ASSESSORS' CONSOLIDATED REPORT ON SYNGENTA PHILIPPINES INC.'s APPLICATION FOR DIRECT USE AS FOOD AND FEED, OR FOR PROCESSING OF COTTON COT102

EXECUTIVE SUMMARY

On December 13, 2019, Syngenta Philippines Inc. submitted cotton COT102 for 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 cotton COT102 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 cotton COT102 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 cotton COT102.

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, except for the SEC expert, the complete dossier submitted by Syngeta Philippines Inc. The SEC expert, on the other hand, was provided with a questionnaire on socio-economic, ethical and cultural considerations that have been addressed by Syngenta Philippines Inc. in relation to their application.

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

STRP ASSESSMENT AND RECOMMENDATION

Based on the documents submitted by the applicant:

A. Host Organism

Cottonseed oil is used as human food. Linters are used as fiber source for humans. The meal by-product is used as animal feed (for ruminants, monogastrics, fishes) to supply additional crude protein. [1] Certain fatty acid forms like sterculic, malvalic and dihydrosterculic acids are found in cotton. These are classified as anti-nutrients.[3] Gossypol is a toxic substance in cottonseed and its by-product meal. Processing of cottonseed detoxifies the gossypol, from free form to bound form. The presence of gossypol limits its use in whole and unprocessed cottonseed as feed materials.[6]

The Codex Alimentarius Commission does not list cotton among foods that require special hypersensitivity labeling. Cotton by-products used as food materials are subjected to processing, thus potential allergens are eliminated. [8]

Cottonseed oil ranks third behind soybean and corn oil for human consumption and currently comprises 5-6% of the total U.S. domestic fat and oil supply. About 56, 36 and 8 % of the oil is used for salad/cooking oil, baking/frying fats, and margarine/other uses, respectively. Cottonseed oil is one of the most unsaturated oils, ranking with canola, corn, soybean, safflower and sunflower seed oils. Its mild, nut like taste makes it highly desirable for use as a salad oil.[2][14] Processed cottonseed meal is also an excellent source of protein for ruminant animals, provided that free gossypol content is kept low and supplementation with lysine is conducted. Also, recent research indicates it can also be used in non-ruminant feed, at a level < 50% of the total protein. [2][15]

B. Transgenic Plant

COT102 Cotton has already been approved for use as food in the United States, Argentina, Australia, Brazil, Canada, China, Colombia, Japan, Korea, Mexico, Philippines and Taiwan. It also has has feed approval in the United States, Argentina, Canada, China, Colombia, Japan, Korea, Mexico, Philippines and Taiwan.

It is not materially different in composition, safety and other relevant parameters from the conventional corn in the market and it is not expected that consumption pattern will change with the introduction of COT102 cotton in the market. The amount of cotton consumed as food is not likely to change as it is today. Although, in general, increase in consumption may be expected due to consistent increase in human population.[16]

C. Donor Organism

The VIP3AA19 protein expressed in COT102 cotton is produced from the modified version of the native vip3Aa1 gene.The gene was cloned from Bt strain AB88. No incidences of oral allergies were reported, and the US EPA has stated that the laboratory animal studies have not shown any potential for allergic reactions to Bt or its components. Thus, VIP3AA19 is not derived from a source known to produce allergenic proteins. [17]

On the other hand, the E. coli strain from where aph4 gene was derived is not included in the list of pathogenic E. coli. And such, has no history of allergenicity. [17] Both E. coli and B. thuringiensis are not known to be toxic. [17]

D. Transformation System

Agrobacterium-mediated transformation method was used. Briefly, Agrobacterium cells, harboring the transformation vector pCOT1, were incubated with cotton (Gossypium hirsutum cv. Coker 312) hypocotyl tissue. The insecticidal vip3Aa19 gene and selectable marker aph4 were delivered as a single T-DNA insert from the pCOT1 plasmid. Later, the incubated tissue was plated onto synthetic culture medium containing antibiotics, including hygromycin B. Plants were regenerated and subsequently analyzed for the presence of Vip3Aa19 protein. [18] Nuclear DNA is the target of genetic modification.[20]

