

**CONSOLIDATED REPORT OF MONSANTO'S SOYBEAN MON89788
APPLICATION FOR DIRECT USE AS FOOD AND FEED, OR FOR PROCESSING**

EXECUTIVE SUMMARY

On May 25, 2017, Monsanto Philippines Inc.'s submitted soybean MON89788 for direct use as food and feed, or for processing to the Bureau of Plant Industry (BPI) under the DOST-DA-DENR-DOH-DILG Joint Department Circular (JDC) No. 1 Series of 2016. After reviewing the Risk Assessment Report and attachments submitted by the applicant, the assessors namely: Scientific and Technical Review Panel (STRP), BPI- Plant Products Safety Services Division (BPI-PPSSD) and Bureau of Animal Industry (BAI), concurred that soybean MON89788 is as safe for human food and animal feed as its conventional counterpart.

The Department of Environment and Natural Resources – Biosafety Committee (DENR-BC), after a thorough scientific review and evaluation of the documents related to Environmental Risk along with the submitted sworn statement and accountability of the proponent, recommended the issuance of a biosafety permit for this regulated event provided that the conditions set by them are complied.

Also, the Department of Health – Biosafety Committee (DOH-BC), after a thorough scientific review and evaluation of documents related to Environmental Health Impact, concluded that soybean MON89788 will not pose any significant risk to health and environment and that any hazards could be managed by the measures set by the department. DOH-BC also recommended for the issuance of biosafety permit for soybean MON89788.

Furthermore, the Socio-economic, Ethical and Cultural (SEC) Considerations expert also recommended for the issuance of biosafety permit for this regulated article after assessing the socio-economic, social and ethical indicators for the adoption of Genetically Modified Organisms.

BACKGROUND

In accordance with Article VII. Section 20 of the JDC, no regulated article, whether imported or developed domestically, shall be permitted for direct use as food and feed, or for processing, unless: (1) the Biosafety Permit for Direct Use has been issued by the BPI; (2) in the case of imported regulated article, the regulated article has been authorized for commercial distribution as food and feed in the country of origin; and (3) regardless of the intended use, the regulated article does not pose greater risks to biodiversity, human and animal health than its conventional counterpart.

The BPI Biotech Office provided the assessors the complete dossier submitted by Monsanto Philippines, Inc.

Upon receipt of the individual reports from the assessors, the BPI Biotech Office prepared this consolidated risk assessment report for the information of the public.

A. STRP, PPSSD, BAI ASSESSMENT

After thorough review of the technical documents submitted by the applicant, the assessors' findings are as follows:

1. Host Organism (Soybean)

Soybean is used as both food and feed. It is processed into a wide variety of food products including soy flour, soy protein, tofu, soy milk, soy sauce, and soybean oil. On the other hand, soybean is the premier supplemental protein source in livestock and poultry ratios due to its availability and nutrient composition.

While it is considered a good source of key nutrients (carbohydrates, fiber, amino acids, fatty acids, minerals and vitamins), a number of anti-nutritional factors including trypsin inhibitor, lectins, phytic acid, phytoestrogen, tannins, goitrogens, and raffinose can also be found in raw soybeans. These components are, however, labile to heat, and are therefore destroyed during cooking and processing.

Soybean is also known as a common allergen. However, this soy allergy is usually outgrown by the age of three years as the immune system matures.

2. Transgenic Plant

Molecular characterization of the inserted DNA confirmed that an intact *cp4 epsps* cassette was integrated at a single site within the soybean genome. Further, biochemical characterization of the expressed CP4 EPSPS in MON89788 demonstrated that the protein is equivalent to the CP4 EPSPS produced from other Roundup Ready crops that already have a long history of safe use. More importantly, assessment of the composition and nutritional profiles of MON89788 has shown that grain and forage are equivalent to and are as safe as those from conventional soybeans.

3. Donor Organisms

The donor organism, *Agrobacterium* sp. strain CP4, is not a known human or animal pathogen and is not known to induce allergenic responses in humans.

To generate MON89788, the developer used plasmid vector PV-GMGOX20. PV-GMGOX20 is approximately 9.7 kb and contains a *cp4 epsps* gene expression cassette delineated by left and right border regions. The T-DNA that is incorporated into the soybean genome is approximately 4.3 kb, and the DNA backbone region that is not incorporated into the soybean genome is approximately 5.4 kb.

