

FACT SHEET

APPLICATION FOR APPROVAL FOR RELEASE OF PRODUCT OF MS11xRF3 *Brassica napus* FOR SUPPLY OR OFFER TO SUPPLY FOR SALE OR PLACING IN THE MARKET

NBB REF NO: JBK(S) 600-2/1/19

The objective of the Biosafety Act 2007 is to protect human, plant and animal health, the environment and biological diversity. Under the Biosafety Act 2007, the National Biosafety Board (NBB) is currently assessing an application for approval submitted by BASF (Malaysia) Sdn. Bhd.

1. What is the application for?

The application is for import and release of MS11xRF3 *B. napus* and its products.

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

The purpose of the import and release is to supply or offer for sale/ placing on the market for direct use as food, feed and processing (FFP) of MS11xRF3 *B. napus* and its products. This means that MS11xRF3 *B. napus* may enter Malaysia as seed, food ingredients for processing or packaging or as finished products ready for distribution or as feed meal for animals. The MS11xRF3 *B. napus* is not intended for cultivation in Malaysia.

3. How has the MS11xRF3 *B.napus* been modified?

The MS11 x RF3 *B. napus* is a hybrid oilseed rape from the genetically modified male sterile MS11 and fertility restorer RF3. The MS11 and RF3 *B. napus* lines were both produced using *Agrobacterium*-mediated transformation of the *Brassica napus* variety N90-740 and cultivar 'Drakkar', respectively.

MS11 *B. napus* was modified to contain the *barnase*, *barstar* and *bar* genes. The *barnase* gene codes for the Barnase protein. Expression of Barnase in the tapetum cells of MS11 *B. napus* results in lack of viable pollen and male sterility. The *barstar* gene codes for the Barstar protein and enhances transformation frequency. The *bar* gene codes for the phosphinothricin acetyl transferase (PAT) protein which confers tolerance to glufosinate-ammonium.

RF3 *B. napus* (fertility restorer line) contains the *barstar* and *bar* genes. Expression of the Barstar protein in the tapetum cells leads to restoration of fertility after crossing to a male sterile MS11 *B. napus* line.

MS11 x RF3 *B. napus* canola was developed by crossing the single parental lines MS11 *B. napus* and RF3 *B. napus* using traditional breeding methods. No new genetic modification was introduced in MS11 x RF3 *B. napus*. MS11 x RF3 *B. napus* plants are fully fertile hybrids which are tolerant to glufosinate-ammonium herbicide.

4. Characteristics of MS11xRF3 *B.napus*

(a) Details of the parent organism

Brassica napus (rapeseed, canola, Oilseed rape) is the result of interspecific cross between plant of *B. oleracea* and *B. rapa*. Oilseed rape has an extended history of cultivation and safe use. *Brassica* is a genus of the *Brassicaceae* (formerly the *Cruciferae*) family, commonly known as the mustard family, which consists of about 375 genera and 3,200 species of plants mainly found in the northern hemisphere.. The genus *Brassica* contains about 100 species, including cabbage, cauliflower, broccoli, oilseed rape, brussels sprouts, turnip, various mustards and weeds. Many *Brassica* species have been cultivated since prehistoric times for their edible roots, stems, leaves, buds, flowers and seeds. Within a species, crops have been developed for different purposes.

(b) Details of the donor organisms

Characteristics of *Bacillus amyloliquefaciens*

Bacillus amyloliquefaciens is the source of the *barnase* and *barstar* genes. It is a commonly found bacterium in the soil ecosystem and is frequently used as a source of industrial enzymes such as α -amylase. *B. amyloliquefaciens* has no known pathogenicity and is used in brewing, bread-making and food industry.

Characteristics of *Streptomyces hygroscopicus*

Streptomyces hygroscopicus is the source of the *bar* gene. It is a safe common soil saprophytic bacterial species not known to be toxic, allergenic or pathogenic to humans and animals. This *Streptomyces* species is widespread in nature and a common part of the living biosphere all over the world.

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

Summary of introduced genetic elements

Gene	Protein expressed	Trait	Introduced into
<i>barnase</i>	Barnase ribonuclease	male sterile	MS11
<i>barstar</i>	Barstar (barnase ribonuclease inhibitor)	male sterile, restored fertility	MS11 and RF3
<i>bar</i>	Phosphinothricin N-acetyltransferase (PAT)	glufosinate-ammonium tolerant	MS11 and RF3

(d) Safety of the expressed protein

Results of the studies conducted using several *in silico* approaches (such as full-length identity search and 8-mer search) to evaluate the potential amino acid sequence homology of the Barnase, Barstar and PAT proteins with known allergens and known toxins did not reveal any allergenic or toxicological findings associated with these proteins.

The human consumption of oilseed rape products is limited to the refined oil. Because virtually no protein is present in the oil extracted from the plants, the potential for human exposure is exceedingly low. Furthermore, the amounts of PAT protein present in seed-meal fed to animals would be too low to cause concern. Additionally, the barnase RNase and its inhibitor encoded by *barnase* and *barstar* genes, respectively, were not detected in dry seeds. As the introduced gene products were not detectable in the refined oil produced from transgenic oilseed rape, there will be no human exposure to these proteins based on normal consumption patterns.

