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Scientific Opinion of the Panel on Genetically Modified Organisms on application (Reference EFSA-GMO-UK-2005-20) for the placing on the market of the insect-resistant and herbicide-tolerant genetically modified maize 59122 x NK603, for food and feed uses, and import and processing under Regulation (EC) No 1829/2003 from Pioneer Hi-Bred International[1]

(Question No EFSA-Q-2005-247)

Opinion adopted on 19 November 2008

GMO PANEL MEMBERS

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SUMMARY

Following a request from Pioneer Hi-Bred International within the framework of Regulation (EC) No 1829/2003 on genetically modified food and feed (EC, 2003) for food and feed uses, import and processing, the Panel on Genetically Modified Organisms was asked to deliver a scientific opinion on the authorisation of the insect-resistant, glyphosate- and glufosinate-tolerant genetically modified 59122 x NK603 maize (Unique Identifier DAS-59122-7xMONØØ6Ø3-6).

In delivering its scientific opinion, the GMO Panel considered the new application EFSA-GMO-UK-2005-20, additional information provided by the applicant (Pioneer Hi-Bred International) and the scientific comments submitted by the Member States. Further information from applications for placing the parental events 59122 and NK603 on the market

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under EU regulatory procedures was taken into account where appropriate. The scope of application EFSA-GMO-UK-2005-20 is for food and feed uses, import and processing of genetically modified 59122 x NK603 maize and all derived products, but excluding cultivation in the EU.

The GMO Panel assessed 59122 x NK603 maize with reference to the intended uses and the appropriate principles described in the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants and derived food and feed and the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants containing stacked transformation events. The scientific assessment included molecular characterization of the inserted DNA and expression of the new proteins. A comparative analysis of agronomic traits and composition was undertaken and the safety of the newly expressed proteins and the whole food/feed was evaluated with respect to potential toxicity, allergenicity and nutritional quality. An assessment of environmental impacts and the post-market environmental monitoring plan were also undertaken.

Maize 59122 was developed to express CRY34Ab1 and CRY35Ab1 proteins rendering maize 59122 resistant to certain coleopteran pests and the PAT (phosphinothricin-N-acetyltransferase) protein which was used as a selectable marker and confers tolerance to glufosinate. Maize 59122 was authorised under Regulation (EC) No 1829/2003 with Commission Decision 2007/702/EC. Maize NK603 was developed to be tolerant to glyphosate by the introduction of the gene coding for 5-enolpyruvylshikimate-3-phosphate synthase from *Agrobacterium* sp. strain CP4 (CP4 EPSPS and CP4 EPSPS L214P). Maize NK603 has received an opinion in favour of its authorisation and was authorised under Directive 2001/18/EC by Commission Decision 2004/643/EC. The use of food and food ingredients from NK603 maize was authorised under Regulation (EC) No 258/97 by Commission Decision 2005/448/EC.

59122 x NK603 maize was produced by crosses between maize inbred lines containing 59122 and NK603 events to combine resistance to certain coleopteran species trait in 59122 and the tolerance to glyphosate in NK603.

The molecular characterisation data established the molecular equivalence and identical copy number of the two inserts present in maize 59122 x NK603 to each single insert present in maize 59122 and maize NK603. Appropriate analyses of the integration sites including the flanking regions in maize 59122 x NK603 and updated bioinformatics analysis of the single events have been performed. Bioinformatics analysis of junction regions in the single events demonstrated the absence of any potential new ORFs coding for known toxins or allergens. The expression of the genes introduced by genetic modification has been sufficiently analysed and the stability of the genetic modification has been demonstrated over several generations. The GMO Panel is of the opinion that the molecular characterisation of the DNA inserts and

flanking regions of maize 59122 x NK603 does not raise any safety concern, and that sufficient evidence for the stability of the genetic modification was provided.

Based on the results of comparative analysis it was concluded that 59122 x NK603 maize is compositionally and agronomically equivalent to its non-GM counterpart and conventional maize, except for the presence of CRY34Ab1, CRY35Ab1, CP4 EPSPS, CP4 EPSPS L214P and PAT proteins in 59122 x NK603 maize. Based on the assessment of data available, including the additional information provided by the applicant in response to the Panel request, for 59122 x NK603 maize, for the single events and for appropriate non-GM controls, the GMO Panel does not see a reason to assume that crossing of NK603 maize and 59122 maize results in an interaction of the newly expressed proteins which causes compositional or agronomic changes. The GMO Panel concluded that the maize 59122 x NK603 is as safe as its non-GM counterpart and that the overall allergenicity of the whole plant is not changed.

A higher stack of 59122 x NK603 maize, 59122 x 1507 x NK603 maize has been studied in a nutritional feeding study with broilers and showed its nutritional equivalence with conventional maize lines. The Panel concluded that this study provides additional evidence that the nutritional properties of maize 59122 x NK603 would be no different from those of the conventional maize.

The application EFSA-GMO-UK-2005-20 concerns food and feed uses, import and processing, but excluding cultivation in the EU. There are no indications of increased likelihood of establishment or survival of feral maize plants in case of accidental release into the environment of 59122 x NK603 seeds during transportation and processing for food and feed uses. Taking into account the scope of the application, both the rare occurrence of sporadic feral plants and the low levels of exposure through other routes indicate that the risk to target and non-target organisms is negligible. The scope of the monitoring plan provided by the applicant is in line with the intended uses of maize 59122 x NK603 since the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects. Furthermore the GMO Panel agrees with the reporting intervals proposed by the applicant in the general surveillance plan.

In conclusion, the Panel considers that the information available for 59122 x NK603 maize addresses the scientific comments raised by the Member States and that it is as safe as its non genetically modified counterpart with respect to potential effects on human and animal health or the environment. Therefore the GMO Panel concludes that 59122 x NK603 is unlikely to have any adverse effect on human or animal health or on the environment in the context of its intended uses.

Key words: GMO, maize, 59122, NK603, glufosinate, glyphosate, human and animal health, environment**,** import, processing, food, feed, Regulation (EC) No 1829/2003, CRY34Ab1, CRY35Ab1, PAT, CP4 EPSPS, CP4 EPSPS L214P.

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BACKGROUND AS PROVIDED BY REGULATION EC (NO) 1829/2003

On 19 September 2005, EFSA received from the Competent Authority of the United Kingdom an application (Reference EFSA-GMO-UK-2005-20), for authorisation of the glyphosate and glufosinate-tolerant insect-resistant genetically modified 59122 x NK603 maize (Unique Identifier DAS-59122-7xMONØØ6Ø3-6), submitted by Pioneer Hi-Bred International within the framework of Regulation (EC) No 1829/2003 on genetically modified food and feed (EC, 2003) for food and feed uses, import and processing.

After receiving the application EFSA-GMO-UK-2005-20 and in accordance with Articles 5(2)(b) and 17(2)b of Regulation (EC) No 1829/2003, EFSA informed the Member States as well as the European Commission and made the summary of the dossier publicly available on the EFSA website. EFSA initiated a formal review of the application to check compliance with the requirements laid down in Articles 5(3) and 17(3) of Regulation (EC) No 1829/2003. On 19 June 2007 EFSA received additional information requested under completeness check and on 20 June 2007 EFSA declared the application as valid in accordance with Articles 6(1) and 18(1) of Regulation (EC) No 1829/2003.

