

FACT SHEET

APPLICATION FOR APPROVAL FOR RELEASE OF PRODUCTS OF DAS-59122-7 MAIZE FOR SUPPLY OR OFFER TO SUPPLY FOR SALE OR PLACING IN THE MARKET

NBB REF NO: JBK(S) 602-1/1/27

The objective of the Biosafety Act is to protect human, plant and animal health, the environment and biological diversity. Under the Biosafety Act, the National Biosafety Board (NBB) is currently assessing an application for approval submitted by DuPont Malaysia Sdn. Bhd. on behalf of Pioneer Hi-Bred International, Inc.

1. What is the application for?

This application is to get approval for the importation and release of DAS-59122-7 maize and its products for use as food, feed and processing.

2. What is the purpose of the import and release?

The purpose of the import and release is to sell DAS-59122-7 maize and its products into the market for direct use as food, feed and for processing. DAS-59122-7 maize is not intended for cultivation in Malaysia.

3. How has DAS-59122-7 maize been modified?

DAS-59122-7 maize has been genetically modified to express proteins, Cry34Ab1 and Cry35Ab1, that act together to provide protection against certain coleopteran insect pests, such as corn rootworm larvae (*Diabrotica spp.*). This protection is conferred by insertion of the *cry34Ab1* and *cry35Ab1* genes, isolated from the soil bacterium, *Bacillus thuringiensis (Bt)*. In countries where corn rootworm species cause significant reductions in maize yield, such as the United States, DAS-59122-7 maize provides farmers with an economical and effective means to control these pests and subsequently meet the global export demand for maize.

DAS-59122-7 maize also contains the *pat* gene, isolated from *Streptomyces viridochromogenes*, which encodes the PAT protein. Expression of the PAT protein in DAS-59122-7 maize confers resistance to glufosinate-ammonium herbicidal active ingredient and serves as a marker to select transformed maize in laboratory.

4. Characteristics of DAS-59122-7 maize

a. Details of the parent organism

Maize is extensively cultivated worldwide and has a long history of safe use. Maize grain and maize-derived products represent staple food and feed for a large portion of the global population (CFIA, 1994). No significant toxicity or allergenicity has been ascribed to any food or feed uses of maize and has been described as a food that is likely to have low allergenicity (OECD, 2002). Maize is not included in the list of food allergy indication of the US Food and Drug Administration (FDA) (US-FDA, 2006). The biology and history of safe use of maize demonstrate that the unmodified organism is safe for human and animal consumption.

b. Details of the donor organism

Bacillus thuringiensis (Bt): donor of the cry34Ab1 and cry35Ab1 genes

Bt is a diverse group of Gram-positive, spore-forming bacteria that has a history of safe use as a pesticide over several decades (US-EPA, 1996). It occurs naturally in the soil and on plants including vegetables, cotton, tobacco, tree crops, and forest crops (Schnepf *et al.*, 1998; Shelton, 2012). Several Cry proteins have been deployed as safe and effective pest control agents in microbial *Bt* formulations for almost 40 years.

Streptomyces viridochromogenes: donor of the pat gene

Streptomyces viridochromogenes is a common soil bacterium that is not considered pathogenic to humans or animals (OECD, 2007) and naturally produces the tripeptide phosphinothricyl-L-alanyl-L-alanine, which was developed as a non-selective herbicide by a German chemical company, Hoechst AG. The *pat* gene, encoding the phosphinothricin acetyl transferase, confers resistance to the phosphinothricin herbicide application (OECD, 1999).

c. Description of the trait(s) and characteristics which have been introduced or modified

DAS-59122-7 maize was generated by the insertion of the *cry34Ab1*, *cry35Ab1*, and phosphinothricin acetyltransferase (*pat*) genes using *Agrobacterium*-mediated transformation. The *cry34Ab1* and *cry35Ab1* genes were both isolated from *Bacillus thuringiensis (Bt)* and encode the Cry34Ab1 and Cry35Ab1

proteins, respectively. The Cry34Ab1 and Cry35Ab1 proteins together comprise an active insecticidal crystal protein that provides protection against certain coleopteran pests including western corn rootworm (WCR, *Diabrotica virgifera virgifera*). The *pat* gene was isolated from *Streptomyces viridochromogenes* and encodes the PAT protein. The PAT protein confers resistance to the herbicidal active ingredient glufosinate.

