

ASSESORS' CONSOLIDATED REPORT ON SIMPLOT PLANT SCIENCE INTERNATIONAL INC.'S LOWERED FREE ASPARAGINE , REDUCED BLACK SPOT AND LOWERED REDUCING SUGARS POTATO E12 APPLICATION FOR DIRECT USE AS FOOD AND FEED, OR FOR PROCESSING

EXECUTIVE SUMMARY

On February 01, 2019 Simplot Plant Science International Inc.'s filed for application of potato E12 direct use as food and feed, or for processing, as original application 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- Biotech Team (BAI-BT), concurred that potato E12 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 the conditions set by DENR 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 potato E12 will not pose any significant risk to the 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 the transformation event.

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 Simplot Plant Science Philippines, Inc. Upon receipt of the individual reports from the assessors, the BPI Biotech Secretariat prepared this consolidated risk assessment report for the information of the public.

STRP, BPI-PPSSD, BAI (Safety Assessment)

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

1. **Host Organism** (*Solanum tuberosum*)

The assessors has reported that potatoes are conventional sources of key nutrients and are also reported to contain ascorbic acid, potassium, carotenoids and antioxidant phenols such as chlorogenic acid. Potato tubers contains trypsin, chymotrypsin, other protease inhibitors as well as lectins (OECD, 2002).

Glycoalkaloids are the reported toxicants contained by potato tubers and are found in its periphery (peeling) and internal part. Other potential toxicants include calystegines; multiple heat-labile proteins that can induce hypersensitivity reactions; patatin which induces allergic reactions; and IgE binding component that neutralize the effects of several proteins (OECD, 2002). Processing of the tubers such washing, peeling, cutting, rinsing and heat treatment usually result in its removal.

Potato is being used as food and according to OECD Consensus Document (2002), cooking is done prior to consumption of potatoes due to the indigestibility of non-gelatinized starch and the presence of anti-nutritional proteins. In the Philippines, unprocessed potatoes consumption is about 1 kg/capita/year (AIS, 2017), and is not a common source of feed. A small fraction of raw potatoes is used as feed for ruminants while cooked potato leftovers from processors and restaurants in some areas are fed to swine.

2. **Transgenic Plant**

The assessors has concurred that potato E12 has been approved for cultivation in the United States and Canada, and as food in several countries and like other potato varieties, potato E12 do not necessarily change the consumption patterns in over all populations or any population or sub-group (Horton et al., 1992). The consumption pattern in the overall population or any population sub-groups will not be changed, as E12 will be used in the same ways as the other conventional chipping potatoes.

3. **Donor Organism**

Solanum tuberosum, and *S. verrucosum* are the donor organisms for the genetic elements in the pSIM1278 plasmid and potatoes have a history of safe use for food consumption by humans

The assessors has verified that the developer provided sufficient information on all regulatory sequences including non-coding spacer elements and promoter sequences used in the transformation of potato E12 were reported.

The transgenic potato E12 contains the insert from pSIM1278 that reduces and down regulates expression of asparagine synthetase (*Asn1*), polyphenol oxidase (*Ppo5*), phosphorylase L (*PhL*) and water dikinase (*R1*) potato enzymes. Asparagine synthetase (*ASN1*) decreases free asparagine levels in potato tubers and causes reduction of acrylamide potential. Polyphenol oxidase (*Ppo*) decreases PPO levels in tubers and reduces black spots. Phosphorylase L (*PhL*) lowers reducing sugars in tubers which contributes to the reduction of acrylamide potential. Lastly, water dikinase (*R1*) also lowers reducing sugars in tubers which contributes to the reduction of acrylamide potential.

4. Transformation System

The introduction method for development of E12 employed *Agrobacterium tumefaciens*-mediated plant transformation using pSIM1278 in which the potato internode segments were transformed with C58-derived *Agrobacterium* strain AGL-1 containing pSIM1278. All the genetic components used were presented, which includes both coding and non-coding regions. The plasmid vector pSIM1278 was described aptly.

No protein-encoding sequences were introduced therefore, no novel proteins were produced. Based on the data presented, potato E12 does not produce any novel protein.

