

Determination of the Safety of Monsanto's
New Leaf[®]Y Potato (Potato Virus Y and Colorado Potato Beetle resistant-potato)
for Direct Use as Food, Feed and for Processing

Food and Feed Safety:

The product dossier on New Leaf[®]Y Potato were reviewed for safety and nutritional differences compared with the conventional potato. 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 potato. At the end of the safety assessment, a conclusion was made that the New Leaf[®]Y Potato Events RBMT15-101, SEMT15-02 and SEMT15-15 are as safe as the conventional corn taking into account dietary impact of any changes in nutritional content or value.

A biosafety permit for New Leaf[®]Y Potato and all progenies derived from crosses of the product with any conventionally-bred potato and potato containing approved-biotech events for direct use as food, feed or for processing was issued to Monsanto Philippines Inc. on December 5, 2003. The permit is valid for five years and shall expire on December 4, 2008 subject to the terms and conditions set forth in DA Administrative Order No. 8, Series of 2002. The said potato event (New Leaf[®]Y Potato Events RBMT15-101, SEMT15-02 and SEMT15-15) 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 Potato Virus Y and Colorado Potato Beetle resistant –potato in the Philippines. Food, Feed and use of its by-products is therefore authorized as of December 5, 2003. The biosafety permit stated that “New Leaf[®]Y Potatoes 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:	New Leaf [®] Y Potato
Applicant:	MONSANTO PHILIPPINES INC. 7 th Floor, Ayala-FGU Center Alabang-Zapote Rd., cor Acacia Avenue Madrigal Business Park Alabang 1770 Muntinlupa City Philippines
Plant Species:	
Name:	Potato (<i>Solanum tuberosum</i> L.)
Parent Material:	Shepody and Russet Burbank varieties
Center of Origin:	Potatoes originated in the Andes mountains of Peru and Bolivia
Toxic Factors/Allergen(s):	Toxicants that are naturally occurring in the potato tubers such as glycoalkaloids, solanine, and chaconine are within the standard levels established previously for potatoes
Trait Description:	Resistance to Potato Virus Y (PVY) and Colorado Potato Beetle (CPB)
Trait Introduction Method:	<i>Agrobacterium</i> -mediated transformation method
Donor Organism:	<i>Bacillus thuringiensis</i> subsp. <i>tenebrionis</i> , source of <i>cry3A</i> which confers resistance to the Colorado potato beetle and DNA sequences corresponding to potato virus Y (PVY) coat protein domains which confers resistance to PVY

Pathogenicity: *Bacillus thuringiensis* subsp. *tenebrionis* is a common soil bacterium that has a long history of safe use as a microbial insecticide with no reported allergenic and toxic responses, establishing basis for the lack of allergenic or toxic concern for the Cry3A protein. *B.t* based products have shown that the proteins produce toxic effects only in the gut of chewing insects and are not activated in human digestive tracts. The *cry3A* encodes for a protein, normally produced by the bacterium during sporulation, which is toxic only to targeted leaf feeding beetles.

Proposed Use: For direct use as food, feed of for processing

II. Background Information

Potato is grown in over 120 countries primarily for human consumption in its processed form. For direct consumption it is cooked before eaten and its processed form are potato fries, crisps and dehydrated potatoes. It is also used as raw material for starch manufacturing. The starch can be used for coating of papers, sizing of cotton and as finishing in textile industry. By-products (pulp, coagulated protein from fruit water) are normally used in animal feeding, but trends exist for food use too. Potatoes are also used for industrial alcohol production. In countries with a significant potato processing industry (for both human food and industrial use), the residues and by-products (peel, trimmings, rejected potatoes, separated pulp and proteins) are used as feedstuff (often after dehydration).

Approximately one-third of the estimated 3 million pounds of chemical insecticides annually applies to potatoes in the United States is targeted for Colorado Potato Beetle (CPB) control. Potato crops infected with mosaic viruses can experience yield reductions of up to 80%. This aphid-vectored virus is a serious concern for certified seed production because virus infected seed tuber lots cannot be certified for sale. Thus, insecticides must be consistently applied at frequent intervals to prevent any aphids from colonizing seed potato fields.

On June 12, 2003, Monsanto Philippines, Inc. submitted an application to the Bureau of Plant Industry requesting for biosafety permit under AO#8 part 5 for New Leaf [®]Y Potato Events RBMT15-101, SEMT15-02 and SEMT15-15 which has been genetically modified for Potato Virus Y and Colorado Potato Beetle resistant.

Monsanto Philippines, Inc. has provided data on the identity of Potato events RBMT15-101, SEMT15-02 and SEMT15-15, a detailed description of the transformation method, data and information on the gene insertion sites, copy number 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 and non-target organisms. Relevant scientific publications were supplied.

New Leaf [®]Y Potatoes events RBMT15-101, SEMT15-02 and SEMT15-15 have been evaluated according to BPI's safety assessment by concerned agencies (Bureau of Animal Industry (BAI), Bureau of Agriculture, Food 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 corn.

The Monsanto Company developed New Leaf [®]Y Potatoes. The genetically modified potato plants are resistant to infection by Potato Virus Y (PVY), causal agent of Rugose Mosaic disease, and to feeding by the CPB. New Leaf [®]Y Potatoes was produced by the introduction of (1) *cry3A*, the gene that produces a protein toxic only to target insects, isolated from the bacteria *Bacillus thuringiensis tenebrionis*, *Btt*; (2) *PVYcp*, the gene for the virus coat (capsid) protein, isolated from a naturally occurring strain of PVY; and (3) *nptII*, the marker gene for plant selection which codes for the Neomycin phosphotransferase (NPTII), isolated from the Tn5 transposon.

