RISK ASSESSMENT REPORT

OF THE GENETIC MODIFICATION ADVISORY COMMITTEE (GMAC)

FOR

AN APPLICATION FOR APPROVAL FOR RELEASE OF PRODUCTS OF SOYBEAN A2704-12

FOR SUPPLY OR OFFER TO SUPPLY

NBB REF NO: JBK(S) 602-1/1/7

APPLICANT: BAYER CO. (MALAYSIA) SDN. BHD.

DATE SUBMITTED: 23 AUGUST 2011

I - Summary of Assessment Process

The Genetic Modification Advisory Committee (GMAC, please refer to ***Appendix 1*** for details of GMAC), under the purview of the National Biosafety Board was given the dossier by the Department of Biosafety on 23 September 2011 for an application for approval for importation for release (sale/placing on the market for direct use as food, feed and for processing (FFP)) of a product of a Living Modified Organism (Soybean A2704-12, herbicide-tolerant soybean). The application was filed by Bayer Co. (Malaysia) Sdn. Bhd. (hereafter referred to as "the applicant"). GMAC members also took the opportunity to obtain further clarification on certain details of the activity. Additional information was also provided by the applicant as requested.

A public consultation for this application was conducted from the 19 December 2011 to 18 January 2012 via advertisements in local newspapers. No technical and scientific issues were raised through the Public Consultation for this application regarding the release.

GMAC had four meetings pertaining to this application and prepared the Risk Assessment Report and Risk Assessment Matrix along with its recommended decision, for consideration by the National Biosafety Board.

II - Background of Application

This application is for approval to import and release products of a Living Modified Organism (Soybean A2704-12, herbicide-tolerant soybean). The aim of the import and release is to supply or offer to supply for sale/placing on the market for direct use as food, feed and for processing (FFP). According to the applicant, there will be no difference in use of product of Soybean A2704-12 compared to conventional soybeans already on the market.

A major food use of soybean is as purified oil, utilized in margarines, shortenings and cooking and salad oils. It is also used in various food products including tofu, soya sauce, simulated milk and meat products. Most soybean meal (97%) is used as a protein supplement in feed rations for livestock (with 46% going to poultry, 32% to swine and 9% each to dairy and beef cattle feed). Industrial use of soybean ranges from the production of yeasts and antibodies to the manufacture of soaps and disinfectants. A sizeable amount is also used in pet food.

The applicant claims that Soybean A2704-12 seeds are substantially equivalent to conventional soybean and provide the same nutritional value as soybean currently being consumed.

Information about Soybean A2704-12

The recipient or parental plant is *Glycine max* (L.) Merr. (Soybean). Soybean is generally regarded as one of the oldest cultivated crops, native to North and Central China. The wild soybean species are endemic in China, Korea, Japan, and Taiwan but do not exist naturally in Malaysia. Soybean is a self-pollinated species, propagated commercially by seed. Artificial hybridization is used in conventional breeding for genetic improvement of the crop cultivar. Cultivated soybean seed rarely display any dormancy characteristics. As the soybean plant is not weedy in character, it does not effectively compete with other cultivated plants.

The Soybean line A2704-12 has been genetically modified to be herbicide tolerant, i.e. tolerant to the herbicide glufosinate ammonium (glufosinate for short), an active ingredient in phosphinothricin herbicides which is used as a weed control option in soybean crops. Glufosinate is a natural compound isolated from two species of *Streptomyces* bacteria. It inhibits the activity of an enzyme, glutamine synthetase, which is necessary for the production of glutamine and for ammonia detoxification. The application of glufosinate leads to reduced glutamine and increased ammonia levels in the plant tissues. This causes photosynthesis to stop and the plant dies within a few days. Glufosinate also inhibits the same enzyme in animals. It is highly biodegradable, has no residual activity, and very low toxicity for humans and wild fauna.

The modified glufosinate ammonium tolerant trait of the soybean A2704-12 is conferred by a synthetic/cloned version of the *pat* gene (which is derived from the common aerobic soil bacterium *Streptomyces viridochromogenes, strain T*ü 494). The *pat* gene encodes the enzyme phosphinotrichin acetyl transferase (PAT), which catalyzes the conversion of L-phosphinothricin (L-PPT) to N-acetyl-L-phosphinothricin in the presence of acetyl CoA as a co-substrate. N-acetyl-L-phosphinothricin does not inhibit glutamine synthetase and thus has no herbicidal activity. The functional expression of the *pat* gene in the plant cell prevents glufosinate ammonium from becoming phytotoxically active.

