

Summary Notification Information Format

A. General information

A1. Details of notification

Notification Number

B/BE/24/V3

Member State

Belgium

Date of Acknowledgement

12 January 2024

Title of the Project

Scientific field evaluation of maize with improved digestibility

Proposed period of release:

15/04/2024 to 31/10/2025

A2. Notifier

Name of the Institute

VIB

A3. Is the same GMPt release planned elsewhere in the Community?

No.

A4. Has the same GMPt been notified elsewhere by the same notifier?

No

B. Information on the genetically modified plant

B1. Identity of the recipient or parental plant

- | | |
|----------------------------|---------------------------------|
| a) family: | <i>Poaceae</i> |
| b) genus: | <i>Zea</i> , section <i>Zea</i> |
| c) species: | <i>Zea mays</i> |
| d) subspecies: | <i>mays</i> |
| e) cultivar/breeding line: | inbred line B104 |
| f) common name: | maize |

B2. Description of the traits and characteristics which have been introduced or modified, including marker genes and previous modifications

The genetically modified maize plants have an altered composition of their cell wall resulting from the presence of mutations in three genes involved in the biosynthesis of lignin. These mutations lead to the inactivation of these genes. The plants have a lowered lignin content in their cell walls

which is expected to contribute to a better digestibility of the maize thereby improving the feed conversion rate.

In first instance plants with a single homozygous mutation in one gene involved in the biosynthesis of lignin were made, creating a pool of plants, each with a homozygous mutation in a different gene involved in the biosynthesis of lignin. Using conventional crossing plants were generated that combine homozygous mutations in three different genes.

B3. Type of genetic modification

Targeted editing of genes using CRISPR-Cas.

B4. In case of insertion of genetic material, give the source and intended function of each constituent fragment of the region to be inserted

There is no donor DNA inserted into the genetic material. The genome editing has resulted in small deletions and in one case one basepair of DNA has been added.

B6. Brief description of the method used for the genetic modification

The plants have been modified using CRISPR-Cas. The CRISPR-Cas machinery was introduced into the maize using *Agrobacterium tumefaciens* mediated genetic modification. This resulted in the stable integration of T-DNA containing the sequences coding for the guide RNA and CAS protein into the genome of the maize. Plants with the desired mutation were selected and the T-DNA was segregated out in the next generations leading to the selection of plants that only contained the desired mutation but no longer contained any foreign genetic material.

B7. If the recipient or parental plant is a forest tree species, describe ways and extent of dissemination and specific factors affecting dissemination

Not applicable.

C. Experimental Release

C1. Purpose of the release

The purpose of the release is to measure the performance of the modified maize plants under normal field conditions and learn whether they also in these conditions have an altered composition of their cell wall. The field trial also has the goal to determine whether the modification would lead to undesired effects such as a higher vulnerability for breaking of their stem during strong winds.

C2. Geographical location of the site

The field trial will take place on grounds belonging to the ILVO research institute in the municipality of Wetteren.

C3. Size of the site (m²)

The trial plot, including non-modified controls, non-modified fertilizer lines and non-modified buffer rows is 684 m².

C4. Relevant data regarding previous releases carried out with the same GM-plant, if any, specifically related to the potential environmental and human health impacts from the release

The plants have not been field tested before.

D. Summary of the potential environmental impact from the release of the GMPts

The environmental impact from the release is expected to be zero. The modified characteristics are not expected to lead to greater weediness or the ability of the maize to establish in non-

agricultural habitats. The modified characteristics are also not expected to change the interaction of the maize with herbivores. It is possible that the altered composition of the cell wall leads to a change in the variety of micro-organisms that live in the maize, but this is not seen as a negative impact.

There will also be no spread of the modified maize to the environment. The male flowers (tassels) of the maize will be removed before they can start shedding pollen, meaning that shedding of modified pollen to the environment is impossible. Also the spread of seeds to the environment is prevented. The modified seeds are well retained in the cobs and all cobs will be carefully hand harvested to prevent any spread of seeds.

E. Brief description of any measures taken for the management of risks

The risk of spread of the modified properties to the environment is mitigated by removing the tassel, thus preventing the spread of modified pollen to non-modified maize plants in the surroundings. The formed modified seeds are, as already stated above, well retained in the cobs and these cobs will be very carefully hand harvested, thus preventing any spread of seeds to the environment. Experience with such field trials in the past 12 years has shown that the way the cobs are harvested effectively prevents any volunteers being formed. The field trial plot is surrounded by a 1.80 m high wire fence to prevent accidental trespassing and accidental removal or spread of GM material.

F. Summary of foreseen field trial studies focused to gain new data on environmental and human health impact from the release

There are no specific studies foreseen to gain new data on the environmental and human health impact from the release other than the study of the phenotype and growth characteristics of the maize.

G. Final report

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H. European Commission administrative information

To be filled in by the Commission

I. Consent given by the Competent Authority:

To be filled in by the Commission.