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SCIENTIFIC OPINION

Scientific Opinion on application EFSA-GMO-UK-2007-41 for the placing on the market of herbicide-tolerant genetically modified cotton MON 88913 for food and feed uses, import and processing under Regulation (EC) No 1829/2003 from Monsanto[1]

EFSA Panel on Genetically Modified Organisms (GMO)[2,3]

European Food Safety Authority (EFSA), Parma, Italy

ABSTRACT

Cotton MON 88913 contains one insert consisting of the CP4 *epsps* expression cassette, providing herbicide tolerance. Bioinformatics-supported identification of hazards linked to the formation of new open reading frames caused by the insertion could not be completed due to the use of an outdated toxin database. Genetic stability studies did not raise safety issues. No biologically relevant differences which would raise safety concerns were identified in the composition or agronomic and phenotypic characteristics of plants and seeds obtained from cotton MON 88913 compared with its conventional counterpart and non-genetically modified reference varieties. Based on all the available information, there are no indications that the newly expressed CP4 EPSPS protein in cotton MON 88913 may be allergenic or toxic. Cotton MON 88913 was found to be as nutritious as commercially available varieties and it is unlikely that the overall allergenicity of the whole plant is changed. There are no indications of an increased likelihood of establishment and spread of feral cotton plants. Considering its intended uses as food and feed, interactions with the biotic and abiotic environment were not considered to be an issue. Risks associated with an unlikely but theoretically possible horizontal gene transfer from cotton MON 88913 to bacteria have not been identified. The monitoring plan and reporting intervals are in line with the intended uses of cotton MON 88913. The EFSA GMO Panel considers, however, that the information available for cotton MON 88913 is not sufficient to reach a final overall conclusion due to the use of an outdated toxin database for bioinformatic analyses.

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KEY WORDS

GMO, cotton MON 88913, Regulation (EC) No 1829/2003, CP4 EPSPS, food and feed safety, environment, import and processing

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[1] On request from the Competent Authority of the United Kingdom for application EFSA-GMO-UK-2007-41 submitted by Monsanto, Question No EFSA-Q-2007-085, adopted on 3 July 2013.

[2] Panel members: Salvatore Arpaia, Andrew Nicholas Edmund Birch, Andrew Chesson, Patrick du Jardin, Achim Gathmann, Jürgen Gropp, Lieve Herman, Hilde-Gunn Hoen-Sorteberg, Huw Jones, József Kiss, Gijs Kleter, Martinus Løvik, Antoine Messéan, Hanspeter Naegeli, Kaare Magne Nielsen, Jaroslava Ovesná, Joe Perry, Nils Rostoks and Christoph Tebbe. Correspondence: gmo@efsa.europa.eu

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SUMMARY

Following the submission of an application (Reference EFSA-GMO-UK-2007-41) under Regulation (EC) No 1829/2003[4] from Monsanto, the Panel on Genetically Modified Organisms of the European Food Safety Authority (EFSA GMO Panel) was asked to deliver a scientific opinion on the safety of the herbicide-tolerant genetically modified (GM) cotton MON 88913 (Unique Identifier MON-88913-8) for import and processing, and for food and feed uses. Cotton MON 88913 was developed to provide tolerance to glyphosate-based herbicides.

In delivering its scientific opinion, the EFSA GMO Panel considered the application EFSA-GMO-UK-2007-41, additional information provided by the applicant and the scientific comments submitted by the Member States. The scope of application EFSA-GMO-UK-2007-41 is for food and feed uses, import and processing of cotton MON 88913 and all derived products, but excludes cultivation in the European Union (EU).

The EFSA GMO Panel evaluated cotton MON 88913 with reference to the intended uses and appropriate principles described in the guidance documents of the Scientific Panel on Genetically Modified Organisms for the risk assessment of food and feed derived from genetically modified plants (EFSA, 2006a, 2011a). The scientific risk assessment evaluation included molecular characterisation of the inserted DNA and expression of target protein. A comparative analysis of agronomic traits and composition was undertaken, and the safety of the new proteins, as individual proteins and in combination, the changed levels of natural constituents and the whole food/feed were evaluated with respect to potential toxicity, allergenicity and nutritional quality. Evaluations of environmental impacts and the post-market environmental monitoring plan were undertaken.

Event MON 88913 was developed to produce a glyphosate-tolerant cotton by the introduction of the *epsps* coding sequence of *Agrobacterium* sp. strain CP4 encoding a 5-enolpyruvylshikimate-3-phosphate synthase protein (CP4 EPSPS). EPSPS is an enzyme active in the biosynthesis of aromatic amino acids necessary for plant growth. The CP4 EPSPS is, in contrast to the plant enzyme, less sensitive to glyphosate, rendering the genetically modified plant tolerant to glyphosate-based herbicides.

The molecular characterisation data establish that the GM cotton MON 88913 contains a single insert consisting of a single copy of the CP4 EPSPS expression cassette. No other parts of the plasmid used for transformation are present in cotton MON 88913. Hazard identification based on the bioinformatic analyses could not be completed because the version of the database used by the applicant for similarity searches of the open reading frames (ORFs) spanning the inserted DNA–genomic DNA junctions to known toxins was outdated (from 2001). Therefore, the EFSA GMO Panel cannot exclude that one of these ORFs might resemble a known toxin not included in this version of the database. Consequently, the EFSA GMO Panel cannot conclude on the safety of these ORFs based on updated information. Genetic stability studies did not raise a safety issue. The levels of the CP4 EPSPS protein in cotton MON 88913 have been sufficiently analysed.

Based on the information available, the EFSA GMO Panel concludes that no biologically relevant differences which would raise safety concerns were identified in the composition or agronomic and phenotypic characteristics of plants and seeds obtained from cotton MON 88913.

The EFSA GMO Panel considered that there are no indications that the CP4 EPSPS protein expressed in cotton MON 88913 may be allergenic or toxic. No biologically relevant differences were identified in the nutritional characteristics of cotton MON 88913 compared with its conventional counterparts as indicated by compositional data. The EFSA GMO Panel concludes that cotton MON 88913 is as

[4] Regulation (EC) No 1829/2003 of the European Parliament and of the Council of 22 September 2003 on genetically modified food and feed. OJ L 268, 1–23.

nutritious as non-GM reference varieties and that it is unlikely that the overall allergenicity of the whole plant is changed.