Unlike the native *pat* gene which has a high G:C content (which is atypical for plants), the nucleotide sequence of the synthetic *pat* gene was altered (while still using codons prefered by plants) via site-directed mutagenesis in order to reduce the high G:C content. These sequence modifications did not result in changes to the amino acid sequence of the PAT enzyme.

A *pat* gene cassette was formed by fusing the synthetic *pat* gene to a 35S promoter and 35S terminator from the cauliflower mosaic virus (CaMV). The cassette was then contained in a Pb2/35saCk plasmid and maintained in *E. coli.* After cloning, selection was carried out using the *beta lactamase* gene (*bla* gene) present in the plasmid backbone as an antibiotic resistant marker. The *bla* gene does not possess any promoter which functions in plants, therefore making it non-functional in the cells of soybeans. Furthermore, prior to transformation, the plasmid was linearized in order to destroy the *bl*a gene, leaving the plasmid with no other plant expressible genes except the *pat* gene. The biolistic transformation

method (particle acceleration/gene gun) was used to transfer the purified plasmid into the soybean genome to produce the A2704-12 soybean line.

III - Risk Assessment and Risk Management Plan

GMAC evaluated the application with reference to the following documents:

(i) CODEX Guideline for the Conduct of Food Safety Assessment of Foods Derived from Recombinant-DNA Plants.

(ii) Roadmap for Risk Assessment of Living Modified Organisms, (according to Annex III of the Cartagena Protocol on Biosafety produced by the *Ad Hoc* Technical Expert Group (AHTEG) on Risk Assessment and Risk Management of the Convention on Biological Diversity).

(iii) The risk assessment and risk management plan submitted by the applicant.

GMAC took cognizance of the following as suggested within the AHTEG guidelines:

(i) That the risk assessment exercise be specific to the details of this particular application

(ii) That the risk assessment exercise be specific to the receiving environment in question, and

(iii) That any risk identified be compared against that posed by the unmodified organism.

A Risk Matrix was prepared based on an assessment mechanism developed by Office of the Gene Technology Regulator, Australia (OGTR, 2009). In applying this matrix, GMAC identified potential hazards, and then added a value/rank for the likelihood of each hazard as well as its consequences. The likelihood of each hazard occurring was evaluated qualitatively on a scale of 1 to 4, with 1 for 'highly unlikely', and 4 for 'highly likely'. The consequences of each hazard, if it were to occur, were then evaluated on a scale of 1 to 4, with 1 for 'marginal' and 4 to denote a 'major consequence'. A value was finally assigned for the overall risk from the identified potential hazard. The general formula: Overall Risk = Likelihood x Consequence was employed. GMAC also proposed risk management strategies for potential hazards, where appropriate. This methodology of assessment follows the procedure of Risk Assessment in Annex III of the Cartagena Protocol on Biosafety.

The Risk Assessment was conducted over a series of four meetings. To start with, the possible pathways to risk/hazard arising from the release of the products were identified and listed. The potential hazards were identified in three main areas:

(i) **Effects on human health**

Issues pertaining to acute toxicity of the novel protein, potential allergenicity, mutagenic/tetragenic/carcinogenic effects, reproductive toxicity, potential transfer of antibiotic resistance genes in the digestive tract, the pathogenic potential of donor microorganisms and nutritional equivalence were examined.

(ii) **Effects on animal health**

Issues pertaining to allergenicity, toxicity, anti-nutritional properties, compromised nutritional content, metabolic breakdown of products, survivability, horizontal gene transfer and animal product contamination were examined.

(iii) **Effects on the environment**

Issues pertaining to unintentional release and planting, weediness, gene transfer to bacteria, accumulation of the PAT protein in the environment, cross pollination and toxic effects on non-target organisms were examined.

Based on the above, a final list of 24 potential hazards was identified and most of the hazards had a rating of 1 (highly unlikely) for the likelihood of the hazard to happen. Although two of the hazards identified had a rating of 2 (unlikely), all 24 hazards had an overall risk rating of 1 (negligible).

The potential risk of A2704-12 soybean was evaluated in equivalence to, and above any potential risk reported for unmodified soybean.

However as a precautionary measure, GMAC recommends that the proposed terms and conditions under section IV should be adhered to.

