Import and processing of genetically modified maize MON89034x1507xNK603xDAS-40278-9

COGEM advice CGM/170131-01

- The present application (EFSA/GMO/NL/2013/112) concerns the authorisation for import and processing for use in feed and food of genetically modified (GM) maize MON89034x1507xNK603xDAS-40278-9;
- Maize MON89034x1507xNK603xDAS-40278-9 expresses the *cry1A.105, cry2Ab2* and *cry1F* genes conferring resistance to certain lepidopteran insects, and the *pat, cp4 epsps, cp4 epsps L214P* and *aad-1* genes conferring tolerance to glyphosate, glufosinate-ammonium, and certain 'aryloxyphenoxypropionate' (AOPP) containing herbicides and auxin acting herbicides;
- COGEM advised positively on the import and processing of the parental lines MON89034, 1507, NK603 and DAS-40278-9 in 2009 (MON89034), 2003 (1507 and NK603) and 2011 (DAS-40278-9);
- In the Netherlands, feral maize populations have never been observed and the appearance of volunteers is rare;
- In the Netherlands, wild relatives of maize have never been observed and hybridisation of maize with other species is therefore not possible;
- The molecular characterisation of maize MON89034x1507xNK603xDAS-40278-9 meets the criteria of COGEM;
- There are no reasons to assume that the introduced traits will allow GM maize MON89034x1507xNK603xDAS-40278-9 to survive in the Dutch environment;
- There are no indications that the introduced traits altered the fitness of maize MON89034x1507xNK603xDAS-40278-9;
- The updated molecular characterisation does not give any indication of a potential environmental risk;
- COGEM is of the opinion that import and processing of maize MON89034x1507x NK603xDAS-40278-9, or potential subcombinations in segregating progeny, 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/NL/2013/112), filed by Dow AgroScience LLC, concerns import and processing of genetically modified (GM) maize MON89034x1507xNK603xDAS-40278-9, for use in feed and food.

Maize MON89034x1507xNK603xDAS-40278-9 is produced by conventional crossbreeding of four GM parental maize lines. The stacked line contains the *cry1A.105*, *cry2Ab2* and *cry1F* genes conferring resistance to certain lepidopteran insects. This maize line also contains the genes *pat*, *cp4 epsps*, *cp4 epspsL214P*, and *aad-1*, conferring tolerance to certain glyphosate, glufosinate-ammonium, and 'aryloxyphenoxypropionate' (AOPP) containing herbicides and certain auxin acting herbicides.

Parental lines MON89034[1], 1507[2,3] and NK603[4], have been authorized for import and processing for use in food and feed in the European Union since 2004 (NK603), 2006 (1507) and 2009 (MON89034). [5] In 2016, EFSA issued a positive opinion on the import and processing of maize line DAS-40278-9[6]. Several stacked lines have also been authorized for import and processing for use in food and feed in the European Union.[7,8,9]

2. Previous COGEM advice

COGEM has previously advised on the import and processing of the parental lines MON89034[10,11], 1507[12,13], NK603[14] and DAS-40278-9[15*], and combinations thereof (MON89034xNK603[16,17], MON89034x1507xNK603[18], and 1507xNK603[19]). The environmental risks of import and processing of the parental and above-mentioned stacked maize lines were considered negligible.[11,12,13,14,15,17, 18,19]

3. Environmental risk assessment

MON89034x1507xNK603xDAS-40278-9 maize contains four inserts. These inserts and the traits they encode may segregate in the progeny of the GM maize line. As a result, the imported kernels (seeds) of this GM maize line may possess all traits that are present in MON89034x1507xNK603xDAS-40278-9 or a combination thereof. Potential risks of MON89034x1507xNK603xDAS-40278-9 maize kernels, and segregates containing subcombinations of this stacked line, are assessed as part of the environmental risk assessment.

