute oral toxicity study of the mEPSPS protein in young laboratory mice (approximately 7.5 weeks of age).

4.2.1 Acute oral toxicity study in mice

Monsanto studies:

T.C. Lee *et al*., 1997. Preparation and Confirmation of Doses for an Acute Oral Toxicity Study (ML-97-195) with Modified Maize 5-Enolpyruvylshikimate-3-Phosphate Synthase (mEPSPS) Protein in Albino Mice.

M.W. Naylor, 1997. Acute Oral Toxicity Study with Modified Maize 5-Enolpyruvylshikimate-3-phosphate Synthase (mEPSPS) Protein in Albino Mice.

Supplies of the mEPSPS protein required for the oral toxicity study in animals were produced in *E. coli* in the laboratory. The protein was partially purified and in a separate study, the applicant conducted experiments to formulate and characterise the mEPSPS prior to its use as a test substance in the toxicity study. Confirmation of the mEPSPS protein concentration, integrity and functional activity was experimentally determined by a series of analyses including a total protein assay, densitometry of colloidal blue stained SDS-PAGE gels, Western blot analysis, and enzyme activity assay. From this information, the applicant determined an appropriate formulation and dose range of mEPSPS suitable for use as the test substance.

In the toxicity study, the mEPSPS was administered by a single oral gavage to ten male and ten female CD-1 mice, at target doses of 5, 15 and 50 mg/kg in a constant volume. Using the results of the equivalence study above, this corresponded to actual doses of 3.7, 11.8 and 45.6 mg/kg respectively. Based on the highest anticipated human exposure of 0.06 mg mEPSPS /kg bw/day calculated for children, 1-6 years of age (in the U.S.), the applicant claims that the highest dose of 45.6 mg mEPSPS/kg bw is at least 500-fold higher than the likely human exposure (Holden, 1997).

A control group of ten mice/sex was administered only the carrier substance used above, at the same delivery volume as the test substance. An additional control group of ten mice/sex was administered Bovine Serum Albumin (BSA) in the same carrier substance at the highest target dose (50 mg/kg) and also in the same delivery volume.

At defined stages throughout the duration of the study, clinical observations were performed for mortality and signs of toxicity, and body weights and food consumption measured. At the termination of the study (day 13-14), animals were sacrificed, examined for gross pathology and numerous tissues were collected. However, no organs were weighed and no tissues were examined microscopically.

The results of the study showed no statistically significant differences in group mean body weights, cumulative weight gains or food consumption in either males or females at any level of either the BSA control or test material, when compared with the respective carrier control group. All animals survived to the end of the study, and there were no clinical signs observed that could be related to the test material. A unilateral corneal opacity was noted in one male mouse at the high dose level of the test material, but this finding was not considered to be treatment related.

In conclusion, there was no evidence of toxicity in mice following a single oral dose of 45.6 mg/kg mEPSPS protein.

4.3 Potential allergenicity of novel protein

Monsanto study:
J.D. Astwood, 1997. Modified Maize 5-Enolpyruvylshikimate-3-Phosphate Synthase (mEPSPS) has no significant Sequence Similarity to known Allergens and Toxins.

Allergic reactions to foods are relatively rare and are generally associated with a small group of well-characterised proteins found in common foods such as milk from dairy cows, wheat, soybeans, fish and tree nuts. For the vast majority of the population, consumption of these foods is without adverse effects.

The mEPSPS gene was derived from corn which is not regarded as an allergenic food (Wright, 1987). No known allergens have ever been confirmed to be present in corn. At the protein level, the mEPSPS enzyme exhibits 99.3% amino acid homology with the wildtype corn EPSPS enzyme which is commonly found in food. The mEPSPS protein is present at low levels in the grain, at approximately 0.01% of the total protein in grain from transgenic corn line GA21.

As an indicator of allergenic potential, the amino acid sequence of the introduced mEPSPS protein was compared with amino acid sequences of known allergens available on public protein databases. Based on published scientific information about the common molecular features of known protein allergens, a sequence match of at least eight contiguous identical amino acids is considered to be a significant degree of homology. When the appropriate comparisons were made, no biologically or immunologically significant sequence similarities were observed between the mEPSPS and at least 219 allergen sequences. It is therefore concluded that the mEPSPS gene introduced into corn does not encode a known allergen and that the mEPSPS protein does not share immunologically significant amino acid sequences with known allergens.

4.3.1 *Digestibility of the mEPSPS*

Monsanto study:
R.S. Sidhu *et al*. 1997. Assessment of the Digestability of Modified Maize 5-Enolpyruvylshikimate-3-phosphate Synthase (mEPSPS) Protein *in vitro* Using Mammalian Digestive Fate Models.

The biochemical profile of the mEPSPS enzyme also provides a basis for allergenic assessment when compared to known protein allergens. Protein allergens must be stable to the peptic and acidic conditions of the digestive system if they are to reach and pass through the intestinal mucosa to elicit an allergenic response.

The purpose of this study was to assess the digestibility of mEPSPS protein *in vitro* using simulated gastric fluid (SGF) and simulated intestinal fluid (SIF) as mammalian digestion models. The method of preparation of the simulated mammalian gastric and intestinal digestive solutions used is described in the United States Pharmacopoeia (1989). The exposure of mEPSPS to SGF and SIF was conducted over a series of timed incubations at 37°C. The products of the digestion were analysed using gel electrophoresis and Western blots.

The results of these experiments demonstrated that the mEPSPS protein was no longer detectable after 15 seconds in the gastric system and within one minute in the intestinal system. Moreover, the results of these simulated digestion experiments showed no evidence for the presence of stable peptide fragments larger than 2 kDa. These results provide evidence that the mEPSPS protein is readily digested in the mammalian digestive tract.

Conclusion

The family of EPSPS enzymes from various plant and microbial food sources are currently part of the human diet and have been consumed over a long period without any health concerns. The data and analyses on the potential for allergenicity of the mEPSPS protein support the conclusions that the protein is not derived from an allergenic food source, does not exhibit sequence similarity with known allergens, and does not exhibit the biochemical characteristics of known protein allergens. Furthermore, the protein is present in relatively low abundance in the grain and demonstrates digestive lability in conditions that mimic human digestion. These results strongly support the conclusion that corn line GA21 expressing mEPSPS does not pose any greater risk than conventional corn, with respect to potential allergenicity.

5. NUTRITIONAL ISSUES

5.1 Compositional analysis

Monsanto studies submitted:

K.M. Magin, B.E. Ledesma, G.J. Rogan, P.R. Sanders and R.S. Sidhu, 1998. Expression and Compositional Analyses of the Roundup Ready® Corn Line GA21 Produced in 1996 Field Trials. Performing Laboratories: Monsanto Life Sciences Company and Covance Laboratories Inc. Report No. MSL-15196, Study 96-01-46-01, CHW Project 6103-199

The safety assessment of foods produced using gene technology entails, in this case, evaluating compositional data from the transgenic corn plant in comparison with equivalent data from the parental (or untransformed) plant line or literature values for the particular crop species. This process involves identifying the key components, including nutrients and any toxicants, characteristic of corn grain and also takes into account the variation in composition due to genetic variability, environmental factors, and post-harvest handling and processing.

Transformed corn line GA21 and its non-transgenic control were grown in 1996 at five field locations in the United States, predominantly in the major corn growing belt. The control plants were the population of non-transgenic negative segregants (that is, plants lacking the mEPSPS gene addition) present in untreated plots of transgenic GA21 corn. The non-transgenic negative segregants were identified using the polymerase chain reaction (PCR) technique. The plants were individually self-pollinated, and the harvested forage and grain analysed in order to determine the compositional profile of the transgenic corn in comparison with that of conventional corn varieties grown commercially.

5.1.1 *Sample collection and preparation*

At the outset of the compositional studies, forage was defined as the entire plant minus the roots collected at the soft dough stage. Four plants were collected at the glyphosate treated plots to provide the treated GA21 samples. Two positive and two negative segregant plants were collected from the untreated plots to provide the untreated GA21 and control samples respectively. Non-transgenic segregants were used as the negative control because these are the same genetic line (isogenic) as the transformed line GA21.

At normal commercial harvest (approximately 25% grain moisture), all self-pollinated ears from up to 12 plants were collected from glyphosate treated plots to provide treated GA21 grain samples. Similarly, all ears from up to 12 self-pollinated plants tagged as positive and negative segregants were collected from untreated plots to provide untreated GA21 and control grain samples, respectively.

5.1.2 *Analyses*

The components measured included proximates (protein, fat, ash, carbohydrates, moisture, acid detergent fibre and neutral detergent fibre), amino acid composition, fatty acids profile, calcium and phosphorus in the grain, and proximates, calcium and phosphorus in the forage. References were provided for all methods used in the analyses. The majority of methods were derived from standard methodology in *Official Methods of Analysis*, Association of Official Analytical Chemists and are validated AOAC International Methods, or American Oil Chemists Society (AOCS) Methods.

5.1.3 *Proximate, calcium and phosphorus analyses*

The corn kernal has been extensively studied for its composition, nutritional properties and feeding value, both as an important stock feed and as human food. It contains approximately 72% starch on a dry basis, is low in fibre, and relatively low

in protein content (approximately 4%-20%). Like other cereal grains, corn is very low in calcium, and low in other minerals including phosphorus, potassium and magnesium. The oil in corn is highly polyunsaturated and rich in linoleic acid (2.9% of the whole corn on a dry basis).

The values for the proximate, calcium and phosphorus analyses of the corn grain are represented in the following summary Table of Results and are expressed as percent dry weight of the sample correcting for the measured moisture content. The mean value of each compositional parameter was calculated from measurements for each sample from each of the five test sites.

Table 1

Component	Control (Untreated) Mean (%)	Untreated Line GA21 Mean (%)	Treated Line GA21 Mean (%)	Literature (Range %)
Protein	10.1	10.0	9.9	6.0-12.0
Total fat	3.6	3.5	3.5	3.1-5.7
Ash	1.3	1.3	1.3	1.1-3.9
ADF	3.7	3.7	3.9	3.3-4.3
NDF	11.7	10.8	11.4	8.3-11.9
Carbohydrates	85.1	85.2	85.2	
Calcium	0.003	0.003	0.003	0.001-0.01
Phosphorus	0.3	0.3	0.29	0.26-0.75
Moisture	14.4	14.2	14.6	7-23

The results of the proximate analyses demonstrate that there are no statistically significant differences in these components between grain from control and untreated or treated glyphosate-tolerant corn line GA21. In addition, the values obtained in the experiments were either within the range in published literature (Watson, 1987; Jugenheimer, 1976) or within previously reported ranges for non-transgenic corn varieties.

