Application for authorization of stacked Bt11 x MIR604 x GA21 maize cultivation in the European Union under Regulation (EC) No 1829/2003

PART II: SUMMARY

This document is complete as of June 2010. Since it is submitted as one part of a regulatory application, which is subject to an on-going regulatory review, it may be subject to later amendment or replacement. The information may also be supplemented with additional material requested by regulatory authorities. As such, it may only be considered properly with reference to those later amendments or supplementary materials and in the context of the dossier as a whole.

A. GENERAL INFORMATION

1. Details of application

a) Member State of application UK
b) Application number Not available at the time of submission
c) Name of the product (commercial and other names) Bt11 x MIR604 x GA21 maize
d) Date of acknowledgement of valid application Not available at the time of submission

2. Applicant

a) Name of applicant Syngenta Seeds S.A.S on behalf of Syngenta Crop Protection AG, Basel
b) Address of applicant Syngenta Seeds S.A.S. 12, chemin de l'Hobit BP 27 F-31790 Saint-Sauveur On behalf of Syngenta Crop Protection AG, Basel Switzerland and all affiliated companies Schwarzwaldallee 215 CH 4058 Basle Switzerland
c) Name and address of the person established in the Community who is responsible for the placing on the market, whether it be the manufacturer, the importer or the distributor, if different from the applicant (Commission Decision 2004/204/EC Art 3(a)(ii)) Bt11 x MIR604 x GA21 maize will be cultivated and used as any other maize in the EU by operators currently involved in these processes.

3. Scope of the application

☐ GM plants for food use

☐ Food containing or consisting of GM plants

☐Food produced from GM plants or containing ingredients produced from GM plants

☐GM plants for feed use

☐ Feed containing or consisting of GM plants

☐Feed produced from GM plants

☐Import and processing (Part C of Directive 2001/18/EC)

☒Seeds and plant propagating material for cultivation in Europe (Part C of Directive 2001/18/EC)

4. Is the product being simultaneously notified within the framework of another regulation (e.g. Seed legislation?)?

Yes ☐	No ☒
If *yes*, specify	

5. Has the GM plant been notified under Part B of Directive 2001/18/EC and/or Directive 90/220/EEC?

Yes ☒ (for release in 2008)	No ☐
If *no*, refer to risk analysis data on the basis of the elements of Part B of Directive 2001/18/EC	

6. Has the GM plant or derived products been previously notified for marketing in the Community under Part C of Directive 2001/18/EC or Regulation (EC) 258/97?

Yes ☐	No ☒
If *yes*, specify	

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

Yes ☒	No ☐

If *yes*, specify

Bt11 x MIR604 x GA21 maize is approved for commercial cultivation in the USA and Canada. Bt11 x MIR604 x GA21 maize is also approved for import in Mexico, Japan, Philippines and Taiwan. A number of other submissions have been made in other countries around the world and these are at different stages in the approval process.

8. General description of the product

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

Bt11 x MIR604 x GA21 maize is a stacked genetically modified (GM) product that has been produced by conventional breeding crosses of:

- Event Bt11 maize (Bt11 maize) which produces a truncated Cry1Ab protein for control of certain lepidopteran pests and a phosphinothricin acetyltransferase (PAT) protein that confers tolerance to herbicide products containing glufosinate ammonium.
- Event MIR604 maize (MIR604 maize) which produces a modified Cry3A (mCry3A) protein for control of certain coleopteran pests and a phosphomannose isomerase (MIR604 PMI) protein, which acts as a selectable marker enabling transformed plant cells to utilize mannose as a primary carbon source.
- Event GA21 maize (GA21 maize) which produces a modified maize 5-enolpyruvylshikimate-3-phosphate synthase enzyme (mEPSPS) that confers tolerance to herbicide products containing glyphosate.

b) Types of products planned to be placed on the market according to the authorisation applied for

This application requests the authorization for production and cultivation of genetically modified Bt11 x MIR604 x GA21 maize in the European Union, including seed production and breeding of the GM maize lines necessary to generate Bt11 x MIR604 x GA21 maize. This corresponds to category 3.2 of Annex II of the EFSA Guidance Document (EFSA, 2006a).

Concurrently, a separate application has been submitted requesting the authorization for production and cultivation of genetically modified Event MIR604 maize in the European Union, including seed production and breeding.

c) Intended use of the product and types of users

It is intended that Bt11 x MIR604 x GA21 maize will be used as any other conventional maize which is cultivated or imported for all food, feed and industrial purposes.

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

The characteristics of Bt11 x MIR604 x GA21 maize and products derived from it are not different from those of its conventional counterpart, apart from the introduced traits of insect tolerance and tolerance to herbicide products containing glufosinate ammonium or glyphosate. Bt11 x MIR604 x GA21 maize has been shown to be as safe and as wholesome as existing varieties of maize. Therefore there are no specific instructions or recommendations for use, storage and handling of Bt11 x MIR604 x GA21 maize.

e) Any proposed packaging requirements

The characteristics of Bt11 x MIR604 x GA21 maize and products derived from it are not different from those of its conventional counterpart. Bt11 x MIR604 x GA21 maize has been shown to be as safe and as wholesome as existing varieties of maize. Therefore there are no specific instructions for packaging.

f) A proposal for labelling in accordance with Articles 13 and Articles 25 of Regulation (EC) 1829/2003. In the case of GMOs, food and/or feed containing or consisting of GMOs, a proposal for labelling has to be included complying with the requirements of Article 4, B(6) of Regulation (EC) 1830/2003 and Annex IV of Directive 2001/18/EC

A proposal for labelling has been included in the application following the guidance provided by EFSA. This includes the labelling requirements outlined by Regulation (EC) No 1829/2003 and Annex IV of Directive 2001/18/EC. Bt11 x MIR604 x GA21 maize grain will therefore be labelled as “genetically modified maize” and products derived from it will be labelled as “containing (or produced from) genetically modified maize”. Since Bt11 x MIR604 x GA21 maize and products derived from it are not different from those of its conventional counterpart, no additional labelling is required.

g) Unique identifier for the GM plant (Regulation (EC) 65/2004; does not apply to applications concerning only food and feed produced from GM plants, or containing ingredients produced from GM plants)

A unique identifier for Bt11 x MIR604 x GA21 maize has been assigned in accordance with Commission Regulation (EC) 65/2004: SYN-BTØ11-1 x SYN-IR6Ø4-5 x MON-∅∅∅21-9.

h) If applicable, geographical areas within the EU to which the product is intended to be confined under the terms of the authorisation applied for. Any type of environment to which the product is unsuited

Bt11 x MIR604 x GA21 maize is suitable for use as any other maize under the terms of the authorisation applied for.