3.1 Aspects of the wild-type crop

Maize (*Zea mays*) is a member of the grass family *Poaceae*. Maize is a highly domesticated crop originating from Central America, but nowadays it is cultivated globally. Maize is predominantly wind pollinated.[20,21] Maize has both male and female flowers that are spatially separated. Female flowers are not attractive to insect pollinators, because they do not produce nectar. As insects do not visit the female flowers, insect pollination of maize is limited. [22]

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.[22,23] After ripening, the kernels remain on the cob and do not shatter naturally.[22,24] In cultivation areas with warmer climatic conditions, the appearance of volunteers can occur the year following maize cultivation due to spilled cobs or kernels. However, these volunteers are usually

[*] In the 2011 COGEM advice, the maize line DAS-40278-9 was incorrectly named DAS-40728-9.

killed by common mechanical pre-planting soil preparation practices.[22] In the Netherlands, the appearance of volunteers is very rare to absent.[25]

Maize is very sensitive to weed competition.[26] During the long process of domestication, maize has lost the ability to survive in the wild.[21] Establishment of maize plants in the wild has never been observed in the Netherlands and COGEM is not aware of any reports of feral maize populations 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 Molecular characterisation

Previously, COGEM evaluated the molecular characterisation of each parental line and considered them adequate.[11,12,13,14,15]

For each parental line, the applicant updated the molecular characterisation and the bioinformatic analyses. According to the applicant, no endogenous genes were disrupted at the insertion site or in the genomic DNA flanking the insertion site, and no biologically meaningful protein sequence similarities with allergens or toxic proteins were detected in these analyses. The molecular characterisation was conducted according to the criteria previously laid down by COGEM.[27]

The applicant states that for parental line 1507 a significant amino acid alignment was found with the Mas1 protein (agropine synthesis reductase) from *Agrobacterium.* According to the applicant, the alignment was limited to half of the Mas1 protein. If expressed, the protein would therefore be incomplete and unlikely to be active. In view of this, and the long history of safe use of maize 1507, COGEM is of the opinion that this finding does not affect the outcome of the environmental risk assessment.

Conclusion: The molecular characterisation of maize MON89034x1507xNK603xDAS-40278-9 is adequate and no indications for potential environmental risks were identified.

3.3 Description of the introduced genes and traits

Introduced genes	**Encoded proteins**	**Traits**
cp4 epsps (two copies)	The 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) enzyme originating from *Agrobacterium tumefaciens* strain CP4[14]	Tolerance to glyphosate containing herbicides, because of a decreased binding affinity for glyphosate
cp4 epsps L214P	A variant of the EPSPS enzyme from *A. tumefaciens* strain CP4[14]	Tolerance to glyphosate containing herbicides, because of a decreased binding affinity for glyphosate
pat	Phosphinothricin N-acetyltransferase (PAT) enzyme originating from *Streptomyces viridochromogenes* strain Tü 494[12,13]	Tolerance to glufosinate-ammonium containing herbicides

aad-1	Aryloxyalkanoate dioxygenase (AAD-1) enzyme originating from *Sphingobium herbicidovorans*[15]	This enzyme can cleave several synthetic auxins, and certain 'aryloxyphenoxy-propionate' (AOPP) herbicides, resulting in a tolerance to several synthetic auxin acting herbicides like 2,4-dichlorophenoxoxyacetic acid' (2,4-D) and to AOPP containing herbicides
cry1A.105	The Cry1A.105 protein is a chimeric protein with domains from different Cry1 proteins from *Bacillus thuringiensis*.[10,11]	Resistance to certain lepidopteran insects
cry2Ab2	Variant of the Cry2Aa protein from *B. thuringiensis* subsp. *kurstaki*[10,11]	Resistance to certain lepidopteran insects
cry1F	The Cry1F protein originating from *B. thuringiensis* subsp. *aizawa*[12,13]	Resistance to certain lepidopteran insects
For a detailed description of the traits see references.		

