SUMMARY NOTIFICATION INFORMATION FORMAT (SNIF)
FOR PRODUCTS CONTAINING GENETICALLY
MODIFIED HIGHER PLANTS (GMHP)

NK603 × MON 810 MAIZE

(06 DECEMBER 2002)

A. GENERAL INFORMATION

1. Details of notification

(a) Member State of notification:

United Kingdom

(b) Notification number:

C/GB/02/M3/03

(c) Name of the product (commercial and other names):

The Monsanto development code for this genetically modified maize is: NK603 × MON 810. The commercial name of this maize derived from maize transformation events NK603 and MON 810 has not yet been established.

This application under Directive 2001/18/EC is for import into the European Union, and use therein of NK603 × MON 810 maize, comprising hybrid maize varieties that are produced by traditional breeding of two genetically modified parental inbred lines of maize, one being derived from transformation event NK603 and the second one derived from event MON 810. The proposed uses of NK603 × MON 810 maize will be the same as for any other maize, but do not include the cultivation in the E.U. of varieties produced by the combination of NK603 and MON 810 maize.

(d) Date of acknowledgement of notification:

17 April 2002

2. Notifier

(a) Name of notifier:

Monsanto Company, represented by Monsanto Europe S.A.

(b) Address of notifier:

Monsanto Europe S.A.
270-272 Avenue de Tervuren
B-1150 Brussels
BELGIUM

Monsanto
800 N. Lindbergh Boulevard
St. Louis, Missouri 63167
U.S.A

(c) Is the notifier domestic manufacturer: [X] importer: [ ]

(d) In the case of an import the name and address of the manufacturer shall be given

Not applicable.

3. General description of the product

(a) Name of the recipient or parental plant and the intended function of the genetic modification

NK603 × MON 810 maize consists of hybrid maize varieties, produced using conventional methods of maize breeding by crossing parental inbred lines of NK603 and MON 810 maize. Genetic modification was used in the development of NK603 and MON 810 maize, but no additional genetic modifications were involved for the production of NK603 × MON 810 hybrids.

NK603 × MON 810 maize expresses CP4 EPSPS proteins derived from *Agrobacterium* sp. strain CP4, which confer tolerance to Roundup®[1] herbicide (containing glyphosate), and it expresses the Cry1Ab protein, derived from *Bacillus thuringiensis* subsp. *kurstaki,* which confers protection from predation by certain lepidopteran insect pests including European corn borer (*Ostrinia nubilalis)* and pink borers (*Sesamia* spp).

The use of NK603 × MON 810 maize plants enables the farmer to utilise Roundup herbicide for effective control of weeds during the growing season and to take advantage of the favourable environmental and safety characteristics of Roundup herbicide. The use of NK603 × MON 810 maize also enables the farmer to effectively control certain lepidopteran insect pests in maize, ensuring maximum realization of yield potential, while removing the environmental burden of the production, packaging and transport of insecticides, previously used to control *Ostrinia nubilalis* and *Sesamia* spp.

(b) Any specific form in which the product must not be placed on the market (seeds, cut-flowers, vegetative parts, etc.) as a proposed condition of the authorisation applied for

This application is for import and use in the E.U. of NK603 × MON 810 maize. The proposed uses of this maize are the same as for any other maize, including use in animal feed. The scope of this notification does not include the cultivation of varieties derived from this maize in the E.U.

(c) Intended use of the product and types of users

The proposed uses of this maize are the same as for any other maize. The primary use of maize is for animal feed. Maize is also processed into valuable food and industrial products.

(d) Any specific instructions and/or recommendations for use, storage and handling, including mandatory restrictions proposed as a condition of the authorisation applied for

NK603 × MON 810 maize has been demonstrated to be substantially equivalent to other maize varieties except for its introduced traits (tolerance to Roundup herbicide and protection from predation by certain lepidopteran insect pests). Grain of NK603 × MON 810 maize will be used and handled in the same manner as current commercial maize varieties.

[1] Roundup® is a registered trademark of Monsanto Technology LLC.

(e) If applicable, geographical areas within the E.U. to which the product is intended to be confined under the terms of the authorisation applied for

NK603 × MON 810 maize is intended for use throughout the E.U.

(f) Any type of environment to which the product is unsuited

NK603 × MON 810 maize varieties may be imported or used in any given environment.

(g) Any proposed packaging requirements

NK603 × MON 810 maize has been shown to be substantially equivalent to other maize varieties. Therefore, NK603 × MON 810 maize grain will be used in the same manner as with other maize and no specific packaging is foreseen.

(h) Any proposed labelling requirements in addition to those required by law

Grain of NK603 × MON 810 maize could be imported into the European Union from a number of world areas, including Africa, The Americas, Asia and Central and Eastern Europe. In accordance with the requirements of Directive 2001/18/EC, Monsanto will undertake a number of measures to ensure that international traders are provided with the necessary information to comply with statutory requirements relating to the placing on the market of NK603 × MON 810 maize grain.