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

Maize is incapable of sustained reproduction outside domestic cultivation and is non-invasive of natural habitats. The characteristics of Bt11 x MIR604 x GA21 maize and products derived from it are not different from those of its conventional counterpart, apart from the intended effect of tolerance to certain lepidopteran and coleopteran insect pests and herbicide products containing glufosinate ammonium or glyphosate.

Bt11 x MIR604 x GA21 maize has been shown to be as safe and as wholesome as existing varieties of maize. Any unintended releases or misuse can be dealt with in the same way as any other conventional maize.

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

1. Complete name

a) Family name Poaceae (formerly Gramineae)
b) Genus *Zea*
c) Species *mays*
d) Subspecies *mays*
e) Cultivar/breeding line or strain Bt11 x MIR604 x GA21 maize
f) Common name Maize; corn

2 a. Information concerning reproduction

(i) Mode(s) of reproduction *Zea mays* is an allogamous plant that propagates through seed produced predominantly by wind-borne cross-pollination. Self pollination of up to 5% may be observed. Male and female flowers are separated on the plant by about 1–1.3m. *Z. mays* has staminate flowers in the tassels and pistillate flowers on the ear shoots. *Z. mays* is a plant with protoandrous inflorescence; however, decades of conventional selection and breeding have produced varieties of maize with protogyny. There is no asexual reproduction in maize.
(ii) Specific factors affecting reproduction The key critical stages of maize reproduction are tasselling, silking, pollination and fertilization. Climatic and drought stress affect pollen viability and silk longevity thus potentially limiting the period of possible cross-pollination. Maize pollen is very sensitive to dehydration as it loses water rapidly. Other factors like rainfall or irrigation inhibit pollen emission because the anther dehiscence is limited by the mechanical layer. In general, maize pollen is only viable for a few hours after

emission. As maize pollen is large and heavy it tends to be deposited close to the source plant and studies have indicated that most maize pollen falls within 5m of the field's edge. In general, such studies have shown that over 98% of maize pollen remains within a radius of 25-50m of the source, although some grains can travel several hundred meters. Climatic conditions also affect grain and seed production, especially under drought conditions during flowering, tasseling and silking. If severe drought occurs during these phenological stages, the grain yield is reduced.

(iii) Generation time

Maize is an annual crop. The generation time from sowing to harvesting varies according to the genetic background and the climate, it can range from as short as 60 to 70 days to as long as 43 to 48 weeks from seedling emergence to maturity.

2 b. Sexual compatibility with other cultivated or wild plant species

Other cultivated plant species: The sexual compatibility of maize with other cultivated plant species is limited to *Zea* species.

Wild plant species*:* No wild relatives of maize are present in Europe. Therefore, maize cannot exchange genes with any other wild species in the EU.

3. Survivability

a) Ability to form structures for survival or dormancy

Maize is an annual crop. Seeds are the only survival structures; they cannot be dispersed without mechanical disruption of the cobs and show little or no dormancy. Natural regeneration from vegetative tissue is not known to occur.

b) Specific factors affecting survivability

Survival of maize is dependent upon temperature, seed moisture, genotype, husk protection and stage of development. Maize cannot persist as a weed. Maize seed can only survive under a narrow range of climatic conditions. Volunteers are killed by frost or easily controlled by current agronomic practices including cultivation and the use of selective herbicides. Maize is incapable of sustained reproduction outside of domestic cultivation and is non-invasive of natural habitats.

4. Dissemination

a) Ways and extent of dissemination

Maize dissemination can only be accomplished through seed dispersal. Seed dispersal does not occur naturally due to the structure of the ear.

b) Specific factors affecting dissemination

Compared to other wind-pollinated species, maize pollen grains are relatively large and therefore settle to the ground rapidly and have usually a short flight range. Although vertical wind movements or gusts during pollen shedding can lift pollen up high in the atmosphere and distribute it over significant distances, concentrations of viable pollen considerably decrease with height and distance from the source. Hence, only low levels of cross-pollination could occur over longer distances under suitable climatic conditions.

5. Geographical distribution and cultivation of the plant, including the distribution in Europe of the compatible species

Maize is the world's most widespread cereal with very diverse morphological and physiological traits; it is grown on approximately 161 million hectares worldwide (2008). Maize is distributed over a wide range of conditions: from latitudes 50º North to 50º South, below sea level of the Caspian plains up to 3000m in the Andes Mountains and from semi-arid regions to arid regions. The greatest maize production occurs where the warmest month isotherms range between 21º and 27º C and the freeze-free season lasts 120-180 days.

The EU is the fourth largest grain maize producer in the world, after the USA, China and Brazil. In the EU-27, grain maize was cultivated on about 8.4 million hectares (2009) with a production of 57 million tonnes (2009). Another major maize product is silage maize produced on about 5.1 million hectares (2008).

There are no wild relatives of maize in Europe.

6. 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 was introduced into Europe in the 15th century by Columbus and is widely grown in the European Union Member States.

7. Other potential interactions, relevant to the GM plant, of the plant with organisms in the ecosystem where it is usually grown, or used elsewhere, 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 to competition from surrounding weeds. Maize is extensively cultivated and has a history of safe use for human food and animal feed. No significant native toxins are reported to be associated with the genus *Zea*.

C. INFORMATION RELATING TO THE GENETIC MODIFICATION

1. Description of the methods used for the genetic modification

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the genetically modified (GM) maize events: Bt11, MIR604 and GA21 through conventional breeding techniques. There was no further genetic modification to produce the stack.

The Bt11, MIR604 and GA21 maize events maize were produced by genetic modification as follows:

- Bt11 maize was produced using protoplast transformation/regeneration
- MIR604 maize was produced via *Agrobacterium*-mediated transformation
- GA21 maize was produced via microprojectile bombardment of maize suspension culture cells.

2. Nature and source of the vector used

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques.

The vectors used to produce Bt11 maize, MIR604 maize and GA21 maize are as follows:

- The Plasmid pZO1502, cut with a *Not*I restriction enzyme, was used to produce Bt11 maize. The plasmid is a derivative of the commercially available plasmid pUC18.
- The Plasmid pZM26, a binary vector used for *Agrobacterium* mediated plant transformation, was used to generate MIR604 maize.
- A *Not*I restriction fragment from the Plasmid pDPG434, was used to transform GA21 maize via microprojectile bombardment transformation. The plasmid is derived from a pSK- vector which is commonly used in molecular biology and is derived from pUC19.