E. Inserted DNA

There is one insertion site that contains 1 copy of vip3Aa19, 1 copy of aph4, 1 Act2 promoter, 1 Ubq3 promoter and 2 copies of NOS terminator. Southern blot analyses were performed using specific probes for each gene. Lanes with added AscI and XmaI restriction enzymes gave positive responses.[20]

Some truncations occurred in the right (24 bp)- and left (19bp)-border regions of the T-DNA. Further, 82 bp of the cotton genomic sequence were deleted upon COT102 T-DNA insertion and 690 bp of cotton genomic sequence is present adjacent to the COT102 insert that is not present in non-transgenic cotton. However, this did not affect the functionality of the T-DNA insert. [20]

F. Genetic Stability

To assess the genetic stability over generations in event COT102 cotton, determination for vip3Aa19 was repeated for three generations (F1, BC1F1and BC4F1) of event COT102 cotton. These were used for Southern blot analyses. These two generations showed the same hybridization patterns for vip3Aa19 gene. The results demonstrated the stability of the insert in COT102 cotton during conventional breeding. Each generation established does prove genetic stability for the transgenic locus of transformation in event COT102 cotton.[22]

Moreover, genotypic data were analyzed to test the hypothesis that the two genes in COT102 cotton are segregating in a Mendelian fashion. The results showed that the genetic traits are behaving in a Mendelian fashion. All F1 plants were positive for the genetic traits, while in BC1F1 and BC4F1, the traits segregated in a 1:1 ratio. [22]

G. Expressed Material

For most tissues and sampling stages, VIP3A concentrations were generally comparable across all locations (where samples were available from more than one location). Leaves had higher VIP3A levels than other individual tissues at all sampling stages (mean values measured ranged from ca. 3 – 22 µg VIP3A/g fresh wt. and ca. 5 – 118 µg VIP3A/g dry wt.), and levels generally declined at the later sampling times. The VIP3A levels in whole-plant samples typically paralleled those in leaves, although they tended to be somewhat lower across all sampling stages (means ranged from ca. 1 – 13 µg VIP3A/g fresh wt. and ca. 1 – 73 µg VIP3A/g dry wt). VIP3A concentrations measured in young leaves (ca. 1 – 19 µg VIP3A/g fresh wt.; ca. 5 – 66 µg VIP3A/g

dry wt.; Table 7) were generally comparable, across sampling stages, to VIP3A concentrations in leaf samples that represented a composite of both young and older leaves. Mean VIP3A protein levels measured in roots were uniformly low (ca. < 0.2 – 2 µg VIP3A/g fresh wt.; ca. < 0.4 – 7 µg VIP3A/g dry wt.) at all times sampled. Mean VIP3A levels in bolls were also low and declined from ca. 1 µg VIP3A/g fresh wt. (ca. 7 - 9 µg VIP3A/g dry wt.) at peak bloom to levels that were generally undetectable or too low to quantify at the pre-harvest stage. In squares, mean VIP3A concentrations at all stages sampled ranged from ca. 2 – 4 µg VIP3A/g fresh wt. (ca. 4 – 17 µg VIP3A/g dry wt.), except for the Georgia location at pre-harvest stage, for which the VIP3A levels were below the LOQ. Mean VIP3A levels in seeds were ca. 3 µg VIP3A/g fresh or dry wt. [23]

H. Toxicological Assessment

The simulated mammalian gastric fluid (SGF) containing pepsin was used to evaluate the susceptibility of vip3Aa19 protein to proteolytic degradation, using Western blot analysis and Na-dodecylsulfate polyacrylamide electrophoresis. Vip3Aa19 was readily digested in SGF. No intact vip3Aa19 was detected after incubation in SGF for 1 min. These results show that vip3Aa19 protein will likely be digested as conventional dietary protein in the typical mammalian gastric conditions.[24]