No additional elements from the transformation vector PV-GMGOX20, linked or unlinked to the intact *cp4 epsps* gene cassette, were detected in the genome of MON89788.

The description of other regulatory sequences for the transformation event was adequately provided in Table IV-1 of the Monsanto Petition to USDA.

4. Transformation System

Soybean MON89788 was produced by *Agrobacterium*-mediated transformation of soybean meristem tissues using the binary vector PV-GMGOX20. The target of the genetic modification is the stable integration of the *cp4 epsps* cassette into the nuclear DNA of the recipient soybean line.

5. The Inserted DNA

Result of the Southern Blot analysis showed that only one copy of the *cp4 epsps* expression cassette was inserted at one locus of the soybean genome. No other genetic elements or backbone sequences of PV -GMGOX20 was detected. Moreover, segregation analyses show heritability and stability of the *cp4 epsps* gene across multiple generations, which corroborate the molecular insert stability analysis.

The organization of the genetic elements within the insert of MON89788 was confirmed by DNA sequence analyses. Results of the PCR confirm that the arrangement of the genetic elements is identical to that in plasmid PV-GMGOX20.

Other than soybean, the *cp4 epsps* gene has also been expressed in several GM crops such as *Gossypium hirtum* L. (cotton), *Beta vulgaris* L. (sugarbeet), *Brassica napus* L. (oilseed rape)-Canola, *Brassica rapa* L. (turnip rape) – Canola, and *Zea mays* L. (maize)

6. Genetic Stability

Southern blot analysis demonstrated the stability of the DNA insert across four generations. Additionally, these generations have been shown not to contain any detectable backbone sequence from plasmid PV-GMGOX20.

Segregation is assessed by Chi-square (χ^2) analysis over 3 generations. Results confirmed that the expected and the observed segregation ratios were identical. This result is consistent with the finding of a single chromosomal insertion of the *cp4 epsps* gene cassette that segregates according to Mendel's laws of genetics.

7. Expressed Material

The CP4 EPSPS protein in the Roundup Ready plant acts on the shikimic acid pathway such that it is able to synthesize aromatic amino acids even in the presence of glyphosate, an ingredient in herbicides.

Using a validated enzyme-linked immunosorbent assay (ELISA), CP4 EPSPS protein levels in over-season leaf (OSL), grain, root, and forage were determined in tissues collected from MON89788 produced in replicated field trials.

The mean CP4 EPSPS protein levels across sites for over-season leaf OSL1, OSL2, OSL3, OSL4, grain, root, and forage were 300, 340, 330, 290, 150, 74, and 220 μ g/g DW, respectively.

8. Toxicological Assessment

Digestibility. Digestibility of CP4 EPSPS was tested with Simulated Gastric Fluid (SGF) using pepsin enzyme. At least 98% of the CP4 EPSPS protein was digested in SGF within 15 seconds. Therefore it cannot be absorbed in the intestines and is unlikely to pose a health concern.

Heat Inactivation. Western blot analysis was used to measure the decrease in immunoreactivity. Results showed that the amount of immune-detectable CP4 EPSPS protein present in MON 89877 was below the limit of detection (LOD) after heat treatment. This represents greater than 97% reduction in the

quantity of detectable CP4 EPSPS compared to the unheated MON89788 samples.

Therefore, processing of ground soybean grain by heat as practiced in commercial facilities reduces the amount of CP4 EPSPS, and should not be a health concern.

Amino Acid Comparison. Using the database TOXIN5 with FASTA sequence alignment tool, the most significant alignment was found to Bacillus cereus sphingomyelinase c precursor protein (Accession No. P11889) with 28.2% amino acid sequence identity over a 131 amino acid overlap and E score of 0.26. However, the % amino acid sequence identity and the E-value are very low to be meaningful.

Acute Oral Gavage. An acute oral toxicity assessment was performed by gavage on 3 groups of 10 mice with a dose of 572 mg/kg BW. No adverse effect was observed in terms of body weight gain, food consumption and gross pathology. It was concluded that NOAEL was 572 mg/kg BW.

Source of Test Protein. E-coli produced CP4 EPSPS was used as the test protein, which has been shown to be equivalent to CP4 EPSPS in MON89788.

9. Allergenicity Assessment

Amino Acid Comparison. Based on the dossier provided by the developer, bioinformatics tool using FASTA alignment tool and an eight-amino acid sliding window search indicated that the CP4 EPSPS protein found in MON89788 has no amino acid sequence similarity to known allergens, gladadins or glutenins.