3. Source of donor DNA, size and intended function of each constituent fragment of the region intended for insertion

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques. There was no further genetic modification to produce the stacked product. The size, source and intended function of each constituent fragment of the regions intended for insertion in each of the single events is described below:

Event Bt11 maize

Vector Component	Approx. Size (bp)	Description
35S	509	Promoter from the cauliflower mosaic virus.
IVS6-ADH1	471	Maize intron sequence from the maize alcohol dehydrogenase gene used to enhance gene expression in maize.
cry1Ab	1848	*cry1Ab* gene, which encodes a Cry1Ab protein that confers resistance to certain lepidopteran insect pests. The *cry1Ab* gene was originally cloned from *Bacillus thuringiensis* var. *kurstaki* HD-1
NOS	253	Polyadenylation region from the nopaline synthase gene from *Agrobacterium tumefaciens*.
35S	418	Promoter from the cauliflower mosaic virus.
IVS2-ADH1	180	Maize intron sequence from the maize alcohol dehydrogenase gene used to enhance gene expression in maize.
pat	552	*Streptomyces viridochromogenes* gene encoding the selectable marker PAT. PAT confers resistance to herbicides containing glufosinate
NOS	253	Polyadenylation region from the nopaline synthase gene from *Agrobacterium tumefaciens*.

Event MIR604 maize

Vector component	**Size (bp)**	**Description**
MTL	2556	Promoter derived from the *Zea mays* (maize) metallothionein-like gene.
mcry3A	1797	A modified *cry3A* gene that confers tolerance to western corn rootworm *(Diabrotica virgifera virgifera)* and related *Diabrotica* species.
NOS	253	Polyadenylation region from the nopaline synthase gene from *Agrobacterium tumefaciens.*
ZmUbiInt	1993	Promoter from *Zea may* polyubiquitin genes
pmi	1176	*E. coli pmi* gene encoding the enzyme phosphomannose isomerase (PMI)
NOS	253	Polyadenylation region from the nopaline synthase gene from *Agrobacterium tumefaciens.*

Event GA21 maize

Vector Component	Approx. Size (Kb)	Description
Rice actin promoter, exon and intron	1.4	5' region of the rice actin 1 gene containing the promoter and first exon and intron provides constitutive expression of the *mepsps* gene in maize.
Optimised transit peptide	0.4	Optimised transit peptide sequence constructed based on transit peptide sequences from maize and sunflower ribulose-1,5-bis phosphate carboxylase oxygenase (RuBisCo) genes.
Modified maize EPSPS gene	1.3	Mutated *epsps* gene, which confers resistance to herbicide products containing glyphosate.
Nos 3' end	0.3	Polyadenylation region from the nopaline synthase gene from *Agrobacterium tumefaciens*.

D. INFORMATION RELATING TO THE GM PLANT

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

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques and produces the following proteins:

- a truncated Cry1Ab protein for control of certain lepidopteran pests.
- a phosphinothricin acetyltransferase (PAT) protein that confers tolerance to herbicide products containing glufosinate ammonium.
- a modified Cry3A (mCry3A) protein for control of certain coleopteran pests.
- a phosphomannose isomerase (MIR604 PMI) protein as a selectable marker.
- a modified maize EPSPS (mEPSPS) protein that confers tolerance to herbicide products containing glyphosate.

2. Information on the sequences actually inserted or deleted

a) The copy number of all detectable inserts, both complete and partial

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques.

The inserts in Bt11 maize and MIR604 maize are present at a single locus and inherited as a single gene in a Mendelian fashion. The insert in GA21 maize is comprised of six contiguous regions derived from the 3.49 kb *Not*I restriction fragment from pDPG434 employed in the generation of GA21 maize (copies 1-6). Copy 1 contains the rice actin promoter that has a 5' deletion of 696 bp, the actin first exon and intron, the optimized transit peptide, the *mepsps* gene and the NOS terminator. Copies 2, 3 and 4 are intact versions of the 3.49 kb *Not*I restriction fragment from pDPG434. Copy 5 contains a complete rice actin promoter, the actin first exon and intron, the optimized transit peptide and the first 288 bp of the *mepsps* gene which ends in a stop codon and does not contain the NOS terminator. Copy 6 contains the rice actin promoter and a truncated actin first exon only and contains no other elements from pDPG434.

In addition to sequencing, southern analysis performed on each of the single events demonstrate the absence of further copies of the insert or vector sequence elsewhere in the genome. In order to assess the integrity of the insert from each individual event during conventional breeding to produce Bt11 x MIR604 x GA21 maize, additional Southern analysis was performed. The predicted DNA hybridization patterns from each individual event were confirmed in Bt11 x MIR604 x GA21

maize, demonstrating preservation of the integrity of the transgenic fragment from each individual event to Bt11 x MIR604 x GA21 maize.

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

Not applicable

c) Chromosomal location(s) of insert(s) (nucleus, chloroplasts, mitochondria, or maintained in a non-integrated form), and methods for its determination

The inheritance pattern of the inserts in Bt11, MIR604 and GA21 maize were analysed and the results showed that insertions had taken place in the nucleus.

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques. It therefore contains the inserts derived from the single events. The presence of the inserts from Bt11, MIR604 and GA21 maize in the stacked product was confirmed by Southern blot analyses.

d) The organisation of the inserted genetic material at the insertion site

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques. The organisation of the inserted genetic material in Bt11, MIR604 and GA21 maize is as follows:

Bt11 maize

Sequencing and southern data have demonstrated that Bt11 maize contains a single DNA insertion with one copy of both the *cry1Ab* and the *pat* genes.

MIR604 maize

Sequencing and southern data have demonstrated that MIR604 maize contains a single DNA insertion with one copy of both the *mcry3A* and the *pmi* genes.

GA21 maize

Sequence analysis of the GA21 maize insert demonstrates that the insert is comprised of six contiguous regions derived from the 3.49 kb *Not*I restriction fragment from pDPG434 employed in the generation of Event GA21 (copies 1-6).

A molecular comparison of the Bt11 x MIR604 x GA21 maize with the single events Bt11, MIR604 and GA21 maize has shown that the inserts are preserved in Bt11 x MIR604 x GA21.

3. Information on the expression of the insert

a) Information on developmental expression of the insert during the life cycle of the plant

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding techniques. Therefore these maize plants produce the transgenic proteins inherited from these single GM maize events: Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS.