The stability of vip3Aa19 at different temperatures was also determined using solutions of VIP3A test substance. After incubation at various temperatures the solutions with test substance were analyzed for bioactivity using the Spodoptera frugiperda (fall armyworm) larvae bioassay, and for immunoreactivity by ELISA. Incubation at 25C and 37C for 30mins had little effect on Vip3Aa19 bioactivity. At 65C and 95C, bioactivity was reduced such that the LC50 and 95% CI could not be estimated due to low larval mortality. ELISA analyses showed little effect of low temperature on immunoreactivity; higher temperatures resulted in higher loss of immunoreactivity, while at 150C and 170C immunoreactivity was completely lost. [24]

Using BLASTP program, the vip3Aa19 protein was compared to sequences in two databases (NCBI) and Syngenta Toxin Database. The comparison would determine its significant similarity to known and putative toxins. Results confirm that vip3Aa19 is not a toxic protein to humans or other mammals, nor does vip3Aa19 share significant sequence similarity with other known or putative protein toxins.[24]

Moreover, acute oral gavage was then done on the microbially-produced VIP3Aa19 test material using male and female mice in a single dose at a level of 5,000mg/kg. This dose has ca. 3,675mg VIP3Aa19 protein/kg. No apparent toxicity was observed in mice, other than a decrease in body weight gain for males on Day 8. No test material-related macro or microscopic findings on sacrificed animals. Terminal body and organ weights did not change in animals receiving 5,000mg/kg of VIP3Aa19 protein.[24]

On the other hand, SGF containing pepsin was also used to evaluate the susceptibility of hygromycin B phosphotransferase to proteolytic degradation, using Na-dodecylsulfate polyacrylamide electrophoresis and Western blot analysis. APH4 from recombinant E. coli was readily digested as conventional dietary protein in the typical mammalian gastric environment. No intact APH4 (MW ca 42 kDa) was detected after incubation in SGF for 1 min.[25]

Temperature stability of APH4 was then evaluated by assessment of immunoreactivity by enzyme-linked immunosorbent assay (ELISA) on incubated solutions of test substance APH4-0102 at 25–200C. At 25-95C for 30mins, no effect on immunoreactivity was detected. Incubation

at 120-150C resulted in substantial decrease in ELISA response with 61-70% loss. At 170-200C for 30 mins resulted in 100% loss of immunoreactivity. Thus, it is concluded that APH4 is unstable upon heating at 120C and above.[25]

A comparison of APH4 protein to sequences in two databases was performed using the Basic Local Alignment Search Tool (BLASTP) program. Results from both database comparisons confirm that APH4 is not a toxic protein, and an acute oral gavage was done using a mouse oral toxicity study. A single gavage dose of APH-0102 test material at 1,828mg/kg BW was administered to male and female mouse. This amounted to 779mg APH4 protein/kg BW. No mortalities occurred during the study. No clinical signs of toxicity were observed in both test and control groups. The estimated LD50 value for pure APH4 protein in male and female mice is >779mg/kg BW.[25]

I. Allergenicity Assessment

A full-length sequence search using FASTA indicated NO sequence similarity greater than 35% shared identity over 80 or more amino acids was observed between the vip3Aa19 amino acid sequence and any entry in the Comprehensive Protein Allergen Resource (COMPARE) database. The results from the exact match search of 8 or more contiguous amino acids showed no alignments between the vip3Aa19 amino acid sequence and sequences in the COMPARE database. Therefore, the results show that vip3Aa19 shares no biologically relevant amino acid sequence similarity to known or putative protein allergens.[24]

Further, the vip3Aa19 protein would be readily digested as the conventional dietary proteins. The protein is unstable to moderate heating and processing; thus, it can be substantially degraded by standard cottonseed processing methods.The mass spectral analysis showed no evidence of any post-translational glycosylation of VIP3A protein. The apparent MW of microbially-produced and plant produced vip3Aa19 as determined by Western blot analysis is consistent with the theoretical weight of 89kDa.[24][26]

