

**SUMMARY NOTIFICATION INFORMATION FORMAT FOR THE PLACING  
ON THE MARKET (IMPORT) OF 1507 MAIZE IN ACCORDANCE WITH  
DIRECTIVE 2001/18/EC**

**A. GENERAL INFORMATION**

**1. Details of notification**

**(a) Member State of notification**

The Netherlands

**(b) Notification number**

C/NL/00/10

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

The product described in notification **C/NL/00/10** consists of grain derived from *B.t.* Cry1F maize line 1507, referred to as 1507 maize, genetically modified to express CRY1F protein, conferring resistance to certain lepidopteran insect pests, and PAT protein, conferring tolerance to glufosinate-ammonium herbicide. The scope of the notification covers import of 1507 maize for use as any other maize including processing and use as food and feed, but not for cultivation.

**(d) Date of acknowledgement of notification**

11th December 2000

**2. Notifier**

**(a) Name of notifier**

Pioneer Hi-Bred International, Inc. as represented by Pioneer Overseas Corporation.

Mycogen Seeds, c/o Dow AgroSciences LLC.

This is a joint notification submitted by Pioneer Hi-Bred and Mycogen Seeds. PioneerHi-Bred is taking the lead for this submission.

**(b) Address of notifier**

Pioneer Overseas Corporation  
Avenue Tedesco, 7  
B-1160 Brussels  
Belgium

Pioneer Hi-Bred International, Inc.  
400 Locust Street, Suite 800  
Des Moines, IA 50309  
U.S.A.

Dow AgroSciences Europe  
European Development Centre  
3 Milton Park, Abingdon  
Oxon OX14 4RN  
United Kingdom

Mycogen Seeds  
c/o DowAgroSciences LLC  
9330 Zionsville Road  
Indianapolis, IN 46268-1054  
U.S.A.

**(c) Is the notifier**

**domestic manufacturer**

Pioneer Hi-Bred and Mycogen Seeds are developers of the technology and producers of 1507 maize seed (inbreds and hybrids). This notification is for the import of products derived from 1507 maize seed, including import of 1507 maize grain produced outside the EU.

**importer**

No.

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

Same as notifier.

**3. General description of the product**

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

Maize (*Zea mays* L.) is extensively cultivated and has a long history of safe use. Production has benefited from many improvements, particularly since the 1920's when maize varieties were developed by conventional breeding between progeny of two inbreds to give hybrid varieties which are superior to open-pollinated varieties in their agronomic characteristics, such as increased grain yield. Today, hybrid maize varieties are used in most developed countries for consistent agronomic performance and production. The product consists of grain derived from 1507 maize, genetically modified to express CRY1F protein, conferring resistance to certain lepidopteran insect pests, and PAT protein, conferring tolerance to glufosinate-ammonium herbicide.

**(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 notification (Ref: C/NL/00/10) is to import grain and products derived from 1507 maize seed (inbreds and hybrids). Cultivation of 1507 maize seed in the EU is being considered separately under notification C/ES/01/01, in accordance with

Directives 90/220/EEC and 2001/18/EC. Therefore, seed products of 1507 maize will not be placed on the EU market for cultivation following the import authorisation under notification **C/NL/00/10**.

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

Products from 1507 maize will be used in a manner consistent with current uses of maize grain and maize products. This will include import of grain and grain products into the EU for storage and processing into food, animal feed and industrial uses.

The cultivation of 1507 maize seed products in the EU is being considered separately under notification **C/ES/01/01** in accordance with Directives 90/220/EEC and 2001/18/EC; and the approval for food use of 1507 maize is also being considered separately in accordance with the Regulation (EC) No 258/97.

Maize, together with rice and wheat, is one of the most important cereal crops in the world with total production of 596.4 million tonnes in 2000 (FAOSTAT Database, 2000). The FAO estimation for the EU maize production in 2000 was 38.4 million tonnes. Majority of grain and forage derived from maize is used for animal feeds, and about 8% of the grain is processed for human food products mainly by wet-milling or dry-milling. Maize grain is also processed into industrial products (11%), such as ethyl alcohol by fermentation and highly refined starch by wet-milling to produce starch and sweetener products. In addition to milling, the maize germ can be processed to obtain maize oil.