Tissues from maize plants derived from Bt11 maize, MIR604 maize, GA21 maize and a breeding stack containing these events (Bt11 x MIR604 x GA21) were analyzed by ELISA to compare the concentrations of Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS.

The analyses were performed on key plant tissues collected from transgenic hybrid plants at different sampling times across the growing season. To control for background effects, the corresponding tissues from a near-isogenic control maize were also analyzed.

Mean Cry1Ab and PAT concentrations were comparable in the tissues of Bt11 maize and Bt11 x MIR604 x GA21 maize. Similarly, mCry3A and MIR604 PMI concentrations were comparable in the tissues of MIR604 maize and Bt11 x MIR604 x GA21 maize and mEPSPS concentrations were comparable in the tissues of GA21 maize and Bt11 x MIR604 x GA21 maize. As expected, no Cry1Ab, PAT, mCry3A, MIR604 PMI or mEPSPS proteins were detected in the near-isogenic control samples. Although some statistically significant differences were seen, these differences were small or not consistent across the growing season. These results support the conclusion that, as expected, transgenic protein expression in Bt11 x MIR604 x GA21 maize is not substantially different from that of the Bt11, MIR604 or GA21 single maize events.

b) Parts of the plant where the insert is expressed

To characterize the range of expression of Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS proteins in Bt11 x MIR604 x GA21 maize plants and the single events, the concentrations of these proteins were determined by ELISA in several plant tissues (leaves, roots, kernels and pollen).

Quantifiable concentrations of Cry1Ab protein were detected in leaves, roots and kernels derived from Bt11 maize and Bt11 x MIR604 x GA21 maize. Very low levels of Cry1Ab expression were detected in the pollen of Bt11 maize and Bt11 x MIR604 x GA21 maize.

Quantifiable concentrations of PAT protein were detected in leaves and roots derived from Bt11 maize and Bt11 x MIR604 x GA21 maize at most stages of development, however no quantifiable levels could be detected in the kernels or pollen.

Quantifiable concentrations of mCry3A protein were detected in leaves, roots and kernels derived from MIR604 maize and Bt11 x MIR604 x GA21 maize. Very low

levels of mCry3A expression were detected in the pollen of MIR604 maize and Bt11 x MIR604 x GA21 maize, but these were not quantifiable.

Quantifiable concentrations of MIR604PMI protein were detected in all MIR604 maize and Bt11 x MIR604 x GA21 maize derived plant tissues.

Quantifiable concentrations of mEPSPS protein were detected in all GA21 maize and Bt11 x MIR604 x GA21 maize derived plant tissues.

4. Information on how the GM plant differs from the recipient plant in

a) Reproduction

No changes in the reproduction compared to near-isogenic conventional maize have been observed in agronomic assessments conducted with Bt11 x MIR604 x GA21 maize.

b) Dissemination

No changes in the dissemination compared to near-isogenic conventional maize have been observed in agronomic assessments conducted with Bt11 x MIR604 x GA21 maize.

c) Survivability

No changes in the survivability compared to near-isogenic conventional maize have been observed in agronomic assessments conducted with Bt11 x MIR604 x GA21 maize.

d) Other differences

No changes in the reproduction, dissemination or survivability compared to near-isogenic conventional maize have been observed in agronomic assessments conducted with Bt11 x MIR604 x GA21 maize.

5. Genetic stability of the insert and phenotypic stability of the GM plant

Molecular analyses showed that the inserts have been stably integrated into the plant's genome in Bt11, MIR604 and GA21 maize.

Bt11 x MIR604 x GA21 maize F_1 seed is produced through conventional breeding involving Bt11, MIR604 and GA21 lines. Bt11 x MIR604 x GA21 seed once planted by growers produces grain (F_2) which is harvested for food, feed or industrial use. Such grain or products entering the commodity chain are not kept for further sowing.

6. Any change to the ability of the GM plant to transfer genetic material to other organisms

a) Plant to bacteria gene transfer

The horizontal gene transfer from GM plants to bacteria with subsequent expression of the transgene is regarded as a highly unlikely event under natural conditions, especially in the absence of selective pressure. No changes in the ability of the Bt11 x MIR604 x GA21 maize, Bt11, MIR604 or GA21 maize to transfer genetic material to other organism are expected compared to conventional maize since no sequences have been introduced to allow this to occur.

b) Plant to plant gene transfer

The genetic modification in the single maize events (Bt11, MIR604 and GA21) is not intended to change any of the typical crop characteristics of maize (except for the tolerance to insect and herbicide products). Observations from field trials have confirmed that the agronomic and phenotypic characteristics of Bt11 maize, MIR604 maize, GA21 maize and Bt11 x MIR604 x GA21 maize have not changed in comparison with near-isogenic controls, and therefore, there is no increase or decrease in the potential for plant-to-plant gene transfer compared to traditional maize. Gene transfer from Bt11 x MIR604 x GA21 maize to other sexually compatible plant species is not possible since maize has no wild relatives in the EU.

7. Information on any toxic, allergenic or other harmful effects on human or animal health arising from the GM food/feed

7.1 Comparative assessment

Choice of the comparator

Stacked maize plants containing Bt11, MIR604 and GA21 maize were compared with relevant control maize lines that had not been genetically modified. Commercial varieties were also included in the comparison where possible.

7.2 Production of material for comparative assessment

a) number of locations, growing seasons, geographical spreading and replicates

To evaluate whether biologically significant changes in composition occurred in Bt11 x MIR604 x GA21 maize plants compared to near-isogenic conventional maize replicate trials were planted at a range of locations in the USA and the EU. The locations of the trial sites were selected to be representative of the range of environmental conditions under which the hybrid varieties are expected to be grown.

b) the baseline used for consideration of natural variations

The levels of multiple nutritive components were compared in maize kernels (grain) or whole plants (forage) from Bt11 x MIR604 x GA21 maize and near-isogenic conventional maize plants grown concurrently. The mean values were also compared with the range of data published in the literature, where data was available.

7.3 Selection of materials and compounds for analysis

Based on guidance of the OECD, grain from transgenic plants and isogenic control plants were analysed for proximates (including starch), minerals, amino acids and selected fatty acids, vitamins, anti-nutrients and secondary metabolites. Forage (whole plants) from transgenic maize plants and isogenic control plants were analysed for proximates and minerals.

No consistent pattern has emerged to suggest that biologically relevant changes in composition or nutritive value of the grain or forage have occurred as an unintended result of the combination of the single events or expression of the transgenes in Bt11 x MIR604 x GA21 maize.

