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KENMERK CGM/171013-01

ONDERWERP Advies import en verwerking van genetisch gemodificeerde herbicidentolerante en insectenresistente maïs MZIR098

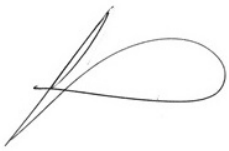
Geachte mevrouw Dijksma,

Naar aanleiding van een vergunningaanvraag voor de import en verwerking van genetisch gemodificeerde maïs MZIR098 (EFSA-GMO-DE-2017-142), ingediend door Syngenta, deelt de COGEM u het volgende mee.

Samenvatting:
De COGEM is gevraagd te adviseren over mogelijke milieurisico's bij import en verwerking van de genetisch gemodificeerde (gg-) maïslijn MZIR098. In deze lijn komen de genen *ecry3.1Ab*, *mcry3A* en *pat* tot expressie, waardoor het gewas resistent is tegen bepaalde plaaginsecten (keverachtigen). Daarnaast is het gewas tolerant voor glufosinaat-ammonium bevattende herbiciden.
Verwildering van maïsplanten is in Nederland nooit waargenomen. Maïsplanten uit gemorst zaad (opslagplanten) worden hier nauwelijks aangetroffen. Bovendien zijn er in Nederland geen wilde verwanten van maïs aanwezig, waardoor de ingebrachte sequenties niet naar andere soorten kunnen verspreiden.
De moleculaire karakterisering van MZIR098 voldoet aan de eisen van de COGEM. Er zijn geen redenen om aan te nemen dat expressie van de ingebrachte genen ervoor zorgt dat deze gg-maïslijn zich zou kunnen vestigen of zou kunnen verwilderen.
Gezien het bovenstaande acht de COGEM de milieurisico's bij import en verwerking van de gg-maïslijn MZIR098 verwaarloosbaar klein.
Omdat een voedselveiligheidsbeoordeling door andere instanties wordt uitgevoerd, heeft de COGEM bij deze vergunningaanvraag de risico's van incidentele consumptie niet beoordeeld.

De door de COGEM gehanteerde overwegingen en het hieruit voortvloeiende advies treft u hierbij aan als bijlage.

Hoogachtend,



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Import and processing of insect resistant and herbicide tolerant genetically modified maize MZIR098

COGEM advice CGM/171013-01

- The present application (EFSA-GMO-DE-2017-142) concerns the authorisation for import and processing for use in feed and food of genetically modified (GM) maize MZIR098;

- Maize MZIR098 was produced by *Agrobacterium tumefaciens* mediated transformation;
- Maize MZIR098 expresses the *ecry3.1Ab* and *mcry3A* genes conferring resistance against certain coleopteran insects, and the *pat-08* gene conferring tolerance to glufosinate-ammonium containing herbicides;

- In the Netherlands, feral maize populations have never been observed and the appearance of volunteers is rare;
- In the Netherlands, wild relatives of maize are absent and hybridisation of maize with other species is therefore not possible;

- The molecular characterisation of maize MZIR098 meets the criteria of COGEM;
- There are no indications that the introduced traits alter the fitness of maize MZIR098 under natural conditions;
- There are no reasons to assume that the introduced traits will allow GM maize MZIR098 to survive in the Dutch environment;

- COGEM is of the opinion that import and processing of maize MZIR098 poses a negligible risk to the environment in the Netherlands;
- COGEM abstains from giving advice on the potential risks of incidental consumption since a food/feed assessment is carried out by other organisations.

1. Introduction

The present application (EFSA-GMO-DE-2017-142), filed by Syngenta, concerns the import and processing of genetically modified (GM) maize MZIR098, for use in feed and food. Maize MZIR098 was produced by *Agrobacterium tumefaciens* mediated transformation, and expresses two different Cry proteins and a phosphinothricin N-acetyltransferase (PAT) enzyme. As a result, maize MZIR098 is resistant against certain insects of the order of Coleoptera and tolerant to glufosinate-ammonium containing herbicides.

2. Previous COGEM advices

COGEM previously issued several advices on the import, processing and cultivation of GM maize lines containing *ecry3.1Ab*, *mcry3A* or *pat* genes in combination with additional transgenic traits. COGEM advised positively on the import, processing and cultivation of Bt11 maize containing the *pat* gene (in combination with the *cry1Ab* gene).[1,2,3] COGEM advised positively on the import and processing of MIR604 maize containing the *mcry3A* gene,[4] and 5307 maize containing the *ecry3.1Ab* gene (both in combination with the *pmi* gene).[5] Furthermore, COGEM advised positively on the import and processing of the stacked event Bt11 x MIR604 maize.[6,7]

3. Environmental risk assessment

3.1 Aspects of the wild-type crop

Maize (*Zea mays*) is a member of the grass family *Poaceae*. It is a highly domesticated crop originating from Central America, but nowadays cultivated globally. Maize is wind pollinated,[8,9] and has both male and female flowers that are spatially separated. Female flowers are not attractive to insect pollinators, because they do not produce nectar. Insect pollination of maize is probably highly limited but cannot be excluded.[10]

Recently the wild relative of maize teosinte, has been reported as a weed in maize fields in Spain[11,12,13] and France.[14,15] In the Netherlands, no wild relatives of maize are present and hybridisation with other species cannot occur.

