

Determination of the Safety of Monsanto's
Sugar beet 77 (Glyphosate-Tolerant Sugar beet)
for Direct Use as Food, Feed and for Processing

Food and Feed Safety:

The product dossier on Glyphosate-Tolerant Sugar beet 77 (GTSB 77) were reviewed for safety and nutritional differences compared with the conventional sugar beet. The focus of the review was on any new or altered expression trait and changes in composition and nutritional content or value relative to the conventional sugar beet. At the end of the safety assessment, a conclusion was made that the Glyphosate-Tolerant Sugar beet 77 is as safe as the conventional sugar beet taking into account dietary impact of any changes in nutritional content or value.

A biosafety permit for Glyphosate-Tolerant Sugar beet 77 and all progenies derived from crosses of the product with any conventionally-bred sugar beet and sugar beet containing approved-biotech events for direct use as food, feed and for processing was issued to Monsanto Philippines Inc. on October 20, 2004. The permit is valid for five years and shall expire on October 19, 2009 subject to the terms and conditions set forth in DA Administrative Order No. 8, Series of 2002. Glyphosate-Tolerant Sugar beet 77 was included in the Lists of Approval Registry (Delisting) being prepared by the Department of Agriculture -Bureau of Plant Industry

This approval is for direct use as food, feed and for processing only. This does not include cultivation of Glyphosate-Tolerant Sugar beet 77 in the Philippines. Food and Feed use of GTSB 77 and its by-products is therefore authorized as of October 20, 2004. The Biosafety Permit (No. 04-018) stated that "Glyphosate-Tolerant Sugar beet 77 is as safe for human food, livestock feed and for processing as its conventional counterparts".

I. Brief Identification of the Genetically Modified Organism (Living Modified Organism)

Designation:	Glyphosate-Tolerant Sugar beet 77
Applicant:	MONSANTO PHILIPPINES, INC. 7 th Floor, Ayala-FGU Center Alabang-Zapote Rd., cor Acacia Avenue Madrigal Business Park Alabang 1770 Muntinlupa City
Plant Species:	
Name:	Sugar beet (<i>Beta vulgaris</i> ssp. <i>vulgaris</i> L.)
Parent Material:	Sugar beet variety A1012 (Propriety material)
Center of Origin:	Mediterranean Basin and Middle East
Allergen(s):	Saponins are triterpenoid glycosides that occur naturally in numerous food and feed crops including beans, peas, potatoes, tea and sugar beet. Hydrolysis of glycoside releases a lipid-soluble sapogenin. The predominant sapogenin in sugar beet is oleanolic acid, whose structure is well characterized. Generally saponins have a bitter and astringent taste and act as a deterrent to foraging. Saponins are actively eliminated during sugar processing and thus do not pose a risk to human health. Analysis for saponins in sugar beet usually consists of liberation of the oleanolic acid, which is quantified by HPLC
Trait Description:	Herbicide tolerance
Trait Introduction Method:	<i>Agrobacterium</i> - mediated transformation
Donor Organism:	<i>Agrobacterium</i> sp. strain CP4, the source of the <i>cp4epsps</i> gene, which confers tolerance to the herbicide glyphosate.

Escherichia coli, source of *uid* gene which encodes β D-glucuronidase (GUS), which serves as a marker for plant transformation.

Orthobacterium anthropi, source of *gox* gene which encodes the glyphosate oxidoreductase enzyme that can degrade the herbicide however it was truncated during transformation and 69% of the gene is fused to sugarbeet DNA resulting in a chimeric gene.

Pathogenicity:

Agrobacterium spp. is non-pathogenic, ubiquitous in nature and are ordinarily present in food derived from plant sources

E. coli is ubiquitous in nature and considered to be non-pathogenic.

Ochrobacterium anthropi is ubiquitous in nature and considered to be non-pathogenic.

Proposed Use:

For direct use as food, feed and for processing

II. Background Information

Sugar beets are grown for the fleshy, bulbous root which normally contains 10-15% sucrose. Sugar beets are processed into products for use in both human and animal food. Sugar, which is used in a variety of foods, is the main product. Common by-products from sugar processing are sugar beet molasses, which is used for production of yeast, chemicals, pharmaceuticals, and mixed cattle feeds; dried sugar beet pulp, which is used in feeding dairy cattle, beef cattle, and sheep; and sugar beet fiber, which is used in human food, as a source of fiber. A very modest amount of sugar beet tops is consumed as animal feed.

Monsanto Philippines Inc. has developed a sugar beet, derived from the Sugar beet A1012 variety, which expresses their proprietary Roundup-Ready™ genes. These confer novel tolerance to glyphosate, the active ingredient of Roundup® herbicide, which can control or suppress economically important weeds in sugar beet production.

On May 29, 2003, Monsanto Philippines Inc. submitted an application to the Bureau of Plant Industry requesting for Biosafety Permit under Administrative Order Number 8 (AO#8) Part 5 for Glyphosate-Tolerant Sugar beet 77 which has been genetically modified for herbicide resistance.

Monsanto Philippines Inc has provided data on the identity of the glyphosate tolerant Sugar beet 77, a detailed description of the modification method, data and information on the gene insertion sites, copy numbers and levels of expression in the plant, the role of the inserted genes and regulatory sequences in donor organisms, and full nucleotide sequences. The novel proteins were identified, characterized, and compared to the original bacterial proteins, including an evaluation of their potential toxicity to livestock. Relevant scientific publications were also supplied.