These data support the conclusion that Bt11 x MIR604 x GA21 maize is compositionally equivalent to conventional maize, apart from the introduced traits of insect and herbicide tolerance. It is expected that the double stack sub-combinations of these three single events will also result in maize products with equivalent composition to conventional maize.

7.4 Agronomic traits

Bt11 x MIR604 x GA21 maize plants were grown concurrently with near-isogenic conventional maize plants in a series of trials across the USA and the EU. Selected agronomic and phenotypic traits were assessed and compared. The results of these trials showed that Bt11 x MIR604 x GA21 maize is agronomically and phenotypically equivalent to conventional maize, apart from the introduced traits.

7.5 Product specification

Maize as a product has a history of safe use for human food and animal feed. No significant native toxins are reported to be associated with the genus *Zea*. The information presented in this application confirms that Bt11 x MIR604 x GA21 maize and products derived from it are not different from those of its conventional counterpart.

7.6 Effect of processing

Bt11 x MIR604 x GA21 maize will be produced and processed in the same way as any non-GM maize and there is no evidence to suggest that the expression of the proteins produced by this maize (Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS) will influence this processing in any way.

7.7 Anticipated intake/extent of use

There are no anticipated changes to the intake/extent of use of maize as a result of the introduction of Bt11 x MIR604 x GA21 maize to the conventional maize supply. It is anticipated that the introduction of Bt11 x MIR604 x GA21 maize will replace some of the maize in existing food and feed products. However, the genetic modification was not intended to change any of the compositional parameters in food and feed and confirmed by the results obtained from the extensive compositional assessment.

7.8 Toxicology

7.8.1 Safety assessment of newly expressed proteins

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding and therefore produces the proteins inherited from the single GM maize events. Potential adverse effects to human and animal health arising from Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS have previously been assessed and it was concluded that the potential toxic effects to humans and animals of these proteins could be considered negligible. A summary is provided below:

- The recipient organism, maize, has a history of safe use throughout the world.
- None of the gene sequences or their donors are known to be pathogenic to humans and no pathogenic sequences have been introduced.
- Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS have no significant amino acid homology to known mammalian protein toxins.
- Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS are unlikely to be allergenic
- Cry1Ab, PAT, mCry3A, PMI and mEPSPS are readily degraded in *in vitro* digestibility assays.
- Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS show no acute oral toxicity in mammalian studies.

7.8.2 Testing of new constituents other than proteins

Maize is a common source of food and feed and has a long history of safe use. Bt11 x MIR604 x GA21 maize has been modified to produce the Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS proteins. No other new constituents apart

from these proteins are expected to be produced in Bt11 x MIR604 x GA21 maize and compositional analyses have confirmed the compositional equivalence of Bt11 x MIR604 x GA21 to conventional maize. Therefore no testing of any other constituent is considered necessary.

7.8.3 Information on natural food and feed constituents

The presence and levels of natural food and feed constituents such as macro- and micronutrients, secondary plant metabolites as well as natural toxins and antinutritional factors have been analysed and compared with non-GM isolines and data from the literature.

These analyses showed that the levels of the components measured had not changed beyond the natural variation in maize. No consistent patterns emerged to suggest that biologically relevant changes in composition or nutritive value of the grain or forage had occurred as an unintended result of the expression of the transgenes.

7.8.4 Testing of the whole GM food/feed

In addition to the compositional analysis, the wholesomeness and safety of Bt11 x MIR604 x GA21 maize was confirmed in a poultry feeding study. The study showed that the transgenic corn had no deleterious effects on broiler chickens.

7.9 Allergenicity

7.9.1 Assessment of allergenicity of the newly expressed protein

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding and therefore expresses the proteins inherited from these three single GM maize events. The allergenic potential arising from Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS have previously been assessed and it was concluded that the allergenic potential to humans and animals of these proteins could be considered negligible. In summary:

- None of the transgenic proteins produced by Bt11 x MIR604 x GA21 (Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS) come from donors known to be a significant cause of food allergy.
- Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS have no biologically significant amino acid homology to known allergens
- Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS are readily degraded in *in vitro* digestibility assays.

From these data, it can be concluded that Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS produced by Bt11 x MIR604 x GA21 maize plants are highly unlikely to be allergenic.

7.9.2 Assessment of allergenicity of the whole GM plant or crop

Maize has been extensively cultivated and has a history of safe use for human food and animal feed. Maize is not considered to be a food crop which causes significant food allergy and the newly expressed proteins in Bt11 x MIR604 x GA21 are very unlikely to be allergenic.

7.10 Nutritional assessment of GM food/feed

7.10.1 Nutritional assessment of GM food

Bt11 x MIR604 x GA21 maize is not intended to change the nutritional status of individuals of populations or to result in products with enhanced functionality. Compositional analysis and whole food safety tests have demonstrated that no unexpected alterations in nutrients and other food components have occurred and that no nutritional imbalances were introduced.

7.10.2 Nutritional assessment of GM feed

Bt11 x MIR604 x GA21 is not intended to change the nutritional status of livestock animals. Compositional analysis and whole food and feed safety tests have demonstrated that no unexpected alterations in nutrients and other food or feed components have occurred and that no nutritional imbalances were introduced.

7.11 Post-market monitoring of GM food/feed

As described in sections 7.1 to 7.10 above, the presence of Bt11 x MIR604 x GA21 maize in food and feed will not result in any nutritional changes, therefore post-market monitoring is not considered necessary.

8. Mechanism of interaction between the GM plant and target organisms (if applicable)

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding. Therefore Bt11 x MIR604 x GA21 maize plants produce the five transgenic proteins inherited from each of the single GM maize events: Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS. The intended use of Bt11 x MIR604 x GA21 maize in the EU is resistance to maize pests such as *Ostrinia nubilalis* (European corn borer; ECB), *Sesamia nonagrioides* (Mediterranean corn borer; MCB) and *Diabrotica virgifera virgifera* (Western Cornrootworm; WCRW) and tolerance to herbicides containing glyphosate. The resistance to corn borers (ECB and MCB) is achieved through the expression of Cry1Ab an insecticidal protein well known for its specificity to insects from the order Lepidoptera. Resistance to WCRW is achieved through the expression of mCry3A, an insecticidal protein with specific activity to insects from the order Coleoptera. The other transgenic proteins produced by Bt11 x MIR604 x GA21 maize, PAT, MIR604 PMI and mEPSPS are not known to have any effects on organisms. Therefore the target organisms for Bt11 x MIR604 x GA21 maize are limited to certain species of Coleoptera and Lepidoptera.