Considering its intended uses as food and feed, interactions with the biotic and abiotic environment were not considered to be an issue by the EFSA GMO Panel. In the case of accidental release into the environment of viable cotton MON 88913 seeds during transport and/or processing, there are no indications of an increased likelihood of spread and establishment of feral cotton plants. Risks associated with an unlikely but theoretically possible horizontal gene transfer from cotton MON 88913 to bacteria have not been identified.

The scope of the PMEM plan provided by the applicant is in line with the intended uses of cotton MON 88913 and the guidance document of the EFSA GMO Panel on PMEM of GM plants (EFSA, 2011b). In addition, the EFSA GMO Panel acknowledges the approach proposed by the applicant to put in place appropriate management systems to restrict environmental exposure in cases of accidental release of viable cotton MON 88913 seeds. The EFSA GMO Panel agrees with the reporting intervals proposed by the applicant in the PMEM plan.

In conclusion, the EFSA GMO Panel considers that the information available for cotton MON 88913 is not sufficient to reach a final overall conclusion due to partially outdated bioinformatic analyses.

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BACKGROUND

On 11 April 2007, EFSA received from the Competent Authority of the United Kingdom an application (Reference EFSA-GMO-UK-2007-41), for authorisation of herbicide-tolerant GM cotton MON 88913 (Unique Identifier MON-88913-8), submitted by Monsanto within the framework of Regulation (EC) No 1829/2003 on genetically modified food and feed for food and feed uses, import and processing.

After receiving the application EFSA-GMO-UK-2007-41 and in accordance with Articles 5(2)(b) and 17(2)b of Regulation (EC) No 1829/2003, EFSA informed the Member States as well as the European Commission and made the summary of the dossier publicly available on the EFSA website. EFSA initiated a formal review of the application to check compliance with the requirements laid down in Articles 5(3) and 17(3) of Regulation (EC) No 1829/2003. On 3 October 2007, the applicant provided EFSA with additional information requested under completeness check (requested on 10 September 2007). On 19 October 2007, EFSA declared the application as valid in accordance with Articles 6(1) and 18(1) of Regulation (EC) No 1829/2003.

EFSA made the valid application available to Member States and the European Commission and consulted nominated risk assessment bodies of the Member States, including the national Competent Authorities within the meaning of Regulation (EC) No 1829/2003 and Directive 2001/18/EC[5] following the requirements of Articles 6(4) and 18(4) of Regulation (EC) No 1829/2003, to request their scientific opinion. The Member State bodies had three months after the date of receipt of the valid application (until 19 January 2008) within which to make their opinion known.

The EFSA GMO Panel carried out a scientific assessment of genetically modified (GM) cotton MON 88913 in accordance with Articles 6(6) and 18(6) of Regulation (EC) No 1829/2003, taking into account the appropriate principles described in the guidance documents of the Scientific Panel on Genetically Modified Organisms for the risk assessment of food and feed derived from genetically modified plants (EFSA, 2006a, 2011a). In addition, the scientific comments of Member States, the additional information provided by the applicant and relevant scientific publications were taken into consideration.

On 20 December 2007, 27 February 2008, 24 June 2008 and on 2 February 2009, the GMO Panel asked for additional data on application EFSA-GMO-UK-2007-41. The applicant provided further information on 21 January 2008, 3 June 2008, 1 September 2008, 15 September 2010, 1 December 2010 and on 3 December 2012.

In giving its opinion on GM cotton MON 88913 to the European Commission, the Member States and the applicant, and in accordance with Articles 6(1) and 18(1) of Regulation (EC) No 1829/2003, EFSA has endeavoured to respect a time limit of six months from the receipt of the valid application. As additional information was requested by the EFSA GMO Panel, the time limit of 6 months was extended accordingly, in line with Articles 6(1), 6(2), 18(1), and 18(2) of Regulation (EC) No 1829/2003.

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

[5] Directive 2001/18/EC of the European Parliament and of the Council of 12 March 2001 on the deliberate release into the environment of genetically modified organisms and repealing Council Directive 90/220/EEC. OJ L106, 1–39.

TERMS OF REFERENCE

The GMO Panel was requested to carry out a scientific assessment of the GM cotton MON 88913 (Unique Identifier MON-88913-8) for food and feed uses, import and processing in accordance with Articles 6(6) and 18(6) of Regulation (EC) No 1829/2003. Where applicable, any conditions or restrictions which should be imposed on the placing on the market and/or specific conditions or restrictions for use and handling, including post-market monitoring requirements based on the outcome of the risk assessment and, in the case of GMOs or food/feed containing or consisting of GMOs, conditions for the protection of particular ecosystems/environments and/or geographical areas should be indicated in accordance with Articles 6(5)(e) and 18(5)e of Regulation (EC) No 1829/2003.

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

ASSESSMENT

1. Introduction

Genetically modified (GM) cotton MON 88913 (Unique Identifier MON-88913-8) was assessed with reference to its intended uses, taking account of the appropriate principles described in the guidance documents of the Scientific Panel on Genetically Modified Organisms for the risk assessment of food and feed derived from GM plants (EFSA, 2006a, 2011a).

The genetic modification in cotton MON 88913 results in the expression of the CP4 EPSPS protein. EPSPS is an enzyme active in the biosynthesis of aromatic amino acids necessary for plant growth. The CP4 EPSPS is, in contrast to the plant enzyme, less sensitive to glyphosate, rendering the GM plant tolerant to glyphosate-based herbicides. The genetic modification in cotton MON 88913 is intended to improve agronomic performance only and it is not intended to influence the nutritional properties, processing characteristics and overall use of cotton as a crop.

The scope of application EFSA-GMO-UK-2007-41 is for food and feed use, import and processing of cotton MON 88913 within the EU. Thus, cotton MON 88913 would be used for the production of cotton products as any commercial cotton variety. Likely uses of cotton MON 88913 include the production of refined oil from seeds, production of cellulose from linters as food or food ingredients and use of cotton seed meal and hulls in animal feed.

2. Issues raised by Member States

The scientific comments raised by Member States are addressed in Annex G of the EFSA overall opinion and have been considered throughout this EFSA GMO Panel scientific opinion.

