



ASSESSMENT REPORT

NOTIFICATION C/ES/00/01 CONCERNING THE PLACING ON THE MARKET OF GENETICALLY MODIFIED MAIZE (NK603 LINE) WITH TOLERANCE TO GLYPHOSATE HERBICIDE, APPLIED BY MONSANTO COMPANY PURSUANT OF COUNCIL DIRECTIVE 2001/18/EC.

Introduction

1. The attached file for the placing on the market of NK603 maize tolerant to glyphosate herbicide, proposed by Monsanto Europe, S.A. contains the information required under Article 13 and Annexes II, IIIB, IV and VII of Directive 2001/18/CE with the risk assessment and appendices which contain the studies on this transgenic plant and its safety for human and animal health or the environment.
2. From the risk evaluation completed it is concluded that there is no reason to believe that there will be any adverse effects on human and animal health or the environment from the import, production, and processing of glyphosate tolerant NK603 maize.
3. Analysis of grain from NK603 maize, as well as the safety evaluation of the introduced proteins, its composition, and the safety of the grain as feed for animals have been completed.

Product description

4. The product is described as:

"Maize (*Zea mays*) line NK603, genetically modified to be tolerant to glyphosate herbicide. The intended uses of this maize are the same as for any other maize, excluding the cultivation in the European Union of varieties derived from maize NK603. This product has been obtained by the introduction of a glyphosate-tolerant 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) gene from *Agrobacterium* sp. strain CP4 (CP4 CPSPS) “.

5. The insertion of the genetic material was performed by particle acceleration technology with a *Mlu*I restriction fragment, isolated in agarose gel from PV-ZMGT32L plasmid, used as vector. This fragment contains two EPSPS genes, derived from *Agrobacterium* sp. strain CP4.

Purpose of the placing on the market

6. Import and processing of NK603 Roundup Ready maize tolerant to glyphosate herbicide and for its use as any other maize, excluding the cultivation in the European Union of varieties derived from maize event NK603. The primary use of maize is for animal feed and it is also processed into valuable food and industrial products. Traders and processors within the European Union will distribute the grain and products from this line.



7. The use of this modified maize for human food is outside the scope of this notification.

ENVIRONMENTAL RISK ASSESSMENT

8. This Application has been evaluated by the Spanish Commission on Biosafety. From the study of the dossier and the evaluation of the resulting total risk associated with the genetically modified maize by import, transport and processing, including use in animal feed, of the grain or other products, it is concluded that the risk of NK603 maize is not significant for human health or the environment.

9. The main aspects considered to evaluate potential associated risks were:

- Capacity to survive, establish and disseminate.
- Potential for gene transfer.
- Genetic and phenotypic stability.
- Expressed products from the inserted sequences.
- Potential adverse effects for human beings and animals: toxic and allergenic effects.
- Interactions with other organisms.

10. *Capacity to survive, establish and disseminate (Increased persistence or invasiveness):*

The introduced trait has not affected the reproductive plant morphology so a change in the establishment, vigour and competitiveness of NK603 line in relation to other commercial varieties is not expected. Except the glyphosate herbicide tolerance, NK603 maize does not show differences in capability of survival with respect to conventional maize.

Zea mays does not behave as an invasive crop because of inherent characteristics of this species such as lack of dormancy, short persistence of the seed in the soil, and frost sensitivity of maize seedlings. In case volunteer maize seedlings appear in cultivated plots, they can be easily controlled with other non-selective herbicides or with tillage.

In the case of this application, the cultivation in the EU of varieties derived from maize event NK603 is not included, and the environmental release could occur during transport, storage and processing of this maize.

Taking into account that modern methods of grain handling minimise losses of grain, the low likelihood of misuse by growers in the EU, and the characteristics of this crop above mentioned, the estimation of the risk of increased persistence or invasiveness is considered as effectively zero.

11. *Potential for gene transfer:*

In the case of this notification, since it does not include the cultivation of NK603, likelihood for gene transfer to other maize crops is extremely low, and in the case of unintentional release of grains, the risk posed by the transfer of the glyphosate tolerance gene is considered that it would have negligible effects for the environment. For the gene flow to occur, it will depend on wind, moisture and temperature



conditions. The existence of natural maize populations in Europe is not known, and the gene transfer to other species is considered remote.