EFSA made the valid application available to Member States and the European Commission and consulted nominated risk assessment bodies of the Member States, including the national Competent Authorities within the meaning of Directive 2001/18/EC (EC, 2001) following the requirements of Articles 6(4) and 18(4) of Regulation (EC) No 1829/2003, to request their scientific opinion. The Member State bodies had three months after the date of receipt of the valid application (until 20 September 2007) within which to make their opinion known.

The GMO Panel carried out a scientific assessment of genetically modified (GM) 59122 x NK603 maize taking into account the appropriate principles described in the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants and derived food and feed (EFSA, 2006a) and the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants containing stacked transformation events (EFSA 2007d).

On 16 November 2007 and 20 June 2008, the GMO Panel asked for additional data on 59122 x NK603 maize. The applicant provided the requested information on 2 April 2008 and 2 September 2008 respectively, and spontaneous information on 14 April 2008. After receipt and assessment of the full data package, the GMO Panel finalized its risk assessment of 59122 x NK603 maize.

The GMO Panel carried out a scientific assessment of the GM 59122 x NK603 maize for food and feed uses, import and processing in accordance with Articles 6(6) and 18(6) of Regulation (EC) No 1829/2003, taking into consideration the scientific comments of the Member States

and the additional information provided by the applicant. Further information from applications for placing the single insert lines on the market under EU regulatory procedures, was taken into account where appropriate.

The single events 59122 and NK603 have been the subjects of earlier assessments and have received EFSA opinions in favour of their authorisation (EFSA, 2007b; 2003a,b). Maize 59122 was authorised under Regulation (EC) No 1829/2003 with Commission Decision 2007/702/EC. Maize NK603 was authorised under Directive 2001/18/EC by Commission Decision 2004/643/EC (EC, 2004a). The use of food and food ingredients from NK603 maize was authorised under the Regulation (EC) No 258/97 (EC, 1997) by Commission Decision 2005/448/EC (EC, 2005).

In giving its opinion on GM 59122 x NK603 maize to the European Commission, the Member States and the applicant, and in accordance with Articles 6(1) and 18(1) of Regulation (EC) No 1829/2003, EFSA has endeavoured to respect a time limit of six months from the receipt of the valid application. As additional information was requested by the EFSA GMO Panel, the time-limit of 6 months was extended accordingly, in line with Articles 6(1), 6(2), 18(1), and 18(2) of Regulation (EC) No 1829/2003.

According to Regulation (EC) No 1829/2003, the EFSA opinion shall include a report describing the assessment of the food and feed and stating the reasons for its opinion and the information on which its opinion is based. This document is to be seen as the report requested under Articles 6(6) and 18(6) of that Regulation and thus will be part of the overall opinion in accordance with Articles 6(5) and 18(5).

TERMS OF REFERENCE BY REGULATION EC (NO) 1829/2003

The GMO Panel was requested to carry out a scientific assessment of the genetically modified 59122 x NK603 maize for food and feed uses and import and processing in accordance with Articles 6(6) and 18(6) of Regulation (EC) No 1829/2003. Where applicable, any conditions or restrictions which should be imposed on the placing on the market and/or specific conditions or restrictions for use and handling, including post-market monitoring requirements based on the outcome of the risk assessment and, in the case of GMOs or food/feed containing or consisting of GMOs, conditions for the protection of particular ecosystems/environments and/or geographical areas should be indicated in accordance with Articles 6(5)(e) and 18(5)e of Regulation (EC) No 1829/2003.

The EFSA GMO Panel was not requested to give an opinion on information required under Annex II to the Cartagena Protocol, nor on the proposals for labelling and methods of detection (including sampling and the identification of the specific transformation event in the food/feed and/or food/feed produced from it), which are matters related to GMO risk management.

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ASSESSMENT

1. INTRODUCTION

The genetically modified (GM) 59122 x NK603 maize (Unique Identifier DAS-59122-7xMONØØ6Ø3-6) was assessed with reference to its intended uses, taking account of the appropriate principles described in the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants and derived food and feed (EFSA, 2006a) and the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants containing stacked transformation events (EFSA 2007d). In its evaluation the GMO Panel also considered the scientific comments that were raised by Member States on application EFSA-GMO-UK-2005-20. The risk assessment presented here is based on the information provided in the application relating to 59122 x NK603 maize submitted in the EU including additional information from the applicant and information on the single events.

2. ISSUES RAISED BY MEMBER STATES

All issues raised by MS are addressed in Annex G of the overall opinion.

3. MOLECULAR CHARACTERISATION

3.1. Evaluation of relevant scientific data

The EFSA GMO Panel guidance document (EFSA, 2006a) states that when events have been combined by the interbreeding of existing approved GM lines, the need for further molecular analysis will depend, on a case-by-case basis, on the nature of the genetic modifications involved.

Having considered the information provided in the application and the comments of the Member States, the GMO Panel requested clarification from the applicant with regard to the results from some Southern analysis. Furthermore, additional information was requested on the integrity of the insertion locus in NK603 and 59122, an update of the bioinformatic analyses for the junction regions of 59122 and NK603, on the plant comparators used in field trials for protein expression and on the choice of statistical tools used to conclude on expression levels of the newly expressed proteins.

3.1.1. Method of production of maize 59122 × NK603

Conventional breeding methods were used to produce maize 59122 x NK603 and no new genetic modification was involved. The two inserts that are present in 59122 x NK603 were derived from maize lines containing two independent events: 59122 and NK603. Each of these GM maize events was the subject of an earlier safety evaluation and separate opinions for each of them have been published (EFSA, 2003a,b; 2007b). Maize 59122 x NK603 combines the insect protection and glufosinate tolerance traits from 59122 with the glyphosate tolerance in NK603.

3.1.2. Description of the single events 59122 and NK603

59122

Maize 59122 was transformed by *Agrobacterium*-mediated gene transfer technology and as a result expresses the *cry34Ab1* and *cry35Ab1* genes from *Bacillus thuringiensis* strain PS149B1, conferring resistance against the corn root borer, and the *pat* coding sequence from *Streptomyces viridochromogenes* resulting in tolerance towards glufosinate herbicides. Molecular characterisation data established that maize 59122 contains a single insert of the T-DNA. The structure of the insert in maize 59122 was determined by Southern analysis and DNA sequencing. No vector backbone sequences were detected. BLAST sequence analysis revealed that flanking regions of the maize event 59122 show significant homology to maize genomic DNA and EST sequences. Updated BLASTn and BLASTx analyses indicated that the DNA in 59122 was inserted 1032 bp downstream of the coding region of a maize pentatricopeptide repeat (PPR) protein, the empty pericarp4 (*emp4*). This PPR protein is essential for seed development in maize. In event 59122 seed development is not affected suggesting that expression of *emp4* was not altered by the insertion. Analysis of ORFs

spanning the two junction regions was performed by bioinformatic analysis and no novel ORFs with sequence similarity to known toxins or allergens were identified. This was confirmed by an updated ORF analysis.

Southern analysis of these plants and maintenance of the phenotype indicated genetic and phenotypic stability of the event 59122 over four generations.