3. Molecular characterisation

3.1. Evaluation of relevant scientific data

3.1.1. Transformation process and vector constructs

Cotton MON 88913 was produced by *Agrobacterium*-mediated transformation of cotton hypocotyl explants of *in vitro* cotton seedlings. Embryogenic cotton callus was selected for glyphosate tolerance, germinated and developed into plants.[6]

The T-DNA introduced in MON 88913 was derived from plasmid PV-GHGT35, containing two CP4 *epsps* expression cassettes in tandem orientation.[7] The first expression cassette is under the regulation of the chimeric *FMV/Tsf1* promoter containing the enhancer sequences from the 35S promoter of the *Figwort mosaic virus* and the promoter from the *Tsf1* gene of *Arabidopsis thaliana*, coding for the elongation factor EF-1 alpha, followed by its leader and intron sequences. The second expression cassette is under the regulation of the chimeric 35S/*act8* promoter containing the enhancer sequences from the *Cauliflower mosaic virus* (CaMV) 35S promoter and the promoter of the actin *act8* gene of *A. thaliana*, followed by its leader (including the first and a part of the second exon) plus the first intron. Both expression cassettes contain the chloroplast transit peptide sequence of the *A. thaliana* EPSPS, the coding sequence for the CP4 EPSPS protein and the transcription termination sequence from the ribulose-1,5-bisphophate carboxylase small subunit (*rbc*) *E9* gene derived from *Pisum sativum* (pea).

[6] Technical Dossier/Section C1.
[7] Technical Dossier/Section C2.

3.1.2. Transgene constructs in the genetically modified plant[8]

Southern analyses, polymerase chain reaction (PCR) analyses (amplifying six overlapping regions of DNA that span the entire length of the insert and flanking regions) as well as sequencing and inheritance studies establish integration of a single and intact copy of the T-DNA into the cotton genome. The absence of additional DNA sequences from the vector PV-GHGT35 in cotton MON 88913 has been confirmed by Southern analysis using probes that cover the entire sequence of the vector PV-GHGT35 backbone, including the appropriate controls.[9]

The nucleotide sequence of the insert in cotton event MON 88913 has been determined in its entirety. The DNA sequence of the insert contains 8 512 base pairs (bp) spanning the entire T-DNA of PV-GHGT35 from the right to the left border region.

Flanking sequences extending 1 231 bp at the 5′ end and 1 029 bp at the 3′ end of the insert were determined. Sequence analysis of the insertion locus indicates that, with exception of 18 bp of cotton genomic DNA which was deleted upon insertion of the T-DNA, the insertion locus remained intact. BLASTN analysis with the flanking regions indicated similarity with transcribed cotton sequences (with unknown function) in a region at least 141 bp downstream from the insertion site.[9] Nine potential new open reading frames (ORFs) were identified in the junction regions, three at the 5′ flank and six at the 3′ flank. Bioinformatic analysis by using an updated (2008) database indicated the absence of any ORF potentially coding for known allergenic proteins.[9] However, similarity searches for known toxic proteins were not performed by using an up-to-date database as requested by the GMO Panel.[10] The search included in the application used a database from 2001 which did not indicate a safety issue.

3.1.3. Information on the expression of the insert[11]

Levels of CP4 EPSPS protein were analysed by enzyme-linked immunosorbent assay (ELISA) in young leaf, overseason leaf at three subsequent growth stages (OSL1, OSL2, OSL3), root, pollen and seed samples collected from field trials carried out at four typical cotton-growing regions in the USA in 2002. In addition, CP4 EPSPS levels were also measured in overseason leaf at four subsequent growth stages (OSL1, OSL 2, OSL3, OSL4) and seed samples of cotton MON 88913 collected from field trials at four locations during one season in Australia in 2003–2004. Considering the scope of the application, CP4 EPSPS levels in seeds are considered the most relevant. The mean expression level in seeds was 340 µg/g dry weight (dw; range 72–580 µg/g dw) in the US samples and 310 µg/g dw (range 260–380 µg/g dw) in the Australian samples.

3.1.4. Inheritance and stability of inserted DNA

Genetic stability of the MON 88913 insert was studied by Southern analysis. The restriction enzyme–probe combinations used were sufficient to conclude that the single-copy insert together with its flanking regions was retained over five generations, indicating stability.[12]

Furthermore, the expected inheritance ratio was observed for the CP4 EPSPS expression over several generations, indicating the presence of a single genetic locus, showing Mendelian segregation.

3.2. Conclusion

The molecular characterisation data establish that cotton MON 88913 contains a single insertion locus consisting of a single copy of the CP4 EPSPS expression cassette. No other parts of the plasmid used for transformation are present in cotton MON 88913. Hazard identification based on the bioinformatic

[8] Technical Dossier/Section D2.
[9] Additional information, June 2008.
[10] Information requests of the GMO Panel, February 2008 and June 2008.
[11] Technical Dossier/Section D3.
[12] Technical Dossier/Section D5.

analyses could not be completed because the version of the database used by the applicant for similarity searches of the ORFs spanning the inserted DNA–genomic DNA junctions to known toxins was out of date (from 2001). Therefore, the EFSA GMO Panel cannot exclude that one of these ORFs might resemble a known toxin not included in this version of the database. Consequently, the EFSA GMO Panel cannot conclude on the safety of these ORFs based on updated information. The stability of the inserted DNA and the introduced trait (herbicide tolerance) was confirmed over several generations. The CP4 EPSPS protein levels were sufficiently quantified in field trials carried out in the USA and in Australia.

4. Comparative analysis

4.1. Evaluation of the relevant scientific data

4.1.1. Choice of comparator and production of material for the comparative analysis[13]

The application EFSA-GMO-UK-2007-41 for food and feed use, import and processing of cotton MON 88913 within the EU presented data on agronomic and phenotypic characteristics, and compositional data on seed of MON 88913 collected in field trials in USA at four locations in 2002 and at four locations in 2004. The field trials compared cotton MON 88913 with a negative segregant that had been isolated, after several stages of backcrossing and selfing of the progeny of the initial transformant, from cotton that was essentially homozygous for the insert in cotton MON 88913. The negative segregant was not considered an appropriate conventional counterpart by the EFSA GMO Panel. Therefore, the applicant was asked to provide a new comparative analysis of agronomic, phenotypic and compositional data using an appropriate conventional counterpart.