In particular, Monsanto will:

a) Inform European and International traders of the approval for import into the European Union of NK603 × MON 810 maize grain that this product is a genetically modified organism and that NK603 × MON 810 maize grain may be present in bulk shipments of maize grain. To that effect the words "Contains genetically modified organisms" shall appear either on a label or in an accompanying document to the maize grain shipment.

b) Provide all traders with the commercial name of the product, any agreed European and/or international unique identifier (see question ***3.*****(j)**) replacing the current unique code (NK603 × MON 810) and any other relevant product information, including procedures for accessing the European public registers of GM organisms.

c) Advise all traders, and other operators using the product, that NK603 × MON 810 maize grain is subject to the traceability and labelling requirements of Directive 2001/18/EC and to the requirements of any Community legislation adopted to regulate the traceability and labelling of GM organisms.

It should be noted that other genetically modified maize products have been approved for import into the European Union and that NK603 × MON 810 maize may be marketed in the European Union in mixture with these previously approved maize products.

(i) Estimated potential demand

(i) in the Community

Imports of maize grain from outside the E.U. are limited in practice by the General Agreement on Tariffs and Trade (GATT) to 2.5 million tonnes per annum.

(ii) in export markets for EC supplies

Not applicable

(j) Unique identification code(s) of the GMO(s)

The code NK603 × MON 810 will uniquely identify hybrid seed and grain of NK603 × MON 810 maize until replaced by an internationally recognised unique identifier. It is proposed that NK603 × MON 810 maize is identified by the combination of MON-00603-6, the unique identifier proposed for maize derived from event NK603, and MON-00810-6 the unique identifier for maize derived from MON 810.

4. Has the GMHP referred to in this product been notified under part B of Directive 2001/18/EC and/or Directive 90/220/EEC?

Yes [X]	**No [ ]**

NK603 × MON 810 maize has been planted for field-testing purposes in France since 2000 (B/FR/00.02.06).

(i) If no, refer to risk analysis data on the basis of the elements of Part B of Directive 2001/18/EC

Not applicable

5. Is the product being simultaneously notified to another Member State?

Yes [ ]	**No [X]**

(i) If no, refer to risk analysis data on the basis of the elements of Part B of Directive 2001/18/EC

Please see questions ***9-11***, ***14-27***, ***29*** and ***30-33***, for risk assessment data, including experience from experiments conducted under 90/220/EEC Part B approvals.

or

Has the product been notified in a third country either previously or simultaneously?

Yes [X]	**No [ ]**

If yes, specify

NK603 × MON 810 has been notified in several world areas, including the U.S.A. where the product was first commercialised in 2002.

6. Has the same GMHP been previously notified for marketing in the Community?

Yes [ ]	**No [X]**
If yes, give notification number and Member State Not applicable	

7. Measures suggested by the notifier to take in case of unintended release or misuse as well as measures for disposal and treatment

Maize is not an invasive plant because it is a weak competitor outside cultivation. For this reason, volunteer maize is not found in non-crop situations, for example, in fence or hedgerows, ditches, and roadsides. In the unlikely event that any of the imported grain were disseminated into the environment, it would be highly unlikely to pose any threat to the environment. In the unlikely event of establishment, volunteer plants could be easily controlled by currently available selective herbicides or by mechanical means. Therefore no specific measures are recommended in case of unintended release (spillage or other means) of NK603 × MON 810 maize.

Misuse of the imported grain as seed remains extremely unlikely since 1) only certified hybrid maize seed can be marketed in the E.U. and 2) the (F2) grain, coming from open-pollinated production fields, will not have the agronomic yield potential and homogeneity of true F1 hybrid seed.

The measures for waste disposal and treatment for NK603 × MON 810 maize products are the same as those for other maize products.

B. NATURE OF THE GMHP CONTAINED IN THE PRODUCT

INFORMATION RELATING TO THE RECIPIENT OR (WHERE APPROPRIATE) PARENTAL PLANTS

8. Complete name

(a)	**Family name** Gramineae
(b)	**Genus** *Zea*
(c)	**Species** *mays* (2n = 20)
(d)	**Subspecies** Not applicable
(e)	**Cultivar/breeding line** NK603 × MON 810 maize
(f)	**Common name** Maize; Corn

***9.*(a) Information concerning reproduction**

(i) Mode(s) of reproduction

Maize (*Zea mays)* reproduces sexually, is a wind-pollinated, monoecious species with separate staminate (tassels) and pistillate (silk) flowers, which encourages natural pollination between maize plants. Wind movements across the maize field cause pollen from the tassel to fall on the silks of the same or adjoining plants. Self-pollination leads to homogeneity of the genetic characteristics within a single plant while cross-pollination combines the genetic traits of many plants.

(ii) Specific factors affecting reproduction, if any

Tasselling, silking, and pollination are the most critical stages of maize development and, consequently, grain yield may ultimately be greatly impacted by moisture and fertility stress. Under conditions of high temperature and desiccation, maize pollen viability is measured in minutes; these conditions may even destroy the tassel before any viable pollen is shed. More moderate conditions can extend the field life of pollen to hours.

(iii) Generation time

Maize is an annual crop with a cultural cycle ranging from as short as 60 to 70 days to as long as 43 to 48 weeks from seedling emergence to maturity.

