



## **Biosafety Clearing-House (BCH)**

## LIVING MODIFIED ORGANISM (LMO)

BCH-LMO-SCBD-116286-5

## ? Decisions on the LMO ? Risk Assessments

LAST UPDATED: 01 OCT 2021

## Living Modified Organism identity

The image below identifies the LMO through its unique identifier, trade name and a link to this page of the BCH. Click on it to download a larger image on your computer. For help on how to use it go to the LMO quick-links

page.



https://bch.cbd.int/database/record?documentID=116286

MON-87427-7 × MON-8746Ø-4 × MON-89Ø34-3 × DAS-Ø15Ø7-1 × MON-87411-9 Drought-tolerant, herbicide-tolerant, insect-resistant maize



Read barcode or type above URL into internet browser to access information on this LMO in the Biosafety Clearing-House @ SCBD 2012

Name

Drought-tolerant, herbicide-tolerant, insect-resistant maize

Transformation event

MON87427 × MON87460 × MON89034 × TC1507 × MON87411

Unique identifier

MON-87427-7 × MON-8746Ø-4 × MON-89Ø34-3 × DAS-Ø15Ø7-1 × MON-87411-9

Developer(s)

### - PERSON: DOW AGROSCIENCES GMBH || BCH-CON-SCBD-104809-2

PERSON

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**RELATED ORGANIZATION** 

- PERSON: BAYER CROPSCIENCE | BCH-CON-SCBD-111462-3

ΕN

#### PERSON

Bayer CropScience Bayer CropScience AG Alfred-Nobel-Str. 50 40789 Monheim am Rhein Monheim am Rhein 40789, Germany Phone: +49 21 73 - 38-0 Website: https://www.cropscience.bayer.com/en, https://www.cropscience.bayer.de/de-DE

#### **RELATED ORGANIZATION**

#### Description

The maize (*Zea mays*) was produced through cross breeding of modified parental maize lines for drought tolerance, herbicide tolerance and insect resistance. For abiotic tolerance, the maize expresses *Bacillus subtillus* cold shock protein to enhance natural abiotic (drought) stress responses. For herbicide tolerance, the maize expresses *Agrobacterium tumefaciens* 5-enolpyruvylshikimate-3-phosphate synthase (glyphosate tolerance - enzyme variant) and *Streptomyces viridochromogenes* phosphinothricin N-acetyltransferase (glufosinate tolerance - enzymatic inactivation). For Lepidoptera tolerance, the maize expresses *Bacillus thuringiensis* Cry1A.105, Cry1F and Cry2Ab2. For Coleoptera resistance, the maize expresses *B. thuringiensis* Cry3Bb1. The maize contains an RNA interference cassette targeting *Diabrotica virgifera virgifera* Snf7 for specific resistance against *D. virgifera virgifera*. Additionally, the maize contains an *Escherichia coli* neomycin phosphotransferase II cassette for kanamycin selection, which was used during transformation of a parental line.

Recipient Organism or Parental Organisms

The term "Recipient organism" refers to an organism (either already modified or non-modified) that was subjected to genetic modification, whereas "Parental organisms" refers to those that were involved in cross breeding or cell fusion.

BCH-ORGA-SCBD-246-6 ORGANISM ZEA MAYS (MAIZE, CORN, MAIZE)

Crops

BCH-LMO-SCBD-104758-3 LIVING MODIFIED ORGANISM | MON-87427-7 - MAIZE MODIFIED FOR TISSUE SELECTIVE GLYPHOSATE TOLERANCE |

Resistance to herbicides - Glyphosate

BCH-LMO-SCBD-103066-6 LIVING MODIFIED ORGANISM | MON-8746Ø-4 - DROUGHTGARD<sup>™</sup> MAIZE Resistance to antibiotics - Kanamycin Tolerance to abiotic stress - Cold / Heat, Drought

BCH-LMO-SCBD-43773-18 LIVING MODIFIED ORGANISM | MON-89Ø34-3 - YIELDGARD<sup>™</sup> VT PRO<sup>™</sup> | Resistance to diseases and pests - Insects - Lepidoptera (butterflies and moths)