5.1.4 *Amino acid composition of corn line GA21*

Corn protein content, and its amino acid ratios, may vary widely due to genetic manipulation by traditional plant breeders and to a lesser degree by crop year, soil fertility, crop management (especially nitrogen fertilisation), and climatic conditions (Wright, 1987).

A modified version of an AOAC International method was used to determine the amino acid composition of the grain from corn line GA21. Eighteen individual amino acids were quantitated using an automated amino acid analyser. Both treated and untreated GA21 grain was compared with the control (PCR identified negative segregant) and with the literature values for commercially grown corn. The results of these analyses appear in the summary table below. The values are expressed as a percent of total amino acids and represent the least squares mean of five samples, one from each test site. The literature range is taken from Watson (1982) and represents values as a percent of total protein (10.1%). The amino acid components indicated

with an asterisk (*) show a statistically significant difference in the mean value when compared to the control.

Table 2

Amino Acid	**Control (Untreated) Mean (%)**	**Untreated Line GA21 Mean (%)**	**Treated *Line GA21* Mean (%)**	**Literature Range (%)⊗**	**Reported Range (%)δ**
Methionine	2.0	2.0	2.0	1.0-2.1	1.3-2.6
Cysteine	2.1	2.1	2.1	1.2-1.6	1.8-2.7
Lysine	3.1	3.0	2.8 *****	2.0-3.8	2.6-3.5
Tryptophan	0.6	0.6	0.6	0.5-1.2	0.4-1.0
Threonine	3.7	3.8	3.8	2.9-3.9	3.3-4.2
Isoleucine	3.6	3.6	3.5 *****	2.6-4.0	3.2-4.3
Histidine	2.8	2.8	2.8	2.0-2.8	2.8-3.3
Valine	4.6	4.6	4.5 *****	2.1-5.2	4.2-3.5
Leucine	12.9	13.1	13.2	7.8-15.2	12.6-15.8
Arginine	4.3	4.1	4.0 *****	2.9-5.9	3.6-5.0
Phenylalanine	5.2	5.1	5.1	2.9-5.7	5.0-6.1
Glycine	3.8	3.7	3.7	2.6-4.7	3.2-4.2
Alanine	7.6	7.6	7.7	6.4-9.9	7.3-8.8
Aspartic Acid	6.7	6.7	6.6	5.8-7.2	6.3-7.5
Glutamic Acid	19.1	19.3	19.4	12.4-19.6	19.5-22.8
Proline	8.7	8.7	8.8	6.6-10.3	8.7-10.1
Serine	5.3	5.3 *****	5.4 *****	4.2-5.5	4.9-6.0
Tyrosine	3.9	3.8 *****	4.0	2.9-4.7	3.7-4.3

⊗Values are percent of total protein (10.1% total protein).
δ Data from five nontransgenic corn lines evaluated in 1993-5, Monsanto field trials.

Statistically significant differences between control and untreated or treated GA21 grain were not observed for a majority of the amino acids tested. The results showed that the mean differences from control in grain for untreated GA21 was significantly different in two amino acids. Serine was increased by 1.2% and tyrosine was decreased by 3.4%.

The mean differences for treated GA21 grain compared with the control were statistically significant for five amino acids. The mean values of four amino acids were lower than the control (lysine –8.6%, arginine –8.2%, valine –3.6% and isoleucine –2.6%). One amino acid, serine, was 2.4% greater in the GA21 line than in the control line. However, inspection of the raw data indicates that these differences are not biologically relevant as all values were either within the ranges published in the literature (Watson, 1982) or within previously reported ranges for non-transgenic corn varieties.

5.1.5 *Fatty acid composition of corn line GA21*

Treated and untreated GA21 grain was compared with respect to fatty acid composition to non-transgenic control samples. Nine different fatty acid types were analysed and the results indicated no statistically significant differences between the values recorded for control and either treated or untreated GA21 grain for any fatty

acid component, with one exception. A minor difference was observed in stearic acid content between the control (mean of 1.9% of total fatty acid) and the treated GA21 line (mean of 1.8% of total fatty acid). The results of the fatty acid composition analysis are presented in the following Table of Results:

Table 3

Fatty Acid Mean (%)	Control Mean (%)	Untreated Line GA21 Mean (%)	Treated Line GA21 Mean (%)	Literature Range* (%)	Reported Range** (%)
Arachidic (20:0)	0.4	0.4	0.4	0.1-2	0.3-0.5
Behenic (22:0)	0.2	0.2	0.2		0.1-0.3
Eicosenoic (20:1)	0.3	0.3	0.3		0.2-0.3
Linoleic (18:2)	58.7	58.6	59.1	35-70	55.9-66.1
Oleic (18:1)	27.4	27.5	27.1	20-46	20.6-27.5
Palmitic (16:0)	9.9	9.9	9.9	7-19	9.9-12.0
Palmitoleic (16:1)	0.2	0.2	0.2		
Stearic (18:0)	1.9	1.9	1.8	1-3	1.4-2.2
Linolenic (18:3)	1.1	1.1	1.1	0.8-2	0.8-1.1

* Watson, 1982
** Data from five nontransgenic corn lines evaluated in 1993-5, Monsanto field trials.

Corn oil is an excellent source of polyunsaturated fatty acids, with a high level of the essential fatty acid linoleic acid (18:2). In addition, it has naturally low levels of the saturated fatty acids, palmitic acid (16:0, 11%) and stearic acid (18:0, 2%). It is known also that corn oil from cooler regions has a higher proportion of unsaturated fatty acids than corn oil from warmer areas, which appears to be an adaptation to climatic conditions. However, genotype has a greater influence on fatty acid composition than any environmental factor. The biochemical variability for fatty acid composition among corn genotypes is known to cover a broad range. Examination of the raw data therefore indicates that the minor observed difference in stearic acid levels in the transgenic corn is neither biologically relevant, since the value was within the ranges published in the literature (Watson, 1982), nor is it an issue for food safety because of broad natural variation.

5.1.6 *Conclusions from compositional analyses*

Comprehensive data from a range of compositional analyses conducted on grain from both untreated and treated corn line GA21 and the non-transgenic control were presented for assessment. The compositional components measured included proximates (protein, fat, ash, carbohydrates, moisture, acid detergent fibre and neutral detergent fibre), amino acid composition, fatty acids profile, calcium and phosphorus.

In addition, data were provided on proximates, calcium and phosphorus in the forage, but as this portion of the plant is not for human consumption, the data were not considered in the assessment process.

The results of the compositional data do not indicate that there are any biologically significant differences between glyphosate-tolerant corn line GA21, either untreated or following treatment with glyphosate, and the non-transgenic control in any of the parameters measured. Some minor statistically significant differences were observed in the amino acid composition of the treated GA21 grain in comparison with the control. The differences were observed for arginine, isoleucine, lysine, valine and serine, but were not considered to be of either biological relevance for commercially grown corn varieties nor of significance in terms of food safety.

Similarly, a minor statistical difference observed in the fatty acid profile specifically in the level of stearic acid in the treated GA21 line compared to the control was not considered to be significant as the value was within the known reported range for commercial corn varieties and is not of concern in terms of food safety.

5.2 Levels of anti-nutrients

Corn contains insignificant levels of anti-nutrient compounds. The levels of trypsin inhibitor in particular are known to be very low (Melville *et al*., 1972; Halim *et al*., 1973) and lectins, carbohydrate binding proteins with haemagglutination activity, have been found at low levels in the endosperm and germ (Newberg and Concon, 1985).

The content of trypsin and chymotrypsin inhibitors is traditionally determined by enzymatic methods, but these methods are very dependent on the concentration of protein, non-protein inhibitors and other factors.

5.3 Ability to support typical growth and well being

In assessing the safety of a genetically modified food, a key factor is the need to establish that the food is nutritionally adequate and will support typical growth and well-being. In most cases, this can be achieved through an understanding of the genetic modification and its consequences together with an extensive compositional analysis of the food. Where, on the basis of available data, there is still concern or doubt in this regard, carefully designed feeding studies in animals may provide further re-assurance that the food is nutritionally adequate. Such studies may be considered necessary where the compositional analysis indicates significant differences in a number of important components or nutrients or where there is concern that the bioavailability of key nutrients may be compromised by the nature of the genetic changes to the food.

In the case of corn line GA21, animal feeding studies using the whole food have not been conducted. The nutritional profile of corn line GA21 was determined by compositional analyses of the major components of the kernel and these were found to be comparable to the conventional control lines. Further studies on the specific protein sequence found that no significant similarity to known allergens or toxins exists. An acute oral toxicity study in mice using variable amounts of mEPSPS as a

single dose, found no evidence to indicate that the protein produces toxic effects in animals. In addition, the mEPSPS protein has been shown to be rapidly degraded in model digestive systems. Finally, the level of dietary exposure to the new protein has been estimated to be very low. The major human food uses for corn are the extensively processed starch and oil fractions yielding high fructose corn syrup and corn oil, neither containing protein. Human exposure to the new protein from whole grain corn in the diet is also considered to be very low due both to its low abundance in the protein fraction of the grain and to the proportionately low percentage of protein in the kernal, compared with the major starch component.

In view of the safety data available for this food and the technical features of the genetic modification, it is considered that, for this application, additional studies on wholesomeness were not essential to the safety assessment process. The donor of the mEPSPS gene and the recipient organism are the same plant species, namely corn (*Zea mays* L.). The modified gene was generated *in vitro*, has been entirely sequenced, and the properties of the encoded protein have been biochemically characterised.

In summary, the data and information available on the genetic change, together with the composition data, provide a sound basis for consideration of the safety of this food without requiring feeding studies in laboratory animals.

6. OTHER MATTERS

6.1 Dietary Exposure

The applicant has provided an estimate of the human dietary exposure to mEPSPS protein. The analysis provides a worst-case estimate of exposure to mEPSPS and assumes no breakdown of the protein during preparation of various corn fractions by wet or dry milling procedures.

Ingestion of products derived from corn line GA21 was estimated to provide approximately 0.02 mg/kg bw/day of mEPSPS for general consumers in the US, where corn products are normally a high dietary component. A concentration of mEPSPS of 0.001% (fresh weight basis) was used in this analysis which was approximately 25% higher than the highest value found in corn grain from 1996 US field trials conducted at five separate locations. Levels of the wildtype corn EPSPS protein in the non-transgenic control line were below the detection limit (0.00015%) of the ELISA method used for analysis. These results demonstrate that, even with a relatively high pattern of usage of corn products, human dietary exposure to mEPSPS is expected to be very low.

Acknowledgements

ANZFA gratefully acknowledges Professor Richard Head of CSIRO Health Sciences and Nutrition, Adelaide, and Professor Geoff Fincher of the Department of Plant Science at the University of Adelaide, for their expert comments on this safety assessment report.