9.(b) Sexual compatibility with other cultivated or wild plant species

The current application is for import and use of NK603 × MON 810 maize in the E.U. Cultivation, the scenario in which this maize would reach sexual maturity in the field, is not within the scope of this notification.

Out-crossing with cultivated *Zea* varieties

In Europe, the potential for genetic transfer and exchange with other organisms is limited to other maize plants. Maize is wind pollinated, and the distance that viable pollen can travel depends on prevailing wind patterns, humidity, and temperature. All maize will inter-pollinate, except for certain popcorn varieties and hybrids that have one of the gametophyte factors (Ga^S, Ga, and ga allelic series on chromosome 4). Maize pollen, therefore, moves freely within an area, lands on silks of the same variety or different varieties, germinates almost immediately after pollination, and within 24 hours completes fertilisation. However, due to its relatively large mass, maize pollen does not move in significant absolute quantities more than a few metres from the crop. Research has shown that in adjacent leaves to a *B.t.* maize field at 0, 1 and 3 m distance from it, pollen deposition decreased significantly. In a study of out-crossing in maize undertaken in France, pollen flow as measured by successful fertilisation of neighbouring maize declined to 1 % at 10 m distance from the source crop.

Out-crossing with wild plant species

Cultivated *Zea mays* exhibits limited compatibility with wild *Zea* species, but these are not present in Europe.

10. Survivability

(a) Ability to form structures for survival or dormancy

Maize is an annual crop and seeds are the only survival structures. Natural regeneration from vegetative tissue is not known to occur. Modern maize cannot survive as a weed. Volunteer maize is not found growing in fencerows, ditches, and roadsides as a weed. Although maize from the previous crop year can over-winter in mild winter conditions and germinate the following year, it cannot persist as a weed. The appearance of maize in rotational fields following the maize crop from the previous year is rare under most European conditions. Maize volunteers are killed in areas that are subject to frosts or they are easily controlled by current agronomic practices including cultivation and the use of selective herbicides.

(b) Specific factors affecting survivability, if any

Maize cannot survive without human assistance and is not capable of surviving as a weed due to past selection in its evolution. In contrast to weedy plants, maize has a polystichous female inflorescence (ear) on a stiff central spike (cob) enclosed in husks (modified leaves). Consequently, seed dispersal of individual kernels naturally does not occur because of the structure of the ears of maize.

Maize grain survival is dependent upon temperature, moisture of seed, genotype, husk protection and stage of development. Freezing temperatures have an adverse effect on maize seed germination. Temperatures above 45°C have also been reported as injurious to maize seed viability.

11. Dissemination

(a) Ways and extent of dissemination

Maize is an annual crop and seeds are the only survival structures. Natural regeneration from vegetative tissue is not known to occur. Seed dissemination is impacted by mechanical harvesting and transport as well as insect or wind damage, which may cause some mature ears to fall to the ground and avoid harvest. Pollen dispersal is influenced by wind and weather conditions.

(b) Specific factors affecting dissemination, if any

In contrast to weedy plants, maize has a polystichous female inflorescence (ear) on a stiff central spike (cob) enclosed in husks (modified leaves). Consequently, seed dispersal of individual kernels does not occur naturally because of the structure of the ears of maize. Seed dissemination is impacted by mechanical harvesting and transport as well as insect or wind damage, all of which may cause some mature ears to fall to the ground, where they could remain un-harvested.

Genetic material can be disseminated by pollen movement. Pollen dispersal is influenced by wind and weather conditions. Measuring about 0.1 mm in diameter, maize pollen is the largest of any pollen normally disseminated by wind from a comparably low level of elevation. Dispersal of maize pollen is influenced by its large size and rapid settling rate.

12. Geographical distribution of the plant

Maize, because of its many divergent types, is grown over a wide range of climatic conditions. The bulk of the maize is produced between latitudes 30° and 55°, with relatively little grown at latitudes higher than 47° latitude anywhere in the world. The greatest maize production occurs where the warmest month isotherms range between 21 and 27 °C and the freeze-free season lasts 120 to 180 days. A summer rainfall of 15 cm is approximately the lower limit for maize production without irrigation.

13. In the case of plant species not normally grown in the Member State(s), description of the natural habitat of the plant, including information on natural predators, parasites, competitors and symbionts

Maize is widely grown in the European Union and represents a significant portion of global maize production. Significant areas of maize production in Europe include the Danube Basin from southwest Germany to the Black Sea along with southern France through the Po Valley of northern Italy.

14. Potentially significant interactions of the plant with other organisms in the ecosystem where it is usually grown, including information on toxic effects on humans, animals and other organisms

There are no known toxic effects of the maize plant to humans, animals or livestock; it has a history of safe use for human food and animal feed. However, maize is known to interact with other organisms in the environment including insects, birds, and mammals. It is susceptible to a

range of fungal diseases and nematode, insect and mite pests. (See also question ***31***.)