The applicant supplied new field trials performed at eight locations within the cotton cultivation areas in the USA in 2010 for studies of agronomic and phenotypic characteristics, and field trials at eight locations in 2011 on harvesting material for compositional analysis. The comparator used in 2010 was the non-GM cotton variety SG 125 and the comparator used in 2011 was the non-GM cotton variety 04X293. Both the conventional counterparts were inbred lines with a genetic background similar to cotton MON 88913. At each location the following materials were grown in a randomised complete block design with four replicates: cotton MON 88913 not treated with glyphosate-based herbicides, the conventional counterpart (SG 125 cotton or 04X293 cotton) not treated with glyphosate-based herbicides and four different non-GM cotton reference varieties not treated with glyphosate-based herbicides, and cotton MON 88913 treated with glyphosate on top of other pesticides. Overall, the field trials in 2010 included eight non-GM cotton reference varieties[14] in order to estimate the natural variation in agronomic and phenotypic characteristics, whereas the field trial in 2011 included 12 non-GM cotton reference varieties[15] in order to estimate the natural variation in compositional characteristics. The materials were characterised by event-specific PCR for the presence or absence of the MON 88913 event. Some samples collected from the eight field trials were contaminated by other GM cotton events, and therefore were excluded from the statistical analysis comparing cotton MON 88913 with its conventional counterpart. Three of the excluded samples were cotton MON 88913 not treated with glyphosate, four were samples of the conventional counterpart and 11 were reference cotton samples.

4.1.2. Compositional analysis[16]

The data from the 2011 field trials were analysed according to the applicable guidance document of the EFSA GMO Panel (EFSA, 2011a). It recommends a test of difference to verify whether the GM plant is different from its conventional counterpart or comparator, and a test of equivalence to verify

[13] Technical dossier/Section D7.2.
[14] The non-GM reference materials were DP565; DP5415; DP493; DP435; ST474; FM989; NM 1517-99; Delta Opal.
[15] The non-GM reference materials were DP565; DP5415; DP493; DP435; ST474; NM 1517-99; Delta Opal; Delta Pine 399; Atlas; All-Tex Xpress; FiberMax 958; SG 125 (in trials with 04X293 as comparator).
[16] Technical dossier/Section D7.3.

whether the characteristics of the GM plant fall within the range of natural variation estimated from the equivalence limits calculated from the reference varieties.

The selection of compositional characteristics of cotton seeds analysed was in accordance with OECD recommendations (OECD, 2009). Harvested delinted cotton seeds were analysed for proximates (ash, fat, moisture, protein and carbohydrate and calories by calculation), fibre fractions (acid detergent fibre (ADF), neutral detergent fibre (NDF), crude fibre (CF) and total dietary fibre (TDF)), amino acids, fatty acids, minerals (calcium, copper, iron, magnesium, manganese, phosphorus, potassium, sodium and zinc), vitamin E and antinutrients (gossypol and cyclopropenoid fatty acids). In total, 65 characteristics were analysed in the cotton seed. Thirteen characteristics[17] for which more than 50 % of the observations below the limit of quantification were excluded from the analysis.

The test of difference for samples not treated with glyphosate-based herbicides identified statistically significant differences between cotton MON 88913 and its conventional counterpart for 40 characteristics in seeds.[18] Cotton MON 88913 not treated with glyphosate-based herbicides had a significantly lower protein content and a lower content of many amino acids than its conventional counterpart. However, the differences were small, except for iron, and were not considered to be biologically meaningful since the biochemical function of the compounds is well known. Significant differences were found for the antinutritional compounds malvalic acid, sterculic acid and free gossypol. The levels of malvalic acid and sterculic acid were lower in cotton MON 88913 than in its conventional counterpart whereas the free gossypol levels were higher in cotton MON 88913 than in its conventional counterpart. The differences were small and were not considered to be biologically meaningful since the biochemical and antinutritional properties of the compounds are well known. The test of equivalence indicated non-equivalence for iron (equivalence category IV). The level of iron in seeds of cotton MON 88913 was 81.17 mg/kg dw (range 43.28–114.47 mg/kg dw), whereas in the conventional counterpart it was 63.22 mg/kg dw (range 44.89–84.95 mg/kg dw) and in the reference varieties it was 65.12 mg/kg dw (range 36.32–105.89 mg/kg dw). The EFSA GMO Panel evaluated the iron content and concluded that no further assessment was needed as the biochemical role of iron is well known, the magnitude of the reported difference lacks relevance from a food and feed safety and nutritional assessment point of view, and the iron level in seeds of cotton MON 88913 sprayed with glyphosate was equivalent (equivalence category I) to the level in the non-GM cotton reference varieties.

The test of difference for samples of MON 88913 sprayed with glyphosate in addition to other pesticides and its conventional counterpart not treated with glyphosate-based herbicides identified statistically significant differences for 43 characteristics in seeds.[19] Cotton MON 88913 sprayed with glyphosate had a significantly lower protein content and a lower content of many amino acids than its conventional counterpart. However, the differences were small and were not considered to be biologically meaningful since the biochemical role of the compounds is well known. Significant differences were found for the antinutritional compounds malvalic acid, sterculic acid and gossypol. The levels of malvalic acid and sterculic acid were lower in cotton MON 88913 than in its conventional counterpart whereas the free gossypol levels were higher in cotton MON 88913 than in the conventional counterpart. The differences were small and were not considered to be biologically meaningful since the biochemical and antinutritional properties of the compounds are well known. The

[17] Additional information, December 2012.

[18] Significant differences were identified for the proximates protein, fat, ash, NDF, TDF, CF, calories and carbohydrates by calculation; the amino acids arginine, aspartic acid, glutamic acid, glycine, histidine, isoleucine, leucine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine; the minerals calcium, magnesium, manganese, potassium and iron; the fatty acids palmitic acid, oleic acid, palmitoleic acid, stearic acid, linoleic acid, linolenic acid, behenic acid, myristic acid, malvalic acid, sterculic acid and arachidic acid; vitamin E and the antinutrient free gossypol.