4. Modification Method

DAS-59122-7 maize was produced by means of *Agrobacterium*-mediated transformation with plasmid PHP17662. The T-DNA region of PHP17662 contains the following three gene cassettes: 1. the maize-optimized *cry34Ab1* gene derived from *Bt*, 2. the maize-optimized *cry35Ab1* gene also from *Bt*, and 3. a modified version of the *pat* gene from *Streptomyces viridochromogenes*.

a. Characterization of the Modification

The molecular characterization of the inserted genes of DAS-59122-7 maize was performed using Southern analyses, phenotypic segregation analyses, DNA sequencing, and bioinformatics analysis. These studies demonstrate that the introduced genes segregate according to Mendel's law of inheritance, are stable across multiple generations, and were integrated at a single point of insertion. The introduced genes in DAS-59122-7 maize express the Cry34Ab1, Cry35Ab1, and PAT proteins.

b. Safety of the expressed protein

The allergenic and toxicity potential of the Cry34Ab1, Cry35Ab1, and PAT proteins were evaluated using a series of studies, including bioinformatics, *in vitro* digestibility, glycosylation analyses, acute protein toxicity, and heat lability analyses. The results of these studies demonstrate that the proteins are unlikely to be allergens or toxins.

Based on the safety data submitted to regulatory agencies in other countries, DAS 591227 maize has been approved for cultivation and/or food and feed uses in thirteen countries. The first authorizations of DAS-59122-7 maize occurred in 2005, and since that time, products containing DAS-59122-7 maize have been consumed globally supporting more than a decade of safe use.

5. Assessment of Risks to Human Health

a. Nutritional Data

A compositional comparison was conducted between DAS-59122-7 maize and a non-GM, near-isoline control maize to determine if DAS-59122-7 maize is as safe and nutritious as other maize varieties, as well as whether it can be used interchangeably with its comparator without negatively affecting human and animal health. Based on trait mode of action, it is not expected that insect- and herbicide-resistant trait proteins would alter the composition of maize.

Samples were processed and key compositional analytes were measured in grain (proximates, fatty acids, amino acids, minerals, vitamins, secondary metabolites, and anti-nutrients) and in forage (proximates and minerals). The analytes selected for the compositional assessment were guided by the recommendations and information in the OECD consensus document on compositional considerations for new varieties of maize ([OECD, 2002a](#)).

Overall, the results of the compositional assessment demonstrated that the nutrient composition of DAS-59122-7 maize grain and forage is comparable to that of non-GM maize and that DAS-59122-7 maize is as safe and nutritious as non-GM maize for use in food and feed.

b. Toxicology

The toxicity potential of the Cry34Ab1, Cry35Ab1, and PAT proteins were evaluated using a series of studies, including bioinformatics, *in vitro* digestibility, glycosylation analyses, acute protein toxicity, and heat lability analyses. The results of these studies demonstrate that the proteins are unlikely to be toxins and not acutely toxic in mice (Hérouet et al., 2005; Juberg et al., 2009; US-EPA, 2010).

c. Allergenicity

The allergenicity potential of the Cry34Ab1, Cry35Ab1, and PAT proteins were evaluated using a series of studies, including bioinformatics, *in vitro* digestibility,

glycosylation analyses, and heat lability analyses. The results of these studies demonstrate that the proteins are unlikely to be allergens.