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

The genetic modification in 1507 maize is not expected to impact the existing processing conditions used for maize, which are well known and have a long history of use. The safety evaluation contained in this notification for the import of 1507 maize provides further verification that no specific conditions of use and handling are required for 1507 maize. Imported 1507 maize products will be used, stored and handled as any other commercial maize products. See **Point A.3.h)** below for labelling of 1507 maize.

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

Not applicable. Grain and derived products from 1507 maize will be imported into the EU and will not be confined to any geographical areas within the EU.

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

This notification is for the import of 1507 maize to the EU. In the case of cultivation (separate notification, ref: **C/ES/01/01**), 1507 maize would be particularly suitable in

environments where there is infestation from lepidopteran insect pests, such as Southern Europe.

**(g) Any proposed packaging requirements**

The packaging, handling, and storage systems that are currently used for maize will apply. The grain and processed products of 1507 maize will be imported and packaged in the same manner as other maize products. See **Point A.3.h)** below for labelling of 1507 maize.

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

Product information to indicate that genetic modification has been used in the development of 1507 maize will be provided. A proposal for labelling of 1507 maize has been developed in accordance with Annex IV of Directive 2001/18/EC. This will enable the grain and grain products derived from 1507 maize to be labelled in accordance with Directive 2001/18/EC when imported to the EU.

**(i) Estimated potential demand**

(i) in the Community

Import of maize grain into the EU for 2000 was about 10.3 million tonnes (FAOSTAT Database, 2002). The 1507 maize grain is expected to be part of these imports.

(ii) in export markets for EC supplies

Maize grain is traded as a commodity and no specific demand for 1507 maize should be expected from export markets for EC supplies.

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

In accordance with the OECD guidance for the designation of a unique identifier for transgenic plants (ENV/JM/MONO(2002)7), the unique identification code assigned to 1507 maize is DAS-Ø15Ø7-1.

**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, 1507 maize has been notified in Italy, France and Spain for field trials under Part B of Directive 90/220/EEC.

Year	Member State	Notification No
1998	Italy	B/IT/98/19
1999	Italy	B/IT/98/19
1999	France	B/FR/99.03.09
2000	Italy	B/IT/98/19
2000	France	B/FR/99.03.09
2000	France	B/FR/00.03.04
2002	Spain	B/ES/02/11

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

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

Yes, a separate notification (Ref: C/ES/01/01) to market 1507 maize, including cultivation, has been submitted to Spain in accordance with Directives 90/220/EEC and 2001/18/EC. In addition, a separate application has been submitted to The Netherlands for food use in accordance with Regulation (EC) No 258/97.

**Or**

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

Yes, an application for registration of 1507 maize has been submitted to the US Environment Protection Agency (EPA). Also, an application for non-regulated status of 1507 maize to the US Department of Agriculture (USDA) was submitted in May 2000, and a notification concerning foods derived from 1507 maize to the US Food and Drug Administration (FDA) was submitted in July 2000. The corresponding permits were granted as follows: by US EPA and FDA on 18<sup>th</sup> May 2001 and by USDA on 14<sup>th</sup> June 2001.

In addition, applications have been submitted to Argentina, Australia/New Zealand, Canada, China, Japan, Korea, Mexico, South Africa, Switzerland and Taiwan. The necessary approvals for import, animal feed use and food safety of 1507 maize in Japan were obtained on 15<sup>th</sup> June, 28<sup>th</sup> May and 8<sup>th</sup> July of 2002, respectively. In Canada permits were granted by Health Canada for novel food use of 1507 maize on 10<sup>th</sup> October 2002 and by the Canadian Food Inspection Agency for animal feed use and environmental release on 11<sup>th</sup> October 2002. Approval for import of 1507 maize for animal feed and food use in South Africa was obtained on 12<sup>th</sup> December 2002.

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

No

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

No specific measures, differing from those for any other commercial maize grain, need to be taken in case of unintended release or misuse or for disposal and treatment. Any unintentional release or misuse of 1507 maize would be limited and highly unlikely to have any adverse effect. Furthermore, and if necessary, such limited release can be controlled by current agronomic practices such as selective use of herbicides (with the exception of glufosinate-ammonium). This is based on the conclusions from the environmental risk assessment (e.r.a.) for the placing on the market of 1507 maize in accordance to Annex II of Directive 2001/18/EC.