Meanwhile, a comparison of APH4 protein to sequences in two databases was performed using the Basic Local Alignment Search Tool (BLASTP) program. Results from both database comparisons confirm that APH4 is not a toxic protein, and that it shares no significant and biologically relevant amino acid sequence similarity with known or putative protein allergens. APH4 was also compared with each of the known or putative allergen sequences using FASTA search and COMPARE database. Results showed no alignments of 8 or more contiguous amino acids between APH4 amino acid sequence (FASTA search) and any entry in the COMPARE database. Thus, APH4 shares no biologically relevant sequence similarity to known or putative allergens. [25]

The APH4 protein is also readily digestible as the conventional protein. It is unstable upon heating at 120C and above as evidenced by loss of immunoreactivity. No glycosylation sequence for either N- or O-linked glycosylation were also identified for APH4 protein in cotton. The MW of APH4 is 42 kDa.[25]

Lastly, According to OECD, the protein content of cottonseed ranges from 21.8 to 34.2%, dry basis. Based on data provided by the applicant, the aph4 content of cottonseed <60 nanogram/g dry weight. This translates to a maximum of 175-275 ng aph4/g protein or 0.000018-0.000028%.[26] On the other hand, the Vip3Aa19 content of cottonseed ranges from 2.72 to 3.23

microgram/g dry weight. This translates to 7.95- 14.82 mcg Vip3Aa19/g protein or 0.0008-0.0015% [26]

J. Nutritional Data

The levels of difference for the proximates of both cotton COT102 and its comparator are well within the confidence interval of 99%. [27] The mineral analyses done in 2001 showed no statistically significant differences in minerals between COT102 and the non-transgenic comparator. In 2002, significant differences were observed for potassium, zinc and copper levels of COT102 and the non-transgenic comparator, but the magnitude of differences were very small.[28]

In addition, there is no statistically significant difference in the amino acid composition of COT102 and the conventional untransformed counterpart. [29] There is also no statistically significant difference in the fatty acid composition of COT102 and the conventional untransformed counterpart.[30]

Lastly, The total gossypol analysis in 2001 showed no statistically significant differences between COT102 and the non-transgenic comparator. Similarly, the analysis in2002 showed no significant differences inn gossypol (free and total) and cyclopropenoid fatty acids) between COT102 and the non-transgenic comparator.[31]

H. Recommendation

Find scientific evidence that the regulated article applied for human food and animal feed use is as safe as its conventional counterpart and shall not pose any significant risk to human and animal health

BPI-PPSSD ASSESSMENT AND RECOMMENDATIONS

Based on the documents submitted by the applicant, BAI made the following assessment:

A. Toxicological Assessment

Digestibility study through SDS PAGE and Western blot analysis demonstrated that Vip3Aa19 protein was rapidly digested upon incubation with simulated gastric fluid (SGF) with pepsin within 1 minute. Based on these analyses, the T50 result is 1 minute.[24]

Heat stability assay through determination of immunoreactivity by ELISA also demonstrated that the concentration of Vip3Aa19 protein decreased by 76% upon subject to 95˚C for 30 minutes. The concentration decreased to limit of quantitation (LOQ) upon subject to heat greater than 95˚C. Loss of functional activity was also observed at temperatures greater than 65˚C.[24]

Further, amino Acid Sequence Comparison with non-redundant protein sequences database using BLASTp showed no significant homology of Vip3Aa19 to any known toxin (BLAST). [24] Acute oral gavage also demonstrated that administration of 5000 mg/kg bw Vip3Aa19 protein in mice did not yield any significant effects on survival, clinical observations, body weight gain or gross pathology.The No Observed Effect Level (NOEL) for Vip3Aa19 is 5000 mg/kg bw.[24]

Meanwhile, digestibility study through SDS PAGE and Western blot analysis demonstrated that APH4 protein was rapidly digested upon incubation with simulated gastric fluid (SGF) with

pepsin within 1 minute and SIF with pancreatin within 5 minutes. Based on these analyses, the T50 result is 5 minutes.[25]