IV - Proposed Terms and Conditions for Certificate of Approval

Based on the 24 potential hazards identified and assessed, GMAC has drawn up the following terms and conditions to be included in the certificate of approval for the release of this product:

a) Any spillage (during loading/unloading) shall be collected and cleaned up immediately with strict adherence to the GMAC approved SOP.

b) Transportation of the consignment from the port of entry to any destination within the country shall be in closed containers.

c) A post-monitoring plan shall be implemented, whereby the approved person shall submit a yearly report to the National Biosafety Board in compliance with procedures for handling any spillage.

d) Should the approved person receive any scientifically proven information that confirms any adverse effect of A2704-12 soybean, the National Biosafety Board authority shall be informed immediately.

e) There shall be clear labeling of the product from importation down to all levels of marketing to state that it is only for the purpose of food, feed and processing and is not to be used as planting material.

V - Other Regulatory Considerations

a) Administrative regulatory arrangements shall be carried out between the Department of Biosafety and the Malaysian Quarantine and Inspection Services (MAQIS) to impose post-entry requirements for accidental spillage involving the GM product.

b) Administrative regulatory arrangements shall be carried out between the Department of Biosafety and the Department of Veterinary Services so that any unanticipated adverse effects in animals caused by any consumption of the GM products shall be reported immediately.

c) Administrative regulatory arrangements shall be carried out between the Department of Biosafety and relevant agencies to ensure clear labeling of the product is implemented.

VI - Identification of issues to be addressed for any subsequent release of this product

No additional issues have been identified that would be important during the assessment of an application for the long-term usage of this product. Continuous monitoring is required from the approved person to report any unanticipated adverse effect caused by the A2704-12 soybean.

VII – Conclusion and Recommendation

GMAC has conducted a thorough evaluation of the application for approval for importation for release (sale/placing on the market for direct use as food, feed and for processing (FFP)) of a product of a Living Modified Organism (Soybean A2704-12, herbicide-tolerant soybean) and has determined that the release of this product does not endanger biological diversity or human, animal and plant health. GMAC recommends that the proposed application for release be **APPROVED WITH TERMS AND CONDITIONS** as listed in section IV - Proposed Terms and Conditions for Certificate of Approval.

VIII - Bibliography

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Appendix I

GENETIC MODIFICATION ADVISORY COMMITTEE (GMAC) MEMBERS INVOLVED IN SPECIFIC RISK ASSESSMENT AREAS FOR THE APPROVAL FOR RELEASE OF PRODUCTS OF SOYBEAN A2704-12 FOR SUPPLY OR OFFER TO SUPPLY

Genetic Modification Advisory Committee (GMAC) members divided the task of looking up more information for the Risk Assessment matrix based on three broad categories. The scope of research aspects for each group is as listed below. Each sub-committee had a nominated leader to coordinate the work and report back to the main GMAC. The respective leader contacted the sub-committee members and discussed the work process with their members. The groupings of GMAC sub-committee members and their assigned tasks are as below:

1. ENVIRONMENT

- Effect on ecology of receiving environment due to unintentional release and planting (e.g. weediness, gene transfer to bacteria, accumulation of the PAT protein in the environment, cross pollination and toxic effects on non-target organisms)
    - **Assoc. Prof. Dr. Mohd. Faiz Foong bin Abdullah (Universiti Teknologi MARA) (Leader)**
    - Dr. Sim Soon Liang (Sarawak Biodiversity Centre)
    - Dr. Martin Abraham (Malaysian Society of Marine Sciences)
    - Madam Atikah binti Abdul Kadir Jailani (Department of Agriculture)

2. HUMAN HEALTH

- Effect on human health (e.g. acute toxicity of the novel protein, potential allergenicity, mutagenic/tetragenic/carcinogenic effects, reproductive toxicity, potential transfer of antibiotic resistance genes in the digestive tract, the pathogenic potential of donor microorganisms and nutritional equivalence)
    - **Madam T.S. Saraswathy (Institute of Medical Research) (Leader)**
    - Dr. S. Ravigadevi (Malaysian Palm Oil Board)
    - Madam Shamsinar binti Abdul Talib (Ministry of Health)
    - Dr. Chow Keng See (Malaysian Rubber Board)
    - Prof. Dr Son Radu (Universiti Putra Malaysia)

3. ANIMAL HEALTH

- Effect on animal health (e.g. allergenicity, toxicity, anti-nutritional properties, compromised nutritional content, metabolic breakdown of products, survivability, horizontal gene transfer and animal product contamination)
    - **Prof. Dr Jothi Malar Panandam (Universiti Putra Malaysia) (Leader)**
    - Dr. Ahmad Parveez bin Hj Ghulam Kadir (Malaysian Palm Oil Board)
    - Dr. Tan Swee Lian (Academy of Science Malaysia)
    - Prof. Dr. Helen Nair (Academy of Science Malaysia)