**B. NATURE OF THE GMHP CONTAINED IN THE PRODUCT INFORMATION RELATING TO THE RECIPIENT OR (WHERE APPROPRIATE) PARENTAL PLANTS**

**8. Complete name**

<b>(a) Family name</b>	Gramineae
<b>(b) Genus</b>	Zea
<b>(c) Species</b>	<i>Zea mays</i> L.
<b>(d) Subspecies</b>	None
<b>(e) Cultivar/breeding line</b>	Line Hi-II
<b>(f) Common name</b>	Maize, corn

**9. a) Information concerning reproduction**

**(i) Mode(s) of reproduction**

As a wind-pollinated, monoecious grass species, self-pollination and fertilisation and, cross-pollination and fertilisation, are usually possible and frequencies of each are normally determined by proximity and other physical influences on pollen dispersal.

**(ii) Specific factors affecting reproduction, if any**

Tasselling, silking, and pollination are the most critical stages of maize development, and grain yield is greatly impacted by moisture and fertility stress. Dispersal of maize pollen tends to be limited, as it is influenced by the large size and rapid settling rate of the pollen.

**(iii) Generation time**

Maize is an annual crop with a cultural cycle ranging from as short as 10 weeks to as long as 48 weeks covering the period of seedling emergence to maturity. This variance in maturity allows maize to be grown over a range of climatic conditions.

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

Maize will intra-pollinate and transfer genetic material between maize except for certain popcorn varieties. The extent of pollination will depend upon prevailing wind patterns, humidity and temperature. It is generally considered that teosinte (*Zea mays* ssp. *mexicana*) is an ancestor of maize. Teosinte is an ancient wild grass found in Mexico and Guatemala and is not present in the EU.

## **10. Survivability**

### **(a) Ability to form structures for survival or dormancy**

During the domestication of maize, many agronomically significant attributes for cultivation have been gained whilst losing its ability to survive in the wild. Maize is a non-dormant annual crop and seeds are the only survival structures. Natural regeneration of maize from vegetative tissue is not known to occur.

### **(b) Specific factors affecting survivability, if any**

Survival of maize seed is dependent upon temperature, moisture of seed, genotype, husk protection and stage of development. Maize seed can only survive under favourable climatic conditions. Freezing temperatures have an adverse effect on germination of maize seed and it has been identified as a major risk in limiting production of maize seed.

## **11. Dissemination**

### **(a) Ways and extent of dissemination**

Maize has a polystichous female inflorescence (ear) on a stiff central spike (cob) enclosed in husks (modified leaves). As a result, seed dispersal of individual kernels does not occur naturally.

### **(b) Specific factors affecting dissemination, if any**

Mechanical harvesting and transport are ways of disseminating grain and insect or wind damage may cause mature ears to fall to the ground and avoid harvest. Regardless of these routes of dissemination, maize cannot survive without human assistance.

## **12. Geographical distribution of the plant**

Maize is grown over a wide range of climatic conditions because of its many divergent types. However, survival and reproduction in maize is limited by cool conditions. 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. Maize has been cultivated in Europe starting in Southern Europe since the 16<sup>th</sup> century.

## **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**

Not applicable as maize has been cultivated in Europe since the 16<sup>th</sup> century.

**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**

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 insect pests, as well as competition from surrounding weeds. Maize is extensively cultivated and has a history of safe use. Maize or derived products of maize are not considered to have harmful characteristics. Maize has no pathogenic characteristics.

**15. Phenotypic and genetic traits**

Maize (*Zea mays* L.) is the only species usually included in the genus *Zea*, of the family Gramineae. It is a highly domesticated agricultural crop with well-characterised phenotypic and genetic traits.

Controlled cross-pollination of inbred lines from chosen genetic pools combines desired genetic traits in a hybrid resulting in improved agronomic performance and yield increase. This inbred-hybrid concept and resulting yield response is the basis of the modern maize seed industry.

**INFORMATION RELATING TO THE GENETIC MODIFICATION**

**16. Description of the methods used for the genetic modification**

The particle acceleration method was used to introduce into maize cells a linear DNA fragment containing the *cry1F* and *pat* coding sequences and the necessary regulatory components (insert PHI8999A). Maize event 1507 expressing the CRY1F protein and the PAT protein was produced, referred to as 1507 maize.