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ATTACHMENT 3

REGULATORY IMPACT ASSESSMENT

Regulatory Impact Assessment

The Authority is required, in the course of developing regulations suitable for adoption in Australia and New Zealand, to consider the impact of various options (including non-regulatory options) on all sectors of the community, including consumers, the food industry and governments in both countries. The regulatory impact assessment will identify and evaluate, though not be limited to, the costs and benefits of the regulation, and its health, economic and social impacts.

Identification of affected parties

1. Governments in Australia and New Zealand
2. Consumers in Australia and New Zealand
3. Manufacturers, producers and importers of food products

Options

Option 1–To prohibit the sale of food produced using gene technology

GOVERNMENT Commonwealth, New Zealand Health Departments, State/Territory Health Departments	**Benefits** • no benefits were identified.	**Costs** • the governments of Australia and New Zealand may be challenged under the WTO to justify the need for more stringent restrictions than apply internationally. • a prohibition on food produced using gene technology in Australia and New Zealand could result in retaliatory trade measures from other countries. • there may be technical problems for AQIS in enforcing such a prohibition at the import barrier.
INDUSTRY Manufacturers, producers and importers of food products	**Benefits** • Some companies may benefit from being able to exploit niche markets for non-GM products overseas.	**Costs** • food manufacturers and producers will be unable to use the processed food fractions from foods produced using gene technology thus requiring the switch to non-GM ingredients and the reformulation of many processed food products. The cost to manufacturers of going non-GM has been estimated to be $A 207m in Australia and $NZ 37m in New Zealand[5]. This is equivalent to 0.51% of turnover in Australia and 0.19% in New Zealand.

[5] Report on the cost of labelling genetically modified foods (2000)

CONSUMERS	**Benefits** • no benefits were identified, however as some consumers perceive GM food to be unsafe, they may perceive prohibition of GM food to provide a public health and safety benefit.	**Costs** • could lead to decreased availability of certain food products. • increased costs to consumers because manufacturers and producers may have to source non-GM ingredients.

Option 2– to permit the sale of food produced using gene technology

GOVERNMENT Commonwealth, New Zealand Health Departments, State/Territory Health Departments	**Benefits** • increased innovation and competitiveness in the food industry will benefit the economy.	**Costs** • minor costs associated with amending the *Food Standards Code.*
INDUSTRY Manufacturers, producers and importers of food products	**Benefits** • food producers and manufacturers will be able to capitalise on the latest technology. • food importers will continue to be able to import manufactured products from overseas markets including the USA and Canada where there is no restriction on the use of food produced using gene technology.	**Costs** • there may be some discrimination against Australian and New Zealand food products in overseas markets that have a preference for non-GM foods (e.g., Japan and the European Union).
CONSUMERS	**Benefits** • consumers may have access to a greater range of food products.	**Costs** • those consumers who wish to avoid GM food may experience restricted choice in food products. • those consumers who wish to avoid GM food may have to pay more for non-GM food.

Conclusion of the regulatory impact assessment

Consideration of the regulatory impact for foods produced using gene technology concludes that the benefits of permitting foods produced using gene technology primarily accrue to the government and the food industry, with potentially a small benefit to consumers. These benefits are considered to outweigh the costs to government, consumers and industry, provided the safety assessment does not identify any public health and safety concerns.

ATTACHMENT 4

WORLD TRADE ORGANISATION AGREEMENTS

With the completion of the Uruguay Round of trade negotiations, the World Trade Organisation (WTO) was created on 1 January 1995 to provide a forum for facilating international trade.

The WTO does not engage in any standard-setting activities but is concerned with ensuring that standards and procedures for assessment of and conformity with standards do not create unnecessary obstacles to international trade.

Two agreements which comprise part of the WTO treaty are particularly important for trade in food. They are the:

- Agreement on the Application of Sanitary and Phytosanitary Measures (SPS); and
- Agreement on Technical Barriers to Trade (TBT).

These agreement strongly encourage the use, where appropriate, of international standards, guidelines and recommendations, such as those established by Codex (in relation to composition, labelling, food additives, veterinary drug and pesticide residues, contaminants, methods of analysis and sampling) and the code and guidelines on hygienic practice.

Both Australia and New Zealand are members of the World Trade Organization (WTO) and signatories to the agreements on the Application of Sanitary and Phytosanitary Measures (SPS agreement) and on Technical Barriers to Trade (TBT agreement). Within Australia, a memorandum of understanding binding all States and Territories to the agreements has been put in place by the Council of Australian Governments (COAG).

The WTO agreements are predicated on a set of underlying principles that standards and other regulatory measures should be:

- based on sound scientific principles;
- developed using consistent risk assessment practices;
- transparent;
- no more trade-restrictive than necessary to achieve a legitimate objective;
- recognise the equivalence of similar measures in other countries; and
- not used as arbitrary barriers to trade.

As members of the WTO both Australia and New Zealand have an obligation to notify the WTO of changes to food standards to enable other member countries of the WTO to make comment. Notification is required in the case of any new or changed

standards which may have a significant trade effect and which depart from the relevant international standard (or where no international standard exists). Matters raised in this proposal may be notified to the WTO as either SPS notifications or TBT notifications, or both.

SPS Notifications

These are primarily health related, and refer to any measure applied:

- to protect animal or plant life from risks arising from the entry, establishment or spread of pests, diseases or disease carrying organisms;

- to protect human or animal life or health from risks arising from additives, contaminants, toxins or disease-carrying organisms in foods, beverages or foodstuffs;

- to protect human life or health from risks arising from diseases carried by animals, plants or products thereof, or from the entry, establishment or spread of pests; and

- to prevent or limit other damage from the entry, establishment or spread of pests.

The Agreement on the Application of Sanitary or Phytosanitary Measures relates to any measure applied to protect animal, plant or human life or health which may directly or indirectly affect international trade. Whether the SPS measure is in the form of a law or mandatory regulation, an advisory guideline, a code of practice or a requirement, it is the purpose of the measure that is important - not its regulatory status. Each WTO member country is entitled to apply SPS measures that are more stringent than the international standards in order to protect the health of its population. In the interests of transparency, each instance of such non-alignment which could result in an impediment to trade must be identified and justified and the documentation of that justification must be readily available

Each member country is also required to apply its methods of risk assessment and management consistently so arrangements under the SPS Agreement do not generate what may really be technical barriers to trade

Under the SPS Agreement, an exporting country can have resort to the WTO's dispute settlement procedures with respect to such a non-alignment. These arrangements mean there is potential for a code of practice to introduce an SPS measure that may bring about non-alignment with international requirements. Such non-alignment would need to be justified scientifically on the grounds that it is necessary to protect human, animal or plant life or health.

TBT Notifications

A technical barrier to trade arises when a mandatory requirement in a country's food regulatory system does not align with the international standard and it is more trade restrictive than is necessary to fulfil a legitimate objective. However, it can be acceptable for a country to have a more stringent requirement than that set internationally for reasons including:

- Maintaining national security;
- Preventing deceptive practices; and
- Protecting human health or safety.

Instances of non-alignment with international standards which could result in trade barriers must be identified and, if questioned, justified. Voluntary codes of practice are not expected to generate technical barriers to trade except where compliance with a code of practice or some aspect of a code of practice is expected. Consequently, it is possible for a voluntary code of practice to be viewed by the WTO as mandatory and subject to all the notification and other provisions applying to mandatory regulations.

The Agreement on Technical Barrier to Trade relates to requirements covering product characteristics or their related processes and production methods. TBT covers measures that are not SPS, such as requirements relating to terminology, symbols, packaging, marking, labelling, food composition and processing methods.

It is considered that these Full Assessments do constitute a potential Technical Barrier to Trade or a Sanitary/Phytosanitary matter. Matters raised in these Full Assessments therefore will be notified to the WTO.

ATTACHMENT 5

SUMMARY OF FIRST ROUND PUBLIC COMMENTS

A362 – FOOD DERIVED FROM GLYPHOSATE-TOLERANT CORN

The Authority received the first six applications for foods produced using gene technology from Monsanto Australia Ltd. Due to commonalities in these applications, a combined Notice of Application (formally referred to as the Preliminary Assessment Report) was advertised on 28 October 1998, which called for public comment on the applications. A total of 58 submissions were received in response to the combined Notice of Application, of which 53 relate to this application.

Jean Adams (Aust)

- does not want these experimental foods in the common food supply until they have been long–term tested for undesirable side–effects related to public health or to environmental damage;
- questions the legality of forcing such genetically modified foods onto the public and the intention to remove labelling of such foods.

Robert Anderson (member of Physicians and Scientists for Responsible Application of Science and Technology)

- knowledge about the nature of the promoter, genes and the type of antibiotic resistance genes is crucial to a proper assessment;
- the applications should be rejected because most of the New Zealand population does not want to eat genetically engineered food. There are real dangers of allergic reactions, the Maori people are opposed to genetic engineering and these products are all an unknown risk to human health because they have not been tested.

Aoraki Greens and the Organic Garden City Trust (NZ)

- opposed to the amendment to the *Food Standards Code* to permit the foods in the applications;
- claim there is no alternative but to decline the acceptance of these products until they are clearly labelled and can be differentiated from their conventional counterparts;
- believe consumer choice is being violated;
- consider that because the science is new, potential problems or long term implications are yet to be made apparent.

Elaine Attwood (Aust)

- supports Option 1 in the combined Preliminary Assessment - that is, to maintain the status quo and not approve any of the six applications;
- re: A338 - considers 4 weeks of laboratory animal testing inadequate and doubts the applicant's claim that the need for herbicide will be reduced. Comments on proposed increase in the MRL for glyphosate;
- re: A355, A362 and A346–genetically modified material will enter the food chain via cotton seed meal and hulls and corn waste being fed to animals;
- re: A363 – canola free of genetic modification would be marketable overseas;
- re: A341 – the results of laboratory feeding studies in rats are of concern. Long

term safety is uncertain and therefore the genetically modified cotton should not be permitted;
- trade considerations should not prevail over consumer rights to have all genetically modified foods labelled as such.