Heat stability assay also demonstrated the effect of temperature to the immunoreactivity of APH4 protien. Mean concentration of APH4 subjected to varying temperatures from 4˚C to 200˚ C was monitored through ELISA. The concentration and relative immunoreactivity of APH4 protein significantly decreased by 70% upon subject to 120˚C. At 170˚C, the protein was undetectable. [25]

Moreover, amino acid Sequence Comparison with non-redundant protein sequences database using BLASTp showed no significant homology of APH4 to any known toxin. BLAST results includes 99% shared identity with serinethreonine protein kinase (SPS1) and thre (3) other sequences from cloning vectors pRR24, pRR14 and pSLS51. [25] Acute oral gavage also demonstrated that administration of 2000 mg/kg bw APH4 protein in mice did not yield any significant effects on survival, clinical observations, body weight gain, food consumption or gross pathology. The No Observed Effect Level (NOEL) for APH4 is 779 mg/kg bw.[25]

B. Allergenicity Assessment

Escherichia coli was used as the source of Vip3Aa19 protein for testing. The E. coli-produced Vip3Aa19 protein has been shown to be equivalent to the plant-produced Vip3Aa19 protein in terms of functional activity, structure, glycosylation and apparent molecular weight.[24] The same bacteria was also used as the source of APH4 protein for testing. Equivalency of the E. coli produced APH4 and the COT102-produced APH4 was not established due to the very low presence of APH4 in COT102. [25]

Bioinformatics analysis using the full-length sequence, an 80-mer sliding window and 8-mer exact match in AllergenOnline.org database did not yield any significant homology of Vip3Aa19 to any know allergen above 35% shared identity. The identity of the Vip3Aa19 protein in COT102 cotton was confirmed through N-terminal sequencing of amino acids and peptide mass mapping analysis of purified proteins. Immunoreactivity of the Vip3Aa19 protein in COT102 was demonstrated by Western Blot. [24] Glycosylation analysis of Vip3Aa19 protein in COT102 demonstrated that the novel protein is not glycosylated .The molecular weight (MW) of HPPD W336 in FG 72 is ~89 kDa.[24][26]

Based on the the mean level of Vip3Aa19 protein in seed of COT102 cotton and the mean dry weight of total protein in seed of COT102, Vip3Aa19 protein comprises 0.0010063% of the total protein.[26]

Meanwhile, bioinformatics analysis using the full-length sequence, an 80-mer sliding window and 8-mer exact match in AllergenOnline.org database did not yield any significant homology of APH4 to any know allergen above 35% shared identity.[25]

The levels of APH4 in COT102 is below LOQ. Hence, physico-chemical properties of APH4 in COT102 were not determined. Glycosylation prediction software predicted that APH4 is not glycosylated.The molecular weight (MW) of APH4 in COT102 is ~38.0 kDa . [25-26]

Based on the the mean level of APH4 protein in seed of COT102 cotton and the mean dry weight of total protein in seed of COT102, APH4 protein comprises 0.0005% of the total protein.[26]

C. Nutritional Composition

Compositional analysis demonstrated that the proximate levels in COT102 cottonseed were not significantly different to the non-transgenic counterpart Coker 312 cottonseed.[27]

Moreover, based on the statistical analyses, any statistical differences between the fatty acid, amino acid, and mineral levels of COT102 cotton and non-transgenic cotton are not biologically relevant. All mean values that has significant differences with the non-transgenic cotton, Coker 312, were within the range of literature values. All mean values outside of the literature range is not significantly different from the non-transgenic cotton, Coker 312.[28]

Lastly, based on the statistical analyses, there were no statistical differences between gossypol, sterculic acid, malvalic acid and dihydrosterculic acid content of COT102 cotton and non-transgenic cotton that can be considered biologically relevant.