**17. Nature and source of the vector used**

No vector was used for the transformation of 1507 maize. As described in the notification, the intended insert consists of a linear DNA fragment, containing the *cry1F* and *pat* coding sequences together with the necessary regulatory components only, which was introduced by particle acceleration for the transformation of 1507 maize. No additional DNA sequences were used for introduction of the insert into 1507 maize.

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

The intended insert PHI8999A contains the plant optimised coding sequences for the *cry1F* and *pat* genes, together with the necessary regulatory components to drive their expression.

The *cry1F* gene (1818 bp; origin: *Bacillus thuringiensis* subsp. *aizawai*) is under the control of the ubiquitin promoter (1986 bp; origin: *Zea mays*) and the ORF25PolyA terminator (714 bp; origin: *Agrobacterium tumefaciens* pTi15995). The function of the CRY1F protein in 1507 maize is to provide resistance against certain lepidopteran insect pests such as the European corn borer.

The *pat* gene (552 bp; origin: *Streptomyces viridochromogenes* strain Tü494) is under the control of the CaMV35S promoter and terminator (554 and 204 bp, respectively; origin: cauliflower mosaic virus). The function of the PAT protein in 1507 maize is to tolerate application of glufosinate-ammonium herbicide.

## INFORMATION RELATING TO THE GMHP

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

The 1507 maize expresses CRY1F protein conferring resistance to certain lepidopteran insect pests, and PAT protein conferring tolerance to glufosinate-ammonium herbicide.

The *cry1F* gene is expressed constitutively by the *ubiZM1(2)* promoter. Expression of CRY1F protein provides control against lepidopteran insect pest damage to the above-ground parts of the maize plant including those parts which are beyond the reach of chemical insecticides. Specifically, the CRY1F protein confers season-long resistance against the European corn borer (*Ostrinia nubilalis*) and certain other lepidopteran pests such as the pink borer (*Sesamia* spp.). It is also highly effective against fall armyworm (*Spodoptera frugiperda*), black cutworm (*Agrotis ipsilon*) and southwestern corn borer (*Diatraea grandiosella*).

The *pat* gene is also expressed constitutively by the CaMV35S promoter. Expression of PAT protein confers tolerance to application of glufosinate-ammonium herbicide. Field trials show that 1507 maize will tolerate field application rates of 1600 g a.i./ha of glufosinate-ammonium herbicide without showing any phytotoxicity symptoms. Tolerance to glufosinate-ammonium herbicide provides for improved weed management.

No other new traits have been introduced into 1507 maize and, in particular, no trait for antibiotic resistance has been introduced. This is confirmed by the molecular characterization (**Point 30(f)** of notification **C/NL/00/10**), protein expression analysis (**Point 38** of notification **C/NL/00/10**), the expected agronomic performance (**Point 42** of notification **C/NL/00/10**), and comparable composition data to other conventional maize (**Annex 1** to notification **C/NL/00/10**).

## 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

The genetic modification in 1507 maize has been characterised in detail by Southern blot and DNA sequence analyses. The analyses have confirmed that the inserted genetic material is integrated into the nuclear genome of the maize plant and consists of an almost full-length copy of the linear fragment used in the transformation (*i.e.*, 6186 bp from the 6235 bp of insert PHI8999A, containing the *cry1F* and *pat* genes together with the regulatory sequences necessary for their expression). In addition, the plant insert contains the following non-functional fragments:

- one fragment (335 bp) of the *cry1F* gene, with no *ubiZM1(2)* promoter sequence, and one fragment (15 bp) of the *cry1F* gene, both located at the 5' end of the almost full-length insert;
- two fragments (201 bp, 138 bp) of the *pat* gene, without regulatory sequences associated, located at the 5' border and, one fragment (188 bp) of the *pat* gene, located at the 3' border;
- one fragment (118 bp) of the polylinker region and *ubiZM1(2)* promoter sequence located at the 5' border;
- one fragment (550 bp) of the ORF25PolyA terminator sequence in inverted position located immediately at the 3' end of the almost full-length insert.