Interactions of the target pests with Bt11 x MIR604 x GA21 maize have been described in the application and an insect resistance management (IRM) plan has been included.

9. Potential changes in the interactions of the GM plant with the biotic environment resulting from the genetic modification

9.1 Persistence and invasiveness

Potential changes in persistence and invasiveness in Bt11 x MIR604 x GA21 maize compared to conventional maize have been assessed. For the comparative assessment of phenotypic and agronomic characteristics of Bt11 x MIR604 x GA21 maize and its corresponding near-isogenic conventional maize, multiple field trials were grown in the USA and in the EU. One of the aims of these trials was to test the hypothesis of no greater persistence or invasiveness of Bt11 x MIR604 x GA21 maize compared with conventional maize. The endpoints measured in these trials were selected to study significant unintended changes in seed dispersal or other traits that might affect the ability of maize to survive without human intervention (such as seedling emergence, plant height, failure to produce an ear, dropping of ears before harvest, grain yield and disease incidence).

These investigations showed that, while some differences between Bt11 x MIR604 x GA21 maize and near-isogenic controls for some of the measured endpoints were statistically significantly different at some locations, there were no consistent trends in the data across locations or hybrids that would indicate that any of these differences were due to the presence of the transgenes. Therefore the results indicate that Bt11 x MIR604 x GA21 maize will not be more persistent or invasive than conventional maize.

In summary, the likelihood that Bt11 x MIR604 x GA21 maize will become more persistent than the recipient or parental plants in agricultural habitats or more invasive in natural habitats can be considered negligible.

9.2 Selective advantage or disadvantage

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding. No new genetic modification has therefore taken place in Bt11 x MIR604 x GA21 maize and, as intended, the plants produce the five proteins inherited from these three single GM events: Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS.

Expression of Cry1Ab and mCry3A, conferring resistance to certain species of Lepidoptera or Coleoptera, in areas of Europe where these are important maize pests, could be considered an advantage over conventional maize. However maize is a highly domesticated plant and cannot survive without human intervention, even in areas without pressure from these target pests. Therefore, expression of Cry1Ab and mCry3A will not increase the chances of maize survival under European conditions and would not confer any selective advantage.

Expression of PAT and mEPSPS could confer advantage to maize plants when herbicide products containing glufosinate ammonium or glyphosate are applied.

However this rarely happens outside agricultural environments. Therefore, expression of PAT and mEPSPS is highly unlikely to confer selective advantage to maize plants.

Expression of MIR604 PMI could only confer an advantage to maize plants growing under conditions where mannose was the only source of carbon, conditions that are highly unlikely in normal soils. Therefore, expression of MIR604 PMI cannot be considered a factor that would confer selective advantage to maize.

In summary, the likelihood that the expression of the Lepidoptera and Coleopteran pest protection traits, the herbicide tolerant traits or the selectable markers in Bt11 x MIR604 x GA21 maize will result in a selective advantage or disadvantage compared with conventional maize can be considered negligible.

9.3 Potential for gene transfer

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding. No new genetic modification has therefore taken place in Bt11 x MIR604 x GA21 maize and, as intended, the plants produce the five proteins inherited from these three single GM events: Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS.

Given the characteristics of these genes and the constructs used, the likelihood that genes from Bt11 x MIR604 x GA21 maize (or from any of the double stack sub-combinations) would become established in the genome of microorganisms in the environment or human and animal digestive tract is very low. In the very unlikely event that such a horizontal gene transfer would take place, no adverse effects on human and animal health or the environment are expected.

Gene transfer from Bt11 x MIR604 x GA21 maize to other sexually compatible plant species is not possible since there are no wild relatives of maize in the EU.

9.5 Interactions of the GM plant with non-target organisms

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding. No new genetic modification has therefore taken place in Bt11 x MIR604 x GA21 maize and, as intended, the plants produce the five proteins inherited from these three single GM events: Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS. Previous ERA conducted for Bt11, MIR604 and GA21 maize concluded that the cultivation of these individual maize events will pose low risk to non-target organisms. It is therefore unlikely that the combination of these individual maize events through conventional breeding techniques will result in a maize product with adverse effects in the environment. To test the hypothesis that the combination of the insecticidal proteins expressed in Bt11 x MIR604 x GA21 maize (Cry1Ab and mCry3A) will not result in interactions of safety concern, a study was conducted where sensitive target species were

exposed to each of the proteins alone or in combination. The results of this study demonstrated that the overall mortality levels in these species were similar when exposed to the proteins alone or in combination. Therefore, the results of this study demonstrate that there are no interactions of safety concern between mCry3A and CrylAb. This allows the conclusion that the combination of Bt11, MIR604 and GA21 through conventional breeding will not result in any stack combination with increased likelihood to cause adverse effects on non-target organisms compared with the individual single events, which have been previously assessed and shown to have low risk to non-target organisms.

9.6 Effects on human health

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding and therefore produces the proteins expressed in these single maize events. Potential adverse effects to humans and animals of the newly expressed proteins Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS were previously evaluated as part of the risk assessments conducted to support the Bt11, MIR604 and GA21 import applications. In addition, a safety assessment of the proteins Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS has been carried out as part of the risk assessment conducted for the authorization of import of Bt11 x MIR604 x GA21 maize and derived food and feed (application EFSA-GMO-UK-2008-56). The conclusions reached by the EFSA GMO Panel were that there are no safety concerns to mammals arising from the proteins Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS.

In summary, the risk to human or animal health as a result of exposure to Bt11 x MIR604 x GA21 maize is negligible.

9.7 Effects on animal health

Bt11 x MIR604 x GA21 maize was produced by combining Bt11, MIR604 and GA21 maize through conventional breeding and therefore produces the proteins expressed in these single maize events. Potential adverse effects to humans and animals of the newly expressed proteins Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS were previously evaluated as part of the risk assessments conducted to support the Bt11, MIR604 and GA21 import applications. In addition, a safety assessment of the proteins Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS has been carried out as part of the risk assessment conducted for the authorization of import of Bt11 x MIR604 x GA21 maize and derived food and feed (application EFSA-GMO-UK-2008-56). The conclusions reached by the EFSA GMO Panel were that there are no safety concerns to mammals arising from the proteins Cry1Ab, PAT, mCry3A, MIR604 PMI and mEPSPS.

In summary, the risk to human or animal health as a result of exposure to Bt11 x MIR604 x GA21 maize is negligible.