NK603

NK603 event was generated by particle bombardment. As a result of the genetic modification NK603 is tolerant to glyphosate herbicides due to the expression of the CP4 *epsps* gene from *Agrobacterium sp*. strain CP4 (CP4 EPSPS and CP4 EPSPS L214P, a variant of CP4 EPSPS containing a proline residue at position 214 instead of leucine).

Molecular analysis showed that NK603 contains a single inserted copy of the DNA present in the construct used for the transformation. This construct contained two adjacent plant gene expression cassettes each consisting of a single copy of the CP4 *epsps* gene with different promoters. The structure of the insert in NK603 was determined by Southern analysis and DNA sequencing. In addition to the two CP4 *epsps* expression cassettes, the insert in NK603 does include some molecular rearrangements at one end of the insert and also includes a fragment of chloroplast DNA. These rearrangements and the insertion of chloroplast DNA do not lead to new traits and are not considered to pose a safety risk. In the unlikely event that a new peptide or protein is produced as a consequence of the insertion event, bioinformatic analysis showed that these would have no homology to known toxins or allergens. An updated BLAST analysis of the flanking DNA sequences indicate that no endogenous open reading frames were disrupted as a consequence of the insertion.

Segregation data for nine generations of line NK603 have demonstrated the stability of the inserted DNA through six generations of crossing and three generations of self-pollination.

3.1.3. Transgenic constructs in the maize 59122 x NK603

59122 x NK603 maize has been obtained by conventional breeding methods between genetically modified 59122 and NK603 maize. No new genetic modification has been introduced in 59122 x NK603 maize. The molecular structures of the DNA inserts present in this maize were investigated using Southern analyses. This involved the use of DNA probes specific for the 59122 and NK603 maize inserts.
In order to confirm the molecular equivalence and identical copy number of the insert present in maize 59122 x NK603 to that present in 59122 maize, samples of genomic DNA were digested with the restriction enzyme *Sac*I and subjected to Southern analysis with the *cry34Ab1* and *cry35Ab1* probes. For comparison with the insert present in NK603, genomic DNA of 59122 x NK603 and NK603 maize plants were digested with the restriction enzyme *Eco*RV

and subjected to Southern analysis with a DNA probe containing the coding region of the CP4 *epsps* gene. Further comparisons were made between maize 59122 x NK603, 59122 and NK603 maize using *Sac*I to digest genomic DNA followed by probing with the *pat* gene. These analyses confirmed the intactness in the hybrid of the 59122 insert (including the right border). Additional information was supplied on the intactness of the left border of the 59122 insert contained in the hybrid by using a 59122 event-specific PCR for the left border junction of the 59122 insert. The intactness of the NK603 insert (including both borders) was also confirmed.

3.1.4. Expression of the introduced genes

Grains for studies of expression levels of the newly expressed proteins Cry34Ab1, Cry35Ab1, PAT, and CP4 EPSPS and CP4 EPSPS L214P were obtained from maize 59122 x NK603 harvested from six field trials in USA and Canada in 2003. Additional expression data from a field trial in 2005 on three locations in Spain were supplied. In this experiment expression levels of the transgenes in roots, leaf, whole plant, stalk, forage grain and pollen in 59122 x NK603 and the respective single events were measured. Plants were either sprayed or unsprayed with glufosinate. The proteins were extracted and quantified using an ELISA (enzyme-linked immunosorbent assay) technique.

Expression levels in grain (ng/mg tissue dry weight) obtained from plants treated with glyphosate and/or glufosinate are summarized in the table below. Levels of all measured proteins were unaffected by herbicide treatment and were in the same range as in the trials in the USA, Canada and Chile.

Table 1. Expression levels in grain (ng/mg tissue dry weight) from plants treated with glyphosate and/or glufosinate.

	59122x NK603		**59122**		**NK603**	
	Mean	Range	Mean	Range	Mean	Range
Cry34Ab1	52	34 - 76	41	30 - 51	NA	NA
Cry35Ab1	2.8	2.1 – 3.7	2.2	1.3 - 3.2	NA	NA
PAT	0.18	0 - 0.46	0.09	0 - 0.18	NA	NA
CP4 EPSPS and CP4 EPSPS L214P	12	7.5 - 15	NA	NA	13	4.6 - 17

NA - Data not available.

3.1.5. Inheritance and stability of inserted DNA

The genetic stability of the inserted DNA in events 59122 and NK603 was demonstrated previously (EFSA, 2003; 2007). In maize 59122 x NK603 the two inserts are combined. The Southern data presented show that the two events are present and the structure of each insert is retained. Furthermore, each of the traits has been conserved in this maize. The GMO Panel is of the opinion that there is no *a priori* reason to expect instability of the transgenes in 59122 x NK603 maize.

3.2. Conclusion

As conventional breeding methods were used in the production of maize 59122 x NK603, no additional genetic modification was involved. Southern and PCR analyses demonstrated that the structures of the 59122 and NK603 events were retained in maize 59122 x NK603. The genetic stability of the integrated DNA has been demonstrated in the single events. Phenotypic analyses demonstrated that the traits were retained in the hybrid.

The expression levels of Cry34Ab1, Cry35Ab1, PAT, and CP4 EPSPS and CP4 EPSPS L214P proteins in maize 59122 x NK603 were measured in grain. Levels of expression of these proteins in grain of 59122 x NK603 have been demonstrated to be comparable with those of the single events.

The Panel concludes that these data do not raise safety concerns.

4. COMPARATIVE ANALYSIS

4.1. Evaluation of relevant scientific data

Having considered the information provided in the application and the Member States comments, the GMO Panel requested from the applicant further information with respect to the compositional analysis data and in particular the statistical differences observed in the levels of amino acids as well as various additional compositional differences. Additional information concerning potential interactions between the newly expressed proteins was also requested by the Panel.

4.2.1. Summary of the previous evaluation of the single events

Maize 59122

59122 maize was compared with non-GM controls with similar genetic background. Whole crops and maize tissues, including kernels, were collected for compositional analysis from field trials. These field trials were carried out during several seasons and at different locations (six locations in Chile (2002-2003), three locations in the USA (2003), two locations in Canada (2003), three locations in Bulgaria (2003 and 2004), and three locations in Spain (2004)). Maize 59122 plants treated with glufosinate-containing herbicide, untreated with glufosinate, and the non-GM control maize were included in these field trials. Based on the results of compositional analysis of samples from a representative range of environments and grown in several seasons, it was concluded that forage and kernels of 59122 maize are compositionally equivalent to those of conventional maize, except for the presence of CRY34Ab1 and CRY35Ab1 and PAT proteins in 59122 maize.

In addition, during field trials over several seasons and at different locations (six locations in Chile (2002-2003), three locations in the USA (2003), two locations in Canada (2003), three locations in Bulgaria (2003), three locations in Spain (2004), and three locations in Bulgaria (2004)) agronomic data did not show indications for unexpected changes of agronomic characteristics and performance (EFSA, 2007b).

