FACT SHEET

APPLICATION FOR APPROVAL FOR RELEASE OF PRODUCTS OF MON 94804 MAIZE FOR SUPPLY OR OFFER TO SUPPLY FOR SALE OR PLACING IN THE MARKET

NBB REF NO: JBK(S) 600-2/1/34

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 Bayer Co. (Malaysia) Sdn. Bhd.

1. What is the application for?

This application is to import and release of MON 94804 maize and all its products for supply or offer to supply for sale or placing in the market. The application does not cover deliberate environmental release (i.e. cultivation) in Malaysia and does not cover any subsequent maize products that result from the use of MON 94804 maize for breeding purposes (*stacked events*[1]).

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

The purpose of the import and release is for direct use as food, feed and for processing (FFP) of MON 94804 maize and its products. This means that MON 94804 maize may enter Malaysia as grain, food ingredients for processing or packaging or as finished products ready for distribution, or as feed meal for animals. The MON 94804 maize is not intended for cultivation in Malaysia.

3. How has MON 94804 maize been modified?

MON 94804 maize was produced by *Agrobacterium tumefaciens*-mediated transformation of maize tissue using the transfer DNA (T-DNA) transformation vector PV-ZMAP527892. The T-DNA contains the *GA20ox_SUP* suppression cassette. The sequence present in the *GA20ox_SUP* suppression cassette in MON 94804 maize contains a miRNA coding gene, *GA20ox_SUP,* that was designed with sequences from *Oryza sativa* (rice) to provide the backbone structure of the initial transcript and sequences from maize to provide an inverted repeat sequence derived from coding sequences of *ZmGA20ox3* and *ZmGA20ox5* genes from *Zea mays* (maize). GA20ox_SUP miRNA produced in MON 94804 maize is processed to a miRNA that causes suppression of gene expression of the targeted *ZmGA20ox* genes within the

[1] An event in the context of a genetically modified organism is defined by the insertion of DNA into the plant genome as a result of a single transformation process. Multiple DNA sequences may be inserted during a single transformation process.

maize plant (Paciorek et al., 2022). This suppression results in the reduction of gibberellic acid (GA)/gibberellin levels in the stalk, leading to a reduction of internode length and consequently reduced overall plant height compared to the conventional control maize.

4. Characteristics of MON 94804 maize

a. Details of the parent organism

The recipient or parental plant is *Zea mays* (maize), also known as corn. Maize has been a staple of the human diet for centuries and is grown in nearly all areas of the globe. It is the largest cultivated crop in the world followed by wheat (*Triticum* sp.) and rice (*Oryza sativa* L.) in total global metric ton production (FAOSTAT, 2022[1]). However, unlike wheat and rice, the majority of maize produced is consumed as animal feed in the form of grain, forage, or silage.

Maize grain and maize-derived products represent staple food and feed for a large portion of the global population (Shiferaw *et al*. 2011). Food uses of maize include processed products from field maize and direct consumption of sweet maize and popcorn. Food products derived from the wet milling process include starch and sweetener products (e.g., high fructose maize syrup) (May, 1987). Food products derived from the dry milling process include maize grits, maize meal, and maize flour (Watson, 1988). Maize oil may be derived from either milling process (Watson, 1988). Maize is used extensively as a livestock feed for reasons that include its palatability, digestibility, and metabolizable energy (Loy and Lundy, 2019) and its relatively low cost (OECD, 2002). Maize grain may be fed whole (Watson, 1988), but in many cases it is ground and mixed with other ingredients to provide a balanced ration (Leath and Hill, 1987). Maize can also be fed as a whole plant silage.

No significant toxicity or allergenicity has been associated 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 known major food allergens described by the United States Food and Drug Administration (FDA) (U.S. FDA, 2006). The biology and history of safe use of maize demonstrate that the parent organism is safe for human and animal consumption.

b. Details of donor organism

Characteristics of rice (*Oryza sativa)*

Rice (*Oryza sativa*) is one of the most important crops in the world serving as a primary food source for more than half of the world's population (Khush, 1997). Generally, rice is

considered to be a safe source of food and feed, and is not considered by allergists to be a common source of allergens. There are very few compounds in rice which are considered unfavourable for human or animal food/feed, and these compounds have not been observed to exist at levels in rice-based foods that would be a concern for food or feed safety (OECD, 2016).