The 1507 maize does not contain the *nptII* gene nor any other detectable fragments from the portion of plasmid PHP8999 that was not intended for transformation of 1507 maize. Maize genomic DNA flanking regions at both the 5' and 3' borders of the 1507 maize insert have been sequenced and characterised in detail. Analysis by PCR amplification has confirmed the presence of both genomic flanking regions in non-GM Hi-II maize used in the transformation of 1507 maize.

### (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, mitochondrion, or maintained in a non-integrated form), and methods for its determination

The inserts are integrated into the maize plant genome as confirmed in the molecular characterisation of 1507 maize by detailed Southern blot analysis and DNA sequencing.

**(d) Copy number and genetic stability of the insert**

As discussed above in **Point 20.(a)**, the actual insert in 1507 maize contains one copy of the almost full-length linear fragment used in the transformation, which includes one functional copy of the complete *cry1F* gene and one functional copy of the complete *pat* gene, together with the regulatory sequences necessary for their expression. The actual insert also contains two non-functional fragments of the *cry1F* gene; three non-functional fragments of the *pat* gene; one non-functional fragment of the polylinker region and *ubiZM1(2)* promoter; and, one non-functional fragment of the ORF25PolyA terminator sequence.

The inserted genetic material in 1507 maize is stable for at least six generations, and the *cry1F* and *pat* genes are inherited as Mendelian dominant genes. Results from Southern blot and DNA sequencing analyses show that the additional non-functional fragments were present in the BC4 backcross generation, **Point 30(f)** of notification **C/NL/00/10**, thus supporting the conclusion that they are genetically linked to the almost full-length insert containing the *cry1F* and *pat* genes.

**(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**

Leaf, pollen, silk, stalk, whole plant, grain and senescent whole plant tissue samples from 1507 maize and a control with comparable background genetics were obtained from field studies conducted during the growing seasons of 1998/99 in Chile, 1999 in France and Italy, and 2000 in France, Italy and Bulgaria. Expression levels of CRY1F and PAT proteins in these tissues were measured using specific Enzyme Linked Immunosorbent Assay (ELISA) developed for each protein. Results show that the CRY1F protein is expressed in all tissues and throughout the development of maize, while the PAT protein was measurable at the V9 developmental stage only.

The characteristics of the CRY1F and PAT proteins expressed in 1507 maize were further examined by Western blot analysis. The CRY1F protein was detected as two bands of approximately 65 and 68 kDa, respectively, which result from limited N-terminal processing of maize expressed CRY1F protein by a plant protease with trypsin-like specificity. No other bands indicative of a partial CRY1F protein or a fusion protein of greater molecular weight were observed.

The PAT protein is known to be a homodimer of approximately 43 kDa in its native form, and it is comprised of two components of approximately 22 kDa. The results of the Western blot analysis of 1507 maize confirmed the presence of the ~22 kDa PAT monomeric form and of the ~43 kDa PAT homodimer in leaf tissue. No other bands indicative of a partial PAT protein or fusion protein of greater molecular weight were observed in 1507 maize.

The genetic modification in 1507 maize results in expression of CRY1F protein conferring resistance to certain lepidopteran insect pests, and PAT protein conferring tolerance to glufosinate-ammonium herbicide. Specifically, the CRY1F protein confers season-long resistance against the European corn borer (*Ostrinia nubilalis*) and *Sesamia* spp. It is also highly effective against fall armyworm (*Spodoptera frugiperda*), black cutworm (*Agrotis ipsilon*) and southwestern corn borer (*Diatraea grandiosella*).

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

Addressed in **Point 21.(a)** above.

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

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

No unexpected changes in pollen production, seed production, seed viability or germination compared to non-GM maize have been observed in field trials of 1507 maize.

**(b) Dissemination**

Maize hybrids have been domesticated to the extent that the seeds cannot be disseminated without human intervention. The 1507 maize plants show no difference in dissemination compared to non-GM maize.

**(c) Survivability**

Cultivated maize has been domesticated to the extent that it can not survive outside managed agricultural environments. Lack of dormancy prevents maize seed to readily survive from one growing season to the next. The genetic modification in 1507 maize results in expression of CRY1F conferring resistance to certain lepidopteran insect pests and expression of PAT conferring tolerance to the herbicide glufosinate-ammonium. The survival characteristics of 1507 maize in the environment remain comparable to those of non-GM maize.