Maize NK603

NK603 maize was compared with a non-GM control with genetic background representative of NK603 maize. The material analysed in the comparative analysis was collected from replicated field trials in the USA (year 1998) and Europe (year 1999). NK603 maize treated with glyphosate and NK603 maize untreated with glyphosate were included in the field trials. With the exception of the glyphosate-tolerance, NK603 maize was found to be morphologically and agronomically similar to the non-GM comparator. With regard to compositional equivalence a total of 51 different parameters including proximates, amino- and fatty acids, minerals (Ca, Cu, Fe, K, Mg, Mn, Na, P, Zn), vitamin E, and trypsin inhibitor were analysed. The levels of the chemical constituents in NK603 maize were either within the range found for the non-GM control or within the ranges reported in published literature. No consistent compositional differences requiring further studies were found. Therefore, it was concluded that NK603 maize for food and feed use is compositionally equivalent to conventional maize, except for the presence of the newly inserted proteins in NK603 maize (EFSA, 2003a,b).

4.2.2 Choice of comparator and production of material for the comparative assessment

During field trials 59122 x NK603 maize was compared to non-GM control maize. The comparator had similar background genetic representative for 59122 x NK603 maize. The

pedigree information provided by the applicant for the non-GM control maize showed that the control represented an appropriate comparator for 59122 x NK603 maize in the field trials.

Replicated field trials were carried out in 2003 (6 locations in the US and Canada) and 2004 (5 locations in Spain, Bulgaria and Hungary). 59122 x NK603 maize grown for compositional analysis either received two applications of glyphosate, two applications of glufosinate ammonium, or one application of glyphosate followed by one application of glufosinate-ammonium. Whole crops (forage) and kernels derived from 59122 x NK603 maize and non-GM control maize were collected from field trials for compositional analysis. 59122 x NK603 maize grown for agronomic analysis received one application of glyphosate followed by one application of glufosinate-ammonium. The GMO Panel considered the fact that treatment of the single events with corresponding target herbicides did not affect their agronomic / compositional characteristics compared to untreated plants. Therefore the GMO Panel accepted the design of the field trials although plots untreated with both target herbicides had not been systematically included in the studies.

In addition, data derived from material obtained from field trials with the single events and appropriate comparators were provided by the applicant (see section 3.2.1). The GMO Panel had previously assessed these data and had concluded that 59122 maize and NK603 maize (treated and untreated with the target herbicides) are agronomically and compositionally equivalent to their respective comparators, except for the newly introduced traits. Therefore, non-inclusion of the single events in the field trials with the stacked event was accepted by the Panel.

The GMO Panel considered the studies and the derived spectrum of data which was available for the comparative agronomic and compositional assessment as sufficient.

4.2.3. Compositional analysis

Compositional data were obtained by analysis of forage and kernels harvested from field trials performed in maize growing regions of North America in 2003 and Europe in 2004. Statistical analysis of supplied data was performed on both individual and combined locations. The GMO Panel is of the opinion, that the selection of compounds follows the recommendations of OECD (2003).

The proximate and mineral analyses (fat, protein, total carbohydrate, fibre, ash, phosphorus, and calcium) of forage from 59122 x NK603 maize were compared to forage from the non-GM control and to typical ranges reported in literature for commercial maize (ILSI, 2006; OECD, 2003). The compositional analysis of kernels of maize 59122 x NK603 maize and its control included proximate analyses (fat, protein, ash, moisture, carbohydrates, fibre), fatty acids (palmitic, stearic, oleic, linoleic, and linolenic acid), amino acids (eighteen amino acids

including aromatic amino acids), minerals (calcium, copper, iron, magnesium, manganese, phosphorus, potassium, sodium, selenium and zinc), vitamins (vitamin B1, vitamin B2, folic acid, β-carotene, vitamin E), phytic acid, raffinose, anti-nutrients (trypsin inhibitor) and other constituents (inositol, furfural, p-coumaric acid, and ferulic acid).

The across location analysis of the compositions of forage and kernels from 59122 x NK603 maize and its control revealed statistically significant differences in some parameters which were observed in both seasons and under different herbicide treatment regimes (*i.e.* for phosphorous in forage, crude protein, amino acids, and ash in kernels). However, none of these differences was consistently observed at each location.

The GMO Panel considered the fact that 59122 x NK603 maize combines two traits conferring tolerance to different herbicides targeting amino acid metabolism. On request by the Panel the applicant presented overview tables summarising levels of crude protein and individual amino acids in the stack, the single events and corresponding non-GM comparators. It was demonstrated that crude protein and amino acid levels in the stack fell well within the respective ranges observed for the single events and/or the non-GM controls. Amino acid levels in the stack calculated as percentage of total amino acids were not consistently different compared to the non-GM control.

In general, the levels of those compounds which were different from the level in the corresponding control were within the literature ranges reported for commercial maize varieties. As the comparison of the level of the various key constituents in 59122 x NK603 maize and its non-GM control did not reveal any statistically significant difference for constituents for which a food safety concern could be foreseen, the GMO Panel accepted that none of the field trial sites was replicated the second year.

4.2.4. Agronomic traits and GM phenotype

During field trials at different locations in North America during the 2003 growing season and in Europe during the 2004 growing season, extensive agronomic data (*e.g.* germination/early population, number of emerged plants, ear height, plant height, early population, final population), were collected for 59122 x NK603 maize (treated with glyphosate and glufosinate) and for the corresponding non-GM control. Some statistically significant differences were detected, *e.g.* for early and final population and plant height. None of these differences were consistently observed over locations and seasons. The GMO Panel concludes that the agronomic performance and phenotypic characteristics of 59122 x NK603 maize are comparable to its non-GM control except for the introduced traits.

4.3 Conclusion

Based on the results of comparative analysis it is concluded that 59122 x NK603 maize is compositionally and agronomically equivalent to its non GM counterpart and conventional maize, except for the presence of CRY34Ab1, CRY35Ab1, CP4 EPSPS, CP4 EPSPS L214P and PAT proteins in 59122 x NK603 maize. Based on the assessment of data available, including the additional information provided by the applicant in response to the Panel request, for 59122 x NK603 maize, for the single events and for appropriate non-GM controls, the GMO Panel does not see a reason to assume that crossing of NK603 maize and 59122 maize results in an interaction of the newly expressed proteins which causes compositional or agronomic changes.

5. FOOD/FEED SAFETY ASSESSMENT

5.1.1. Summary of the previous evaluation of the single events

Maize 59122

P. fluorescens produced CRY34Ab1 and CRY35Ab1 proteins and *E.coli* produced PAT protein were used for toxicity studies after it had been demonstrated experimentally that they were equivalent to those extracted from leaf material of maize event 59122. The transgenic CRY34Ab1, CRY35Ab1 proteins induced no adverse effects in acute and repeated dose (28 day) oral toxicity studies in mice and they are rapidly degraded in simulated gastric fluid and inactivated during heat treatments. The PAT protein is expressed at very low levels in maize 59122 and also did not cause adverse effects in acute and repeated dose (14 day) oral toxicity studies in mice and rats respectively. In addition it is rapidly degraded in simulated gastric fluid. The sequence of the newly expressed CRY34Ab1, CRY35Ab1 and PAT proteins did not show any significant similarity with the sequences of known toxins or allergens. With regard to animal studies with the whole product, there were no indications of adverse effects in a 90-day rat feeding study using diets containing maize 59122 grains. In addition, nutritional data comprising a target animal feeding study with maize 59122 grains on broilers indicate that maize 59122 is nutritionally equivalent to the non-GM comparator. These animal studies therefore further supported the findings of the compositional analysis of no effect beyond the intended introduction of the CRY34Ab1, CRY35Ab1 and PAT proteins (EFSA, 2007b).