Australian GeneEthics Network
- Monsanto's proposals should all be rejected as inadequate;
- there should be pre–market human testing to provide data for a precautionary approach on safety and nutritional efficacy;
- there should be full labelling of all approved foods in keeping with the Ministerial decision;
- there should be public review of the MRLs for Roundup in these foods;
- there should be public review of the toxicity of the quantities of Bt toxins likely to enter the human and animal food supplies, taking cultural, social, ethnic and age diversity into account;
- an adverse reactions register should be established to enable systematic monitoring of any impacts of these foods;
- all proposals should be submitted for GMAC assessment and recommendation including an updated and public review of Bt cotton and Roundup Ready soy for environmental and health impacts;
- GMAC's assumption that AQIS regulations would keep imported soy out of the Australian environment does not apply to the other commodities, and the geographical limits and performance of Bt cotton need public review;
- Monsanto has not studied the dietary implications of these products and presents no evidence that the company considered the diversity of diets among different cultures, social or ethnic groups;
- RR soy and corn crops are very different in containing novel DNA, proteins at elevated levels, and new levels of synthetic chemical residue not in food before;
- RR canola and cotton seed oils are so extensively processed before human consumption that no DNA or proteins will remain. This ignores, for example, the use of whole seeds for sprouting, the inclusion of whole seeds in uncooked foods, and the cold pressing of oils;
- Bt cotton and corn are substantially equivalent to parental lines in composition, safety and wholesomeness, yet Bt has never been in the food supply in such quantities before;
- the toxicological studies of RR foods are brief and insufficient as no chemical residue studies are cited, proteins created by inserted genes have only been checked against known protein toxins and allergens, no human, and very few animal testing of the products has been done, whole genetically engineered soybean, corn, canola or cotton were not checked in simulated gastric and intestinal fluids;
- no toxicological studies were carried out on the Bt crops as Monsanto asserts that "regulatory agencies world-wide have determined that the use of registered B.t.k products pose no significant risks to human health, non-target organisms or the environment." Believes this is grossly misleading as it refers to the topical use of a whole organism which quickly disappears from the environment following spraying, whereas Bt crops express large amounts of toxin throughout their systems.

Berylla (NZ)

- these foods will be in 60–80% of all processed foods therefore freedom to choose will be compromised;
- as these foods will also be fed to animals, choices will be restricted even further and if the animals were eaten then the degree of risk will increase;
- support the submissions of the Natural Law Party and Clive Elwell.

Willi Borst (NZ)
- wants all genetically modified foods to be labelled and if not they should all be banned;
- concerned about antibiotic resistance, viral recombination and environmental pollution;
- all genetically modified food should be deemed unsafe until proven otherwise;
- submits that ANZFA not amend the *Food Standards Code* to permit foods derived from genetically modified crops.

Jim Chapple (NZ)
- strongly opposed to all six applications on the grounds that approval of these foods may create a market monopoly for the applicant in the supply of agrochemicals and that gene technology is potentially unsafe;
- very strongly objects to the term "substantially equivalent" as a play on words;
- genetically modified foods are not identical to their conventional counterpart and therefore all such products must carry labelling.

Commerce Commission (NZ)
- no issues raised by the applications on which the Commission has any comments.

Consumers' Association of South Australia Inc. (Aust)
- supports comments made by Elaine Attwood.

Clive Elwell (NZ)
- the applications should be rejected because Maori people find genetic engineering in conflict with their beliefs and values, the overwhelming majority of people in Australia and New Zealand do not want to eat genetically modified food, there is a danger of allergic reactions, and genetically modified food is insufficiently tested and so cannot be regarded as safe for human consumption;
- the foods cannot be sufficiently tested because it is impossible to carry out appropriate tests; the tests that are carried out are limited and inappropriate.

Consumers' Federation of Australia Inc.
- not supportive of these applications being approved at this stage;
- questions the safety of soya milk as infant food because of the presence of trypsin inhibitor and other anti–nutrients after heat processing, and also the presence of isoflavones;
- refers to a reference (no details supplied) which has shown that the isoflavone levels may differ from the levels in conventional soybeans;
- application A338 does not provide sufficient evidence of anti-nutrients to prove that the soybeans are safe for processing into infant formula. In light of this, interprets ANZFA's safety assessment guidelines as requiring a full toxicological and nutritional assessment of the soybeans. Believes these

concerns are serious enough to warrant a recall of foods containing Roundup Ready soy ingredients;

- no evidence is presented by the applicant on glyphosate residues in A338, A362, and A363, despite a specific requirement to do so in ANZFA's safety assessment guidelines;
- does not accept the assertion by the applicant that there is only one novel protein in the Roundup Ready soybeans;
- does not believe that testing for homology of protein structure is a sufficient test for allergenicity. At the very least these foods should be fed to human volunteers in closely monitored trials before they are released generally;
- traces of the introduced proteins could be present in cold–pressed oils at levels sufficient to precipitate allergic reactions, if there is an allergic potential. At the very least, such oils should carry precautionary labels warning of the possibility of allergic reactions;
- the approval of Roundup Ready maize will facilitate even greater use of high fructose corn syrups in Australian processed foods. The end result of this could well be that consumption of high energy products by Australians will rise and that the current excessive levels of nutritional diseases such as obesity, diabetes and heart disease will increase further;
- ANZFA needs to be satisfied that anti–nutrient levels in canola are safe and that they will not rise over time;
- expresses concern about the decreased weight gain by laboratory rats in the first week of a 4 week feeding trial with INGARD cotton seed. Believes that further feeding trials on a range of animals should be performed before this product is released;
- approval of foods produced using gene technology should be deferred until a national coordinating system for regulatory approvals is in place so that a global assessment of their likely impacts can be made;
- a system for monitoring adverse reactions to these foods should be established before they are released into the diet of Australians;
- approval and release of these foods should not occur until the system of labelling agreed to by Health Ministers is established;
- Australia should not be bullied by other countries to accept their exports of unsegregated mixtures of genetically modified and non–modified foods.

Francela Davies (NZ)

- concerned about the addition of food additives in the form of genetically engineered foods that have not been given adequate testing of their benefits or side effects to human health;
- wants ANZFA to address the long term effects of the consumption of foreign proteins, antibiotic resistant marker genes and viruses;
- the applications should be rejected because there is no evidence that these foods are contributing anything positive to the food supply or the environment.

Food Technology Association (FTA) Victoria Inc.

- the risk assessment must be completed and reported to ANZFA stakeholders prior to any decision on the Applications;
- it is unclear from Standard A18 as to the labelling that would apply to these products;
- wants to know what special conditions might apply to these products;

- the option to not permit the sale of these foods is the preferred option;
- the application needs more detail and background information such as a Full Assessment report, details on special conditions and labelling and a complete risk assessment.

Friends of the Earth (NZ)

- share the same concerns as expressed in the submission of the Natural Law Party and Clive Elwell;
- glyphosate has not been included among the residues tested, and there is no awareness of any program that monitors for glyphosate residues in food;
- Treaty of Waitangi obligations have not been considered in ANZFA processes;
- the New Zealand Bill of Rights provides that no New Zealand may be subjected to experimentation without providing informed consent, therefore full disclosure of all transgenic foods and ingredients via labelling is the only way this can begin to be achieved;
- Monsanto has not done any long term studies on health effects;
- submit that ANZFA should approve these foods for a period of 6 months only conditional on a requirement for immediate, prominent labelling of all food products and a warning logo. This should be followed by a moratorium on any further approval of genetically engineered foods.

Noeline Gannaway (NZ)

- supports labelling of all food containing genetically engineered products;
- there may be risks of toxic or allergic reactions;
- oppose the transfer of genetic material between different species as unethical and potentially unsafe.

Goodman Fielder (Aust)

- is fully supportive of developments in the agri–food industry through the application of gene technologies provided that consumer benefits are clearly defined and communicated;
- urges ANZFA to undertake wide consultation with all affected parties, including growers, crushers (in the case of oilseeds), food industry users and consumers before these modified plants are introduced.

Nathan Green (NZ)

- objects vehemently to the further introduction of genetically modified foods, specifically the applications by Monsanto;
- there have not been sufficient tests to prove safety;
- NZ should exploit the GMO free market opportunities;
- there has not been adequate public debate on the introduction of genetically modified foods;
- does not agree with the concept and use of substantial equivalence.

Mike and Jeanne Gregory (NZ)

- the public has not been properly consulted or informed by Government or ANZFA on the introduction of genetically modified foods;
- strongly opposed to genetically modified foods on grounds that these are not adequately tested;

- there is significant and growing scientific concern worldwide about the technology and the processes undertaken to evaluate the safety of genetically modified foods;
- NZ would have a market advantage if genetically engineered foods were prohibited altogether.

Martin Hartman and Cornelia Baumgartner (NZ)
- object to genetically modified foods;
- call for mandatory labelling of all genetically modified foods.

Karen Hunt (NZ)
- demands that all genetically modified foods be labelled;
- states that consumer rights are violated if products are deemed substantially equivalent and consequently are not subject to mandatory labelling.

InforMed Systems Ltd (NZ)
- the transfer of EPSPS genes to soybean, maize, cotton and canola is acceptable;
- the transfer of the gox gene to canola and the use of the cry1Ac gene are also acceptable;
- noted that no mention was made of any maker genes in the applications for soybeans, corn or canola;
- noted that the nptII gene is used in cotton and one insect resistant corn variety. Considers that there are remaining questions with regard to the use of antibiotic resistance genes. It would be reassuring if independent biomedical advice were available to reassure us that this does not pose a risk to the future use of these or related antibiotics in the management of human disease;
- notes that none of the modified plants provides any nutritional or functional benefit for the consumer. It is unfortunate that the first applications should not demonstrate benefits to the consumer, who may thus feel that failure to permit the use of such foods will have no measurable effect on them.

Oraina Jones (NZ)
- genetically engineered foods have not been adequately tested for their benefits or side effects to human health;
- what are the long term effects of the consumption of foreign proteins, antibiotic resistant marker genes and viruses;
- questions whether Monsanto supplied any evidence of long term trials;
- requests that the application be declined as the foods are not contributing in any way to the food supply or environment.

Michael Karas (Aust)
- is opposed to applications A338, A355, A362 and A363 because they are for herbicide resistant crops;
- is concerned about the potential for Roundup residues to be increased in human food supply;
- is concerned about the out–crossing of herbicide resistant crops to create ‘super–weeds’.

Colin Kell (NZ)
- criticises some of the wording used in the preliminary assessment report;

- claims that genetically altering food decreases the nutritional value;
- the application provides no proof that glyphosate does not cause long term effects;
- there has been insufficient testing of these genetically modified foods;
- balanced information on genetic modification needs to be made available and the rights of everyone taken into consideration;
- imported commodities should be segregated at source;
- the applications do not indicate the source of the genes being used - believes that genes from fish and animals are being used which is unethical and against nature.

Janine Kelly (NZ)
- concerned about the depth of investigation into the safety of genetically modified foods and the apparent lack of concern by regulatory authorities for the opinions of informed members of the general public and some scientists;
- ANZFA puts too much faith in the integrity of companies who are producing genetically modified foods;
- urges ANZFA to consider the long–term implications of allowing the sale of genetically modified foods;
- if they are allowed, they should all be labelled.

Kristen Khaine (NZ)
- consumer rights include the choice not to eat any genetically modified foods, therefore labelling is of paramount importance;
- trade barrier issues are secondary to public health and safety.

Hilde and Kristin Knorr (Aust)
- advocate a prohibition on genetically modified foods altogether, but otherwise strongly demand mandatory labelling.