#### **(d) Other differences**

Maize does not exhibit any weedy tendencies and is non-invasive in natural ecosystems. Based on the agronomic data, there is no evidence for altered survival, multiplication, or dissemination of 1507 maize in the environment as compared to non-GM maize. In addition, the inserted traits do not alter the phenotype of maize in a way that would confer a fitness advantage for maize outside managed agricultural environments.

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

The potential for gene transfer to sexually compatible plant species will be negligible because this notification (C/NL/00/10) is for the import of 1507 maize and not for cultivation of 1507 maize seed products. Therefore, any release of 1507 maize into the environment will be unintentional, limited and highly unlikely to have any adverse effect. Furthermore, and if necessary, such limited release can be easily controlled by the application of current management practices used for the control of unintentional releases of maize plants, such as the application of non-selective herbicides with the exception of glufosinate-ammonium. In any case, there are no sexually compatible wild or weedy relatives of *Zea mays* known to exist in the EU, which eliminates the possibility of potential gene transfer to such species.

Transfer of transgenes originating from 1507 maize to bacteria is a negligible concern. There is no known mechanism for, or definitive demonstration of, DNA transfer from plants to microbes under natural conditions. Even if horizontal gene transfer were to take place, transfer of the *cry1F* or *pat* gene from 1507 maize does not represent a risk to human or animal health nor is it of consequence as a plant pest risk. The *nptII* gene coding for resistance to the antibiotic kanamycin is not present in 1507 maize.

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

##### **(i) Toxicity**

The genetic modification in 1507 maize results in expression of CRY1F and PAT proteins. The CRY1F protein has specific toxicity against certain lepidopteran insect pests (target organisms). An acute toxicity study with CRY1F protein in mice has confirmed the safety of the CRY1F protein to human and animal health. No mortality, toxicity or adverse clinical signs were observed at the highest dose tested of 5050 mg of test material per kg of body weight which was equivalent to 576 mg of pure CRY1F protein per kg of body weight. In addition, there is no evidence for CRY proteins originating from *Bacillus thuringiensis* to have harmful effects on the health of humans and animals.

The safety in terms of toxicity for the PAT protein has already been determined in detail during the assessment of glufosinate-ammonium tolerant maize. The *pat* gene was originally obtained from *Streptomyces viridochromogenes* strain Tü494 which has no known toxic or pathogenic potential. Toxicity studies carried out on rats and mice containing up to 50000 and 5000 mg/kg body weight respectively, have confirmed the absence of any adverse treatment-related clinical signs.

In addition, a poultry feeding study over a period of 42 days has been carried out confirming that there are no statistically significant differences on mortality, body weight gain or feed conversion between chickens fed a diet containing grain from 1507 maize or from non-GM maize.

#### (ii) Allergenicity

The most important factor to consider in assessing allergenic potential is whether the source of the gene being introduced into plants is known to be allergenic. Neither *Bacillus thuringiensis* (the source of the *cry1F* gene) nor *Streptomyces viridochromogenes* (the source of the *pat* gene) have a history of causing allergy. Also, both donor organisms are common soil bacteria.

The assessment of the allergenic potential of the CRY1F and PAT proteins has been made following the recommendations and the application of the decision-tree from FAO/WHO. The analyses have consisted of amino acid sequence comparison with known allergens, rapid degradation in simulated gastric fluids, relatively low level of expression, lack of glycosylation and thermolability. The results confirm that CRY1F and PAT proteins do not pose any significant risk of being a potential allergen.

#### (iii) Ecotoxicity studies

The absence of toxicity of the CRY1F protein to non-target and beneficial organisms has been thoroughly assessed in multiple dietary toxicity studies including green lacewing larvae, ladybird beetle, beneficial parasitic Hymenoptera *Nasonia vitripennis*, Monarch butterfly larvae, honey bees, earthworm, collembola and daphnia. In addition, levels of beneficial arthropods in field plots of 1507 maize were found to be comparable to those observed in non-GM maize.

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

A detailed safety evaluation concerning all possible feed applications of 1507 maize and feed products derived from 1507 maize (processed and non-processed) has been carried out (Annex 1 of notification C/NL/00/10).