[19] Significant differences were identified for the proximates protein, fat, ash, NDF, TDF, CF, calories and carbohydrates by calculation; the amino acids arginine, aspartic acid, glutamic acid, glycine, histidine, isoleucine, leucine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine and valine; the minerals calcium, magnesium, manganese, potassium and iron; the fatty acids palmitic acid, oleic acid, palmitoleic acid, stearic acid, linoleic acid, linolenic acid, behenic acid, myristic acid, malvalic acid, sterculic acid and arachidic acid; vitamin E and the antinutrient free gossypol.

test of equivalence indicated non-equivalence for potassium (equivalence category III). The level of potassium in cotton seeds from MON 88913 was 1.00 % dw (range 0.86–1.13 % dw), whereas for its conventional counterpart it was 1.04 % dw (range 0.93–1.15 % dw) and for the reference varieties it was 1.06 % dw (range 0.89–1.23 % dw). The EFSA GMO Panel concluded that no further assessment was needed as the biochemical role of potassium is well known and the magnitude of the differences were small and reported levels lack relevance from a food and feed safety and nutritional assessment point of view.

The GMO Panel considered the total set of compositional data supplied and the outcome of the statistical analysis comparing cotton MON 88913, its conventional counterpart and the set of non-GM cotton varieties, and concludes that no biologically relevant differences were identified between the composition of delinted seeds obtained from cotton MON 88913 and its conventional counterpart and other non-GM cotton reference varieties, except for the newly introduced trait.

4.1.3. Agronomic and phenotypic characteristics[20]

Based on data collected at eight field trial locations in the USA in 2010, the applicant performed a comparative assessment of the phenotypic and agronomic characteristics of cotton MON 88913 (in the cotton SG 125 background) and its conventional counterpart (cotton SG 125). There were 26 phenotypic and agronomic characteristics evaluated.[21] MON 88913 was also evaluated for environmental interaction, namely plant response to abiotic stress, disease and arthropod damage.

The test of difference of phenotypic and agronomic characteristics for cotton MON 88913 not treated with glyphosate-based herbicides identified small but statistically significant differences between cotton MON 88913 and its conventional counterpart for four endpoints (stand count at 14 days after planting (DAP), stand count at 30 DAP, final stand count at harvest and fibre micronaire). The same four characteristics and, in addition, seed cotton yield differed between cotton MON 88913 and its conventional counterpart when cotton MON 88913 was sprayed with glyphosate on top of other pesticides. However, the test for equivalence indicated equivalence for all the analysed characteristics except for mainstem nodes per plant and nodes to first fruiting branch (equivalent more likely than not) and fibre elongation (equivalent more likely than not for MON 88913 not sprayed with glyphosate and non-equivalent more likely than not for MON 88913 sprayed with glyphosate). For these characteristics, no difference was found between cotton MON 88913 (sprayed or non-sprayed with glyphosate) and its conventional counterpart.

Data on environmental interaction of cotton MON 88913 compared with the conventional counterpart were obtained in materials that was not treated with glyphosate. Comparable responses to abiotic stressors such as cold, compaction, drought, flood, frost, hail, nutrient deficiency and wind were observed. There were also no differences observed between cotton MON 88913 and the conventional counterpart for any of the diseases or arthropod damage.

4.2. Conclusion

Based on the information available, the EFSA GMO Panel concludes that no biologically relevant differences which would raise safety concerns were identified in the composition or agronomic and phenotypic characteristics of plants and seeds obtained from cotton MON 88913.

[20] Technical dossier/Section D7.4.

[21] Phenotypic and agronomic characteristics evaluated were stand count at 14 days after planting (DAP), stand count at 30 DAP, final stand count at harvest, plant height at 30 DAP, plant height at harvest, nodes above white flower (NAWF) at three different growth stages, seed cotton yield, number of mainstem nodes, number of nodes to first fruiting branch, number of bolls per plant, number of first-position bolls per plant, number of vegetative bolls per plant, percentage of first-position bolls retained by the plant, percentage of first-position bolls compared with total bolls, seed index, number of seeds per boll, number of mature seeds per plant, number of immature seeds per plant, weight per boll, fibre micronaire, fibre elongation, fibre strength, fibre length and fibre uniformity.

5. Food/feed safety assessment

5.1. Evaluation of relevant scientific data

5.1.1. Effects of processing[22]

Data on transgenic and conventional cottons other than MON 88913 were provided to support the claim that processed oil contains no detectable level of proteins[23] or DNA[24] and that cotton linters contain no detectable level of proteins (Sims et al., 1996).

Since no biologically relevant differences were identified in the composition or agronomic and phenotypic characteristics of plants and seeds obtained from cotton MON 88913, the effect of processing on cotton MON 88913 is not expected to be different from that on conventional cotton.

5.1.2. Toxicology[25]

5.1.2.1. Toxicological assessment of the newly expressed protein

The EFSA GMO Panel has previously evaluated the safety of the CP4 EPSPS protein in the context of several previous applications for the placing on the EU market of GM crops, and no concerns were identified (e.g. EFSA 2006b, 2008, 2010b, 2012).

In the current application (EFSA-GMO-UK-2007-41), an updated bioinformatic analysis[26] of the amino acid sequences of the CP4 EPSPS protein was provided. No significant similarities to known toxic proteins were found. The EFSA GMO Panel is of the opinion that no data have emerged which call for a revision of this conclusion.

Acute oral toxicity testing

The CP4 EPSPS protein produced in *Escherichia coli* was tested in mice in an acute oral toxicity study by oral gavage at dosages up to 572 mg/kg bw; no adverse effects were observed (Harrison et al., 1996). As demonstrated by N-terminal sequence, the amino acid sequence of the *E. coli* produced and the cottonseed-extracted CP4 EPSPS is identical.

The EFSA GMO Panel is of the opinion that acute toxicity testing of the newly expressed proteins is of little additional value for the risk assessment of the repeated consumption of food and feed from GM plants by humans and animals.

5.1.2.2. Toxicological assessment of new constituents other than proteins and/or changed levels of natural constituents

No new constituents other than CP4 EPSPS was deliberately introduced and expressed in cotton MON 88913. No relevant changes in the composition of cotton MON 88913 were detected in the comparative compositional analysis (see Section 4.1).