5. Inserted DNA Genetic Stability

The assessors has confirmed that the southern blots have demonstrated that a single genomic locus where the pSIM1278 T-DNA was integrated. The presence of a single band indicates a single insertion site at a single locus. The location of the restriction site is validated and confirmed by sequencing the regions of the junctions, PCR amplification and Southern blot analysis of the insert.

Based on the Southern Blot Analysis, deletion of 24 bp was observed in the LB and 119 bp in the RB. The assessors has verified that the deletions observed in the E12 insert do not have the potential for creating novel chimeric ORFs.

Furthermore, the assessors agree with the evidence provided by the developer provided sufficient information that elements of the pSIM1278 DNA inserts are specific to potato and have not been used in approved crops other than potato.

6. Genetic Stability

The assessors has reported that the Southern blot analysis on G0 and G3 plants showed consistent banding pattern demonstrating the that the integrity of the insert was maintained across vegetative propagation. Cultivated potato plants are cultivated genetically and phenotypically stable even after several propagations. Furthermore, vegetative propagation does not alter processes responsible for genetic variation, meiosis, recombination and segregation.

7. Expressed Material

The assessors has stated that potato E12 does not produce a novel protein. The T-DNA is designed to down-regulate the *Asn*, *Ppo*, *PhL*, and *R1* genes in tubers leading to reduced expression of the *ASN*, *PPO*, *PhL*, and *R1* proteins. Northern blot analysis showed reduced expression of *Asn* and *Ppo* in the tuber and *Asn* in the flower. Expression of *Asn*, *Ppo*, *PhL*, and *R1* genes in the leaves, stems and roots were unchanged.

8. Toxicological and Allergenicity Assessment

The assessors have reported that toxicological and allergenicity assessment was conducted by identifying thirty unique open reading frames (ORFs) and conducting a bioinformatics analysis. Identification of the open reading frames (ORFs) in E12 and conduct of bioinformatic analysis are applied to assess allergenicity of potato E12. A 30 unique start-to-stop ORFs were identified within the E12 insert and associated with the junction regions surrounding the insert. The bioinformatic analysis involved examination on the homology between ORFs and known putative allergens and demonstrated that there are no ORFs which showed homology with known toxins and allergens. Additionally, glycoalkaloid content was similar to control Russet Burbank potatoes.

9. Nutritional Data

According to the assessors, there were no differences observed for any proximates analyzed with the mean values of the proximates in potato E12 were all within the tolerance interval and combined literature range. Comparison with nine potato varieties namely Atlantic, Chieftain, IdaRose, Ranger Russet, Red Norland, Russet Burbank, Snowden and 2 propriety varieties were performed. These varieties were grown under the same environmental conditions as potato E12 and upon comparison, it was demonstrated that the data derived from potato E12 proximate analysis were within the tolerance interval or range for commercial varieties.

No differences were observed for vitamins and minerals between E12 and Russet Burbank. Some amino acid levels differed between E12 and Russet Burbank however, the mean and range values for all amino acids fell within the combined literature range, indicating that E12 was comparable to conventional potato varieties. Any nutrient like Vit B3, Vit B6, Vit C, copper, magnesium and potassium in E12 potato are comparable with reference non-transgenic varieties reported in literature. Differences in the amounts of amino acid contents of the potato E12 compared to the reference potato Russet Burbank do not necessarily imply a biologically relevant activity in the transgenic E12 potato.

The assessors reported that this implies that genetic transformation does not alter the nutrient components of the transformation event E12 and it is of substantial

equivalence to the reference non-transgenic commercial varieties. Comparable levels of key nutrients in E12 and in conventional non-transgenic potatoes explain the substantial equivalence of the transgenic E12 potato in terms of its safe and nutritious character as food as with that of the reference non-transgenic potato.

Furthermore, no significant difference in glycoalkaloid content, although slightly lower content in E12. All mean glycoalkaloid values for the E12 event fell within the tolerance interval or combined literature range. Processing, involving washing, peeling, cutting, rinsing and heat treatment usually result in removal of the outer skin where there is higher concentration of alkaloids. Sprouted and green potatoes have as much alkaloid in the tuber tissues and are not usually consumed.