The conclusions obtained confirm that feed products from 1507 maize are substantially equivalent to, nutritionally equivalent to, and as safe as, feed products

derived from commercially available (non-GM) maize. This is based on compositional analyses comprising protein, fiber, carbohydrates, ash, minerals, fatty acids, amino acids, vitamins, secondary metabolites and anti-nutrients in grain samples from 1507 maize; the nutritional equivalence shown in a poultry feeding study; and, the detailed safety evaluation of the CRY1F and PAT proteins expressed in 1507 maize.

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

This notification is for the import of 1507 maize and does not include cultivation in the EU. Therefore, there is no likelihood of any significant interaction between 1507 maize plants and target organisms. The cultivation of 1507 maize seed in the EU is being considered separately under notification **C/ES/01/01** in accordance with Directives 90/220/EEC and 2001/18/EC.

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

This notification is for the import of 1507 maize and does not include cultivation in the EU. Therefore, there is no likelihood of any significant interaction between 1507 maize plants and non-target organisms. The cultivation of 1507 maize seed in the EU is being considered separately under notification **C/ES/01/01** in accordance with Directives 90/220/EEC and 2001/18/EC.

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

The 1507 maize can be detected by placing small amounts of the glufosinate-ammonium herbicide on leaves of maize plants. Maize plants with the expression of the PAT protein will be those with leaves that do not show any necrosis at point of herbicide application. Alternatively, maize plants can be sprayed with glufosinate-ammonium herbicide, and those that survive will be expressing the PAT protein.

Plant parts of 1507 maize can also be analysed by ELISA to detect the proteins expressed by the *cry1F* and *pat* genes. Additionally, an insect bioassay with sensitive lepidopteran insect species such as European corn borer (*Ostrinia nubilalis*) can be used to identify maize plants expressing the CRY1F protein.

A PCR detection method to confirm the molecular identity of 1507 maize has been developed. The PCR method can also be used to confirm presence of 1507 maize for the purposes of labelling products containing or derived from 1507 maize. The detection method and seed of 1507 maize as reference material is provided to the Dutch regulatory authority. The PCR detection method and reference material will also be available to a central body operating under the auspices of the EU regulatory authorities and the European Commission.

A combination of the techniques described above can be used to identify 1507 maize.

## **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 GMO(s) (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)**

This notification is to import grain and products derived from 1507 maize seed. The grain and other products derived from 1507 maize will be imported, stored and processed for use in food, animal feed and industrial products in the same way as other conventional maize. This notification does not include the cultivation of 1507 maize in the EU and therefore the environmental impact from import of 1507 maize will be negligible as summarised in the environmental risk assessment (e.r.a.).

As intended by the genetic modification in 1507 maize, specific advantages for the agricultural environment have been conferred to 1507 maize: resistance to certain lepidopteran pests, such as the European corn borer and tolerance to glufosinate-ammonium herbicide. However, maize is highly domesticated, to the extent that it cannot become established as a feral species outside the agricultural environment, and the specific advantages introduced in 1507 maize do not confer any selective advantage to the maize plants in the natural environment, i.e. outside the agricultural environment. In addition, the scope of this notification for import of 1507 maize excludes cultivation, and the e.r.a. has determined that any risks to human and animal health or the environment from the placing on the market (import) of 1507 maize is as negligible as for any conventional maize.

### **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 notification is to import grain and products derived from 1507 maize seed and does not include cultivation in the EU. Therefore, there will be no significant direct and/or indirect interactions of 1507 maize with target organisms. The cultivation of 1507 maize seed in the EU is being considered separately under notification C/ES/01/01 in accordance with Directives 90/220/EEC and 2001/18/EC.

**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**
- (b) Effects on biodiversity in other habitats**
- (c) Effects on pollinators**
- (d) Effects on endangered species**

This notification is for the import of 1507 maize and does not include cultivation in the EU. Therefore, there will be no environmental impact resulting from potential interactions of 1507 maize with non-target organisms. The cultivation of 1507 maize seed in the EU is being considered separately under notification **C/ES/01/01** in accordance with Directives 90/220/EEC and 2001/18/EC.

**C. INFORMATION RELATING 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**

B/IT/98/19

**(b) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

**(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)**

No adverse effects on human health and the environment observed.