5.1.3. Animal studies with the food/feed derived from GM plants

(a) Sub-chronic toxicity study[27]

The applicant has provided a repeated-dose 90-day toxicity study in rats using cottonseeds from of cotton MON 88913 as a component of the diet. Groups of 20 male and 20 female Sprague–Dawley

[22] Technical dossier/Section D7.1.6.
[23] Study report MSL-16577.
[24] Study reports MSL-16090 and MSL-16554.
[25] Technical dossier/Section D7.2.
[26] Additional information, December 2012.
[27] Technical dossier /Section D.7.8.4 and study report WIL-50285.

Crl:CD(SD)IGS BR rats, individually housed, were *ad libitum* fed diets containing 2 % MON 88913 and 3 % of the negative segregant MON 88913(–) (test group, verified by PCR) or 5 % MON 88913 (test group) for a period of 90 consecutive days. The control group received diets containing 5 % of the negative segregant MON 88913(–). This study is not further considered because of the use of the negative segregant as the control.

(b) Catfish feeding study

A eight-week feeding study[28] was performed in channel catfish (*Ictalurus punctatus*) fed diets containing meal from GM cotton MON 88913 (test group, confirmed by PCR), a negative segregant (control group), and four non-GM cotton reference varieties (reference group: SG125, DP565, ST580, HS12) at a 20 % inclusion level. For each treatment, 100 catfish were divided over five aquariums with 20 fish each. Feed consumption was measured and observations of mortality and behaviour were made daily; weights were measured at the beginning, after four weeks and at the end of the experiment at eight weeks. After the experiment, eight fish per aquarium were used to prepare fillets, which were pooled for compositional analysis (moisture, crude protein, crude fat, ash), yielding five pooled fillet samples per treatment group.

Since all diets were designed to deliver the same nutrition, the expectation was that channel catfish in the three groups would show essentially the same performance characteristics. The results showed that feed consumption, weight gain, feed conversion ratio, visceral fat (percentage of body weight), fillet composition, survival and behaviour of fish fed the diet containing the toasted cottonseed meal MON 88913 did not significantly differ from those in fish fed the other diets. The EFSA GMO Panel concluded that this study did not allow the detection of unintended effects because of the absence of an isogenic control, but did show that cotton MON 88913 is as nutritious as other non-GM cotton reference varieties.

(c) Other studies

Whole cottonseed was included in diets for lactating dairy cows at about 10 %. Milk production was not affected by MON 88913 in comparison with a non-GM but genetically similar cotton (Castillo et al., 2004).

5.1.4. Allergenicity[29]

The strategies used when assessing the potential allergenic risk focus on the characterisation of the source of the recombinant protein, the potential of the newly expressed protein to induce sensitisation or to elicit allergic reactions in already sensitised persons and whether the transformation may have altered the allergenic properties of the modified plant.

5.1.4.1. Assessment of allergenicity of the newly expressed protein

A weight-of-evidence approach is followed, taking into account all of the information obtained with various test methods, since no single experimental method yields decisive evidence for allergenicity (EFSA, 2006a, 2011a; *Codex Alimentarius*, 2009).

The CP4 *epsps* gene originates from *Agrobacterium* sp. CP4, a soil microorganism that is not known to be allergenic.

In the current application (EFSA-GMO-UK-2007-41), an updated bioinformatic analysis[30] of the amino acid sequences of the CP4 EPSPS protein using the criterion of 35 % identity in a window of 80 amino acids revealed no significant similarities to known allergens. In addition, the applicant

[28] Technical dossier/Section D7.10 and study report NW-2003-050.
[29] Technical dossier/Section D7.9 and Additional information received in June 2008, August 2008, December 2010 and December 2012.
[30] Additional information, December 2010 and December 2012.

performed an analysis[30] searching for matches of eight contiguous identical amino acid sequences between the CP4 EPSPS protein and known allergens which confirmed the outcome of the previous bioinformatic analysis.

The studies on resistance to degradation of the CP4 EPSPS protein by proteolytic enzymes presented in the current application have been previously risk assessed by the EFSA GMO Panel (EFSA, 2012).

The EFSA GMO Panel has evaluated the safety of the CP4 EPSPS protein in the context of several previous applications and no concerns in relation to its allergenicity were identified (e.g. EFSA, 2006b, 2008, 2010b, 2012).

Based on all the available information, the EFSA GMO Panel considered that there are no indications that the newly expressed CP4 EPSPS protein in cotton MON 88913 may be allergenic.

5.1.4.2. Assessment of allergenicity of the whole GM plant

According to the guidance documents of the EFSA GMO Panel for the risk assessment of food and feed derived from GM plants (EFSA, 2006a, 2011a), when the plant receiving the introduced gene is known to be allergenic, the applicant should test any potential change in the allergenicity of the whole GM plant by comparing the allergen repertoire with that of its appropriate comparator(s).

Cotton has not been considered to be a common allergenic food[31] and only few cases of food allergy to cottonseed have been reported (Atkins, 1988; Malanin and Kalimo, 1988; O'Neil and Lehrer, 1989), all of which were to foods with cottonseed flour as the offending ingredient. Cottonseed protein appears to contain allergen(s) of relevant potency. However, the main cottonseed product in human food, industrially processed cottonseed oil, is highly purified and contains very low amounts of proteins. Also, in cellulose from cottonseed linters for use as food or food ingredients, the protein level is considered to be very low.

In the context of the present application and based on the available information, the EFSA GMO Panel considers that there is no evidence that the genetic modification might significantly change the overall allergenicity of cotton MON 88913.

5.1.5. Nutritional assessment of food/feed derived from GM plants

The intended trait of cotton MON 88913 is herbicide tolerance, with no intention to alter the nutritional parameters. The outcome of the composition analysis (see Section 4.1.2) confirmed the nutritional equivalence of the food and feed products derived from cotton MON 88913. The introduction of these products into the food and feed chain is, therefore, expected to have no nutritional impact compared with its conventional counterpart and non-GM cotton varieties.

The compositional data indicating nutritional equivalence between the GM cotton MON 88913 and non-GM reference cotton varieties (see Section 4.1.2) were confirmed by a feeding study with channel catfish (see Section 5.1.3) and dairy cattle (Castillo et al., 2004). In accordance with the EFSA guidance document (EFSA, 2006, 2011a), the EFSA GMO Panel concludes that cotton MON 88913 is as nutritious as other cotton varieties commercially available.