BCH-LMO-SCBD-14841-13 LIVING MODIFIED ORGANISM | DAS-Ø15Ø7-1 - HERCULEX™ I MAIZE | Resistance to diseases and pests (Insects, Lepidoptera (butterflies and moths)), Resistance to herbicides (Glufosinate)

## BCH-LMO-SCBD-108881-1 LIVING MODIFIED ORGANISM | MON-87411-9 - MAIZE MODIFIED FOR HERBICIDE TOLERANCE AND INSECT RESISTANCE |

Monsanto | Resistance to diseases and pests (Insects, Coleoptera (beetles), Western corn rootworm (Diabrotica virgifera), Northern corn rootworm (Diabrotica barberi)), Resistance to herbicides (Glyphosate)

## Characteristics of the modification process

Vector

#### PV-ZMAP1043; PV-ZMAP595; PV-ZMIR245; PHI8999A; PV-ZMIR10871

Techniques used for the modification

#### Cross breeding

Genetic elements construct



## Introduced or modified genetic element(s)

Some of these genetic elements may be present as fragments or truncated forms. Please see notes below, where applicable.

BCH-GENE-SCBD-100366-6 CAMV ENHANCED 35S PROMOTER

ΕN

#### Promoter

### BCH-GENE-SCBD-100359-7 HSP70 INTRON | (MAIZE, CORN)

Intron

#### BCH-GENE-SCBD-100365-6 CHLOROPLAST TRANSIT PEPTIDE 2 | (THALE CRESS)

Transit signal

#### BCH-GENE-SCBD-14979-7 5-ENOLPYRUVYLSHIKIMATE-3-PHOSPHATE SYNTHASE GENE

Protein coding sequence | Resistance to herbicides (Glyphosate)

#### BCH-GENE-SCBD-100269-8 NOPALINE SYNTHASE GENE TERMINATOR

Terminator

#### BCH-GENE-SCBD-101415-9 TI PLASMID LEFT BORDER REPEAT

Plasmid vector

#### BCH-GENE-SCBD-100364-5 RICE ACTIN 1 GENE PROMOTER | (RICE)

Promoter

### BCH-GENE-SCBD-100355-6 RICE ACTIN 1, INTRON | (RICE)

Intron

#### BCH-GENE-SCBD-103065-7 COLD SHOCK PROTEIN GENE

Protein coding sequence | Tolerance to abiotic stress (Cold / Heat, Drought)

#### BCH-GENE-SCBD-103067-9 TRANSCRIPT 7 GENE 3' UNTRANSLATED REGION

Terminator

#### BCH-GENE-SCBD-103069-3 LOXP RECOMBINATION SITE

recombination site

#### BCH-GENE-SCBD-100287-7 CAMV 35S PROMOTER

Promoter

#### BCH-GENE-SCBD-15001-5 NEOMYCIN PHOSPHOTRANSFERASE II | (BACTERIA)

Protein coding sequence | Resistance to antibiotics (Kanamycin)

#### BCH-GENE-SCBD-101416-6 TI PLASMID RIGHT BORDER REPEAT

Plasmid vector

## BCH-GENE-SCBD-100354-6 5' UNTRANSLATED LEADER FROM CHLOROPHYLL A/B-BINDING PROTEIN | (WHEAT)

Leader sequence

#### BCH-GENE-SCBD-43771-9 CRY1A.105 | BACILLUS THURINGIENSIS - BT, BACILLUS, BACTU

Protein coding sequence | Resistance to diseases and pests (Insects, Lepidoptera (butterflies and moths))

#### BCH-GENE-SCBD-100356-6 HEAT SHOCK PROTEIN 17.3 TERMINATOR | (WHEAT)

Terminator

#### BCH-GENE-SCBD-101507-5 FMV 34S PROMOTER

Promoter

## BCH-GENE-SCBD-100360-4 TRANSIT PEPTIDE AND FIRST INTRON OF RUBISCO SSU | (MAIZE, CORN) Transit signal

### BCH-GENE-SCBD-14988-7 CRY2AB2 | BACILLUS THURINGIENSIS - BT, BACILLUS, BACTU

Protein coding sequence | Resistance to diseases and pests (Insects, Lepidoptera (butterflies and moths))