Susie Lees (NZ)
- not enough information has been provided in these applications;
- the public do not want to eat these products;
- if the products are approved, we will be at risk of unknown toxins and allergens.

Margaret and Leonard Krohn (Aust)
- opposed to genetically modified foods on the grounds that insufficient scientific testing has been done and the effects on public health are unknown.

C. Lamprecht (Aust)
- concerned about the possible detrimental health effects of genetically modified foods;
- concerned about increased pesticide residues in food;
- advocates full mandatory labelling of all genetically modified foods.

Hannah Levy (Aust)
- strongly opposed to genetically modified foods because of the limited knowledge concerning the risks associated with the technology;
- demands full labelling.

Mahikari Australia

- strongly advocates the mandatory labelling of all foods or food ingredients produced using gene technology to allow consumer choice;
- disagrees with validity of "substantial equivalence" as a basis for labelling because of a lack of scientific rigor;
- completely opposed to all six applications because of the potential long term risks;
- concerned about increased levels of glyphosate in food;
- considers gene technology unethical;
- considers the outcomes of gene technology scientifically unpredictable because of the possibility that DNA can readily transfer between species.

Nadine McRae and others (NZ)

- opposes all of the six applications on the grounds that gene technology is unpredictable, unsafe and harmful to the environment;
- demands that all food with some genetically modified food content be labelled.

National Council of Women of Australia

- requests that ANZFA maintain the status quo and not amend Standard A18 to permit the sale of the indicated foods;
- no deliberations on applications should be made under this Standard until the situation with labelling is resolved;
- there is no mention of monitoring pesticide residue increase in the final product as a result of a greater tolerance to what is an obvious need to increase the pesticide used;
- for the soybean applications there should be absolutely no doubt about the safety of the source of the soybean if it is to be used in the Australian food supply;
- only two out of the six foods have been tested by feeding to laboratory animals and then only for 6 weeks;
- no evidence was provided about herbicide residue levels in any of the soybean foods despite there being an application to increase the MRL for glyphosate in soybeans;
- although the CP4 EPSPS protein may be inactivated on processing, the application does not take into account the use of raw soybeans to grow sprouts. This could represent an allergy problem and therefore the foods should be labelled;
- ANZFA has not taken into consideration the considerable consumer backlash that is occurring;
- there must be scientific certainty that humans are not exposed to any newly expressed proteins;
- objects to the commercial-in-confidence aspects of A362;
- concerned about the feeding of genetically modified seeds to animals as this is another source for these products entering the human food supply;
- there is no justification for using glyphosate–tolerant canola;
- Australia should be able to prohibit the import of genetically modified foods if it wishes;
- if ANZFA allows genetically engineered foods to be imported into Australia unlabelled, consumers will be affected by a lack of choice.

Natural Law Party (NZ)

- in the absence of a moratorium on genetically modified food, demands labelling of all genetically modified foods on the grounds that there has been no long term pre–market testing or screening for risk factors associated with this technology and that unlabelled products deprive individuals of their basic freedom of choice;
- rejects the notion of substantial equivalence on the grounds that differences at the DNA level make them substantially different;
- concerned about the potential for increased glyphosate levels;
- the effects of glyphosate on health and on phytoestrogens in genetically engineered soy has not been addressed;
- genetically engineered soy contains genes from a virus, a soil bacterium and from petunia, none of which has been in our food before;
- the technology is being introduced in the total absence of an informed public debate about the general acceptance of GMO technology;
- believe that there is significant potential for environmental or health disasters associated with the current introduction of this technology. Believes that serious liability implications exist and need to be explored;
- recommends that, until long term independent safety and risk assessment studies on genetic technology in food production have been completed and their safety to human health and the ecosystems that support human life is established, approvals for these foods should be declined;
- no further applications should be considered until proper public debate has occurred.

New Zealand Nutrition Foundation

- submission identical to InforMed Systems Ltd

Office of Regulation Review (Aust)

- comments on the preparation of the RIS for the full assessment report;
- ANZFA should discuss, in the background section of the report, why products such as the Roundup Ready soybeans, which previously entered the commercial markets without segregation from the non–transgenic counterpart, now require an approval process. Questions whether the regulation is to address health and safety and/or consumer information concerns;
- the problem section of the RIS should outline the characteristics of food produced using gene technology and why these characteristics might give rise to the need to list special conditions. The RIS should specifically canvass the possible special conditions which could apply and fully discuss the varying costs and benefits that each set of conditions entails;
- the material present in the sections on potential regulatory impacts and identification of affected parties should be summarised in the RIS in matrix form;
- when the RIS is fully developed it will need to include a conclusion section which summarises the views elicited from the main affected parties, a conclusion and recommendation option section which states what the preferred option is and why this option was accepted and the others rejected, and an implementation and review section which outlines how the proposal will be administered, implemented and enforced.

Martin Oliver (Aust)

- opposes all six applications on the grounds that the long term safety of eating foods from herbicide tolerant or insect resistant crops has not been adequately established;
- all genetically modified foods should be labelled in order for consumers to choose;
- claims that the foods have not been tested for any health impact on humans.

The Pacific Institute of Resource Management/Revolt Against Genetic Engineering (NZ)

- all genetically modified food should be labelled so that there can be post-market monitoring for new allergens or toxic effects in consumers;
- strongly opposed to the technology because of a range of concerns about public health and safety;
- raised a number of concerns in relation to Application A338 specifically:
  - the bacterial EPSPS is unlike any protein that humans have ever eaten and there is no reliable method for predicting its allergenic potential;
  - a major allergen, trypsin inhibitor was found to be 26.7% higher in transgenic soybeans;
  - the compositional analyses of the soybeans were not done on soybeans that had been treated with the herbicide;
  - there were significant increases compared to controls in the milk fat of cows fed transgenic soybeans; and
  - the applicant did not submit any data on glyphosate residues in the transgenic soybeans.

Sara Parsons (NZ)

- objects to the applications because she is a vegetarian;
- it is harmful to be introducing genetically modified soybeans, corn, canola oil and cottonseed into the NZ food chain;
- these products are a threat to the safety and well being of animals and humans and are of no benefit to society;
- the testing of genetically modified foods on animals and the harm that may be caused to animals in the wider environment is unacceptable;
- the lack of labelling of genetically modified foods means that NZ consumers have no way of making appropriate choices if they wish to avoid eating such foods which may cause allergic reactions and offend ethical beliefs.

Eric Phimister (NZ)

- is concerned about the importation of unlabelled genetically modified food;
- does not wish to consume soybeans with a higher pesticide level than the previously allowed maximum. This alone should make it not substantially equivalent.

Marja Rouse (Aust)

- opposes all six applications on the grounds that the genetically engineered crops pose a major environmental hazard and human health hazard;
- claims that the technology promotes unsustainable farming practices;
- believes consumers have the fundamental right to be informed about all the ingredients in foods and therefore demands mandatory labelling;

- the safety assessment for the applications should not be based on information provided by the applicant in these cases, as the company has a vested interest in having the applications approved.

Dean Scahill (NZ)
- is opposed to the foods which are the subject of Monsanto's applications on the grounds that the costs in terms of potential risk to health, risk to organic crop contamination, and current inability of consumers to identify such foods, greatly outweighs the benefits;
- if NZ remains GMO–free is represents an opportunity to create a niche market;
- a labelling system should be developed which would provide consumers with a choice so that they can retain the right to not eat genetically modified food should they choose;
- ANZFA should address the large public concern associated with the introduction of genetically modified foods onto the market.

Emma Subue-Timson (Aust)
- opposed to foods produced using gene technology on the grounds that the technology contravenes nature.

Christine Taylor (Aust)
- opposes all applications because of the presence of new genes, new proteins and increased herbicide residues in genetically modified foods;
- concerned about the potential for herbicide resistance genes to transfer to other plant species, creating undesirable effects.

Bridget Thrussell (NZ)
- supports regulatory option 1- to not permit the sale of any of the foods in the applications;
- no long term safety tests have been done;
- worried about antibiotic resistance increasing because of the antibiotic resistance marker genes in A355;
- concerned about gene transfer between Roundup Ready canola and other Brassicas.

E.M. Trevelyan (NZ)
- does not believe that genetically modified foods can be assessed as safe because of the possibility of "gene flow";
- crops containing the Bt gene will inevitably lead to resistant insect populations;
- envisages an enormous marketing advantage to NZ if it maintains a clean, green image by not allowing genetically modified food onto the market;
- all genetically modified food products should be labelled.

Richard van Wegen (Aust)
- supports the restricted use of genetically modified plants for food production;
- strongly supports mandatory labelling as a democratic right to make informed decisions about food purchases.

Arnold Ward (Aust)
- opposed to all applications on the grounds that long term safety has not been

established;
- ANZFA only concerns itself with public safety rather than adopting a 'holistic' approach which takes into consideration the broader issues to do with genetic engineering
- Roundup herbicide contains other chemicals which are harmful. Considers that the acceptable daily intake of glyphosate does not take into account the higher toxicity of the surfactant POEA in Roundup, on individuals with increased susceptibility such as children, immune compromised individuals or the elderly;
- notes examples of scientific evidence which show glyphosate can increase levels of plant oestrogens, which are known to affect humans. Very concerned about the potential health effects, particularly in children, of higher levels of oestrogens;
- feeding experiments in cows indicate a change in the milk fat production in animals fed on Roundup Ready soybeans versus non-transgenic soybeans;
- where resistance to Bt toxin occurs because of a widespread use of insect resistant crops, this would mean that organic farmers, who now rely on Bt formulations, could lose an important pest control agent;
- expresses concern about the possibility of recombination and horizontal gene transfer resulting in environmental catastrophies;
- glyphosate does not degrade in soils as efficiently as claimed by the applicant;
- all transgene products should be given the same testing applicable to pharmaceuticals;
- the seeds from genetically engineered crops could spread due to natural disasters;
- plant viruses can acquire viral DNA from a transgenic plant;
- Bt cotton is not very effective in controlling bollworm infestations;
- calls for a moratorium of 10 years on the introduction of genetically modified foods.

Joyce Weatherhead (NZ)
- opposes approval for the applications on the grounds that genetically modified foods have not undergone an independent scientific testing;
- calls for a moratorium on genetically modified foods in NZ for ethical and religious reasons;
- demands mandatory labelling of all genetically modified foods;
- believes that approval for herbicide resistant soybeans will result in a huge increase in the level of contaminating herbicides in foods derived from these crops.