**(a) Notification number**

B/FR/99.03.09

**(b) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

**(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)**

No adverse effects on human health and the environment observed.

**(a) Notification number**

B/ES/02/11

**(b) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

**(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)**

No adverse effects on human health and the environment observed.

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

**(a) Release country**

Argentina.

**(b) Authority overseeing the release**

Secretary of Agriculture.

**(c) Release site**

Pergamino area, 3 sites; Buenos Aires Province.

**(d) Aim of the release**

Efficacy trials and hybrid registration.

**(e) Duration of the release**

One season.

**(f) Aim of post-release monitoring**

Control of potential volunteers.

**(g) Duration of post-release monitoring**

One season.

**(h) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

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

No adverse effects on human health and the environment observed.

**(a) Release country**

Brazil

**(b) Authority overseeing the release**

CTNBio

**(c) Release site**

One site.

**(d) Aim of the release**

Research.

**(e) Duration of the release**

One season.

**(f) Aim of post-release monitoring**

Control of potential volunteers.

**(g) Duration of post-release monitoring**

One season.

**(h) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

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

No adverse effects on human health and the environment observed.

**(a) Release country**

Chile.

**(b) Authority overseeing the release**

Ministry of Agriculture.

**(c) Release site**

Four sites.

**(d) Aim of the release**

Research.

**(e) Duration of the release**

One season.

**(f) Aim of post-release monitoring**

Control of potential volunteers.

**(g) Duration of post-release monitoring**

One season.

**(h) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

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

No adverse effects on human health and the environment observed.

**(a) Release country**

South Africa.

**(b) Authority overseeing the release**

Ministry of Agriculture.

**(c) Release site**

One site.

**(d) Aim of the release**

Research.

**(e) Duration of the release**

One season.

**(f) Aim of post-release monitoring**

Control of potential volunteers.

**(g) Duration of post-release monitoring**

One season.

**(h) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

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

No adverse effects on human health and the environment observed.

**(a) Release country**

U.S.A.

**(b) Authority overseeing the release**

USDA and EPA.

**(c) Release site**

Multiple sites.

**(d) Aim of the release**

Research.

**(e) Duration of the release**

Five seasons.

**(f) Aim of post-release monitoring**

Control of potential volunteers.

**(g) Duration of post-release monitoring**

One season.

**(h) Conclusions of post-release monitoring**

The 1507 maize plants performed as expected, with no evidence of any unintentional morphological or phenotypical characteristics. In particular, there was no evidence of enhanced weediness of 1507 maize.

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

No adverse effects on human health and the environment observed.

**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**

The environmental risk assessment (e.r.a.) concluded that the introduced traits, characteristics and interaction with the environment of imported 1507 maize will not give rise to any identified adverse effect to human and animal health or the environment. Therefore, the risk to human and animal health or the environment arising from the placing on the market (import) of 1507 maize is as negligible as for any conventional maize, concluding that 1507 maize is as safe as any commercially available maize regarding human and animal health or the environment.

Case-specific monitoring should only be carried out in those cases where potential adverse effects have been identified in the e.r.a. Due to the absence of identified adverse effects in the e.r.a., case-specific monitoring is not applicable to the placing on the market (import) of 1507 maize.

As detailed in the e.r.a., any exposure to the environment from the import of 1507 maize will be limited to any unintended release of 1507 maize e.g. via spillage during transportation of the grain. However, this limited exposure will not give rise to any adverse environmental impact, and if necessary, can be easily controlled by the application of current agricultural practices used for the control of volunteer maize plants, such as tilling or the application of non-selective herbicides with the exception of glufosinate-ammonium.

Therefore, application of established routine surveillance practices (e.g. the monitoring of agricultural cultivars or plant protection products) for the general surveillance of the occurrence of unanticipated adverse effects to conventional agricultural practice due to the placing on the market of 1507 maize as imports is not necessary.

However and since the majority of imported maize is used for animal feed purposes, general surveillance might assist in confirming the safety of animal feed products derived from 1507 maize with a view to safeguarding against any unanticipated effects. If this is considered necessary, we can discuss with the relevant national Competent Authorities and associated bodies the co-ordination of a surveillance network, where appropriate, within the framework of their routine surveillance of animal feed products derived from commercial maize.