3.4 Phenotypic and agronomic characteristics

Previously, COGEM evaluated the phenotypic and agronomic characteristics of each parental line of MON89034x1507xNK603xDAS-40278-9, and found no deviations influencing the outcome of the environmental risk assessment.[10,15,28,29]

The applicant analysed the phenotypic and agronomic characteristics of MON89034x1507x NK603xDAS-40278-9. The introduced traits do not give reason to assume that the parental lines have an altered survivability compared to conventional maize. The applicant noted a difference in plant height between the stacked line and the isoline and a likely difference between the stacked line and reference varieties. However, this is just one of many characteristics that determine plant fitness. Therefore, COGEM is of the opinion that there are no indications to assume that the introduced traits in MON89034x1507x NK603xDAS-40278-9 allow maize to survive or establish in the Dutch environment.

Conclusion: MON89034x1507xNK603xDAS-40278-9 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 new general surveillance plan as part of the PMEM. COGEM has published several recommendations for further improvement of the general surveillance (GS) plan[30,31] but considers the current GS plan adequate for import and processing of maize MON89034x1507xNK603xDAS-40278-9.

6. Overall conclusion

COGEM is of the opinion that import and processing of maize MON89034x1507x NK603xDAS-40278-9, and potential subcombinations in segregating progeny, 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.

References

1. European Commission (2009). Commission Decision of 30 October 2009 authorising the placing on the market of products containing, consisting of, or produced from genetically modified maize MON 89034 (MON-89Ø34-3) pursuant to Regulation (EC) No 1829/2003 of the European Parliament and of the Council (2009/813/EC). Official Journal of the European Union 5.11.2009 L 289/21-24
2. European Commission (2006). Commission Decision of 3 March 2006 authorising the placing on the market of food containing, consisting of, or produced from genetically modified maize line 1507 (DAS-Ø15Ø7-1) pursuant to Regulation (EC) No 1829/2003 of the European Parliament and of the Council (2006/197/EC). Official Journal of the European Union 9.3.2006 L 70/82-86
3. European Commission (2005). Commission Decision of 3 November 2005 concerning the placing on the market, in accordance with Directive 2001/18/EC of the European Parliament and of the Council, of a maize product (*Zea mays* L., line 1507) genetically modified for resistance to certain lepidopteran pests and for tolerance to the herbicide glufosinate-ammonium (2005/772/EC). Official Journal of the European Union 5.11.2005 L 291/42-44
4. European Commission (2015). Commission Implementing Decision of 24 April 2015 authorising the placing on the market of genetically modified maize NK603 (MON-ØØ6Ø3-6) and renewing the existing maize NK603 (MON-ØØ6Ø3-6) products, pursuant to Regulation (EC) No 1829/2003 of the European Parliament and of the Council (EU/2015/684). Official Journal of the European Union 30.4.2015 L 112/6-10
5. International service for the acquisition of agri-biotech applications (ISAAA). GM approval database. http://www.isaaa.org/gmapprovaldatabase/default.asp (bezocht 26 januari 2017)
6. European Food Safety Authority (EFSA) (2016). Scientific Opinion on an application by DOW AgroSciences LLC (EFSA-GMO-NL-2010-89) for placing on the market the genetically modified herbicide-tolerant maize DAS-40278-9 for food and fee d uses, import and processing under Regulation (EC) No 1829/2003. The EFSA Journal 14(12):4633
7. European Commission (2010). Commission Decision of 28 July 2010 authorising the placing on the market of products containing, consisting of, or produced from genetically modified maize MON89034xNK603 (MON-89Ø34-3xMON-ØØ6Ø3-6) pursuant to Regulation (EC) No 1829/2003 of the European Parliament and of the Council (2010/420/EU). Official Journal of the European Union 29.7.2010 L 197/15-18
8. European Commission (2007). Commission Decision of 24 October 2007 authorising the placing on the market of products containing, consisting of, or produced from genetically modified maize 1507xNK603 (DAS-Ø15Ø7-1xMON-ØØ6Ø3-6) pursuant to Regulation (EC) No 1829/2003 of the