12. *Genetic and phenotypic stability:*

Molecular analysis, expression data, and progeny segregation over nine generations of line NK603 have demonstrated:

- The genome of NK603 maize contains a single insertion of the integrated DNA;
- This insert contains one copy of the DNA fragment used in transformation (PV-ZMGT32L), which contains two *cp4 epsps* gene cassettes;
- ;
- The genome of NK603 maize line does not contain any detectable plasmid backbone DNA fragments, including *ori* and the *nptII* coding sequences,
- The insert is inherited as a single dominant gene in a Mendelian fashion.

13. *Expressed products from the inserted sequences:*

The insert products of NK603 maize are CP4 EPSPS and CP4 EPSPS L214P proteins. Two nucleotide changes have occurred in the second of the two *cp4 epsps* encoding regions of the plant insert compared with the plasmid, one of which is silent and the other resulting in a single amino acid change in the expressed protein, indicated by the suffix L214P. The expression of the CP4 EPSPS proteins is expected to occur throughout the whole plant since the rice actin and CaMVe35S promoters have been shown to drive constitutive expression of the encoded protein in this genetically modified maize. The amino acid sequence of CP4 EPSPS proteins is comparable to EPSPS enzymes which are commonly found in a wide variety of food and feed sources and which have a long history of safe use.

14. *Potential adverse effects for human beings and animals:*

NK603 maize is substantially equivalent to traditional maize except for the introduced trait, which confers tolerance to the Roundup herbicide. The known function and ubiquity of EPSPS proteins represent a history of safe use for animal and human consumption, as occurs with the CP4 EPSPS protein expressed in the glyphosate tolerant soybean placed on the market in the European Union since 1996. Regarding CP4 EPSPSL214P protein it has been demonstrated that it is structurally and functionally equivalent to the CP4 EPSPS protein.

Acute toxicity studies in mice, rapid degradation of the CP4 EPSPS proteins and loss of enzymatic activity in simulated human gastric and intestinal fluids, as well as lack of amino acid sequence similarity to toxins and allergens, demonstrate that the likelihood of potential adverse effects to humans and animals exposed or consuming NK603 maize, is negligible.

Compositional analysis of NK603 maize grain has demonstrated substantial equivalence with traditional maize. This is supported by lack of differences shown in the results of feeding NK603 to broiler chickens compared with birds fed with NK603 parental line (B73HTxLH82) or with five commercial reference



lines, as well as repeated dose feeding studies in rats, which contribute to determine the composition and nutritional equivalence.

15. *Interactions with other organisms:*

Since the intended use of NK603 maize is for import and use in the European Union, including feed use but not including the planting of NK603 maize varieties, no environmental impacts are expected. Nevertheless, regarding unintentional release and possible adverse effects on non-target organisms, in different deliberate release field trials carried out in Spain and in other countries, no significant differences have been observed on herbivore insects, insect and fungal pests of this glyphosate tolerant maize.

Identification and labelling:

16. Bulk mixtures of maize grain import: the words “Contains genetically modified organisms” shall appear either on a label or in an accompanying document to the maize grain shipment.

17. All relevant grain traders will be provided with the commercial name of the product, as well as with the unique identifier to comply with the requirements of Directive 2001/18/EC, and any other relevant information of the product, including the procedures for accessing the European public registers of GMOs. The Unique Identifier proposed by Monsanto for NK603 maize is **MON-00603-6**, which has already been notified to the United States Department of Agriculture (USDA) in order to be included in the OECD “Biotrack Product Database”.

Monitoring Plan

18. Taking into account that the intended use of the placing on the market of this product is the import and processing, including feed use, the monitoring plan is based on the identification, surveillance and traceability of NK603 maize grain, providing information to traders and processors of bulk mixtures of maize grain, in order to identify any adverse effects on the environment or human health. Furthermore, it is foreseen to inform the European feed industry directly, as well as, to inform the interested operators about the safety and general characteristics of the product. Operators in the feed chain will be requested to inform the relevant authorities of any adverse effects on animal health reported to them through farmers or national feed associations, which they consider to be attributed to the feed use of NK603 maize grain.

19. **Conclusion:** The Spanish Biosafety Commission, after examining and considering the existing information and the data provided by Monsanto Company, estimates that for the uses considered and at the current scientific and technical “State of the art”, there is no scientific evidences which indicate any risk for human and animal health or the environment of NK603 maize.