Characteristics of maize (*Zea mays*)

Maize (*Zea mays*) has been a staple of the human diet for centuries, and processed corn are consumed in a multitude of food and animal feed products. A thorough description of the anti-nutrients present in maize has been included in an OECD consensus document (OECD, 2002), which concluded that maize is not considered a common allergenic food and there have been few reports of allergenic reactions to the consumption of maize products.

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

The MON 94804 maize plant has been modified to be shorter than the conventional control maize. The presence of inverted repeat transcripts in MON 94804 maize that has been recognized by endogenous RNA interference (RNAi), resulting in down-regulation of the endogenous GA biosynthetic genes, *ZmGA20ox3* and *ZmGA20ox5* (Paciorek et al., 2022). This suppression results in the reduction of gibberellic acid/gibberellin levels in the stalk, leading to a reduction of internode length and consequently reduced overall plant height compared to the conventional control maize. MON 94804 maize can provide agronomic and environmental benefits, including reduced lodging and green snap, season-long crop access using standard ground equipment, and potential for improved environmental sustainability with more precise, well -timed, and “as needed” mid to late -season application of agrochemicals (e.g., fungicide) and/or key nutrients (e.g., nitrogen). This introduced trait will be useful for cultivation (agricultural benefit) for corn growing countries such as in North America and South America.

5. Modification Method

MON 94804 maize was produced by *Agrobacterium tumefaciens*-mediated transformation of maize tissue using the transfer DNA (T-DNA) transformation vector PV-ZMAP527892. The T-DNA contains the *GA20ox_SUP* suppression cassette, expressing an inverted repeat sequence designed to target endogenous maize *gibberellic acid 20 oxidase* (*GA20ox*) genes, *ZmGA20ox3* and *ZmGA20ox5*. The expressed inverted repeat transcript is recognized by the endogenous RNA interference (RNAi), resulting in down-regulation of the targeted *GA20ox* gene expression.

The T-DNA that was inserted also contained the *cp4 epsps* gene derived from *Agrobacterium* sp. strain CP4, as a selectable marker cassette to identify successful transformants. However, as it was flanked by *loxP* sites, it was subsequently removed from the genome using *Cre/lox* technology[2]. A selectable marker-excised line was selected as MON 94804 maize. Subsequently, segregation, selection and screening were used to isolate those plants that contained the *GA20ox_SUP* suppression cassette and lacked plasmid vector backbone and the *cp4 epsps* selectable marker sequences from PV-ZMAP527892 and any sequences from the *cre* gene-containing vector, PV-ZMOO513642. Characterization of the DNA insert in MON 94804 maize was conducted using a combination of sequencing, polymerase chain reaction (PCR), and bioinformatic analyses. The results of this characterization demonstrate that MON 94804 maize contains one copy of the intended T-DNA containing the *GA20ox_SUP* suppression cassette that is stably inherited over multiple generations and segregates according to Mendelian principles of inheritance.

a. Characterization of the modification

The expressed inverted repeat transcript is recognized by the endogenous RNA interference (RNAi) machinery, resulting in down-regulation of the targeted *GA20ox* gene expression. The suppression of targeted genes resulted in the reduction of bioactive GA levels in the stalk internode, leading to a reduction of stalk internode length, which consequently reduced overall plant height without affecting the reproductive potential when compared to the control maize (Paciorek et al., 2022).

b. Safety of the expressed RNAi

MON 94804 does not contain any recombinant gene encoding a protein. RNAs are different from proteins regarding the overall potential for toxicity and allergenicity. The GA20ox_SUP miRNA expressed in MON 94804 maize poses negligible risks to humans and animals based on the ubiquitous nature of RNAi suppression utilizing endogenous RNAs in a wide variety of plant species consumed by humans and animals. There is also a long history of safe consumption of RNAs from a range of sources (Rodrigues and Petrick, 2020), and the lack of toxicity or allergenicity of dietary RNA (Petrick et al., 2016).