5.1.6. Post-market monitoring of GM food/feed

No biologically relevant compositional, agronomic and phenotypic changes were identified in cotton MON 88913 when compared with its conventional counterpart and non-GM reference cotton varieties. Furthermore, the overall intake or exposure is not expected to change because of the introduction of

[31] Directive 2007/68/EC of the European Parliament and of the Council of 27 November 2007 amending Annex IIIa to Directive 2000/13/EC of the European Parliament and of the Council as regards certain food ingredients. OJ, L310, 11–14.

cotton MON 88913 into the market. The EFSA GMO Panel therefore considers that post-market monitoring (EFSA, 2006a, 2011a) of the food/feed derived from cotton MON 88913 is not necessary.

5.2. Conclusion

The EFSA GMO Panel considered that there are no indications that the CP4 EPSPS protein expressed in cotton MON 88913 may be allergenic or toxic. No biologically relevant differences were identified in the nutritional characteristics of cotton MON 88913 compared with its conventional counterparts as indicated by compositional data. The EFSA GMO Panel concludes that cotton MON 88913 is as nutritious as non-GM reference varieties and that it is unlikely that the overall allergenicity of the whole plant is changed.

6. Environmental risk assessment and monitoring plan

6.1. Evaluation of relevant scientific data

The scope of application EFSA-GMO-UK-2007-41 is for food and feed uses, import and processing of cotton MON 88913 and does not include cultivation. Considering the intended uses of cotton MON 88913, the environmental risk assessment is concerned with indirect exposure, mainly through ingestion by animals, and their manure and faeces, leading to exposure of gastrointestinal tract and soil microorganisms, and with the accidental release into the environment of viable cotton MON 88913 seeds (e.g. during transport and/or processing).

Cotton MON 88913 has been developed to be tolerant to glyphosate-based herbicides by the expression of the EPSPS protein (see Section 1). As the scope of the present application excludes cultivation, environmental concerns in the EU related to the use of glyphosate-based herbicides on the GM cotton do not apply.

6.1.1. Environmental risk assessment

6.1.1.1. Unintended effects on plant fitness due to the genetic modification[32]

Gossypium herbaceum and *Gossypium hirsutum* are highly domesticated crops which have been grown in southern Europe since the 19th century, giving rise to feral plants which can occasionally be found in the same area (Todaro 1917; Davis, 1967). In the EU,[33] cotton is cultivated in Greece, Spain and on about 700 hectares in Bulgaria (USDA, 2009). The main cultivated cotton (*G. hirsutum*), which has been present in southern Europe since the 19th century, is an annual self-pollinating crop. In the absence of insect pollinators (such as wild bees, honeybees, bumblebees), cotton flowers are self-pollinating, but when these pollinators are present, low frequencies of cross-pollination can occur (McGregor, 1959; Moffett and Stith, 1972; Moffett et al., 1975; Van Deynze et al., 2005).

Pollen and cottonseed dispersal are potential sources of vertical gene flow to cross-compatible wild cotton relatives, to other cotton varieties and to occasional feral cotton plants. However, in Europe, there are no cross-compatible wild relatives with which cotton can hybridise. Because cotton pollen is very large (120–200 μm), heavy and sticky, wind-mediated dispersal of pollen to other cotton varieties is considered negligible (Vaissiere and Vinson, 1994). In addition, cross-pollination percentages rapidly decrease with increasing distance from the pollen source (Umbeck et al., 1991; Kareiva et al., 1994; Llewellyn and Fitt, 1996; Xanthopoulos and Kechagia, 2000; Van Deynze et al., 2005, 2011; Zhang et al., 2005; Hofs et al., 2007; Llewellyn et al., 2007; Heuberger et al., 2010).

Seeds are the only survival structures. However, seed-mediated establishment of cotton and its survival outside of cultivation in Europe is mainly limited by a combination of absence of a dormancy phase, low competitiveness and susceptibility to diseases and cold climate conditions (Eastick and

[32] Technical Dossier/Section D4 and Additional information, December 2012.
[33] http://epp.eurostat.ec.europa.eu/portal/page/portal/agriculture/data/database

Hearnden, 2006). In regions where cotton is widely grown, such as Australia, the risk of GM cotton being associated with ferality along transportation routes, or weediness on dairy farms where raw cottonseed is used as feed, has been shown to be negligible (Addison et al., 2007). Adequate soil moisture is an additional factor affecting the survival of feral cotton seedlings. Since the general characteristics of cotton MON 88913 are unchanged relative to its conventional counterpart, the inserted herbicide tolerance trait is not likely to provide a selective advantage outside cultivation in Europe.

In accordance with its guidance document on the environmental risk assessment of GM plants (EFSA, 2010a), the EFSA GMO Panel follows a weight of evidence approach in collating and assessing appropriate information from various data sources (e.g. molecular, compositional, agronomic and phenotypic data from field trials performed by the applicant, literature) in order to assess the likelihood of unintended effects on the environment. The applicant provided molecular, compositional and agronomic and phenotypic data that are assessed by the EFSA GMO Panel in Sections 3 and 4, respectively.

The applicant presented agronomic and phenotypic data gathered over a series of field trials conducted across 14 and 5 locations in the USA in 2002 and 2004, respectively. At the request[34] of the EFSA GMO Panel to clarify the choice of comparator as well as the herbicide treatments of field trials, the applicant provided additional agronomic and phenotypic data from eight locations in the USA in 2010. These field trials performed in 2010 fulfil the requirements specified in the current guidance document of the EFSA GMO Panel for the risk assessment of food and feed from GM plants (EFSA, 2011a). There were 26 phenotypic and agronomic characteristics evaluated (for further details, please see Section 4.1.3).

For cotton MON 88913 conventionally treated, statistically significant differences between the GM cotton and its comparator were observed for four characteristics (i.e. stand count at 14 DAP, stand count at 30 DAP, final stand count at harvest and fibre micronaire). For cotton MON 88913 treated with glyphosate-based herbicides, the same four characteristics and the seed cotton yield were significantly different from the non-GM comparator. Stand count was higher for cotton MON 88913, which might be an indication of a higher ability to survive. In the absence of information on germination, it is impossible to assess whether it is due to a potential unintended effect or to differences in seedlots.