Western Australian Food Advisory Committee
- a safety assessment of the foods is lacking along with the absence of any supporting scientific evidence;
- post–market monitoring to confirm the results of risk assessment and establish evidence of a safe history of use is an unacceptable alternative to a full scientific evaluation, with the results being available for public scrutiny;
- the claim that CP4 EPSPS is destroyed in heat processing requires independent scientific validation and it is unclear from ANZFA's papers whether this evidence has been provided and reviewed;
- insufficient evidence has been provided in the discussion document to support claims that these products are safe or that the Authority has undertaken a

rigorous analysis or comprehensive scientific evaluation of these products;
- the issue of decreased availability of food choices in the marketplace listed under both Options 1 and 2 is not nearly as important as the safety issue;
- given the heightened public concern about genetically modified foods it is essential that scientific information relating to compositional variance due to novel gene expression, toxicology, potential for allergenicity, nutritional and dietary properties for each of the foods proposed by Monsanto, is publicly available;
- the Committee recommends the adoption of Option 1 at this time.

S. and L. Wintergraas
- ANZFA should stop all genetically engineered foods from entering into any food products in NZ, as it will destroy the clean green image;
- ANZFA is not able to guarantee safety of these foods - cites DDT, nuclear power and antibiotics as examples;
- ANZFA should protect the consumer, not big business.

SUMMARY OF SECOND ROUND PUBLIC COMMENTS

The Authority received four applications from Monsanto Australia Ltd. (A346, A355, A362, A363) and one from Dupont/Pioneer (A387) for foods produced using gene technology. A draft Risk Analysis Report (formally referred to as the Full Assessment Report) was released for a 10 week period of public consultation on 19 June 2000. At the end of the public comment period (30 August) a total of 24 submissions had been received.

1. J Coburn (New Zealand)
- does not want these experimental foods in the common food supply until they have been long–term tested for undesirable side–effects related to public health;
- questions the fairness of the ANZFA response to the concerns expressed in the first round of public submissions;
- comments on the risk of spread of antibiotic resistance due to the use of antibiotic resistance marker genes;
- objects to any trace of herbicide residues in general;
- submits that the Draft Regulatory Impact Assessment is flawed.

2. Commerce Commission (New Zealand)
- concerned that labelling and advertising of GM foods is not misleading or deceptive;
- concerned whether by-products from the processing of GM foods could be fed to animals.

3. National Genetic Awareness Alliance (Australia)
- believe that there has been no independent scientific research conducted by ANZFA in the risk assessment process;
- request ANZFA to set up analytical techniques to measure DNA or protein in GM-crop-derived oils;
- object to the use of viral promotors such as Cauliflower Mosaic Viral

Promotor (CaMV);
- believe that GM crops yield less than conventional crops and require more herbicides.

4. InforMed Systems (Aust)
- comment that the use of antibiotic resistance marker genes should be phased out;
- the absence of any perceived benefit to the consumer of the modifications is not relevant to their safety, but can only increase public resistance to the technology.

5. Food Technology Association (Australia)
- recommend long-term feeding trials;
- question whether all "novel chemicals" have been "identified/discovered".

6. Australian GeneEthics Network
- recommends rejection of all applications on GM food;
- Monsanto's proposals should all be rejected as inadequate;
- questions the relevance of substantial equivalence;
- believes ANZFA should adopt the precautionary principle in its risk assessment process;
- suggest that insect-resistant crops should be considered an insecticide;
- labelling of GM food should be encouraged;
- believes the research conducted by Ewen and Pusztai should be considered in the assessments;
- objects to the use of antibiotic resistance marker genes;
- believes precautionary principle should be applied to the use of viral promotor sequences;
- recommend long-term feeding studies be undertaken;
- believe that gene silencing and its potential repercussions are not fully understood;
- state that ANZFA needs to take into account the variation in diet between different cultural and ethical groups.

7. Environmental Health Branch – Department of Human Services (South Australia)
- state that the approval of a food produced using gene technology for human consumption in Australia should not depend on the GMO from which it is derived being cleared for general release in Australia. However, if clearance for general release of a GM crop is sought from GMAC and rejected, ANZFA should take account of the reasons for rejection in assessing any application received by the Authority in relation to any food produced using gene technology for human consumption derived from the GM crop.
- applicants should identify which food products produced using genetic modification contain novel DNA and/or protein so labelling requirements can be determined;
- draft variations to Standard A18 should be specific as to which foods are permitted.

8. National Council of Women of Australia

- believe that the safety evaluation used by ANZFA is not the best suited to the evaluation of GM food;
- do not support the concept of substantial equivalence;
- objects to the use of antibiotic resistance marker genes;
- believe that animal feeding studies and human feeding studies should be conducted before GM foods are approved;
- post-market surveillance should be carried out since there is no long-term history of safe use of novel foods;
- all food derived from, or processed using genetic engineering, whether any DNA or protein remains in the finished product or not, should be labelled.
- believe that many statements made in the reports are not decisive;
- believe that the public's concerns are being over ridden by trade and other commercial interests;
- want the Office of the Gene Technology Regulator to be the overall regulator on all gene technology matters;
- believes that ANZFA does not deal with the issue of potential allergenicity appropriately; unknown allergens are not tested for;
- states that the Regulatory Impact Assessments for the applications are misleading and that there are no benefits to consumers;
- warn about the risks of using viral promotors such as CaMV;
- support continuing public consultation and information regarding gene technology;
- object to any chemical residues in foods.

9. Australian Competition & Consumer Commission

- the Commission believes that consumers have a right to purchase products that reflect their own personal preferences. Consumers must be able to rely on disclosures on packaging in order to make purchasing decisions;
- the Commission recommended the delay in the approval of the applications until the ANZFSC labelling decision of 28 July;
- column 2 of the table to clause 2 of Standard A18 could be used to require positive initiatives be undertaken by the applicant such as public information about the food in question or GMOs generally.

10. Institute of Environmental Science & Research Limited (New Zealand)

- ESR believe the ANZFA safety assessment process is consistent with current international "best practice" for this area;
- ESR's review of the data supporting the applications for approval of GM foods concluded that there was no reason to disagree with the ANZFA assessment that these foods are safe for human consumption;
- Greater toxicological testing is desirable to improve the data supporting the safety of GM foods, although it is acknowledged that there are practical difficulties in testing whole foods.

11. Consumers' Institute of New Zealand Incorporated

- expressed concerns over whether ANZFA had established that the evidence provided by the applicant has not been superceded by subsequent research;
- audit processes should be established to ensure that if new knowledge suggests

there is any risk associated with the foods that approval can quickly be withdrawn;
- ongoing monitoring of the long-term effects of the foods should also be established;
- expressed concern on the lack of independent verification of testing carried out by the developers of the products;
- believe the concept of substantial equivalence is not rigorous;
- comment that the language in the risk analysis documents gives the impression that uncertainty remains about the products;
- believe that GM foods should be treated in the same way as medicines in relation to tests required to establish safety.

12. G C Morgan (New Zealand)
- raised concerns regarding the use of pesticides on crops;
- comments that there is little evidence of any benefit of the introduction of GM foods to the consumer.

13. Consumers' Association of South Australia Inc.
- strongly support the submission of the National Council of Women.

14. Dieticians Association of Australia
- supports full labelling of GM food;
- comments that the broader environmental impacts of GM foods are not being addressed in the evaluations of the applications;
- believes that the recent decision on labelling of foods produced using gene technology should be extended further to require labelling of purified foods from GM sources, such as oils from glyphosate tolerant canola, even if there are no nutritional or safety concerns with the food;
- noted that very few of the studies that are relied upon in the evaluations have been published in peer-reviewed journals;
- comment that for a number of the applications there are no feeding studies.

15. I P Hancox (New Zealand)
- expressed general concerns regarding the environmental impact of GM foods.

16. P Gilgenberg (New Zealand)
- expressed general concerns regarding the safety of GM foods.

17. Food Technology Association of Victoria Inc (Australia)
- recommend long-term feeding trials;
- labelling of foods noting the presence of a GMO should only apply where more than 1% of any food contains a GMO present.

18. Canberra Consumers Incorporated (Australia)
- comments that none of the reports were peer reviewed;
- expressed concern over the use of antibiotic resistance marker genes;
- recommend long-term feeding trials;
- expressed concerns that GM foods used as stock feeds for animals are not safe for livestock.

19. National Council of Women of New Zealand (Te Kaunihera Wahine O Aotearoa)

- the Council recommends that where substantial differences are detected in GM foods these products must be labelled;
- the Council advocate that an adequately funded independent scientific body to evaluate data be established as soon as possible.

20. University of Auckland, Food Science Postgraduate Programme

- recommend long-term studies be conducted
- expressed concern over the use of antibiotic resistance marker genes.

21. Monsanto Australia Limited

- there are many formulations of the herbicide Roundup and not all have the surfactant POEA in them;
- some formulations e.g. Roundup Biactive is actually registered for use in waterways because the surfactant is approved with a good aquatic toxicological profile;

22. Arnold Ward (Australia)

- believes that ANZFA largely ignores submissions from the general public and is in league with large biotechnology companies;
- believes that there is a conspiracy between ANZFA and the US government and the FDA regarding the introduction of GM foods;
- wants ANZFA to exercise the precautionary principle and not approve GM food until it is proven to be safe;
- states that GM foods may not be as nutritional as conventional foods;
- objects to the concept of substantial equivalence;
- recommends long-term feeding studies in animals and human studies be conducted;
- recommends caution on the use of promotors such as CaMV;
- recommends that GM foods be treated the same as drugs in terms of testing requirements.

23. Carolyn Kitson

- recommends that ANZFA guidelines for data requirements on safety of GM foods be consistent for every application.

24. Ministry of Health (New Zealand)

- considers the ANZFA safety assessment process is consistent with international “best practice” in this area and that all the applications were subject to this process;
- in relation to the assessments themselves, and by way of summary, MoH agree with the conclusion reached in each assessment, that these foods are safe for human consumption;
- the concentration of newly expressed proteins were generally very low as the refinement processes involved removal of these proteins;
- consider that the applications closely considered the potential allergenicity of the newly expressed proteins on the basis of the physical and chemical nature

of these proteins, and the similarity of their amino acid sequence with known allergens;
- compositional analyses of the nutrients in control and GM food indicated no substantial differences in the levels of major nutrients; and
- toxicological effects of the modified foods were evaluated (although the estimated dietary intakes of the newly expressed proteins were not determined).

The following submissions were received after the end of the consultation period of 30 August 2000:

Public Health Association of Australia (PHAA) (part submission received 28 September 2000)

- believe that all studies submitted by industry should first be published by peer reviewed journals before undergoing the regulatory process;
- believe that there is a conflict of interest in an applicant company doing its own safety assessments and studies should be reproduced by independent laboratories;
- comment that the statistical analyses on the compositional studies is inadequate;
- contend that these foods undertake at least thorough animal testing, and at least the first phase of the four phases of a clinical trial before being released;
- contend that the issue of likely horizontal gene transfer has not been adequately resolved.