Detailed information about the safety of RNAi can be obtained from the Department of Biosafety.

[2] Removal of selectable markers from the genome by Cre-lox recombination is an efficient way to circumvent any effects of marker expression and is widely used in plants, mouse cell lines, yeast, etc. (https://en.wikipedia.org/wiki/Cre-Lox_recombination)

6. Assessment of risks to human health

a. Nutritional data

Data obtained from compositional analyses conducted on the grain and forage of MON 94804 maize showed that there were no statistically significant differences in 52 of the 61 components statistically analyzed. For the 9 components that showed statistically significant difference between MON 94804 maize and the conventional control, the mean difference between MON 94804 maize and the conventional control was less than the range value (maximum value minus the minimum value) of the conventional control (Geng et al., 2021). In addition, the MON 94804 maize mean component values were found to be within the natural variability of these components as published in literature and/or the AFSI Crop Composition Database (AFSI-CCDB) (Geng et al., 2021). Therefore, these statistically significant differences are not considered biologically relevant. This data supports the statement that MON 94804 maize is compositionally equivalent to conventional maize.

Detailed MON 94804 maize composition analysis information can be obtained from the Department of Biosafety.

b. Toxicology

As mentioned above, MON 94804 does not contain any recombinant gene encoding a protein. Several studies observed that plant-derived miRNAs were significantly degraded in the gastrointestinal tract after ingestion (Dickinson et al., 2013; Huang et al., 2018; Snow et al., 2013). Therefore ingested plant-derived miRNAs are considered to not be toxic (Rodrigues and Petrick, 2020). Additionally, based on the ubiquitous nature of RNAi suppression utilizing endogenous RNAs in a wide variety of plant species consumed by humans and animals, the long history of safe consumption of RNAs from a range of sources (Rodrigues and Petrick, 2020), and the lack of toxicity of dietary RNA (Petrick et al., 2016), the GA20ox_SUP miRNA expressed in MON 94804 maize poses negligible risks to humans and animals.

Detailed safety of RNAi information can be obtained from the Department of Biosafety.

c. Pathogenicity

There is no evidence of human or vertebrate pathogenicity for any of the donor organisms of the coding DNA sequences present in MON 94804 maize. DNA has always been present

in feed and food, and upon consumption, is quickly degraded to shorter sequences of nucleic acids by nucleases present in the gastrointestinal tract of humans and animals.

d. Allergenicity

As mentioned above, MON 94804 does not contain any recombinant gene encoding a protein. All known food allergens are proteins and there is no evidence to suggest that ingested nucleic acids including miRNAs can be allergenic (U.S. FDA, 1992).

Information about MON 94804 and allergenicity can be obtained from the Department of Biosafety.

7. 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 MON 94804 maize and its products from countries where it is already approved and commercially grown, and that may enter Malaysia as grain, food ingredients for processing or packaging or as finished products ready for distribution, or as feed meal for animals.

8. What is the emergency response plan?

MON 94804 maize and food and feed products derived from it have been assessed as being as safe as its conventional corn. Should adverse effects be reported and verified, appropriate follow up action would be taken to investigate these, and if verified, appropriate actions taken.

a. First aid measures

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

b. Accidental release measures

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

c. Handling and storage

No special handling and storage procedures are required for this product. MON 94804 maize and its products may be handled and stored as any conventional maize products.

d. Disposal

The same measures for waste disposal and treatment as for conventional maize are valid for MON 94804 maize.

9. 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 in this Fact Sheet. Detailed MON 94804 safety study can be obtained from the Department of Biosafety. 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 to express your concerns 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 11 February 2025 and written submissions are required before/by that date. Submissions must be addressed to:

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

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