Maize NK603

E.coli produced CP4 EPSPS and CP4 EPSPS L214P proteins were used for toxicity studies after it had been demonstrated experimentally that they were equivalent to those extracted from maize event NK603. No toxicity of the CP4 EPSPS and CP4 EPSPS L214P proteins was observed in acute oral toxicity studies in mice. These proteins are quickly degraded in

simulated gastric fluid. Bioinformatics studies demonstrated that the CP4 EPSPS proteins show no homology to known toxic and allergenic proteins. A 13 week toxicity study in rats indicated no toxicity and a 42-day feeding study on broiler chickens showed that NK603 maize is not nutritionally different from its genetically closely related non-GM maize or commercial maize varieties included in the study. The nutritional equivalence of NK603 maize to commercial maize varieties were confirmed in feeding studies on Angus-continental cross steers, Holstein dairy cows and growing-finishing pigs of two breeds. These animal studies therefore further supported the findings of the compositional analysis of no effect beyond the intended introduction of the CP4 EPSPS and CP4 EPSPS L214P proteins (EFSA 2003a,b).

5.1.2. Product description and intended use

The scope of application EFSA-GMO-UK-2005-20 includes the import and processing of 59122x NK603 maize and its derived products for use as food and feed. Thus, possible uses of 59122x NK603 maize include the production of animal feed, but it is also processed into valuable food products, including *e.g.* starch, syrups and oils.

The genetic modification of 59122 x NK603 maize is intended to improve agronomic performance only and is not intended to influence the nutritional properties, processing characteristics and overall use of maize as a crop.

5.1.3. Stability during processing

Since 59122 x NK603 maize is compositionally equivalent to conventional maize, except for the newly expressed traits (see Section 4.3), the stability of constituents during processing is not expected to be different from conventional maize varieties.

5.1.4. Toxicology

5.1.4.1. Toxicological assessment of expressed novel proteins

The transgenic CRY34Ab1, CRY35Ab1 proteins and PAT protein expressed in the parental maize line 59122 as well as the CP4 EPSPS and CP4 EPSPS L214P proteins present in the NK603 parental maize have been assessed previously (EFSA, 2003a,b, EFSA 2004, EFSA 2005a,b,c,d, EFSA2006c, EFSA 2007b,c) and no adverse effects have been observed.

The Panel is not aware of any new information that would change this conclusion.

No new genes in addition to those occurring in the parental maize lines have been introduced in 59122 x NK603 maize. Given the functional properties of the proteins and the results of the compositional analysis (section 4.3), the Panel concludes that interactions between the expressed proteins are unlikely.

5.1.4.2. Toxicological assessment of new constituents other than proteins

No new constituents other than the CRY34Ab1, CRY35Ab1, PAT, CP4 EPSPS and CP4 EPSPS L214P proteins are expressed in 59122 x NK603 maize and no relevant changes in the composition of 59122 x NK603 maize were detected by the compositional analysis.

5.1.5. Toxicological assessment of the whole GM food/feed

The genetically modified maize events 59122 and NK603 have previously been found as safe as the conventional counterpart for human and animal consumption (EFSA, 2003a,b; 2007b). A molecular characterization undertaken on maize 59122 x NK603 identified no altered stability of the two events (see section 3.2) when these were brought together by crossing, and expression analysis of the proteins CRY34Ab1, CRY35Ab1, PAT, CP4 EPSPS and CP4 EPSPS L214P, revealed no change in their protein expression levels. As also no indication for interaction between the newly expressed proteins was found and the composition of maize 59122 x NK603 is comparable with that of non-GM maize hybrids and the single events, the GMO Panel is of the opinion that the maize 59122 x NK603 is as safe as the non GM counterpart and conventional maize. Therefore, the Panel has found no reason to ask for additional animal safety studies with the whole GM food/feed.

5.1.6. Allergenicity

The strategies used when assessing the potential allergenic risk focus on the characterisation of the source of the recombinant protein, the potential of the newly expressed protein to induce sensitisation or to elicit allergic reactions in already sensitised persons and whether the transformation may have altered the allergenic properties of the modified food. A weight-of-evidence approach is recommended, taking into account all of the information obtained with various test methods, since no single experimental method yields decisive evidence for allergenicity (CAC, 2003; EFSA, 2006a). An assessment of any potential for increased allergenicity to humans and animals due to the stacked events should be provided.

5.1.6.1 Assessment of allergenicity of the newly expressed proteins

The proteins present in the maize 59122 x NK603 have been assessed previously and it was found unlikely that they are allergenic (EFSA, 2003a,b; EFSA 2004; EFSA 2005a,b,c,d, EFSA2006c; EFSA 2007b,c). Based on the information provided in the compositional analysis (section 4.2.3), the GMO Panel considers unlikely that potential interactions occur that might change the allergenicity of the expressed proteins.

5.1.6.2 Assessment of allergenicity of the whole GM plant or crop

The issue of a potential increased allergenicity of 59122 x NK603 maize does not appear relevant to the Panel since maize is not considered a common allergenic food. Food allergies to maize are of low frequency and mainly occur in populations of specific geographic areas. Rare cases of occupational allergy to maize dust have been reported. There is no reason to expect that the use of GM maize will significantly increase the intake and exposure to maize. Therefore a possible over-expression of any endogenous protein, which is not known to be allergenic, would be unlikely to alter the overall allergenicity of the whole plant or the allergy risk for consumers.

5.1.7. Nutritional assessment of GM food/feed

The applicant has provided a nutritional study on broilers using the triple stacked event 59122 x 1507 x NK603 maize as test material. The Panel is aware that this does not directly pertain to the double stacked event 59122 x NK603 maize featured in this opinion, but it considers that this study provides relevant supplementary information.

The 42-day poultry feeding study consisted of seven treatment groups. Three groups received grains from 59122 x 1507 x NK603 maize treated with glufosinate, glyphosate or both herbicides respectively, one group received grains from maize 091 (a non-GM maize with comparable background genetics to 59122 x 1507 x NK603 maize), and the other 3 groups grains from different conventional non-GM maize varieties.

Each treatment consisted of 12 replicates each of which contained 10 broilers (5 male and 5 female), which gave 120 broilers/treatment. Study diets were formulated according to National Research Council (NRC) requirements. Animal performance on the various diets were evaluated by measuring mortality, weight gain, organ and carcass yields (post chill) for thighs, breasts, wings, leg, abdominal fat, kidney and liver, and feed efficiency.

Change in female's relative kidney weight was observed in one of the test treatments i.e. test maize treated with glyphosate followed by glufosinate. Given the high range of variability in relative kidney weight observed and the fact that in this particular treatment the females

relative kidney weight was within the natural range of variability calculated, the Panel considered that this change is unlikely to be of biological relevance.

Thus, the broiler feeding study with the higher stack supported the results of the comparative compositional analysis that showed that 59122 x NK603 maize is compositionally and nutritionally equivalent to non-GM maize comparators.