Prevalence in Food. The amount of the EP4 EPSPS protein in grain is about 0.037% of the total protein in grain.

Serum Screening. The assessment of Human IgE Biding to soybean MON 8977 was conducted to determine the binding levels of IgE antibody from soy allergic patients to protein extracts prepared from seeds of soybean MON89788, a non-transgenic control A3244, and 24 commercial soybean lines. The results showed that the levels of endogenous soybean allergens in MON89788 and non-transgenic control lines are comparable to each other and to the level in commercial varieties.

10. Nutritional Data

Compositional analyses were conducted on the grain and forage tissues of MON89788 and A3244 grown in three replicated plots at each of five field sites across the U.S. during 2005. In addition, 12 conventional soybean varieties were also included as references where three varieties were grown at each of two sites and two varieties were grown at each of three sites for a total of 12 references.

Statistical differences were observed for forage moisture, and grain daidzein, glycitein and Vitamin E. However, the differences from mean levels of MON89788 were generally small and within the ranges in conventional reference values. Thus, the reported statistical differences are unlikely to be biologically relevant.

The assessors found MON89788 to be compositionally equivalent, and as safe as conventional soybeans.

B. DENR BC (for Safety of Event to the Environment)

The assessment of the DENR BC focused on the potential environmental impact of Soybean MON89788.

The project description report (PDR) discussed the specified environmental management plan indicating the possible risk and harm to the environment and non-target organisms as well as the mitigating measures and contingency plan of the proponent. Upon evaluation of the submitted PDR and environmental risk assessment (ERA), the Committee noted that the chances of unintended release or planting of the regulated article is very minimal and will not cause any damaging and lasting effects because the receiving environment (areas near the port, roads, railways, etc.) is not conducive for plant growth/germination.

Based on the following observations, the Committee concluded that the regulated article is safe as its conventional counterpart, and its direct use whether for food, feed and/or processing will not cause any significant adverse effect on the environment (land, air, and water) and non-target organisms:

1. Genetic stability in the transgenic crop is ensured such that no unintended horizontal gene transfer shall occur to unrelated species.
2. The protein product produced by the transgenic crop will degrade upon exposure to the natural environment and general conditions (i.e. high temperatures (60 degrees Celsius and above), varying pH, enzyme digestion, etc.); and
3. The protein products will not increase the weediness potential of the transgenic crop.

The DENR BC recommended for the approval and issuance of a biosafety permit for Direct Use as FFP of Soybean MON89788 given the condition:

The importer shall ensure the proper and secure packaging of the regulated article for transport and the safety and durability of the transport vehicle, for prevention of any possible spillage or unintended release during transport/import. The BPI shall conduct inspection of the shipment at the port of entry.

C. DOH-BC (for Environmental Health Safety)

The DOH-BC assessed the potential environmental health impact of soybean MON89788 using the technical dossier provided by the applicant. After thorough review and evaluation of these documents, the following are the observations of the DOH-BC:

1. Specifically, soybean MON89788 does not require changes in the usual practices in unloading and loading, hauling, transport and storage, and processing; and
2. On the Risk to Health Matrix, the proponent/applicant rated the phases/stages of activities a Very Low Incident/Exposure Potential Rating.

Given these findings, the Committee concluded that the GM product is as safe as its conventional counterpart, and shall not pose any significant risk to human and animal health, and the environment.

The DOH BC recommended the approval of the regulated article with the following conditions:

1. Strict monitoring of the regulated article from port of entry to the trader's/ importers storage/warehouse as stated in Section 32 of the JDC No. 1 series, 2016; and
2. The BPI to include in the issuance of permit for the release of this product the following conditions:
 - a) Any spillage (during unloading and loading/hauling and transport unloading and storage) shall be collected and cleaned up immediately;
 - b) Transportation of the consignment from the port of entry to any destination within the country shall be in closed containers; and
 - c) There shall be clear instructions stating that the product is only for the purpose of direct use as food, feed or for processing, and is not to be used as planting material.

D. SEC Expert (for Socio-economic Consideration)

With regard to the significance of the soybean MON89788 in terms of production, consumption and trade, the SEC expert found the applicant's answers acceptable. Specifically, the applicant emphasized that the intended use of the regulated article was solely for importation. Similarly, approval of soybean MON89788 will help maintain global trade of soybean products that are imported into the Philippines.