6. Assessment of Risks to the Environment

Not applicable. This application does not cover an environmental release. This application is to facilitate the importation of DAS-59122-7 maize and its products for use as food, feed and processing.

7. What is the Emergency Response Plan?

On the basis of rigorous testing and multiple comprehensive evaluations, DAS-59122-7 maize has been demonstrated to be safe to humans, animals, non-target organisms and beneficial insects, and that the forage and grain of DAS-59122-7 maize are as safe and nutritious as conventional maize varieties. DAS-59122-7 maize also has a history of safe use and has been authorized for use as food/feed and/or cultivation by regulatory authorities in thirteen different countries. There is no present need to invoke an emergency response plan for potential adverse health effects.

Given that the scope of this application is limited to facilitate importation of DAS-59122-7 maize and its products for use as food, feed and processing, a response plan for potential unintended release into the environment via spillage during transportation of the grain will include a multiparty investigation of the report together with the competent national agencies and the implementation of mitigating measures jointly agreed upon for confirmed cases. This unlikely unintended release can be controlled with current agronomic measures taken to control other commercially available maize, such as selective use of herbicides (with the exception of glufosinate-ammonium), and manual or mechanical removal of plants.

a. First Aid Measures

No special first aid measures are required in response to exposure to this product.

b. Accidental Release Measure

This application is to facilitate the importation of DAS-59122-7 maize and its products for use as food, feed and processing. Accidental release to the environment from the import of DAS-59122-7 maize will be limited to unintended spillage during transportation of the grain. However, survival and reproduction of maize is limited by extreme environmental conditions (heat stress, drought, excessive rainfall, etc.) (Shaw, 1988). Populations of maize are unlikely to

survive outside managed agricultural environments (Shaw, 1988). Although plants may occasionally grow in uncultivated fields or occur as volunteers, maize generally does not sustain reproduction outside of cultivation (CFIA, 1994).

The unlikely unintended release of DAS-59122-7 maize to the environment can be controlled with current agronomic measures taken to control other commercially available maize, such as selective use of herbicides (with the exception of glufosinate-ammonium), and manual or mechanical removal of plants.

c. Handling and Storage

On the basis of rigorous testing and multiple comprehensive evaluations, DAS-59122-7 maize has been demonstrated to be safe to humans, animals, non-target organisms and beneficial insects, and that the forage and grain of DAS-59122-7 maize are as safe and nutritious as conventional maize varieties.

Considering this, recommendations for storage and handling of DAS-59122-7 maize will be no different from conventional, non-transgenic maize.

d. Disposal Consideration

On the basis of rigorous testing and multiple comprehensive evaluations, DAS-59122-7 maize has been demonstrated to be safe to humans, animals, non-target organisms and beneficial insects, and that the forage and grain of DAS-59122-7 maize are as safe and nutritious as conventional maize varieties.

Considering this, measures for waste disposal and treatment of DAS-59122-7 maize will not be different from conventional, non-transgenic maize.

8. How can I comment on this application?

Any member of the public may submit their comments or queries on publicly notified information about the application. Before submission of comments or queries, the person should review the information provided. Your comments and queries on any possible impacts/risks to the health and safety of the people and the environment that may be posed by the proposed release are appreciated. The submission to the comments or queries should be prepared carefully as it will be given the same scrutiny as the application by the NBB. The submission of comments and clarifications of queries should contribute to the NBB's assessment. Even if the submission is not science-based, and focuses on cultural or other values, it should still be developed in the form of a well-founded argument. Please note that the consultation period closes on 2nd December 2015

and written submissions are required by that date. Submissions must be addressed to:

Director General

Department of Biosafety

Ministry of Natural Resources and Environment

Level 1, Podium 2, Wisma Sumber Asli No. 25, Persiaran Perdana, Precinct 4, 62574 Putrajaya, MALAYSIA.

E-mail: biosafety@nre.gov.my

Fax: 03-88904935

Please include your full name, address and contact details in your submission.

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