The petitioner/applicant published the Public Information Sheet (PIS) of the said application on two widely circulated newspapers (Malaya and Daily Tribune) on July 16, 2003 for public comment/review. BPI received no comment 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

The transgenic Shepody (SEMT15-02 and SEMT15-15) and Russet Burbank (RBMT15-101) potato lines were created through two separate *Agrobacterium*-mediated transformation events in which the transfer DNA (T-DNA) contained the *cry3A* gene encoding the Cry3A protein from *B. thuringiensis subsp. tenebrionis* and the *PVYcp* gene encoding CP from PVY-O.

1. the *cry3A* gene from *Bacillus thuringiensis* (Bt) which confers resistance to Colorado potato beetle (CPB);
2. the coat protein gene, *PVYcp*, from PVY which confers resistance to the virus; this was isolated from a naturally occurring strain of PVY; and
3. the *nptII* gene which confers resistance to kanamycin. The expression of NPTII activity was used as a selectable trait for screening transformed plants for the presence of the *Cry3A* and PVY-O CP genes. The *nptII* gene was isolated from the Tn5 transposon.

Colorado Potato Beetle

The novel lines SEMT15-02, SEMT15-15 and RBMT15-101 produce a version of the insecticidal protein, Cry3A, derived from *Bacillus thuringiensis*, as well as the coat protein (CP) from the ordinary (O) strain of potato virus Y (PVY-O). Delta-endotoxins, such as the Cry3A protein expressed in SEMT15-02, SEMT15-15 and RBMT15-101 potatoes, act by selectively binding to specific receptors localized on the brush border midgut epithelium of susceptible insect species. Following binding, cation-specific pores are formed that disrupt midgut ion flow and thereby cause paralysis and death. Cry3A is insecticidal only to Coleopteran insects and its specificity of action is directly attributable to the presence of specific receptors in the target insect. There are no receptors for delta-endotoxins of *B. thuringiensis* on the surface of mammalian intestinal cells, therefore, livestock animals and humans are not susceptible to these proteins.

Potato Virus Y Resistance

PVY is the type member of the potyvirus group and is an aphid-transmissible RNA virus that commonly infects potato, causing serious disease and economic loss. The introduced viral sequences in SEMT15-02, SEMT15-15 and RBMT15-101 do not result in the formation of any infectious particles, nor does their expression result in any disease pathology. The genetically modified potato cultivars exhibit the trait of resistance to infection and subsequent disease caused by PVY through a process that is related to viral cross-protection

Kanamycin Resistance

The kanamycin-resistance gene, isolated from the bacterium *E. coli*, codes for an enzyme (NPTII) phosphorylates kanamycin, rendering it inactive and imparting resistance to the antibiotic. The *nptII* gene was used as a selectable marker to facilitate the selection of transformed cells in the laboratory.

Safety of the Expressed Proteins

The Cry3A and NPTII proteins have been thoroughly tested for potential mammalian toxicity and shown to be non-toxic to humans and non-target organisms and non-allergenic to people through history of safe use as well as through numerous safety studies. Both Cry3A and NPTII proteins are rapidly digested in gastric fluids and do not contain amino acid sequences matching those of known allergens or toxins. *PVYcp* is assumed not to be an allergen since it is commonly consumed in the food supply with no documented ill effects. In conventional potatoes, PVY replicates in potato plant tissues; therefore humans and animals have routinely consumed PV virions and PVY capsid proteins in potatoes without deleterious health effects. There is a long history of using spores of *Bacillus thuringiensis*, which contain insecticidal proteins, in commercial agriculture globally; these products have been demonstrated safe for humans and the environment. NPTII protein is produced by microorganisms normally present in mammalian intestines.

IV. Nutritional Composition (Compositional Analysis)

The key constituents in potatoes appropriate for compositional analysis to establish substantial equivalence are total solids, sugars (reducing or dextrose and sucrose), protein and vitamin C are equivalent to parental variety; well within the expected range for commercially produced potato tubers and within the published ranges for white potatoes.

The nutritional equivalence of New Leaf[®] Y Potatoes to conventional potatoes was confirmed in feeding studies with rats which included clinical and histological evaluations.

V. Anti-Nutritional Factors

Anti-nutrients or toxicants called steroidal glycoalkaloids are found in most *Solanum* species. However in the New Leaf[®] Y Potatoes Events, level of glycoalkaloids is comparable to those found in non-transgenic potatoes thus no significant adverse results noted.

Potato tubers contain several protease inhibitors that inhibit the activity of trypsin, chymotrypsin and other proteases, thus decreasing the digestibility and the biological value of the ingested protein. Protease inhibitors in potatoes are largely inactivated by boiling and other thermal processes. Serious anti-nutritional reactions could occur, however, if raw or inadequately cooked potatoes are consumed or fed.

Lectins are (glyco)proteins which occur in virtually all living organisms and have the common property of binding to specific carbohydrate structures on cell surfaces, e.g. on intestinal or blood cells. Some lectins found in beans are known to cause serious health effects when ingested by humans and animals. As lectins are inactivated during heating, only consumption and feeding of raw or inadequately cooked potatoes may cause adverse effects. Negative effects of lectins on animal's health and their performance are not yet known in detail.

VI Regulatory Decision

After reviewing the scientific data and information relevant to the application of Monsanto Philippines Inc., it is concluded that New Leaf[®] Y Potatoes and all progenies derived from crosses of the product with any conventionally-bred potato, and potato 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.