5.1.8. Post-market monitoring of GM food/feed

The risk assessment concluded that no data have emerged to indicate that 59122 x NK603 maize is any less safe than its non-GM comparator and the parental maize lines. In addition, 59122 x NK603 maize is, from a nutritional point of view, equivalent to conventional maize. Therefore, and in line with the Guidance document (EFSA, 2006a), the GMO Panel is of the opinion that post-market monitoring of the food/feed derived from 59122 x NK603 maize is not necessary.

5.2. Conclusion

The newly expressed CRY34Ab1, CRY35Ab1 and PAT proteins expressed in the parental maize line 59122 as well as the CP4 EPSPS and CP4 EPSPS L214P present in the NK603 parental maize have been assessed previously and no adverse effects have been observed.

Given the functional properties of the proteins and the results of the compositional analysis, the Panel concludes that interactions between the transgenic proteins expressed in 59122 x NK603, which might impact on food and feed safety, are unlikely.

A higher stack of 59122 x NK603 maize, 59122 x 1507 x NK603 maize has been studied in a nutritional feeding study with broilers and showed its nutritional equivalence to conventional maize lines. The Panel concluded that this study provides additional evidence that the nutritional properties of maize 59122 x NK603 would not be different from those of the conventional maize varieties.

In conclusion the Panel considers that 59122 x NK603 maize is as safe as its non GM counterpart and that the overall allergenicity of the whole plant is not changed and concludes that 59122 x NK603 maize is unlikely to have any adverse effect on human and animal health in the context of its intended uses.

6. ENVIRONMENTAL RISK ASSESSMENT AND MONITORING PLAN

6.1. Evaluation of relevant scientific data

The scope of application EFSA-GMO-UK-2005-20 is for food (*e.g*. syrup, starch, oil) and feed (e.g. meal, oil) uses, import and processing of maize 59122 x NK603 and does not include cultivation. Considering the intended uses of maize 59122 x NK603, the environmental risk assessment is concerned with indirect exposure through manure and faeces from the gastrointestinal tracts mainly of animals fed on the GM maize and with accidental release into the environment of GM seeds during transportation and processing.

As the scope of the present application excludes cultivation, environmental concerns related to the use of glufosinate-ammonium and/or glyphosate herbicides on maize 59122 x NK603 apply only to imported and processed maize products that may have been treated with those herbicides in the countries of origin. The GMO Panel is aware that the risk assessment of active substances falls within the scope of Directive 91/414/EEC concerning the placing of plant protection products on the market (EC, 1991**).**

6.2.1. Evaluation of the single events

Maize NK603 and 59122

Maize NK603 and 59122 have been developed for tolerance to herbicides respectively against glyphosate and glufosinate. Tolerance to glyphosate in maize NK603 and glufosinate-ammonium in maize 59122 is conferred respectively by the 5-enolpyruvylshikimate-3-phosphate synthase gene from *Agrobacterium* sp. strain CP4 (CP4 EPSPS) and by phosphinothricin-N-acetyltransferase (PAT) gene from *Streptomyces viridochromogenes*. In addition maize 59122 was transformed to be protected against specific coleopteran (*e.g*. Western corn rootworm larvae (*Diabrotica virgifera virgifera* LeConte)) pests by producing Cry34Ab1 and Cry35Ab1 proteins from *Bacillus thuringiensis* (see Section 3.).

The assessed dossiers for maize 59122 (application EFSA-GMO-NL-2005-12 under Regulation (EC) No 1829/2003) and maize NK603 (notification C/ES/00/01 under Directive 2001/18/EC and a notification under Article 4 of Novel Food Regulation (EC) No 258/97) concerned import and processing of maize NK603 for food and feed uses. The GMO Panel was of the opinion that maize NK603 and 59122 are as safe as conventional maize. Therefore their placing on the market for food and feed uses as well as processing is unlikely to have an adverse effect on human or animal health or, in that context, on the environment (EFSA, 2003a, b; 2007b).

A post-market environmental monitoring plan, including general surveillance of maize NK603, was proposed by the applicant and accepted by the GMO Panel. The GMO Panel also agreed with the post-market environmental monitoring plan provided by the applicant for maize 59122 since the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects. Both monitoring plans were subject to some recommendations to restrict seeds of GM maize entering cultivation, as the latter requires specific approval under Directive 2001/18/EC or Regulation (EC) No 1829/2003.

6.2.2. Environmental risk assessment

6.2.2.1. Potential unintended effects on plant fitness due to the genetic modification

Maize is highly domesticated and generally unable to survive in the environment without cultivation. Maize plants are not winter hardy in many regions of Europe, they have lost their ability to release seeds from the cob and they do not occur outside cultivated land or disturbed habitats in agricultural landscapes of Europe, despite cultivation for many years.

The herbicide tolerance traits can only be regarded as providing a selective advantage for the GM maize plant where and when glufosinate-ammonium and/or glyphosate herbicides are applied. Similarly, insect resistance against certain coleopteran pests provides a potential advantage in cultivation under infestation conditions. However survival of maize outside cultivation in Europe is mainly limited by a combination of low competitiveness, absence of a dormancy phase, susceptibility to diseases and to cold climate conditions. Since these general characteristics of this GM maize are unchanged, the inserted traits, namely insect resistance and herbicide tolerance, are not likely to provide a selective advantage outside cultivation in Europe. Therefore it is considered very unlikely that volunteers of maize 59122 x NK603 or its progeny will differ from conventional maize varieties in their ability to survive until subsequent seasons or to establish feral populations under European environmental conditions.

In addition to the field trials carried out with the parental GM maize NK603 and 59122 (EFSA, 2003a; 2007b), field trials with maize 59122 x NK603 were carried out at 6 locations (5 in US and 1 in Canada) in 2003. The field data provided in the application do not show increased fitness and invasiveness or enhanced weediness, except in the presence of glufosinate-ammonium and/or glyphosate herbicides. In addition to the data presented by the applicant, the GMO Panel is not aware of any scientific report of increased spread and establishment of maize 59122 x NK603 and of any change in survival capacity, including over-wintering.

Since maize 59122 x NK603 has no altered survival, multiplication or dissemination characteristics except in the presence of glufosinate-ammonium or glyphosate herbicides and/or target organisms, the GMO Panel is of the opinion that the likelihood of unintended

environmental effects as a consequence of spread of genes from this GM maize will not differ from that of maize 59122 or NK603, or of conventional maize varieties.

6.2.2.2. Potential for gene transfer

A prerequisite for any gene transfer is the availability of pathways for the transfer of genetic material, either through horizontal gene transfer of DNA, or vertical gene flow via seed dispersal and cross-pollination.

(a) Plant to bacteria gene transfer

Current scientific data (see EFSA, 2004b; EFSA 2007a for further details) suggest that gene transfer from GM plants to micro-organisms under natural conditions is extremely unlikely, and its establishment would occur primarily through homologous recombination in micro-organisms .

In the case of accidental release and establishment of maize 59122 x NK603 in the environment, exposure of micro-organisms to transgenic DNA derived from GM maize plants would take place during natural decay of GM plant material and/or pollen in the soil of areas where GM plants might establish.