15. Phenotypic and genetic traits

Maize (*Zea mays*) is a wind-pollinated, monoecious species with separate staminate (tassels) and pistillate (silk) flowers, which encourages the natural out-crossing between adjacent maize plants. Self-pollination leads to homogeneity of the genetic characteristics within a single plant while cross-pollination combines the genetic traits of many plants. This inbred-hybrid concept and resulting yield response is the basis of the modern seed maize industry.

INFORMATION RELATING TO THE GENETIC MODIFICATION

16. Description of the methods used for the genetic modification

NK603 × MON 810 hybrid maize was created using conventional breeding techniques, by crossing two genetically modified inbred maize lines derived from the maize events NK603 and MON 810, respectively, so no additional transformation was involved.

17. Nature and source of the vector used

No genetic modification was involved in the production of NK603 × MON 810 maize. NK603 × MON 810 maize was produced by conventional breeding of two genetically modified inbred parental maize lines, derived from transformation events NK603 and MON 810. As described in the respective application dossiers for these single-trait maize lines, NK603 maize was generated using a particle acceleration transformation system and a DNA fragment, containing *cp4 epsps* genes encoding glyphosate-tolerant 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) enzymes from *Agrobacterium* sp. strain CP4, and MON 810 maize was produced by the integration of a DNA sequence, which contains the *cry1Ab* gene derived from *Bacillus thuringiensis* subsp. *kurstaki*, encoding the insecticidal Cry1Ab protein. The *cp4 epsps* and *cry1Ab* genes are inherited in the nuclear genome of NK603 × MON 810 maize.

18. Size, source [name of donor organism(s)] and intended function of each constituent fragment of the region intended for insertion

NK603 × MON 810 maize results from a single conventional cross of the inbred parental lines NK603 maize and MON 810 maize, which are made homozygous in their respective inserted sequences. By crossing NK603 and MON 810 maize, NK603 × MON 810 maize inherits the inserted DNA fragments from both its parental lines. The individual components and the function of the inherited sequences are given in Tables 1 and 2.

Table 1. DNA components of the inserted fragment inherited from NK603 maize

Genetic Element	Source	Size (kb)	Function
First *cp4 epsps* gene cassette			
P-ract1/ ract1 intron	*Oryza sativa*	1.4	Contains promoter, transcription start site and first intron.
ctp 2	*Arabidopsis thaliana*	0.2	Encodes chloroplast transit peptide, which directs the CP4 EPSPS protein to the chloroplast
cp4 epsps	*Agrobacterium* sp. strain CP4	1.4	Encodes glyphosate-tolerant CP4 EPSPS protein
NOS 3'	*Agrobacterium tumefaciens*	0.3	Ends transcription and directs polyadenylation of the mRNA.
Second *cp4 epsps* gene cassette			
e35S	Cauliflower mosaic virus	0.6	Promoter
Zmhsp70	*Zea mays L.*	0.8	Stabilizes the level of gene transcription.
ctp 2	*Arabidopsis thaliana*	0.2	Encodes chloroplast transit peptide, which directs the CP4 EPSPS protein to the chloroplast
cp4 epsps l214p	*Agrobacterium* sp. strain CP4	1.4	Encodes glyphosate-tolerant CP4 EPSPS L214P protein
NOS 3'	*Agrobacterium tumefaciens*	0.3	Ends transcription and directs polyadenylation of the mRNA.

Table 2. DNA components of the inserted fragment inherited from MON 810 maize

Genetic Element	Source	Size (kb)	Function
e35S	Cauliflower mosaic virus	0.6	Promoter
Zmhsp70	*Zea mays* L.	0.8	Stabilizes level of gene transcription.
cry1Ab	*Bacillus thuringiensis*	3.5	Encodes Cry1Ab protein, which targets specific lepidopteran insect pests

INFORMATION RELATING THE GMHP

19. Description of the trait(s) and characteristics, which have been introduced or modified

NK603 × MON 810 *hybrid* maize consists in the combination, by traditional breeding, of two genetically modified parental *inbred* lines, derived from maize transformation events NK603 and MON 810, respectively.

NK603 × MON 810 maize expresses CP4 EPSPS proteins, which impart tolerance to glyphosate (N-phosphonomethyl-glycine), the active ingredient in the non-selective herbicide Roundup. EPSPS is an enzyme involved in the shikimic acid pathway for aromatic amino acid biosynthesis in plants and microorganisms. CP4 EPSPS enzymes have been shown to have significantly reduced affinity for glyphosate herbicide when compared with the wild-type maize enzyme, and to retain catalytic activity in the presence of the inhibitor glyphosate. Therefore, when maize plants expressing the CP4 EPSPS proteins are treated with glyphosate, the plants are unaffected since the continued action of the tolerant CP4 EPSPS enzymes provides for the plant's need for aromatic amino acids.

NK603 × MON 810 maize also expresses the Cry1Ab protein, which provides the maize plant of protection from certain lepidopteran insect pests, including European Corn Borer (*Ostrinia nubilalis*) and pink borers (*Sesamia* spp.). The insecticidal activity of the Cry1Ab protein is specific to predation by the larvae of the targeted lepidopterans.