D. Recommendation

Find scientific evidence that the regulated article applied for human food use is as safe as its conventional counterpart and shall not pose any significant risk to human and animal health

BAI ASSESSMENT AND RECOMMENDATIONS

Based on the documents submitted by the applicant, BAI made the following assessment:

E. Toxicological Assessment

The Vip3Aa19 underwent simulation for mammalian gastric fluid, which contains 2mg/ml NaCl and 2600 units/ml of pepsin with a pH of 1.2 ± 0.05. After 2 minutes of subjecting the Vip3Aa19 to gastric fluid, SDS-PAGE detected a molecular weight of approximately 3 kDa,. In western blot technique results showed no cross reaction between Vip3Aa19 and anti Vip3Aa19 antibody after 2 minutes of incubation in SGF.[24] The Vip3Aa19 protein was then subjected to heat (65oC) at 30 minutes which lead to a decrease in immunoreactivity of 66% as detected using ELISA.[24]

A comparison of the Vip3Aa19 protein to sequences in a database was performed using the Basic Local Alignment Search Tool for Proteins (BLASTP) program. The comparisons were used to determine whether the Vip3Aa19 protein (Vip3Aa19) amino acid sequence showed significant similarity to known and putative toxins. The protein Vip3Aa19 is 99.9% identical in amino acid sequence to the native Vip3Aa1 protein identified in Bacillus thuringiensis strain AB88.[24]

The acute oral toxicity of VIP3A-0100 was evaluated in male and female mice when administered as a single gavage dose. There were no recorded mortality at 5000mg/kg except for the decrease in body weight gain on males on the 8th day.[24]

Meanwhile, the APH4 also undergone simulation for mammalian gastric fluid and mammalian intestinal fluid, which contain 2mg/ml NaCl and 2600 units/ml of pepsin with a pH of 1.2 ± 0.05. and 50mM potassium phosphate monobasic and 10 mg/ml pancreatin with pH of 7.5 ± 0.05 respectively. As shown in the SDS-PAGE gel, APH4 almost digested completely at 30 minutes of incubation in SGF. Likewise, Western blot technique showed that there was no immuno-reactive response visible after 1 minute of incubation in SGF. For SIF, SDS-PAGE gel showed evident bands at approximately 20 kDa after 2 hrs of incubation and were still visible at 48 hrs of incubation. Western blot technique results showed no cross reaction between APH4 and anti APH4 antibody after 5 minutes of incubation in SIF.[25]

The APH4 protein was also subjected to heat (120oC) at 30 minutes which lead to a decrease in immunoreactivity of 70% as detected using ELISA.[25]

A comparison of the APH4 protein to sequences in a database was performed using the Basic Local Alignment Search Tool for Proteins (BLASTP) program. In the FASTA search, no sequence

similarity greater than 35% shared identity over 80 or more amino acids was observed between the APH4 amino acid sequence and any entry in the COMPARE database.[25]

Lastly, A group of five male and five female APfCD-1 mice received two oral doses of 914 mg APH4-0102/kg, approximately two hours apart to provide a total test substance dose of 1828 mg APH4-0102/kg. No treatment-related effects were seen in mice following an oral dose of 1828 mg APH4-0102/kg bodyweight. [25]

F. Allergenicity Assessment

The source of test protein is a recombinant Escherichia coli. Equivalency of microbial produced protein from plant produced protein were compared with respect to identity, integrity, insecticidal activity, and glycosylation status.[24]

Expression studies showed that Vip3A protein comprises only 0.0010063% of the total weight of corn seed kernels. Using the data from USDA (2019), protein content is composed of about 32/59% of the total seed weight, thus 1g seed contains about 0.3259 g protein and 1 g of protein in seed is contained in 3.068 g seed. With the given values, it can be derived that Vip3A protein constitutes ~0.001006304% or ~0.0010 % of total protein in COT102 cotton seed, thus, the dietary exposure to Vip3A proteins from COT102 cotton seed is only minimal[26]

A comparison of the APH4 and Vip3Aa19 protein to sequences in a database was performed using the Basic Local Alignment Search Tool for Proteins (BLASTP) program. In the FASTA search, no sequence similarity greater than 35% shared identity over 80 or more amino acids was observed between the APH4 amino acid sequence and any entry in the COMPARE database.Likewise, there were also no homology with any known allergens between Vip3Aa19 and any entry in the database. It 99.9% identical in amino acid sequence to the native Vip3Aa1 protein identified in Bacillus thuringiensis strain AB88. [25]