Australian Food and Grocery Council (received September 2000)

- The AFGC supports approval of each the applications A346, A355, A362, A363 and A387 on the basis that they do not raise any public health and safety concerns;
- Labelling of these foods should be according to the 28 July decision of the ANZFSC to enable consumers to make an informed choice.

ATTACHMENT 6

GENERAL ISSUES RAISED IN PUBLIC COMMENTS

The majority of submissions received in response to the Section 14 Gazette Notice, expressed general views against the use of gene technology and asserted that food produced using this technology is unsafe for human consumption. A number of general issues were raised in these submissions and are addressed below.

1. *The safety of genetically modified foods for human consumption*

A majority of submitters raised the issue of public health and safety in relation to food produced using gene technology. In particular, it was stated that there has been inadequate testing of genetically modified foods, that there is limited knowledge concerning the risks associated with the technology and that there may be potential long–term risks associated with the consumption of such foods.

Evaluation

It is a reasonable expectation of the community that foods offered for sale are safe and wholesome. In this context, 'safe' means that there is a reasonable certainty of no harm. As with other aspects of human activity, the absolute safety of food consumption cannot be guaranteed. Conventionally produced foods, while having a long history of safe use, are associated with human disease and carry a level of risk which must be balanced against the health benefits of a nutritious and varied diet.

Because the use of gene technology in food production is relatively new, and a long history of safe use of these foods has yet to be established, it is appropriate that a cautious approach is taken to the introduction of these foods onto the market. The purpose of the pre–market assessment of a food produced using gene technology under Standard A18 is to establish that the new food is at least as safe as existing foods. The comprehensive nature of the scientific safety assessment, undertaken on a case-by-case basis, for each new modification is reflective of this cautious approach.

The safety assessment focuses on the new gene product(s), including intentional and unintentional effects of the genetic modification, its properties including potential allergenicity, toxicity, compositional differences in the food and it's history of use as a food or food product.

Foods produced using gene technology are assessed in part by a comparison with commonly consumed foods that are already regarded as safe. This concept has been adopted by both the World Health Organisation (WHO)/Food and Agriculture Organisation (FAO) and the Organisation for Economic Cooperation and Development (OECD). The Authority has developed detailed procedures for the safety assessment of foods produced using gene technology that are consistent with international protocols developed by these bodies.

2. *The need for long-term feeding studies*

A number of submissions were concerned about the lack of long-term toxicity studies on genetically modified foods.

Evaluation

Animal studies are a major element in the safety assessment of many compounds, including pesticides, pharmaceuticals, industrial chemicals and food additives. In most cases, the test substance is well characterised, of known purity and of no nutritional value, and human exposure is generally low. It is therefore relatively straightforward to feed such compounds to animals at a range of doses (some several orders of magnitude above expected human exposure levels) in order to identify any potential adverse effects. Establishing a dose-response relationship is a pivotal step in toxicological testing. By determining the level of exposure at which no adverse effects occur, a safe level of exposure for humans can be established which includes appropriate safety factors.

By contrast, foods are complex mixtures of compounds characterised by wide variations in composition and nutritional value. Due to their bulk, they can usually be fed to animals only at low multiples of the amounts that might be present in the human diet. Therefore, in most cases, it is not possible to conduct dose-response experiments for foods in the same way that these experiments are conducted for chemicals. In addition, a key factor to be considered in conducting animal studies on foods is the need to maintain the nutritional value and balance of the diet. A diet that is poorly balanced will compromise the interpretation of any feeding study, since the effects observed will confound and usually override any small adverse effect which may be related to a component or components of the food. Identifying any potentially adverse effects and relating these to an individual component or characteristic of a food can, therefore, be extremely difficult. Another consideration in determining the need for animal studies is whether it is appropriate from an ethical standpoint to subject experimental animals to such a study if it is unlikely to produce meaningful information.

If there is a need to examine the safety of a newly-expressed protein in a genetically-modified food, it is more appropriate to examine the safety of this protein alone in an animal study rather than when it is part of a whole food. For newly-expressed proteins in genetically-modified foods, the acute toxicity is normally examined in experimental animals. In some case, studies up to 14 days have also been performed. These can provide additional re-assurance that the proteins will have no adverse effects in humans when consumed as part of a food. Such experiments can provide more meaningful information than experiments on the whole food. Additional re-assurance regarding the safety of newly-expressed protein can be obtained by examining the digestibility of the new protein in *in vitro* assays using conditions which simulate the human gastric system.

3. *Substantial equivalence*

A number of submitters expressed concern regarding the use of the concept of substantial equivalence as part of the assessment process. Some rejected the premise

of substantial equivalence on the grounds that differences at the DNA level make foods substantially different.

Evaluation

Substantial equivalence embodies the concept that, as part of the safety assessment of a genetically modified food, a comparison can be made in relation to the characteristics and properties between the new food and traditionally-produced food. This can include physical characteristics and compositional factors, as well as an examination of the levels of naturally occurring allergens, toxins and anti-nutrients.

This allows the safety assessment to focus on any significant differences between the genetically modified food and its conventionally produced counterpart. Genotypic differences (i.e. differences at the DNA level) are not normally considered in a determination of substantial equivalence, if that difference does not significantly change the characteristics for composition of the new food relative to the conventional food.

The concept of substantial equivalence allows for an evaluation of the important constituents of a new food in a systematic manner while, recognizing that there is general acceptance that normally consumed food produced by conventional methods is regarded by the community as safe. It is important to note that, although a genetically modified food may be found to be different in composition to the traditional food, this in itself does not necessarily mean that the food is unsafe or nutritionally inadequate. Each food needs to be evaluated on an individual basis with regard to the significance of any changes in relation to its composition or to its properties.

The concept of substantial equivalence was first espoused by a 1991 Joint Consultation of the Food and Agricultural Organisation (FAO) and the World Health Organisation (WHO) where it was noted that the '*comparison of a final product with one having an acceptable standard of safety provides an important element of safety assessment*.' The concept has been internationally recognised and embraced as a valuable tool in the safety assessment of foods produced using gene technology. The OECD also advocates an approach to safety assessment based on substantial equivalence as being '*the most practical to address the safety of foods and food components derived through modern biotechnology*.'

4. *The nutritional value of food produced using gene technology*

A small number of submitters expressed concern that the genetic alteration of food decreases its nutritional value.

Evaluation

The assessment of food produced using gene technology by ANZFA entails an exhaustive evaluation of analytical data on any intentional or unintentional compositional changes to the food. This assessment encompasses the major constituents of the food (fat, protein, carbohydrate, fibre, ash and moisture) as well as the key nutrients (amino acids, vitamins, fatty acids). There is no evidence to suggest that genetic modification *per se* reduces the nutritional value of food.

In the future, genetic modification may be used intentionally to improve the nutritional value of food. In this regard, GM foods may be able to assist in addressing the general nutritional needs of the community and also specific dietary needs of sub-populations.

5. *Potential toxins and allergens*

Some submitters expressed concerns about the risks of the introduction of new toxins or allergens.

Evaluation

This issue is considered in detail as part of the safety assessment conducted on each new genetic modification applied to a food or commodity crop. New toxins or allergens may be introduced into food by either gene technology or by traditional breeding techniques, or by altered production processes. It is also possible to use these techniques to develop foods specifically where such compounds are significantly reduced or eliminated. One advantage of gene technology, in comparison with these other methods, is that any transferred genes are well characterised and defined, thus the possibility of developing a food with a new toxic or allergenic compound is likely to be reduced.

6. *Antibiotic resistance*

Some submitters raised concerns about increased antibiotic resistance resulting from the use of gene technology. Some felt that it would be reassuring if independent biomedical advice were available to reassure the public that the use of antibiotic resistance markers does not pose a risk to the future use of antibiotics in the management of human disease.

Evaluation

The human health considerations in relation to the potential for the development of antibiotic resistance depend on the nature of the novel genes and must be assessed on a case-by case basis. This issue arises because of the use of antibiotic resistance marker genes in the generation of genetically modified plants. In some circumstances, antibiotic resistance genes are linked to the gene of interest, to enable the initial selection of the engineered cells in the laboratory. Those cells that contain the antibiotic resistance marker gene, and hence the gene of interest, will be able to grow in the presence of the antibiotic. Those cells that failed the transformation process are eliminated during the selection procedure.

Concern has arisen that ingestion of food containing copies of antibiotic resistance genes could facilitate the transfer of the gene to bacteria inhabiting the gut of animals and humans. It is argued that these genes may then be transferred to disease causing bacteria and that this would compromise the therapeutic use of these antibiotics.

In 1993, the World Health Organisation Food Safety Unit considered this issue at a Workshop on the health aspects of marker genes in genetically modified plants. It was concluded at that Workshop that the potential for such gene transfers is effectively

zero, given the complexity of the steps required. Since this time, several separate expert panels (Report to the Nordic Council, Copenhagen 1996; Advisory Committee on Novel Foods and Processes, UK 1994, 1996; The Royal Society, UK 1998) and numerous scientific papers published in peer reviewed journals have also considered the available evidence on this issue. It is generally agreed that the presence and subsequent transfer of an intact functional gene from transgenic food to micro-organisms in the human intestine is an extremely unlikely event. Furthermore, if this were to occur, bacteria would not normally retain the resistance genes unless there was an environment for positive selection. The majority of these genes provide for resistance to antibiotics whose use is confined to the laboratory and are not considered to be of major therapeutic use in humans.

Antibiotic resistant bacteria are naturally occurring, ubiquitous and normally inhabit the gut of animals and humans. There is a general consensus that the transfer of antibiotic resistance genes is much more likely to arise from this source and from associated medical practices, rather than from ingested genetically modified food. Even so, at the recent OECD Conference (GM Food Safety: Facts, Uncertainties, and Assessment) held in Edinburgh on 28 February – 1 March 2000, there was general consensus that the continued use of antibiotic marker genes in GM food crops is unnecessary given the existence of adequate alternatives, and should be phased out.

7. Transfer of novel genes

Some submitters have expressed concern that the transfer of any novel gene may be a health concern.

Evaluation

It is extremely unlikely that novel genetic material will transfer from GM foods to bacteria in the human digestive tract because of the number of complex and unlikely steps that would need to take place consecutively. It is equally unlikely that novel genetic material will transfer from GM foods to human cells via the digestive tract. In considering the potential impact on human health, it is important to note that humans have always consumed large amounts of DNA as a normal component of food and there is no evidence that this consumption has had any adverse effect on human health. Furthermore, current scientific knowledge has not revealed any DNA sequences from ingested foods that have been incorporated into human DNA. Novel DNA sequences in GM foods comprise only a minute fraction of the total DNA in the food (generally less than 0.01%) and are therefore unlikely to pose any special additional risks compared with the large amount of DNA naturally present in all foods.

8. *Viral recombination*

Some submitters expressed concern about the long term effects of transferring viral sequences to plants.