Maize requires warm conditions in order to grow and does not tolerate prolonged cold and frost.[10,16] In cultivation areas with warm climatic conditions, the appearance of volunteers can occur the year following maize cultivation due to spilled cobs or kernels. However, these volunteers are usually killed by common mechanical pre-planting soil preparation practices.[10]

Maize is very sensitive to weed competition.[17] During the long process of domestication, maize has lost the ability to persist in the wild.[9] A soil seed bank, small seeds, and an extended period of flowering and seed production are characteristics often observed in persistent weeds.[18] Maize lacks all these characteristics. After ripening, the seeds (the kernels) adhere to the cob and do not shatter naturally.[10,19] Consequently, seed dispersal is severely hampered.

During field observations in Austria some volunteers and maize plants were observed in non-agricultural habitats.[20] In the Netherlands, the appearance of volunteers is very rare, although maize plants occasionally have been observed outside agricultural fields.[21] COGEM is not aware of any reports of feral maize populations in the Netherlands or elsewhere in Europe.

Conclusion: In the Netherlands, feral maize populations do not occur and hybridisation of maize with other species is not possible.

3.2 Description of the introduced genes and traits

In GM maize MZIR098 three expression-cassettes have been introduced. The cassettes contain the *ecry3.1Ab*, *mcry3A* and *pat-08* genes.

The *mcry3A* gene is a modified *cry3A* gene from *Bacillus thuringiensis* subsp. *tenebrionis*, which was optimized to adapt to the preferred codon usage for maize. Additionally, a consensus cathepsin G protease recognition site was incorporated. The modification provides an enhanced insecticidal toxicity, particularly to *Diabrotica* species (corn rootworm).

The *ecry3.1Ab* gene consists of a fusion between the 5′ end of the *mcry3A* gene and the 3′ end of a synthetic *cry1Ab* gene. The original *cry1Ab* gene was derived from *Bacillus thuringiensis* subsp. *kurstaki*. Its sequence was optimized to adapt to the preferred codon usage for maize. At its 5′ end, the *ecry3.1Ab* gene carries a 67-bp oligomer extension. The next 459 encoded amino acid residues are identical to those of mCry3A, followed by 172 amino acid residues identical to those of Cry1Ab.

The *pat-08* gene was obtained from *Streptomyces viridochromogenes* and codon-optimized to enhance expression. Furthermore, the *Xma*I restriction enzyme site was removed.

Introduced genes	**Encoded proteins**	**Traits**
ecry3.1Ab	A chimera of a variant of the Cry3A protein from *B. thuringiensis* subsp. *tenebrionis* (mCry3A),[22,23,24] and the Cry1Ab protein from *B. thuringiensis* subsp. *kurstaki* [22,25,26,27]	Resistance against certain coleopteran insects
mcry3A	A variant of the Cry3A protein from *B. thuringiensis* subsp. *tenebrionis* [22,23,24]	Resistance against certain coleopteran insects
pat-08	A variant of the phosphinothricin N-acetyltransferase (PAT) enzyme from *S. viridochromogenes* strain Tü 494[28,29]	Tolerance to glufosinate-ammonium containing herbicides
For a detailed description of the traits see references		

3.3 Molecular characterisation

Southern blot analyses showed that MZIR098 contains one copy of the insert at a single integration locus and demonstrated the absence of plasmid pSYN17629 backbone sequences.

The applicant determined the sequence of the maize MZIR098 insert and the adjacent flanking regions, and confirmed that no rearrangements or base pair (bp) changes occurred within the insert. The right border of the MZIR098 insert along with 10 bp of non-coding sequence and 10 bp from the left border are deleted. The remainder of the MZIR098 insert is intact. According to the applicant no effect on the functionality of the transgenes is expected, since no functional elements were deleted.

A sequence comparison with the conventional maize control revealed that at the insertion site 24 bp are deleted from the maize genome. According to the applicant, bioinformatic analyses indicate that 1000 bp upstream and 1000 bp downstream of the insert no endogenous maize genes are present. According to the applicant it is therefore unlikely that endogenous genes are disrupted at the MZIR098 insertion site.

The sequences spanning the insertion site at the 5' and 3' flanking regions and the entire insert were screened for potential newly created open reading frames (ORFs) from stop to stop codon. According to the applicant, no significant sequence similarities between the putative products of

these ORFs and any known or putative allergenic proteins or mammalian toxin proteins were detected in these bioinformatics analyses. The molecular characterisation was conducted according to the criteria previously laid down by COGEM.[30]

Conclusion: The molecular characterisation of maize MZIR098 is adequate and no indications for potential environmental risks were identified.

3.4 Phenotypic and agronomic characteristics

The applicant analysed the phenotypic and agronomic characteristics of MZIR098. The introduced traits do not give reason to assume that MZIR098 has an altered survivability compared to non-GM conventional counterparts and commercial reference maize hybrids under natural conditions. Therefore, COGEM is of the opinion that there are no indications that the introduced traits allow maize MZIR098 to survive or establish in the Dutch environment.

Conclusion: MZIR098 does not have an increased potential for the establishment of feral populations in the Netherlands.

4. Food/feed assessment

This application is submitted under Regulation (EC) 1829/2003, therefore a food/feed assessment is carried out by EFSA and national organisations involved in the assessment of food safety. In the Netherlands, RIKILT carries out a food and/or feed assessment for Regulation (EC) 1829/2003 applications. The outcome of the assessment by other organisations (EFSA, RIKILT) was not known when this advice was completed.

5. Post-market environmental monitoring (PMEM)

The applicant supplied a general surveillance plan as part of the PMEM. COGEM has published several recommendations for further improvement of the general surveillance (GS) plan,[31,32] but considers the current GS plan adequate for the import and processing of maize MZIR098.

6. Overall conclusion

COGEM is of the opinion that import and processing of maize MZIR098 poses a negligible risk to the environment in the Netherlands. COGEM abstains from giving advice on the potential risks of incidental consumption since other organisations carry out a food/feed assessment.

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