Food and feed products derived from the GM maize could contain transgenic DNA. Therefore micro-organisms in the digestive tract of humans and animals may be exposed to transgenic DNA.

The *cry34Ab1* and *cry35Ab1* genes are under the control of eukaryotic promoters (see Section 3) with limited, if any, activity in prokaryotes in the unlikely event of horizontal gene transfer.

The *pat* gene is a component of soil microbial populations (Hérouet *et al.*, 2005) and the *cry34Ab1/cry35Ab1* genes, which occur naturally in bacterial populations (Schnepf *et al.*, 2005), were cloned from naturally occurring *Bacillus thuringiensis*. Taking into account the origin and nature of the *cry34Ab1/cry35Ab1*, *cp4 epsps* and *pat* genes and the lack of selective pressure in the intestinal tract and/or the environment, the likelihood that horizontal gene transfer of the *cry34Ab1/cry35Ab1, cp4 epsps* and *pat* genes would confer selective advantage or increased fitness to micro-organisms is very limited. For this reason it is very unlikely that genes from maize 59122 x NK603 would become transferred and established in the genome of micro-organisms in the environment or human and animal digestive tract. In the very unlikely event that such horizontal gene transfer would take place, no adverse effects on human and animal health or the environment are expected, as no principally new traits would be introduced or expressed in microbial communities.

(b) Plant to plant gene transfer

The extent of cross-pollination to other maize varieties will mainly depend on the scale of accidental release during transportation and processing. For maize, any vertical gene transfer is limited to other *Zea mays* plants as populations of sexually compatible wild relatives of maize are not known in Europe (OECD, 2003).

The flowering of occasional GM plants originating from accidental release occurring during transportation and processing is unlikely to disperse significant amounts of GM maize pollen to other maize plants.

Herbicide tolerance and insect resistance provide agronomic advantages in cultivation where and when the specific herbicides are applied and target organisms are present. However survival of maize outside cultivation in Europe is mainly limited by a combination of low competitiveness, absence of a dormancy phase, susceptibility to diseases and to cold climate conditions. Since these general characteristics of this GM maize are unchanged, herbicide tolerance and insect resistance are not likely to provide selective advantages outside cultivation in Europe. Therefore, as for any other maize varieties, GM maize plants would only survive in subsequent seasons in the warmer regions of Europe and are not likely to establish feral populations under European environmental conditions (see Section 5.2.1.1).

In conclusion, since maize 59122 x NK603 has no altered survival, multiplication or dissemination characteristics, except when cultivated in the presence of glufosinate-ammonium or glyphosate and/or the specific target organisms, the GMO Panel is of the opinion that the likelihood of unintended environmental effects as a consequence of spread of genes from this maize in Europe will not differ from that of maize 59122 or NK603, or of conventional maize varieties.

6.2.2.3. Potential interactions of the GM plant with target organisms

Maize 59122 was transformed to co-express Cry34Ab1 and Cry35Ab1 proteins from *Bacillus thuringiensis*. This binary insecticidal toxin is made of two components, the Cry34Ab1 and the Cry35Ab1 proteins, acting together in the control of certain coleopteran pests, such as the western corn rootworm (*Diabrotica virgifera virgifera* LeConte), the northern corn rootworm (*D. Barberi* Smith & Lawrence) and the southern corn rootworm (*D. undecimpunctata howardi* Barber) (Masson *et al*., 2004).

A study showed that Cry35Ab1 protein alone is not active against corn rootworm larvae and that Cry34Ab1 alone causes mortality and growth inhibition to corn rootworm larvae, but for maximal insecticidal activity both the Cry34Ab1 and Cry35Ab1 proteins are required. The binary protein formulation enhances the insect toxicity (Herman *et al*., 2002). The hypothetical

mode of action for this kind of association (*i.e.* binary toxins) is that Cry34Ab1 is responsible for specific binding to receptors on the insect midgut epithelium while Cry35Ab1 is active on membrane pore formation (de Maagd *et al*., 2003).

Considering that the intended uses of maize 59122 x NK603 specifically exclude cultivation, the environmental exposure is limited to exposure through manure and faeces from the gastrointestinal tracts mainly of animals fed on the GM maize as well as to the accidental release into the environment of 59122 x NK603 seeds during transportation and processing and subsequently to potential occurrence of sporadic feral plants. Thus the level of exposure of target organisms to Cry34Ab1 and Cry35Ab1 proteins is likely to be extremely low and of no ecological relevance.

6.2.2.4. Potential interactions of the GM plant with non-target organisms

Considering the intended uses of maize 59122 x NK603, the environmental risk assessment is concerned with indirect exposure through manure and faeces from the gastrointestinal tracts mainly of animals fed on the GM maize and with accidental release into the environment of GM seeds during transportation and processing.

The GMO Panel assessed whether CRY proteins might potentially affect non-target organisms by entering the environment in manure and faeces from the gastrointestinal tracts mainly of animals fed on this maize. Data supplied by the applicant (Herman *et al.*, 2003) and literature on other Cry proteins (Ahmad *et al*., 2005 and references therein; Lutz *et al*., 2005) suggest that most CRY proteins are degraded by the enzymatic activity in the gastrointestinal tract so that only low amounts of CRY proteins would remain intact to pass out in faeces. There would subsequently be further degradation of these proteins in the manure and faeces due to microbial processes. In addition, other sources of environmental exposure for example soil and water, and disposal of organic wastes are likely to be very low and localized (Baumgarte & Tebbe, 2005; Hopkins & Gregorich, 2003).

In conclusion the GMO Panel considers that the level of exposure of any potential non-target organisms to the CRY proteins expressed in maize 59122 x NK603 in combination with the PAT and CP4 EPSPS proteins is likely to be very low and of no ecological relevance.

6.2.2.5. Potential interaction with the abiotic environment and biogeochemical cycles

This point was not considered an issue by the Member States or by the GMO Panel. The level of exposure would be so low that potential effects on the abiotic environment and biogeochemical cycles are unlikely.

6.2.3. Monitoring

The objectives of a monitoring plan according to Annex VII of Directive 2001/18/EC are to confirm that any assumption regarding the occurrence and impact of potential adverse effects of the GMO, or its use, in the environmental risk assessment are correct and to identify the occurrence of adverse effects of the GMO, or its use, on human health or the environment which were not anticipated in the environmental risk assessment.

Monitoring is related to risk management, and thus a final adoption of the monitoring plan falls outside the mandate of EFSA. However, the GMO Panel gives its opinion on the scientific quality of the monitoring plan provided by the applicant (EFSA, 2006b). The potential exposure to the environment of maize 59122 x NK603 would be through manure and faeces from the gastrointestinal tracts mainly of animals fed on the GM maize or through accidental release into the environment of GM seeds during transportation and processing.

No specific environmental impact of this GM maize was indicated by the environmental risk assessment and thus no case specific monitoring is required.

The general surveillance plan provided by the applicant includes i) the description of an approach involving operators (i.e. grain traders and maize processors involved in the handling and use of viable maize 59122 x NK603) to report to the applicant any potential unanticipated adverse effect of GMOs on human health and the environment, ii) a coordinating system newly established by EuropaBio, iii) the use of networks of existing surveillance systems. The applicant will submit a general surveillance report on an annual basis. In case of adverse effects altering the conclusions of the environmental risk assessment, the applicant will immediately inform the European Commission.