20. Information on the sequences actually inserted/deleted/modified

(a) Size and structure of the insert and methods used for its characterisation, including information on any parts of the vector introduced in the GMHP or any carrier or foreign DNA remaining in the GMHP

NK603 × MON 810 maize is produced by traditional breeding of two genetically modified inbred parental lines, one derived from maize transformation event NK603 and the other one derived from event MON 810. No additional genetic modification was applied.

Southern blot analysis using NK603 × MON 810 maize confirmed the presence of the NK603 and MON 810 events in NK603 × MON 810 maize. In addition, there are no indications for the presence of additional (fragments) of both insertions.

The size and structure of the sequences, actually inserted in the parental maize lines NK603 and MON 810, and inherited in NK603 × MON 810 hybrid maize are given in Tables 1 and 2 on page 10.

(b) In case of deletion, size and function of the deleted region(s)

Not applicable.

(c) Location of the insert in the plant cells (integrated in the chromosome, chloroplasts, mitochondria, or maintained in a non-integrated form), and methods for its determination

By conventional crossing of the NK603 and MON 810 inbred maize lines, the inserted sequences from each parental line are inherited in the nuclear genome of the resulting NK603 × MON 810 hybrid. The presence of these inserted DNA sequences has been confirmed by Southern blot analysis of NK603 × MON 810 maize, in comparison with both of the parental lines, NK603 and MON 810, as well as with non-transgenic maize controls.

(d) Copy number and genetic stability of the insert

NK603 × MON 810 hybrid seed (F1) is produced by a single cross of the NK603 and MON 810 parental inbred lines (made homozygous for event NK603 or MON 810, respectively) by traditional breeding. Thereby, each parental line passes on a single copy of its inserted DNA sequence to the resulting NK603 × MON 810 F1 hybrid seed, which is sown by the grower.

The single-trait modified maize lines NK603 and MON 810 each contain a single copy of the respective transformation fragment, which is stably integrated into the nuclear maize genome. The single traits are inherited as single dominant genes in a Mendelian fashion. This has been confirmed by Southern blot analyses and by studies of the inheritance pattern of these traits in maize.

The harvested (F2) grain of NK603 × MON 810 maize is marketed by the grower for food, feed or industrial use and is not used for further breeding. Therefore, since NK603 × MON 810 hybrid maize seed exists only for a single generation, there is no opportunity for its stability to be compromised.

(e) In case of modifications other than insertion or deletion, describe function of the modified genetic material before and after the modification as well as direct changes in expression of genes as a result of the modification

Not applicable.

21. Information on the expression of the insert

(a) Information on the expression of the insert and methods used for its characterisation

The amounts of the CP4 EPSPS and Cry1Ab proteins, present in plant tissues of NK603 × MON 810 maize, were measured using ELISA detection methods.

The mean level of CP4 EPSPS proteins in grain samples from NK603 × MON 810 maize was 12.7 μg/g fw. The mean level of Cry1Ab protein in NK603 × MON 810 maize grain was 0.73 μg/g fw. As expected, the levels of CP4 EPSPS and Cry1Ab proteins in tissue samples from non-transgenic control maize, which does not contain the *cp4 epsps* or *cry1Ab* genes, were below the limit of detection.

(b) Parts of the plant where the insert is expressed (*e.g.* roots, stem, pollen, etc.)

The expression of the CP4 EPSPS and Cry1Ab proteins is expected to occur throughout the whole plant since the *rice actin* and CaMV *e35S* promoters have been shown to drive constitutive expression of the encoded protein in genetically modified maize.

22. Information on how the GMHP differs from the recipient plant in

(a) Mode(s) and/or rate of reproduction

Except for the combined tolerance to Roundup and the protection from predation by certain Lepidoptera, no differences have been observed in the reproductive capability, dissemination or survival characteristics of NK603 × MON 810 maize when compared to conventional hybrid maize varieties. Based on the observational data collected in field trials since 2000, no differences between NK603 × MON 810 maize and its non-transgenic counterpart have been noted when comparing maize plant morphology, seedling emergence and vigour, disease susceptibility, or insect damage (other than protection from the targeted lepidopteran insect pests). The introduced traits had no influence on maize plant and reproductive morphology and hence no changes in seed dissemination and survival capacity would be expected. No phenotypic differences, except for Roundup tolerance and insect-protection, have been observed in all the test environments, when comparing NK603 × MON 810 maize to conventional maize varieties.

This conclusion is consistent with the data demonstrating that NK603 × MON 810 maize grain is substantially equivalent to conventional maize grain, with respect to composition, nutritional value and safety.

Finally, it should be noted that cultivation of NK603 × MON 810 maize in the E.U. is not within the scope of this application.

(b) Dissemination

Please see question ***22.(a)***.

(c) Survivability

Please see question ***22.(a)***.

(d) Other differences

Please see question ***22.(a)***.