(e) Utilization of *Brassica napus*

The *Brassica napus* (oilseed rape) provide 14% of the world's edible vegetable oil and are third most important source of edible oil after soybean and palm. The world demand for edible oils and more recently for biodiesel has led to a rapid growth in the production of most oilseeds with total oil produced increasing by about 4% each year. The percentage growth in the world *Brassica* seed oil production has increased some 60% between 1995-1999 and 2005-2009. The elimination of nutritionally undesirable components – erucic acid (C22-1) from the seed oil (0-varieties) and glucosinolates from the seed meal (00-varieties) – has greatly expanded the market for the seeds and its derived products (oil and meal).

These improvements were obtained through breeding and have revalued the *B. napus* oil and meal for respectively human consumption and livestock feeding. *B. napus* oil has found numerous applications in the edible food industry. Though its sensory properties are most revealed on utilization in the liquid state in the form of salad and cooking oils, the oil is also very acceptable in hydrogenated products such as shortenings, margarines, dressings, mayonnaises and products that are preferentially stored under refrigerator conditions (e.g. butterine). The nutritional quality of the oil has been more intensively studied in both animal and chemical studies than most other vegetable oils.

5. Assessment of risks to human health

(a) Nutritional data

Composition analyses were conducted to determine levels of key nutrients and anti-nutrients of MS11xRF3 *B. napus* and compare those results to the non genetically

modified (non-GM) conventional counterpart and six non-GM reference *B. napus* varieties (Annex 12: Jeffries et al., 2916 M-566827-01-1).

Comparison of key nutrient and anti-nutrient levels of MS11 x RF3 *B. napus* grain (not treated and treated with trait-specific herbicide) to the non-GM conventional counterpart revealed statistically significant differences for 19 analytes of the 57 examined. However, the means of all 19 analytes were within the range of the reference varieties and the tolerance intervals. Therefore, the statistically significant differences are not considered biologically relevant. Based on the comparative assessment, nutrient and anti-nutrient levels in *B. napus* grain produced by plants derived from MS11 x RF3 *B. napus* are comparable to that of the conventional counterpart and commercial *B. napus* reference varieties.

(b) Toxicology

Since only the processed oil from transgenic MS11, RF3, or MS11xRF3 hybrids derived therefrom are available for human consumption, and the processing removes proteinaceous material, there are no additional toxicity concerns regarding this product. This was further assessed by searching for amino acid sequence homologies with known protein toxins, and by examining the physiochemical characteristics of the introduced RNase (*barnase*), RNase inhibitor (*barstar*), and PAT (*bar*) proteins. No homologies with potential toxins were observed. The MS11xRF3 *B.napus* is considered as safe and as nutritious as its non-GM counterpart.

(c) Allergenicity

Since only the processed oil from transgenic MS11, RF3, or MS11xRF3 hybrids derived therefrom are available for human consumption, and the processing removes proteinaceous material, there are no additional allergenicity concerns regarding this product. This was further assessed by searching for amino acid sequence homologies with known protein allergens, and by examining the physiochemical characteristics of the introduced RNase (*barnase*), RNase inhibitor (*barstar*), and PAT (*bar*) proteins. No homologies with potential allergens were observed. The MS11xRF3 *B.napus* is considered as safe and as nutritious as its non-GM counterpart.

6. Assessment of risks to the environment

The application does not cover an environment release. The application is intended only to cover the import of the MS11xRF3 *B.napus* products from countries where the said oilseed rape event is already approved and commercially grown, and that may enter Malaysia as foodstuffs or as feed or for further food processing.

7. What is the emergency response plan?

The seed derived from MS11xRF3 *B.napus* may be imported for processing. However, the MS11xRF3 *B. napus* products that likely enter into Malaysia are in highly processed forms like refined oil or oilseed rape meal. The seed may be viable but would not thrive as Malaysia conditions are not optimum for oilseed rape growth.

Seed derived from MS11xRF3 *B. napus* is compositionally equivalent to those from conventional oilseed rape. The plants behave agronomically in the same way as conventional oilseed rape except showing the intended tolerance to the herbicide glufosinate. Should adverse effects be reported and verified, appropriate follow up action would be taken to investigate these and if verified appropriate action taken.

(a) First aid measures

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

(b) Accidental release measures

No special measures are required in response to an accidental release. Spilled seed should be swept, scooped or vacuumed in a manner that avoids dust generation and dust-related hazards.

(c) Handling and storage

No special handling procedures are required for this product. MS11xRF3 *B. napus* seeds and its products may be handled and stored as any conventional oilseed rape.

(d) Disposal considerations

The same measures for waste disposal and treatment as for conventional oilseed rape are valid for seed derived from MS11xRF3 *B. napus*.

8. How can I comment on this application?

Any member of the public may submit their comment or queries on publicly notified information about the application. Before submission of comments or queries, the person should review the information provided. Your comments or 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 of the comments or queries should be prepared carefully as it will be given the same scrutiny as the application by the National Biosafety Board (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 8th June 2020 and written submissions are required before/by that date. Submissions must be addressed to:

Director General
Department of Biosafety
Ministry of Environment and Water

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