### BCH-GENE-SCBD-100362-7 UBIQUITIN GENE PROMOTER | (MAIZE, CORN)

Promoter



#### Notes regarding the genetic elements present in this LMO

#### DNA insert from MON87427 PV-ZMAP1043

Transcription of 5-enolpyruvylshikimate-3-phosphate synthase (*cp4-epsps*) from *Agrobacterium tumefaciens* commences from the *Cauliflower mosaic virus* (CaMV) enhanced 35S promoter and ends at the *A. tumefaciens* nopaline synthase (*nos*) gene terminator. The transcript contains a *Zea mays* heat shock protein 70 (*hsp70*) intron, *Arabidopsis thaliana* Nterminal chloroplast transit peptide sequence, and *cp4-epsps*. The CaMV enhanced 35S promoter-*hsp70* combination promotes gene expression in female and vegetative tissues, but not in male reproductive tissues (pollen microspores and tapetum).

#### Note:

- Southern blot analyses indicate that a single copy of the T-DNA was inserted at a single site in the parental maize genome and no plasmid vector backbone sequences were detected to have been integrated. DNA sequencing analyses further indicated that the expected T-DNA sequences were integrated.

-The *cp4-epsps* coding sequence is the codon optimized coding sequence of the *aroA* gene from *Agrobacterium sp.* strain CP4 encoding CP4 EPSPS.

## DNA insert from MON87460 vector PV-ZMAP595

The T-DNA insert contains the following gene cassettes: *Bacillus subtillus* cold shock protein (*cspB*) and *Escherichia coli* neomycin phosphotransferase II (*nptII*).

ΕN

Transcription of *cspB* is under control of the *Oryza sativa* actin 1 promoter and *Agrobacterium tumefaciens* transcript 7 gene 3' untranslated region. The transcript initially contains an *O. sativa* actin 1 intron for enhanced gene expression of *cspB*. The sequence is removed (spliced) prior to protein translation. Constitutive expression of *cspB* is expected due to the actin promoter.

Transcription of *nptll* is under control of the *Cauliflower mosaic virus* (CaMV) 35S promoter and *A. tumefaciens* nopaline synthase terminator. High levels of transcription are expected due to the CaMV promoter.

## Note:

- The coding sequence of *cspB* has been codon optimized for optimal expression within plant cells.

- Southern blot analysis indicated that no vector backbone sequences were inserted into the parental genome

- Southern blot analysis indicated that the parental genome contains a single insertion
- Sequencing analyses confirm the Southern blot analyses.
- A 22 base pair deletion of genomic DNA at the insert-to-plant DNA junction occurred.

- *loxP* sites can be found in the parental genome and could potentially allow for the excision of the *nptll* cassette by CRE recombinase.

## DNA insert from MON89034 vector PV-ZMIR245

Two insecticidal protein expression cassettes were inserted into the genome. *Bacillus thuringiensis cry1A.105* expression is under the control of the CaMV 35S enhanced promoter, which first transcribes wheat (*Triticum aestivum*) 5' untranslated region of the chlorophyll a/ b-binding protein (*cab*) and a rice actin 1 intron before transcribing *cry1A.105*. Transcription terminates at the wheat heat shock protein 17.3 terminator. Expression of the *B. thuringiensis cry2Ab2* starts at the *Figwort mosaic virus* 34S promoter, which transcribes the *Zea mays* heat shock protein 70 (*hsp70*), then the *Z. mays* transit peptide and the *cry2Ab2* coding sequence, before terminating at the *nos* terminator.

## Note:

- The Cry2Ab2 coding sequence was modified for optimal expression in plants.

- South blot analysis confirmed that single insertions of both *cry2Ab2* and *cry1A.105*, as well as no vector backbone were present and in the parent.

- A deletion removed the duplicated enhancer elements compared to the original CaMV 35S enhanced promoter in PV-ZMIR245.

- The selectable marker, *nptll*, cassette was bred out of the parental line and became not associated with this transformation event.

## DNA insert from TC1507 vector PHI8999A

DNA fragment PHI8999A contains two adjacent plant gene expression cassettes for *Bacillus thuringiensis cry1F* and *Streptomyces viridochromogenes pat*.