23. Potential for transfer of genetic material from the GMHP to other organisms

NK603 × MON 810 maize, like all other maize, is not sexually compatible with any indigenous or introduced plant species present in Europe, except for other cultivated maize. Therefore, there is no potential for gene transfer from NK603 × MON 810 maize to wild plant species and there would be only limited likelihood for gene transfer to other maize crops if NK603 × MON 810 maize were to be cultivated in the E.U. In the case of this notification, however, since it does not include the cultivation of NK603 × MON 810

maize varieties in the E.U., the likelihood of this occurrence is negligible and the risk posed by this transfer, and hence by NK603 × MON 810 maize, would also be negligible.

24. Information on any harmful effects on human health and the environment, arising from the genetic modification

The assessment of the human and animal health safety of NK603 × MON 810 maize was conducted based upon an extensive characterisation of the introduced traits and based upon comparison of this maize and its parental lines with traditional, nontransgenic counterparts.

Neither the host plant, maize, nor the donor organisms of the introduced sequences, are known to be harmful for human or animal health or the environment. NK603 × MON 810 maize has been shown to be equivalent to other maize varieties, apart from tolerance to the herbicide Roundup by expression of CP4 EPSPS proteins and protection from predation by certain lepidopteran insect species by expression of Cry1Ab protein.

CP4 EPSPS proteins belong to a family of EPSPS enzymes that are commonly found in a wide variety of food sources, which have a long history of safe use. Moreover, (1) there were no indications of acute toxicity in mice administered CP4 EPSPS protein by oral gavage, which is consistent with (2) the rapid degradation of the CP4 EPSPS proteins and loss of enzymatic activity in simulated human gastric and intestinal fluids; (3) the CP4 EPSPS proteins are not homologous to known protein toxins or allergens; (4) the proteins are present at very low levels in NK603 × MON 810 maize, and finally, (5) these proteins are from a family of proteins with a long history of safe consumption, including the CP4 EPSPS protein present in Roundup Ready soybean.

Similarly, the human and animal health safety of the Cry1Ab protein has been established based upon the following considerations: (1) no indications of acute toxicity in mice administered Cry1Ab protein by oral gavage, (2) rapid degradation and loss of insecticidal activity under conditions which simulate mammalian digestion, (3) no amino acid sequence similarity to known toxins, other than *B.t.* proteins, and no immunologically relevant sequence similarity with known allergens, (4) very low dietary exposure, and (5) a history of safe use as MON 810 maize varieties expressing the same protein have been approved, marketed and consumed since 1996 in several world areas including the E.U. Finally, the Cry1Ab protein was demonstrated to be highly selective for certain insects, with no toxicity to other types of living organisms such as mammals, fish, birds or invertebrates.

In addition, the compositional and nutritional equivalence of grain and forage from NK603 × MON 810 maize and traditional maize have been established by compositional analysis. Finally, the wholesomeness of NK603 × MON 810 maize grain has been confirmed in a highly sensitive feeding study using broiler chickens.

In conclusion, on the basis of the extensive characterisation of the introduced traits, the history of safe use of the introduced proteins and the host plant, maize, the compositional and nutritional equivalence of NK603 × MON 810 maize versus traditional maize, and the absence of growth performance effects in a broiler feeding study, it is concluded that NK603 × MON 810 maize grain, containing the NK603 and MON 810 inserts, is as safe and nutritious as traditional maize hybrids.

25. Information on the safety of the GMHP to animal health, where the GMHP is intended to be used in animal feedstuffs, if different from that of the recipient/parental organism(s)

There is no difference between NK603 × MON 810 maize and the recipient organism in terms of safety to animals (see question ***24***).

26. Mechanism of interaction between the GMHP and target organisms (if applicable), if different from that of the recipient/parental organism(s)

The CP4 EPSPS proteins expressed in NK603 × MON 810 maize are glyphosate-tolerant EPSPS enzymes involved in the shikimate pathway of the plant, thereby conferring tolerance to Roundup herbicide. This introduced trait is of agronomic relevance and does not have any target organisms.

Unlike unmodified maize plants, NK603 × MON 810 maize expresses the Cry1Ab protein protecting this maize (in a cultivation scenario) from the European Corn Borer (*Ostrinia nubilalis*) and pink borers (*Sesamia* spp.). Non-target organisms are not affected, since the insecticidal action of the Cry1Ab protein is limited to the targeted lepidopteran insect pests. Furthermore, the scope of this notification does not include the cultivation of NK603 × MON 810 maize varieties in the E.U.

27. Potentially significant interactions with non-target organisms, if different from the recipient or parental organism(s)

Maize is known to interact with other organisms in the environment including insects, birds, and mammals, and it is susceptible to a range of fungal diseases and nematode, insect and mite pests. The interaction of NK603 × MON 810 maize with other organisms has been shown to be no different from traditonal maize, except for the introduced trait conferring protection from certain insect pests. The interaction of this maize with non-target organisms is, however, not different from traditional maize. Moreover, this application is for import and use in the E.U. of NK603 × MON 810 maize. The scope of this notification does not include the cultivation in the E.U. of NK603 × MON 810 varieties. (See also question ***31***.)