G. Nutritional Composition

Based on the submitted results of proximate analysis done in 2001 (Moisture, Fat, Protein, Crude Fiber and Ash) and in 2002 (Moisture, Fat, Protein, Total Dietary Fiber, Acid Detergent Fiber, Neutral Detergent Fiber, Ash, and Carbohydrates), there were no statistical differences between the COT 102 and Coker 312 (non-transgenic comparator).[27]

The analysis of the samples during 2001 showed no significant differences between the COT 102 and Coker 312 (non-transgenic comparator) on mineals, vitamins and amino acid composition. However, samples that were analyzed in 2002, significantly lower ($p<0.05$) values of potassium, zinc, copper, serine and lysine were shown in COT 102 compared to Coker 312 (non-transgenic comparator).[28] However, both amino acids' mean levels in were still within the range of values reported in the literature.

Lastly, based on the submitted results of gossypol analysis done in 2001 and 2002, there were no statistical differences between the COT 102 and Coker 312 (non-transgenic comparator).[31]

H. Recommendation

Find scientific evidence that the regulated article applied for animal feed use is as safe as its conventional counterpart and shall not pose any significant risk to human and animal health

DENR Biosafety Committee

After a comprehensive review and evaluation of the documents and scientific evidence from literature submitted by Syngenta Philippines, Inc. concerning its application for Direct Use as FFP of Cotton (COT102), hereunder are the observations and appropriate actions:

1. The regulated article is considered as safe for direct use as food, feed or for processing and is less likely to pose a threat to the environment, particularly on biodiversity and non-target organisms. The genetically modified crop is substantially similar to its conventional counterpart except for the novel insect resistance trait [2];

2. The VIP3A insect control protein is not toxic to other non-target vertebrates and invertebrates, even to other insect orders both beneficial and pests that are not lepidopteran [33]. Thus, the cottonseeds are unlikely to cause harm to local wildlife; and

3. The project description report (PDR) discusses the specified environmental management plan indicating the possible risk and harm to the environment particularly on biodiversity and non-target organisms as well as the mitigating measures and contingency plan. In case of accidental release, cotton is less likely to persist in a non-dome sticated environment while its survival and dispersal is limited by trampling and grazing, in the unlikely case of its persistence [32].

Based on the review and evaluation, the DENR-BC considered the regulated article safe to the environment, particularly on biodiversity and non-target organisms, and hereby submits the technical report concerning the application of Syngenta Philippines, Inc. for Biosafety Permit for direct use as food, feed, or for processing of Cotton (COT102).

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 and environment.

The following are the observations and recommendations:

1. Scientific pieces of evidence from Toxicity studies and references, find that the regulated article will not cause significant adverse health effects to human and animal health.

2. Dietary exposure to the regulated article is unlikely to result in allergic reaction.

3. The regulated article is as safe as food or feed derived from conventional cotton varieties.

4. The regulated article is not materially different in nutritional composition from that of the non-traiisgenic com or the conventional cotton.

5. It 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 FFP and is not to be used as planting materials.

Based on the above considerations and with the submitted sworn statement and accountability 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 Cotton Cot102

SEC Expert

Based on SEC expert review of the SEC questionnaire answered by the applicant:

The rationale for the favorable SEC assessment of these 2 cotton events lie on the fact that to date, the Philippines is not a cotton producer. It imports raw cotton which is comprised of raw lint for fabrics and cotton seeds for animal feeds. The focus of the SEC assessment is the trade of raw cotton which is further processed into limited fabric production and seed cotton for animal feeds. GM cotton for direct use is favorable, economically to the Philippines. Hence, the approval for SEC endorsement.

Recommendation

The SEC expert has recommended for the approval and issuance of the biosafety permit of the GM product.

References

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