Transcription of *cry1F* is directed by the promoter and first exon and intron of the maize (*Zea mays*) ubiquitin gene and terminates at the *Agrobacterium tumefaciens* ORF25 terminator.

Transcription of the *pat* gene commences from the *Cauliflower mosaic virus* (CaMV) 35S promoter and ends at the CaMV 35S terminator.

## Note:

- The coding sequence of both genes has been optimized to achieve a high level of expression in maize.

The sequences of the complete *cry1F* and *pat* are identical to those in the original plasmid.
The CRY1F protein includes the F604K (phenylalanine to lysine at position 604) amino acid substitution, which was introduced to create a specific restriction site for cloning purposes.

## DNA insert from MON87411 vector PV-ZMIR10871

The MON87411 genome contains three cassettes: an RNA interference (RNAi) cassette targeting *Diabrotica virgifera virgifera, Bacillus thuringiensis cry3Bb1* and *Agrobacterium tumefaciens* 5-enolpyruvylshikimate-3-phosphate synthase (*cp4-epsps*).

Transcription of the RNAi cassette commences from the *Cauliflower mosaic virus* 35S enhanced promoter and terminates at the *Pisum sativum* ribulose bisphosphate carboxylase small chain 2 terminator. The transcript initially contains a *Zea mays* heat shock protein 70 intron, which contributes to enhanced expression in vegetative tissues of the plant, and two partial coding sequences of the *D. virgifera virgifera* Snf7p gene, which encodes the SNF7 subunit of the ESCRT-III complex. The two Snf7p sequences are in an inverted orientation, separated by a 150-nucleotide intervening sequence, which allows base pairing between the inverted sequences and hairpin RNA formation post-transcription, which then triggers an RNAi response. Due to RNAi processing, small interfering RNA molecules (roughly 21-23 nucleotides in length) will be produced and thus no translation into protein will occur from this cassette.

Transcription of the *cry3Bb1* is under control of the *Z. mays* physical impedance induced protein promoter and *Triticum aestivum* (wheat) heat shock protein 17.3 terminator. The transcript also contains a wheat 5' untranslated leader from chlorophyll a/b-binding protein and *Oryza sativa* actin 1 intron for enhanced expression of the transgene. Expression of *cp4-epsps* is under control of an *O. sativa* alpha tubulin promoter and terminator. The transcript additionally contains *Arabidopsis thaliana* chloroplast targeting peptide 2 to sequester the protein to the chloroplast.

### Note:

- Sequencing, PCR and bioinformatic analyses indicate that a single, intact insertions of the three gene cassettes occurred in the parental line.

- No plasmid backbone was detected.

- The ubiquitous expression of *cp4-epsps* overcomes the female-specific expression from the MON87427 genome.

For more information, kindly refer to the parental LMO records.

## LMO characteristics

Modified traits

Resistance to diseases and pests

Insects

Coleoptera (beetles)

Lepidoptera (butterflies and moths)

Resistance to herbicides Glufosinate Glyphosate Resistance to antibiotics Kanamycin Neomycin Tolerance to abiotic stress Cold / Heat Drought Selectable marker genes and reporter genes

Common use(s) of the LMO

Food Feed

## **Detection method(s)**

External link(s)

? MON-87427-7 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) ( *English* )

? MON-8746Ø-4 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) ( *English* )

? MON-89Ø34-3 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) ( *English* )

? DAS-Ø15Ø7-1 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) ( English )

? MON-87411-9 - EU Reference Laboratory for GM Food and Feed (EURL-GMFF) ( *English* )

## **Additional Information**

Other relevant website addresses and/or attached documents

? EUginius - MON87427 x MON87460 x MON89034 x DAS1507 x MON87411 ( English )

## BCH-LMO-SCBD-116286-5

# **Further Information**

Questions about the Cartagena Protocol on Biosafety or the operation of the Biosafety Clearing-House may be directed to the Secretariat of the Convention on Biological Diversity. Secretariat of the Convention on Biological Diversity 413 rue Saint-Jacques, suite 800 Montreal, Québec, H2Y 1N9 Canada Fax: +1 514 288-6588 Email: secretariat@cbd.int