Glyphosate-Tolerant Sugar beet 77 (GTSB77) has been evaluated according to BPI's safety assessment by concerned agencies {Bureau of Animal Industry (BAI), Bureau of Agriculture, Fisheries and Product Standards (BAFPS)} and a Scientific Technical Review Panel (STRP). The process involves an intensive analysis of the nature of the genetic modification together with a consideration of general safety issues, toxicological issues and nutritional issues associated with the modified sugar beet.

The petitioner/applicant published the said application on two widely circulated newspapers (Malaya and Daily Tribune) on June 30, 2003 for public comment/review. BPI did not receive any comments on the petition during the 30-day comment period.

Review of results of evaluation by the BPI Biotech Core Team in consultation with DA-Biotechnology Advisory Team (DA-BAT) completed the approval process.

III. Description of Novel (Introduced) Traits

Glyphosate-Tolerant Sugar beet 77 (Trade name: Roundup-Ready Sugar beet) and all sugar beet lines/hybrids derived from this Event contain the CP4 EPSPS coding sequence from *Agrobacterium* sp. CP4 strain. The CP4 EPSPS sequence encodes for the production of the naturally-occurring CP4 EPSPS protein that renders the Glyphosate-Tolerant Sugar beet 77 tolerant to glyphosate, a herbicide known to be environmentally-compatible. The EPSPS enzyme is present in the shikimic acid pathway for the biosynthesis of aromatic amino acids in plants and microorganisms. Inhibition of this enzyme by glyphosate leads to a deficiency in the production of aromatic acids and lack growth in plants. The aromatic amino acid biosynthetic pathway is not present in mammalian, avian or aquatic animals. This explains the selective activity in plants and contributes to the low risk to human health and the environment from the use of glyphosate according to label recommendations.

Monsanto transformed sugar beet line A1012 with a disarmed *Agrobacterium tumefaciens* double border plant transformation plasmid to produce glyphosate tolerant sugar beet 77. The plasmid contained the following genes within the T-DNA border sequences of the vector: (1) the 5-enolpyruvylshikimate-3-phosphate synthase (*cp4 epsps*) gene from *Agrobacterium* sp. strain CP4; (2) the *uidA* gene, which encodes beta-D-glucuronidase (GUS), from *E. coli*; (3) a glyphosate oxidoreductase gene (*gox*) from *Ochrobactrum anthropi*; and (4) a neomycin phosphotransferase gene (*nptIII*) from transposon *Tn5* which provides resistance to aminoglycoside antibiotics, such as neomycin and kanamycin. In addition, the plasmid contained a bacterial selectable marker gene, *aad*, which provides resistance to spectinomycin and streptomycin, as well as two origins of replication (*ori-V* and *ori-322*) to permit replication and maintenance of the plasmid in bacteria.

Expression of CP4 EPSPS and GOX are controlled by the figwort mosaic virus (FMV) promoter and targeted to the chloroplast by a chloroplast targeting sequence from *Arabidopsis thaliana*. Expression of GUS is controlled by the cauliflower mosaic virus 35S promoter.

Safety of the Expressed Proteins

It has been documented that the refined sugar derived from sugar beet does not contain plant endogenous proteins or DNA. Likewise, refined sugar derived from GTSB 77 has been shown not to contain any detectable CP4 EPSPS, GUS or Protein 34550. It does not contain detectable DNA from the *cp4epsps* gene. CP4 EPSPS, GUS or Protein 34550 shows no significant amino acid similarity to any allergen or toxins.

They do not share characteristics of known protein allergens, such as stability to digestion on acidic condition, heat stability and high concentration in food.

IV. Nutritional Composition (Compositional Analysis)

Under the same agronomic condition as conventional counterpart cotton, the CP4 EPSPS does not alter the quantity and quality of the key components. The nutritional equivalence and wholesomeness of Sugar beet 77 with regards to the key nutrients including proximates, amino acids and fatty acids composition as well as antinutrients are comparable to conventional sugar beet.

The nutritional equivalence of GTSB77 compared to conventional sugar beet was confirmed in numerous feeding studies with sheep and dairy cattle which included clinical and histological evaluations.

V. Anti-Nutritional Factors

Antinutrients are not present in sugar beet, although one component, saponin is considered to act as a deterrent to foraging animals due to its astringent taste. Saponins are triterpenoid glycosides that have a broad biological activity and occur naturally in numerous food and feed crops including sugar beets. It is known to be present in small amounts in sugar beet. The level of this component in GTSB 77 is comparable to the levels in conventional sugar beet.

VI. Regulatory Decision

After reviewing the scientific data and information relevant to the application of Monsanto Philippines Inc., it is concluded that Glyphosate-Tolerant Sugar beet 77 and all progenies derived from crosses of the product with any conventionally-bred sugar beet and sugar beet containing approved-biotech events for direct use as food or feed or for processing is as safe and substantially

equivalent to its unmodified counterpart, and is therefore approved for direct use as food, or feed or for processing. Monsanto shall duly inform the public of this approval by way of publishing in any one (1) of the top three (3) leading newspapers in the country that imports of this product is covered by conditions for approval as provided in Department of Agriculture Memorandum Circular No. 8, Series of 2003.