9.8 Effects on biogeochemical processes

The potential effects of Bt11 x MIR604 x GA21 maize to soil NTOs have been assessed and the conclusions from this assessment are that the cultivation of Bt11 x MIR604 x GA21 maize will not result in harmful effects on soil organisms either in Bt11 x MIR604 x GA21 maize fields or in surrounding environments. Given the specificity of Cry1Ab to Lepidoptera and of mCry3A to Coleoptera, the lack of adverse effects on soil organisms, the low levels of exposure in soil and the rapid degradation of these proteins in soil, the risk that Bt11 x MIR604 x GA21 cultivation will result in adverse effects on soil organisms or soil processes can be considered low.

9.9 Impacts of the specific cultivation, management and harvesting techniques

The likelihood that cultivation of Bt11 x MIR604 x GA21 maize in the EU results in management practices different to those currently used in conventional maize that could result in adverse environmental effects has been assessed. The intended effect of Bt11 x MIR604 x GA21 maize is the protection of maize from pests such as ECB, MCB and WCRW and tolerance to herbicides containing glyphosate. This includes an assessment of the potential environmental effects of the uses supported by Syngenta of herbicides containing glyphosate on Bt11 x MIR604 x GA21 maize.

The conclusion is that cultivation, management and harvesting techniques used for Bt11 x MIR604 x GA21 maize in the EU will not result in harmful environmental effects.

10. Potential interactions with the abiotic environment

The genetic modifications introduced in Bt11, MIR604 and GA21 maize are not intended to change any of the maize crop characteristics. The comparative safety assessments conducted for Bt11 x MIR604 x GA21 maize showed that the composition and phenotypic and agronomic characteristics of these maize products are comparable to those of conventional maize apart from the intended traits. The potential adverse effects of these traits on non-target organisms and biogeochemical processes have also been assessed and the conclusions are that the risks will be low. Therefore, given the specificity and mode of action of the newly expressed proteins, the lack of adverse effects on non-target organisms and soil organisms, the low levels of exposure in soil and the rapid degradation of these proteins in soil, the risk that interactions between Bt11 x MIR604 x GA21 maize with the abiotic environment result in adverse effects to the environment can be considered low.

11. Environmental monitoring plan (not if application concerns only food and feed)

The scope of this application includes production and cultivation of genetically modified Bt11 x MIR604 x GA21 maize in the European Union, including seed production and breeding of the GM maize lines necessary to generate Bt11 x MIR604 x GA21 maize.

Environmental exposure to Bt11 x MIR604 x GA21 maize could occur through cultivation and in the event that small amounts of grain of Bt11 x MIR604 x GA21 maize found their way into the environment in the EU. However, the survival of grain or seed outside of an agricultural environment would be very unlikely as maize is a highly domesticated plant and cannot survive without human intervention, especially under normal European climatic conditions. This grain, if germinated, could be easily controlled using any of the current agronomic measures taken to control other commercially available maize.

An environmental risk assessment (ERA) has been conducted taking into account the scope of this application. Comparison of Bt11 x MIR604 x GA21 maize with conventional maize has been used as a baseline. The conclusions of this ERA confirm that the effects to the environment arising from the use of Bt11 x MIR604 x GA21 maize can be considered as negligible as those from any other commercial maize.

11.1 General (risk assessment, background information)

As required by Article 5(5)(b) and 17(5)(b) of Regulation (EC) No. 1829/2003 the proposed monitoring plan for Bt11 x MIR604 x GA21 maize has been developed according to the principles and objectives outlined in Annex VII of Directive 2001/18/EC and Decision 2002/811/EC establishing guidance notes supplementing Annex VII to Directive 2001/18/EC. The structure of the monitoring plan also takes into account the guidance on presentation of applications provided in the Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of genetically modified plants and derived food and feed.

An ERA for each of the individual GM maize used to produce Bt11 x MIR604 x GA21 maize has previously been conducted as part of the application for the import and use under Regulation (EC) No 1829/2003. The conclusion of these risk assessments was that the adverse effects to the environment arising from the import and use of Bt11, MIR604 or GA21 maize could be considered as negligible as those from any other commercial maize. An environmental risk assessment (ERA) has been conducted also taking into account the scope of this application. Comparison of Bt11 x MIR604 x GA21 maize with conventional maize has been used as a baseline. The conclusions of this ERA confirm that the effects to the environment arising from the cultivation of Bt11 x MIR604 x GA21 maize will be no different to those from any other commercial maize.

11.2 Interplay between environmental risk assessment and monitoring

In general two types of environmental monitoring can be described:

a. case-specific monitoring, designed to evaluate potential adverse effects linked to the genetic modification, identified in the ERA

b. general surveillance, which is aimed to identify adverse unforeseen effects that were not anticipated in the environmental risk assessment.

An ERA has been conducted in accordance with Annex II of Directive 2001/18/EC and takes into account the recent Guidance Document of the Scientific Panel on Genetically Modified Organisms for the risk assessment of GM plants containing stacked transformation events to evaluate potential adverse effects of Bt11 x MIR604 x GA21 maize on human and animal health and the environment. The conclusions of this ERA confirmed that the potential risks to human and animal health or the environment arising from the placing on the market of Bt11 x MIR604 x GA21 maize can be considered negligible. Therefore, a case-specific monitoring plan is not considered necessary under the scope of this application.

Although Syngenta considers that the potential for development of resistance to target insects is not an environmental risk, Syngenta understands that this would represent a threat to the technology, therefore Syngenta has developed an IRM plan for Bt11 x MIR604 x GA21 maize to delay the development of resistance in target pests.

A general surveillance plan based on Annex II of the Directive 2001/18/EC is outlined below.

11.3 Case-specific GM plant monitoring (approach, strategy, method and analysis)

The main objective of case-specific monitoring is to determine the significance of any adverse effects identified in the ERA. The ERA conducted for Bt11 x MIR604 x GA21 maize, confirmed that the potential for adverse effects on human and animal health or the environment can be considered negligible.

The EFSA GMO Panel has identified the possible development of resistance of target organisms to Cry1Ab and mCry3A proteins as a potential risk due to large scale cultivation and/or long term exposure. Although Syngenta considers that the potential for development of resistance to target insects is not an environmental risk, Syngenta understands that this would represent a threat to the technology, therefore IRM plans to delay the development of resistance of the target pests to Cry1Ab and mCry3A has been developed.

11.4 General surveillance of the impact of the GM plant (approach, strategy, method and analysis)

The objective of general surveillance is to identify unforeseen adverse effects of the GM plant or its use, on human health and the environment, which were not predicted in the risk assessment. The scope of this application includes cultivation and seed production of Bt11 x MIR604 x GA21 maize.