28. Description of detection and identification techniques for the GMHP, to distinguish it from the recipient or parental organism(s)

Southern blot or polymerase chain reaction (PCR) techniques may be employed for the detection and identification of the inserted sequences. Specific ELISA methods have been developed and could be used to detect expression of CP4 EPSPS or Cry1Ab proteins in individual plants and their grains. Alternatively, plants can also be sprayed with Roundup herbicide for the detection of the glyphosate tolerance and an insect bioassay using sensitive lepidopteran insect species such as tobacco hornworm (*Manduca sexta*), cabbage looper (*Trichoplusia ni*) or European Corn Borer (*Ostrinia nubilalis*) may be used to identify plants expressing the Cry1Ab protein or trait.

NK603 × MON 810 maize will be detectable using either of the event-specific PCR methods for detection of the introduced DNA also present in the respective parental lines derived from the NK603 and MON 810 modification events.

INFORMATION ON THE POTENTIAL ENVIRONMENTAL IMPACT FROM THE RELEASE OF THE GMHP

29. Potential environmental impact from the release or the placing on the market of GMOs (Annex II, D2 of Directive 2001/18/EC), if different from a similar release or placing on the market of the recipient or parental organism(s)

Analysis of the characteristics of NK603 × MON 810 maize has shown that the likelihood of potential adverse effects on human health and the environment in the European Union, resulting from its *import* and *use* as any other maize, including use in animal feed but not including the cultivation of NK603 × MON 810 varieties, is consistently negligible. Therefore, the overall environmental risk posed by the GMHP is also negligible, and strategies for risk management for NK603 × MON 810 maize would be the same as for traditional maize.

In the regions from which NK603 × MON 810 maize will be imported, its *cultivation* is expected to positively benefit farmers and the environment:

- The use of Roundup in maize is significant as it enables the farmer to take advantage of the herbicide's favourable environmental and safety properties. Glyphosate, the active ingredient in Roundup herbicide (1) has limited mobility as it binds rapidly and tightly to a wide variety

of soils and sediments, (2) is non-persistent as it is readily metabolised, (3) has a low potential to move into surface or groundwater and (4) does not accumulate in and presents minimal risk to terrestrial and aquatic animals including birds, mammals, fish and invertebrates. NK603 × MON 810 maize benefits the farmer by providing (1) an additional broad-spectrum weed control option in maize, (2) a new herbicidal mode of action for in-season maize weed control, (3) increased flexibility to treat weeds on an "as needed" basis, (4) cost-effective weed control and (5) an excellent fit with conservation tillage systems. In turn, a number of environmental benefits arise from the use of conservation tillage including (1) improved soil quality, (2) improved water infiltration, and therefore reductions in erosion and runoff of nutrients to surface water, (3) improved wildlife habitat, (4) increased carbon retention in the soil, and (5) reduced fuel use and encouragement of sustainable agricultural practices.

- Moreover, cultivated NK603 × MON 810 maize is also protected against predation by certain economically damaging lepidopteran insect pests, of which the European Corn Borer (*Ostrinia nubilalis*) and pink borers (*Sesamia* spp.) are prevalent in parts of Europe. The use of NK603 × MON 810 maize will enable the farmer to effectively control these maize pests, ensuring maximum realization of yield potential, while removing the environmental burden of the production, packaging and transport of insecticides, previously used to control *Ostrinia nubilalis* and *Sesamia* spp.

30. Potential environmental impact of the interaction between the GMHP and target organisms (if applicable), if different from that of the recipient or parental organism(s)

This application is restricted to import of grain and excludes cultivation of NK603 × MON 810 varieties in the E.U. The likelihood of potential adverse effects on human health and the environment in the European Union, resulting from the *import* of this maize and its *use* as any other maize, is consistently negligible.

31. Possible environmental impact resulting from potential interactions with non-target organisms, if different from that of the recipient or parental organism(s)

(a) Effects on biodiversity in the area of cultivation

As this application is for import and use of NK603 × MON 810 maize in the E.U., not including the cultivation in the E.U. of NK603 × MON 810 maize varieties, no adverse effects are expected on biodiversity and non-target organisms in the environment, including pollinators and endangered species.

(b) Effects on biodiversity in other habitats

Please see question ***31.(a)***.

(c) Effects on pollinators

Please see question ***31.(a)***.

(d) Effects on endangered species

Please see question ***31.(a)***.

C. INFORMATION RELATED TO PREVIOUS RELEASES

32. History of previous releases notified under Part B of the Directive 2001/18/EC and under Part B of Directive 90/220/EEC by the same notifier

(a) Notification number

NK603 × MON 810 maize has been planted in the E.U. for field-testing purposes (agronomic performance, efficacy, selectivity, yield assessment) and compositional analysis since 2000.

The Part B notification number for trial permits, obtained in France, is B/FR/00.02.06.

(b) Conclusions of post-release monitoring

The conclusions of the French field trials with NK603 × MON 810 maize, as well as the E.U. trials with the single-trait parental lines NK603 maize and MON 810 maize, relate to the assessment of agronomic performance, efficacy and selectivity, yield potential, residues determination, compositional analysis and breeding. Post-release surveillance confirmed that these modified maize lines and their progeny are unlikely to cause any adverse effects to human or animal health or to the environment.