10. Recommendation

Upon review of the provided materials of SPS International, Inc. and other literatures, the BPI-PPSSD, BAI and STRPs find scientific evidence that the regulated article applied for direct use as food and feed or for processing is as safe as its conventional counterpart and shall not pose greater risk to human and animal health. Weight of evidences approach indicate that Potato E12 is as safe as its conventional counterpart with regards to substantial equivalence and food safety. History of safe use is attributed on the host organism (*Solanum tuberosum*) and donor organisms (*Solanum tuberosum* and *S. verrucosum*) which are not known to be toxic or allergenic to humans and animals. The BPI-PPSSD, BAI and STRPs recommend for the approval of Potato E12's application for biosafety permit for direct use as food and feed or for processing.

DENR Biosafety Committee (Environmental Safety)

The DENR-BC has reported that the direct use of the regulated article whether or food, feed or for processing will not cause any significant diverse effect on the environment and biodiversity. The inserted genes, inverted repeats derived from various *Solanum* species (*S. tuberosum*, and *S. verrucosum*), for dsRNAs that dupresses the expression of the targeted genes (Ppo, Asn, Phl, and R1) via RNAi pathway. The inserted genes are dsRNAs, and like other forms of RNA, has no known cases of being an allergen or toxin (FAO-WHO, 1991);

Based on the reproductive biology of potato, terminal pores of anthers must be vibrated (buzz pollination) to release the pollen grains (Plaisted, 1980). Bumblebees exhibit buzz pollination but prefer to target fertile pollen. Pollen and ovule sterility occur frequently in *S. tuberosum*, thus the spread of pollen grains via insect pollination is also reduced (Vasil, 1964). The role of wind in pollination is also minor, thus the spread of pollen grains via wind pollination is unlikely (White, 1983); and

The project description report (PDR) stated that "Potato products, including chips, are not viable for purposes of cultivation. Mitigating measures for unintended release or unauthorized planting are therefore not needed." Thus, the chances of unintended release or planting are therefore not needed." Thus, the chances of unintended release or planting of the regulated article is minimal and will not cause any damaging effects.

Also, Solanum sp. Rarely exist as a wild plant and are cultivated in areas with adequate rainfall or irrigation due to its sensitivity to drought stress (Canadian Food Inspection Agency, 2015).

After a comprehensive review and evaluation of the documents including the scientific evidence from references and literature submitted by Simplot Plant Science International Inc. its application for Direct Use as food and feed or for processing of potato E12, the DENR-BC considered the regulated article safe to the environment particularly on biodiversity , and hereby recommends for the approval of application of SPS International, Inc. for Biosafety Permit for direct use as food, feed or for processing of Potato E12.

DOH Biosafety Committee (Environmental Health Safety)

After a thorough review and evaluation of the documents provided by the proponent, SPS International, Inc., through the Bureau of Plant Industry (BPI), in support of their application for approval for Direct Use as Food, Feed or for Processing of Potato E12, the DOH Biosafety Committee find that the regulated article applied for Direct Use as Food, Feed or for Processing (FFP) is safe as its conventional counterpart and shall not pose any significant risk to human and animal health.

The DOH-BC has reported that scientific pieces of evidences from Toxicity studies and references, find that the regulated article will not cause significant adverse health effects to human and animal health. Dietary exposure to the regulated article of unlikely to result in allergic reactions. The regulated article is not materially different from nutritional composition from that of the non-transgenic potato or the conventional potato.

The DOH-BC is suggested that the Bureau of Plant Industry (BPI) ensure that there shall be clear instructions that the product is only for the purpose of direct use for food and fed or for processing and is not t be used as planting materials.

Based on the above considerations and with the submitted sworn statement of the proponent, we hereby submit our evaluation to BPI relative to the application of a Biosafety Permit for Direct Use as food, feed or for processing (FFP) of Potato E12 and recommends its approval.

SEC Expert (Socio-Economic Considerations)

According to the SEC Expert, the approval to import potato E12 may not affect our domestic production since food processing and institutional food chain were already importing white potatoes. In addition, since the event will not be propagated domestically, there will be no significant effects to the domestic production and trade of white potatoes.

The entry of potato E12 is not expected to affect current consumption. Empirical studies show that food choice of consumers are influenced by a large number of factors. It is

expected that potato E12 will have to have little effect to the patterns of Philippine trade because of its insignificant share in the overall trade of the country.

After a thorough and scientific review and evaluation of the documents provided by SPS international Inc., relevant to Potato E12, the SEC Expert recommends the approval and issuance of biosafety permit for direct use as food and feed or for processing.