European Parliament and of the Council (2007/703/EC). Official Journal of the European Union 31.10.2007 L 285/47-51

9. European Commission (2013). Commission Implementing Decision of 6 November 2013 authorising the placing on the market of products containing, consisting of, or produced from genetically modified maize MON89034 × 1507 × NK603 (MON-89Ø34-3 × DAS-Ø15Ø7-1 × MON-ØØ6Ø3-6) pursuant to Regulation (EC) No 1829/2003 of the European Parliament and of the Council (2013/648/EU). Official Journal of the European Union 13.11.2013 L 302/38-43
10. COGEM (2007). Import and processing of maize MON89034. COGEM advise CGM/071022-02
11. COGEM (2009). Molecular characterization of maize MON89034. COGEM advise CGM/090126-01
12. COGEM (2003). Insect resistant and glufosinate ammonium tolerant transformation event 1507 maize. COGEM advise CGM/030115-01 [in Dutch]
13. COGEM (2015). Renewal of the authorization for import and processing of genetically modified maize line 1507. COGEM advise CGM/150928-01
14. COGEM (2003). Markttoelating 'NK603 maize tolerant to glyphosate'. COGEM advise CGM/030319-08 [in Dutch]
15. COGEM (2011). Import and processing of genetically modified maize DAS-40728-9. COGEM advice CGM/110510-01
16. COGEM (2007). Import and processing of MON89034xNK603. COGEM advise CGM/071022-03
17. COGEM (2009). Additional advice on the import and processing of MON89034xNK603. COGEM advise CGM/091020-01
18. COGEM (2009). Import and processing of genetically modified MON89034x1507xNK603 maize. COGEM advice CGM/090930-01
19. COGEM (2005). Import and processing of insect resistant and herbicide tolerant maize 1507xNK603. COGEM advise CGM/050526-01
20. Hin CJA (2001). Landbouwkundige risico's van uitkruising van GGO-gewassen. Centrum voor Landbouw en Milieu (CLM)
21. Treu R & Emberlin J (2000). Pollen Dispersal in the Crops Maize (*Zea mays*), Oil Seed Rape (*Brassica napus* ssp. *oleifera*), Potatoes (*Solanum tuberosum*), Sugar Beet (*Beta vulgaris* ssp. *vulgaris*) and Wheat (*Triticum aestivum*). Evidence from Publications. Soil Association
22. Andersson M & Carmen de Vicente M (2010). Gene flow between crops and their wild relatives. The John Hopkins University Press, Baltimore, Maryland, The United States of America
23. Miedema P (1982). The effect of low temperature on *Zea mays*. Advances in Agronomy 35: 93-128
24. Organisation for Economic Cooperation and Development (OECD) (2003). Consensus Document on the Biology of *Zea mays* ssp. *mays* (Maize)
25. van de Wiel CCM *et al*. (2011). Crop volunteers and climate change. Effects of future climate change on the occurrence of maize, sugar beet and potato volunteers in the Netherlands. COGEM research report 2011-11
26. CAB International (2007). Crop Protection Compendium. *Zea mays* (maize). CD-ROM edition, Wallingford

27. COGEM (2014). Reconsideration of het molecular characterisation criteria for marketing authorisation of GM crops. COGEM topic report CGM/140929-02
28. COGEM (2006). Cultivation of herbicide tolerant maize line NK603. COGEM advise CGM/060704-01
29. COGEM (2003). Insect resistant and glufosinate ammonium tolerant transformation event 1507 maize. COGEM advise CGM/030919-04 [in Dutch]
30. COGEM (2010). General Surveillance. COGEM report CGM/100226-01
31. COGEM (2015). Advice on improving the general surveillance of GM crops. COGEM advise CGM/150601-02