(c) Results of the release in respect to any risk to human health and the environment (submitted to the Competent Authority according to Article 10 of Directive 2001/18/EC)

Post-release general surveillance from environments inside and outside the E.U. has shown that NK603 × MON 810 maize and its single-trait parental lines, NK603 and MON 810 maize, are unlikely to pose any risk of adverse effects to human or animal health or to the environment.

33. History of previous releases carried out inside or outside the Community by the same notifier.

(a) Release country

In the European Union, NK603 × MON 810 maize has been planted for field-testing in France since 2000 (see question ***32***). This maize is commercially grown in the United States since 2002. In addition, the two parental single-trait maize varieties, NK603 and MON 810 maize, have been extensively tested in the field prior to their approval and subsequent commercialisation in several countries around the world.

(b) Authority overseeing the release

France: Ministry of Agriculture

U.S.A.: United States Department of Agriculture and Environmental Protection Agency

(c) Release site

In the European Union, NK603 × MON 810 maize has been evaluated in field trials at several locations in France since 2000.

(d) Aim of the release

In the E.U. NK603 × MON 810 maize has been released for field-testing of agronomic performance (efficacy, selectivity, yield assessment), testing of agronomic equivalence and for compositional analysis. In addition, field tests in the U.S.A. were carried out to study protein expression.

NK603 × MON 810 maize is commercially grown in the U.S.A. since 2002.

(e) Duration of the release

Please see question ***33.*****(a)**.

(f) Aim of post-releases monitoring

No adverse effects of the GMHP have been identified (see question ***29***). This indicates that a requirement for case-specific post-release monitoring is not appropriate, which is consistent with approvals granted in other world areas.

NK603 × MON 810 maize has been commercialized alongside stewardship programmes involving downstream stakeholders in the use of this maize, in order to ensure the implementation of good agricultural practice in its cultivation and to ensure a channel of communication in the unlikely event that unanticipated adverse effects might occur.

No unanticipated effects have been observed in other world areas since the commercialization of NK603 × MON 810 maize and its single trait parental maize lines, nor during the many years of field-testing inside and outside the E.U.

(g) Duration of post-releases monitoring

Please see question ***33.*****(f)**.

(h) Conclusions of post-release monitoring

Please see question ***33.*****(f)**.

(i) Results of the release in respect to any risk to human health and the environment

Multi-year field-testing and post-marketing experience provided no evidence that grain and derived products from NK603 × MON 810 are likely to cause any adverse effects to human or animal health and the environment.

D. INFORMATION RELATING TO THE MONITORING PLAN – IDENTIFIED TRAITS, CHARACTERISTICS AND UNCERTAINTIES RELATED TO THE GMO OR ITS INTERACTION WITH THE ENVIRONMENT THAT SHOULD BE ADDRESSED IN THE POST COMMERCIALISATION MONITORING PLAN

1. Confirmation that any assumptions regarding the occurrence and impact of potential adverse effects of the GMO or its use in the e.r.a. are correct.

An environmental risk assessment (e.r.a.) of NK603 × MON 810 maize was undertaken in the context of the scope of the notification, that is, for consent for import and use in the E.U. as any other maize, including feed use but not including the cultivation of NK603 × MON 810 maize varieties. The e.r.a. shows that the overall risk to the environment posed by NK603 × MON 810 maize is negligible, relative to:

- Persistence or invasiveness
- Selective advantage
- Potential for gene transfer
- Impact on target organisms
- Impact on non-target organisms
- Effects on biogeochemical processes
- Changes in agricultural practice

Moreover, the e.r.a. has demonstrated that risk to human and animal health posed by NK603 × MON 810 maize is negligible, relative to:

- Persons in proximity or contact with the release
- Animal health and the consumption of derived products

The conclusions of the e.r.a. are derived from the results of scientific studies, rather than major assumptions. On this basis, it is proposed that no case-specific monitoring is indicated.

2. Identification of the occurrence of adverse effects of the GMO or its use on human health or the environment which were not anticipated in the e.r.a.

Since the notification of NK603 × MON 810 maize is for consent for import only, and since the majority of use of this maize will be for animal feed, it follows that unanticipated affects are most likely to be manifested as a result of this use.

Therefore, it is proposed:

- To provide traders and processors of bulk mixtures of maize grain, likely to contain NK603 × MON 810 maize grain, with product information about this maize. Traders and processors will be requested to inform the relevant authorities of any adverse effects on the environment or human health, which they consider to be attributable to NK603 × MON 810 maize grain.
- To inform the European feed industry directly, by way of a public announcement, of the consent for placing on the market of NK603 × MON 810 maize at such time as it appears in the Official Journal of the *Rapporteur* Member State. Monsanto will also offer to meet with interested operators to discuss 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 attributable to the feed use of NK603 × MON 810 maize grain.

- For the duration of the authorisation of NK603 × MON 810 maize, to immediately inform the Commission and the Competent Authorities for Directive 2001/18/EC, of any reports of adverse effects, which come to the attention of Monsanto, so that any reports can be further investigated by the appropriate authorities.