Evaluation

This is an issue that is commonly raised because some of the genes that are transferred to plants use a plant virus promoter. Promoters are controlling DNA sequences which act like a switch and enable the transferred genes to be expressed (i.e. to give rise to a protein product) in a plant cell. The routine use of these viral promoters is often confused with research which has shown that plant virus genes, which have been transferred into plants to render them virus–resistant, may recombine with related plant viruses that subsequently infect the plant, creating new viral variants. This research demonstrates that there may be a greater risk to the environment if viral genes are transferred to plants because it may lead to the generation of new plant virus variants capable of infecting a broader range of plants. This is a matter that will be addressed by the Genetic Manipulation Advisory Committee (GMAC) on a case–by–case basis when it assesses such plants.

However, the presence of plant viruses, plant virus genes or plant virus segments in food is not considered to pose any greater risk to human health as plant viruses are ubiquitous in nature and are commonly found in food eaten by animals and humans. Plant viruses are also biologically incapable of naturally infecting human or animal cells.

9. ***Labelling of foods produced using gene technology***

A majority of submissions focussed on this issue. Specifically, the submissions called for the labelling of all foods produced using gene technology, regardless of whether they are substantially equivalent to conventional foods. The submitters based their demands for full labelling on the presumption that all foods produced using gene technology are unsafe and on consumer "right to know" arguments. It was stated that full labelling was the only means of identification of foods produced using gene technology available to consumers.

Evaluation

The existing Standard A18 already makes provision for mandatory labelling of genetically modified foods that are substantially different from their conventional counterparts. However, ANZFA is committed to implementing the in-principle decision of ANZFSC Health Ministers of August 1999 to require labelling of all genetically modified foods, including those that are substantially equivalent in composition to the unmodified form. In conjunction with a task force of officials from State and Territory Health Departments and the New Zealand Ministry of Health, ANZFA developed draft revision to Standard A18 in October 1999 that requires labelling of other categories of genetically modified foods. At the Ministers request this draft was circulated for public review and a cost-benefit analysis of full labelling was commissioned. The task force considered both public comments and the cost-benefit analysis in finalising their recommendations to Ministers, which were delivered in May 2000. Ministers are to meet to resolve the issue in July 2000 following whole-of-government consideration of the issue. It is therefore expected that, following a decision and legal amendments to the standard, labelling requirements will be implemented that will apply to all current and subsequent applications.

10. The need for post marketing surveillance of genetically modified foods

A number of submitters have commented on the need for post-market surveillance of genetically modified food consumption.

Evaluation

Surveillance of potential adverse or beneficial effects of GM foods is seen by many as a logical follow-up to the initial scientific risk assessment. Nevertheless, it is recognised that there are limitations to the application of epidemiology studies, particularly in relation to food components. A key requirement for post-market surveillance systems is that a clear hypothesis be identified for testing. Establishing a system for the surveillance of potential health effects of exposure to novel foods requires monitoring of the consumption patterns of novel foods in the population, and health effects in both "exposed" and "non-exposed" individuals/populations, so that risk estimates can be derived. For any such monitoring system to be useful, there needs to be a range of exposures, otherwise, any variation in health outcome would be unexplainable by that exposure. Variations in exposure could be apparent over time (temporal trends), space (geographical trends) or both.

Availability of robust data on consumption of the foods in question is vital in order to establish a surveillance system. The other side of the equation is the need for access to data on population health outcomes. Such a system could also be used to identify potential positive health outcomes, such as improved nutritional status or lower cholesterol levels. The availability of linked basic data (e.g. date of birth, sex, geographical location), and the ability to correlate with demographic data, could potentially offer the means of establishing links with food consumption.

The possibility of setting up a post-market health surveillance system for novel foods, including GM foods, has been examined by the UK's Advisory Committee on Novel Foods and Processes (ACNFP). Recognising the many difficulties involved in developing such a system, an initial feasibility study to look at the available data and its usefulness has been proposed. Work is currently being commissioned; when completed in 18 months, it will be subject to peer review. If such a feasibility study suggests that post-market surveillance is practical, methods and details concerning data collection will be determined in the UK, but common strategies might be able to be harmonised internationally in order to minimise the use of resources while maximising the reliability of the final results. This is an area that ANZFA will be monitoring closely, along with international regulatory bodies such as the OECD Taskforce for the Safety of Novel Foods and Feeds.

11. ***Public consultation and information about gene technology***

A number of submitters were concerned that the public has not been properly consulted or informed by government or ANZFA on the introduction of foods produced using gene technology. Some submitters urged to undertake wider consultation with all affected parties including growers, the food industry and consumers before these food commodities are introduced, and to ensure that adequate consultation is undertaken as part of its assessment process.

Evaluation

The issue of gene technology and its use in food has been under consideration in Australia since 1992. The Agreement between the Governments of Australia and New Zealand for a joint food standard setting system, however, did not occur until 1995, and the New Zealand community therefore had not been consulted on this matter by the Authority until after that time. Consequently, the proposed standard (the current Standard A18) underwent only one round of public comment in New Zealand at which time significant objections were raised by the New Zealand community to the use of gene technology in food production. Many New Zealand consumers, both in these submissions, and in previous submissions to the Authority, have expressed the view that there has been insufficient consultation and a consistent lack of information about gene technology.

Although Standard A18 came into force in May 1999, the public have a continuous and ongoing opportunity to provide comment in relation to applications under the standard. ANZFA's statutory process for all applications to amend the *Food Standards Code* normally involves two rounds of public comment. Furthermore, all the documentation (except for commercial in confidence information) relating to these applications is available in the public domain, including the safety assessment reports. There is ample evidence that the provision of such information by ANZFA has already significantly stimulated public debate on this matter.

In addition, other government departments including the Environmental Risk Management Authority (ERMA), are potential sources of information about gene technology available to consumers in New Zealand. ERMA is a statutory authority set up by the New Zealand Government to administer the *Hazardous Substances and New Organisms (HSNO) Act 1996*, and has responsibility for assessing the risks to the environment from genetically modified organisms. This body has been assessing applications for the approval of genetically modified organisms since July 1998 and this has involved a number of public meetings.

In response to the concerns raised in public submissions with regard to gene technology and GM foods, ANZFA is in the process of preparing a public discussion paper on the safety assessment process for GM foods. This will be widely available and may assist in addressing some of the concerns raised by the public. Other government and industry bodies are also addressing the broader concerns in relation to gene technology.

12. *Maori beliefs and values*

Some New Zealand submitters stated that Maori people find genetic engineering in conflict with their beliefs and values and that, out of respect to Maori, no genetically modified foods should be allowed into New Zealand until a wider discussion, both within Maori and non–Maori, is held.

Evaluation

This issue was also raised during consideration of the proposal for the establishment of Standard A18. At that time, it was stated that the likely implications for Maori regarding genetically modified organisms surround the issues of the rights of Maori to the genetic material from flora and fauna indigenous to New Zealand and the release into the environment of genetically modified organisms. The *HSNO Act 1996* requires that these matters be considered by ERMA.

13. *Environmental concerns and the broader regulatory framework*

A number of submitters have raised concerns that genetically modified crops may pose a risk to the environment.

Evaluation

These issues are considered in the assessment processes of GMAC in Australia and the Environmental Risk Management Authority (ERMA) in New Zealand. The Authority does not have the mandate to assess matters relating to environmental risks resulting from the release of food produced using gene technology into the environment. However, links exist between ANZFA and other regulatory agencies in both Australia and New Zealand, and a large degree of information sharing occurs. ANZFA would not recommend the approval of a food produced using gene technology if the genetically modified organism from which it was derived did not have the appropriate clearance for general release from either GMAC (or its successor) or ERMA, as appropriate.

The regulatory system in Australia will comprise the existing regulators with a legal remit to cover some aspects of GM products (such as imports, food, agricultural and veterinary chemicals):

- the Australia New Zealand Food Authority (ANZFA)
- the Therapeutic Goods Administration (TGA)
- the National Registration Authority for Agricultural and Veterinary Chemicals (NRA)
- the National Industrial Chemicals Notification and Assessment Scheme (NICNAS)
- the Australian Quarantine and Inspection Service (AQIS).

Similarly, various other departments and agencies play their role in the regulatory process in New Zealand:

- the Ministry of Agriculture and Fisheries (MAF)
- the Ministry of Health (MoH)
- the Ministry of Research, Science and Technology (MoRST)

In Australia a new Office of the Gene Technology Regulator (OGTR) will complement the existing arrangements. OGTR will supersede the existing arrangements under the Genetic Manipulation Advisory Committee (GMAC), which

advises on research and environmental release of GMOs. OGTR will regulate all GMOs and any 'gap' products (i.e. products for which no other regulator has responsibility).

All GM food is assessed and regulated by the Australia New Zealand Food Authority (ANZFA) under the direction of Commonwealth, State and Territories Health Ministers and the New Zealand Health Minister, sitting as Australia New Zealand Food Standards Council (ANZFSC).

There will be an interface between ANZFA and OGTR. Consequential amendments proposed to the ANZFA Act arising from the draft Gene Technology Bill 2000 will establish a statutory interface between OGTR and ANZFA. This will involve amendments to the ANZFA Act requiring the Authority to advise OGTR of recommendations to ANZFSC regarding the standard for foods produced using gene technology (currently Standard A 18).

14. Maximum residue levels of agriculture/veterinary chemicals

A number of submitters have raised concerns that residues of agricultural and veterinary chemicals in genetically modified (e.g. herbicide tolerant) crops may pose a health risk.

Residues of these chemicals can only legally be present if the chemical has been registered for use in Australia and/or New Zealand, and it has been demonstrated that the residue at specified levels does not lead to adverse health impacts. The concentration of a chemical residue that may be present in a food is regulated through maximum residue limits (MRLs). The MRL is the highest residue concentration that is legally permitted in the food. Food products have to meet the MRL, whether or not they are derived from genetically modified organisms. The MRL does not indicate the chemical residue level that is always present in a food, but it does indicate the highest residue level that could result from the registered conditions of use.

It is important to note that MRLs are not direct public health and safety limits but rather, are primarily indicators of appropriate chemical usage. MRLs are always set at levels lower than, and normally very much lower than, the health and safety limits. The MRL is determined following a comprehensive evaluation of scientific studies on chemistry, metabolism, analytical methods and residue levels. In Australia, the National Registration Authority (NRA) applies to ANZFA to amend the MRLs in the Food Standards Code and the application is considered by ANZFA through its legislated decision making processes. In New Zealand MRLs are set by the Ministry of Health, generally following a request from, and in collaboration with, the Ministry of Agriculture and Forestry. Only following demonstration that the use of agricultural and veterinary chemicals will not result in unsafe residues will the MRL enter into food law through its inclusion in either the Food Standards Code in Australia, or the Food Regulations (1984) in New Zealand.