It is intended that Bt11 x MIR604 x GA21 maize will be used as any other maize in the EU, hence background information relating to conventional practices in the cultivation have been considered in the development of the monitoring plan. In relation to the cultivation of Bt11 x MIR604 x GA21 maize, farmers are considered to be the most valuable source of information because they are regularly in contact with that part of the receiving environment occupied by the GM crop.

The majority of cultivated Bt11 x MIR604 x GA21 maize material will be used for feed purposes. Therefore, traders and processors as well as the European feed industry serve as a good focal point to address questions related to any unanticipated effects that might be associated with the use of Bt11 x MIR604 x GA21 maize.

Syngenta is committed to inform farmers, traders, processors and the European feed and food industry with details on the safety of Bt11 x MIR604 x GA21 maize and to establish a communication network where unforeseen effects can be reported. If unusual observations are reported, more focussed in-depth studies can be carried out in order to determine cause and relationship with the specific event. Final decisions on whether any identified effects are significant can only be made if causality is clear and endpoints are determined.

Although not a formal part of the surveillance plan, there is an extensive information network, with global reach, which will provide additional information on possible adverse effects arising from the use of GM crops. These include new and rapid means of access to information from across the globe through telecommunications, the media and Internet access. Through these means, many groups, including agronomists, ecologists, health professionals, and the general public now have unprecedented access to reports on the use world-wide of GM crops. In addition, electronic discussion sites, for example those of WHO, OECD, FAO, and consumer organisations, are valuable sources of information and communication for professionals and, in many cases, the general public.

11.5 Reporting the results of monitoring

The applicant/consent holder is responsible, under Regulation (EC) No 1829/2003, to inform the Commission of the results of the surveillance. Consistent with the EFSA guidance, the applicant will submit a General Surveillance Report containing information related to the monitoring on an annual basis.

12. Detection and event-specific identification techniques for the GM plant

The Bt11 x MIR604 x GA21 maize described in this application has been produced by combining the GM maize events: Bt11, MIR604 and GA21 through conventional breeding techniques. There was no further genetic modification to produce the stack. As such the detection methods developed for the single events should be appropriate for use on the stacked event.

Methods for detection of Bt11, MIR604 and GA21 maize have been developed by Syngenta. The proposed methods are real-time quantitative TaqMan® PCR based on specific detection of the genomic DNA of these events. There is no reason to suspect that the detection methods developed for the single events should not work on Bt11 x MIR604 x GA21 maize and all the sub-combinations of the individual GM maize events. However, in order to confirm the applicability of these methods, Syngenta has tested the methods on Bt11 x MIR604 x GA21 maize.

This information was submitted to the DG JRC-EURL for testing of the single event methods on the stacked product. The applicability of the single event detection methods on the stack has been validated by the DG-JRC-EURL and the verification report can be found on the DG-JRC EURL website
http://gmo-crl.jrc.ec.europa.eu/summaries/Bt11xMIR604xGA21_val_report.pdf

E. INFORMATION RELATING TO PREVIOUS RELEASES OF THE GM PLANT AND/OR DERIVED PRODUCTS

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

Syngenta has carried out field trials of Bt11 x MIR604 x GA21 maize in Spain, Czech Republic, Slovakia and Romania.
a) Notification number Spain: B/ES/08/34, B/ES/09/43 Czech Republic: B/CZ/09/01 Slovakia: B/SK/09/01 Romania: B/RO/08/04, B/RO/09/15
b) Conclusions of post-release monitoring No unexpected effects or observations have been detected to date. No adverse effects on human health or the environmental have been observed or reported during these releases The results of these field trials confirm the safety of the deliberate release of Bt11 x MIR604 x GA21 maize into the environment in the E.U.
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 unexpected effects or observations have been detected. No adverse effects on human health or the environmental have been observed or reported during these releases Final reports of the releases can be found at the JRC web page http://gmoinfo.jrc.it/

2. History of previous releases of the GM plant carried out outside the Community by the same notifier

a) Release country Bt11 x MIR604 x GA21 is approved for cultivation in the USA and Canada. Syngenta has conducted research field trials with this maize in the USA and Argentina.

b) Authority overseeing the release US EPA, USDA, Conabia
c) Release site Various release sites across the USA and Argentina.
d) Aim of the release Research and development
e) Duration of the release Varied depending on the aim of the trial
f) Aim of post-releases monitoring Control of volunteers
g) Duration of post-releases monitoring Varied depending on the aim of the trial, typically one year
h) Conclusions of post-release monitoring The occurrence of volunteers after planting Bt11 x MIR604 x GA21 field trials was no different to other maize
i) Results of the release in respect to any risk to human health and the environment No evidence of adverse effects to human health or the environment has been found.

3. Links (some of these links may be accessible only to the competent authorities of the Member States, to the Commission and to EFSA):

a) Status/process of approval The status and process of approval can be found on the EFSA website: http://www.efsa.europa.eu/EFSA/ScientificPanels/GMO/efsa_locale-1178620753812_GMOApplications.htm
b) Assessment Report of the Competent Authority (Directive 2001/18/EC) An application for approval of Bt11 x MIR604 x GA21 under the Directive 2001/18/EC has not been made by Syngenta.
c) EFSA opinion An EFSA opinion on Bt11 x MIR604 x GA21 maize was not available at the time of submission. EFSA opinions, once available can be found at http://www.efsa.europa.eu/EFSA/ScientificPanels/GMO/efsa_locale-1178620753812_GMOApplications.htm
d) Commission Register (Commission Decision 2004/204/EC) The Commission register of GM Food and Feed can be found at http://ec.europa.eu/food/food/biotechnology/authorisation/index_en.htm
e) Molecular Register of the Community Reference Laboratory/Joint Research Centre The Community Reference Laboratory webpage is http://gmo-crl.jrc.it/
f) Biosafety Clearing-House (Council Decision 2002/628/EC) Information relating to the Biosafety clearing house can be found at: http://bch.biodiv.org/
g) Summary Notification Information Format (SNIF) (Council Decision 2002/812/EC) An application for approval of Bt11 x MIR604 x GA21 maize under Directive 2001/18/EC has not been made by Syngenta, however a link to this Summary and the Summary supplied as part of the Application EFSA-GMO-UK-2008-56 under Regulation (EC) No 1829/2003 may be found at: http://www.efsa.europa.eu/EFSA/ScientificPanels/GMO/efsa_locale-1178620753812_GMOApplications.htm