The GMO Panel is of the opinion that the scope of the monitoring plan provided by the applicant is in line with the intended uses of maize 59122 x NK603 since the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects. The GMO Panel agrees with the reporting intervals proposed by the applicant in the general surveillance plan. The GMO Panel advises that appropriate management systems should be in place to restrict seeds of maize 59122 x NK603 entering cultivation as the latter requires specific approval under Directive 2001/18/EC or Regulation (EC) No 1829/2003.

6.3. Conclusion

The scope of the application includes food and feed uses, import and processing of maize 59122 x NK603 and excludes cultivation. Considering the intended uses of maize 59122 x NK603, the environmental risk assessment is concerned with indirect exposure through manure and faeces from the gastrointestinal tracts mainly of animals fed on the maize 59122 x NK603 and with accidental release into the environment of 59122 x NK603 seeds during transportation and processing.

Maize is highly domesticated and not able to survive in the environment without cultivation. There are no indications of increased likelihood of establishment or survival of feral maize plants in case of accidental release into the environment of 59122 x NK603 seeds during transportation and processing for food and feed uses. Taking into account the scope of the application, both the rare occurrence of sporadic feral plants and the low levels of exposure through other routes indicate that the risk to target and non-target organisms is negligible.

The scope of the monitoring plan provided by the applicant is in line with the intended uses of maize 59122 x NK603 since the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects. Furthermore the GMO Panel agrees with the reporting intervals proposed by the applicant in the general surveillance plan.

CONCLUSIONS AND RECOMMENDATIONS

The GMO Panel was requested to carry out a scientific risk assessment of the maize 59122 x NK603 for food and feed uses, import and processing and all derived products.

The GMO Panel is of the opinion that the molecular characterisation provided for maize 59122 x NK603 produced by conventional breeding is sufficient for the safety assessment. The bioinformatics analysis of the inserted DNA and the flanking regions of the single events 59122 and NK603 does not raise any safety concern. The expression of the genes introduced by genetic modification has been sufficiently analysed and the stability of the genetic modification has been demonstrated over several generations. The GMO panel considers that the molecular characterization does not indicate any safety concern.

Based on the results of comparative analysis it was concluded that 59122 x NK603 maize is compositionally and agronomically equivalent to conventional maize, except for the presence of CRY34Ab1, CRY35Ab1, CP4 EPSPS, CP4 EPSPS L214P and PAT proteins in 59122 x NK603 maize. Based on the assessment of data available, including the additional information provided by the applicant in response to the Panel request, for 59122 x NK603 maize, for the single events and for appropriate non-GM controls, the GMO Panel does not see a reason to assume that crossing of NK603 maize and 59122 maize results in an interaction of the newly expressed proteins which causes compositional or agronomic changes. The GMO Panel concluded that the maize 59122 x NK603 is as safe as its non GM counterpart and that the overall allergenicity of the whole plant is not changed.

There are no indications of increased likelihood of establishment or survival of feral maize plants in case of accidental release into the environment of 59122 x NK603 seeds during transportation and processing for food and feed uses. Taking into account the scope of the application, both the rare occurrence of sporadic feral plants and the low levels of exposure through other routes indicate that the risk to target and non-target organisms is negligible.

The scope of the monitoring plan provided by the applicant is in line with the intended uses of maize 59122 x NK603 since the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects. Furthermore the GMO Panel agrees with the reporting intervals proposed by the applicant in the general surveillance plan.

In conclusion, the GMO Panel considers that information available for maize 59122 x NK603 addresses the comments raised by the Member States and considers that it is unlikely that maize 59122 x NK603 will have any adverse effect on human and animal health or on the environment in the context of its proposed uses.

DOCUMENTATION PROVIDED TO EFSA

1. Letter from the Competent Authority of the United Kingdom, dated 16 September 2005, concerning a request for the placing on the market of 59122 x NK603 maize in accordance with Regulation (EC) No 1829/2003.

2. Acknowledgement letter, dated 29 September 2005, from EFSA to the Competent Authority of the United Kingdom (Ref. SR/KL/jq (2005) 1160).

3. Letter from EFSA to applicant, dated 28 November 2006, with request for clarifications under completeness check (Ref. SR/CP/shv (2006) 1846713).

4. Letter from Applicant to EFSA, dated 18 January 2007, providing EFSA with an updated version of the Application EFSA-GMO-UK-2005-20 submitted by Pioneer Overseas Corporation under Regulation (EC) No. 1829/2003.

5. Letter from EFSA to applicant, dated 02 April 2007, with a request for clarifications under completeness check (Ref. SR/SM/DC/CP/shv (2007) 2067264).

6. Letter from Applicant to EFSA, dated 16 April 2007, providing EFSA with an updated version of the Application EFSA-GMO-UK-2005-20 submitted by Pioneer Overseas Corporation under Regulation (EC) No. 1829/2003.

7. Letter from EFSA to Applicant, dated 29 May 2007, with a request for clarifications under completeness check (Ref. SR/AC/shv (2007) 2165108).

8. Letter from Applicant to EFSA, dated 31 May 2007, providing EFSA with clarifications on the letter sent by EFSA on 29 May 2007 (Ref. SR/AC/shv (2007) 2165108).

9. Letter from EFSA to Applicant, dated 04 June 2007, with a request for clarifications under completeness check (Ref. SR/AC/shv (2007) 2174598).

10. Letter from Applicant to EFSA, dated 19 June 2007, providing EFSA with clarifications on the letter sent by EFSA on 04 June 2007.

11. Letter from EFSA, dated 20 June 2007, delivering the 'Statement of Validity' for Application EFSA-GMO-UK-2005-20, 59122 x NK603 maize submitted by submitted by Pioneer Overseas Corporation under Regulation (EC) No. 1829/2003 (Ref. SR/AC/shv (2007) 2210466).

12. Letter from Applicant to EFSA, dated 27 June 2006, providing additional copies of the Valid Application EFSA-GMO-UK-2005-20.

13. Letter from EFSA, dated 16 November 2007, with request for additional information (Ref. SR/AC/shv (2007) 2512245).

14. Letter from Applicant to EFSA, dated 20 December 2007, informing about the timeline for the submission of the additional information requested.

15. Letter from Applicant to EFSA, dated 31 January 2008, informing about the timeline for responses for the submission of the additional information requested.

16. Letter from Applicant to EFSA, dated 31 March 2008, responding to the request for additional information of 16 November 2007.

17. Letter from Applicant to EFSA, dated 15 April 2008, providing additional information.

18. Letter from EFSA to Applicant, dated 20 June 2008, with request for additional information (Ref. PB/AC/shv (2008) 3107685).

19. Letter from Applicant to EFSA, dated 30 July 2008, informing about the timeline for responses for the submission of the additional information requested.

20. Letter from Applicant to EFSA, dated 01 September 2008, responding to the request for additional information of 20 June 2008 (Ref. PB/AC/shv (2008) 3107685.

21. Letter from EFSA to Applicant, dated 20 October 2008, re-starting the clock for Application EFSA-GMO-UK-2005-20 (Ref. PB/AC/shv (2008) 3385686.

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