However, average values from the test material fell within the ranges determined by the non-GM reference varieties and the test for equivalence indicated equivalence of GM cotton MON 88913 (treated and not treated with glyphosate-based herbicides) for the aforementioned analysed characteristics. Therefore, the EFSA GMO Panel is of the opinion that cotton MON 88913 does not differ from conventional cotton in terms of persistence or invasiveness and that observed differences do not raise any environmental safety concern.

Even though they did not reveal significant differences between cotton MON 88913 and its conventional counterpart, other endpoints did not fall with equivalence limits of non-GM reference varieties (i.e. mainstem nodes per plant, nodes to first fruiting branch, fibre elongation).

The mainstem nodes per plant and nodes to first fruiting branch were indicated to be equivalent more likely than not. In the absence of differences between cotton MON 88913 and its conventional counterpart, there is no altered persistence or invasiveness of cotton MON 88913.

Fibre elongation was indicated to be non-equivalent more likely than not.

Therefore, in accordance with the EFSA guidance document for the risk assessment of food and feed from GM plants (EFSA, 2011a), the EFSA GMO Panel considered the rationale provided by the

[34] Information request of the GMO Panel, February 2009.

applicant in case of likely non-equivalence of the GMO with non-GM reference varieties. The EFSA GMO Panel agreed with the applicant that a change in fibre elongation was unlikely to contribute meaningfully to altered persistence or invasiveness of GM cotton MON 88913.

Considering the intended uses of the GM cotton which excludes cultivation, the EFSA GMO Panel did not consider the increase in fibre elongation in cotton MON 88913 to be an environmental issue.

Other than the data presented by the applicant, the EFSA GMO Panel is not aware of any scientific report of increased fecundity, persistence (volunteerism) or ferality of GM cotton in regions where it is cultivated (Eastick and Hearnden, 2006; Bagavathiannan and Van Acker, 2008). Neither is there any information to indicate change in the survival capacity (including overwintering) of GM cotton. In case of accidental release of GM cotton seeds and establishment into the environment, cotton MON 88913 plants will have a selective advantage only in the presence of glyphosate-based herbicides.

Therefore, accounting for the intended uses of the GM cotton and the comprehensive dataset considered in the aforementioned weight of evidence approach, the EFSA GMO Panel concludes that it is unlikely that cotton MON 88913 has any enhanced fitness characteristics that will change its persistence and survival following accidental release into the environment of viable GM cotton seeds.

6.1.1.2. Potential for gene transfer[35]

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

(a) Plant-to-bacteria gene transfer

Genomic plant DNA is a component of several food and feed products derived from cotton. It is well documented that DNA present in food and feed becomes substantially degraded during processing and digestion in the human or animal gastrointestinal tract. However, a low level of exposure of fragments of ingested DNA, including the recombinant fraction of such DNA, to bacteria in the digestive tract of humans, domesticated animals and other environments exposed to the GM plant or plant material is expected.

Current scientific knowledge of recombination processes in bacteria indicates that horizontal transfer of non-mobile, chromosomally located DNA fragments between unrelated organisms (such as plants to bacteria) is not likely occur at detectable frequencies under natural conditions (for further details, see EFSA, 2009).

A successful horizontal transfer would require stable insertion of the transgene sequences into a bacterial genome and that a selective advantage is conferred on the transformed host. The only mechanism known to facilitate horizontal transfer of non-mobile, chromosomal DNA fragments to bacterial genomes is homologous recombination. This requires the presence of stretches of DNA sequences that are similar in the recombining DNA molecules and, in addition to substitutive gene replacement, facilitates the insertion of non-homologous DNA sequences if their flanking regions share sequence similarity with bacterial sequences in the recipient.

Cotton MON 88913 was developed through *Agrobacterium tumefaciens*-mediated transformation and contains the recombinant CP4 *epsps* gene originating from *Agrobacterium* sp. strain CP4. The absence of additional DNA sequences from the vector PV-GHGT35 in cotton MON 88913 plants has been confirmed by Southern analysis (see Section 3.1.2). The flanking regions of the recombinant gene insert do not contain sequences of the right or left border regions of the Ti-plasmid of *A. tumefaciens* of sufficient length to facilitate HGT.

[35] Technical Dossier/Section D6.

Although *A. tumefaciens* is not considered to be prevalent in the main receiving environments (i.e. the gastrointestinal tract of humans and animals), considering the intended uses of cotton MON 88913 as food and feed, bacteria of the genus *Agrobacterium* and *A. tumefaciens* are considered to occur in soil.

On a theoretical basis (i.e. in the absence of experimental evidence of horizontal gene transfer in GM food and feed derived from cotton MON 88913 or any other GM plant), it can be assumed that, as an extremely rare event, homologous recombination may occur in the environment between the recombinant *epsps* gene and its natural variants as they may occur in *Agrobacterium*. Such recombination events would only replace natural variants (i.e. substitutive recombination) and are therefore unlikely to provide any new property connected to a selective advantage for the recipient organisms (EFSA, 2009).

In cotton MON 88913, the *epsps* genes in the two cassettes are under the control of plant virus promoters. The activity of plant virus promoters in unrelated organisms such as bacteria cannot be excluded. However, in the unlikely event that the *epsps* genes and their regulatory elements are taken up by bacteria, no selective advantage is anticipated, because *epsps* genes already occur in various bacterial species in the environment.

The expected presence of genetically diverse natural variants of EPSPS protein-encoding genes as they occur in *Agrobacterium* sp. CP4, the use of regulatory sequences optimised for expression in eukaryotes, and the absence of any plausible selective advantage, suggest that it is highly unlikely that the *epsps* genes are transferred from cotton MON 88913 and establish in the genome of bacteria in the digestive tracts of humans or animals fed cotton MON 88913 (EFSA, 2009).

In addition to homology-based recombination processes, illegitimate recombination that does not require similarity between the recombining DNA molecules is theoretically possible. However, transformation rates for illegitimate recombination are considered to be 10^{10}-fold lower than for homologous recombination (Hülter and Wackernagel, 2008; EFSA, 2009). Illegitimate recombination events have not been detected in studies that have exposed bacteria to high concentrations of GM-plant DNA (EFSA, 2009). Thus, this process, compared with homologous recombination, is considered not to contribute significantly to horizontal gene transfer events.

Owing to the natural occurrence of *epsps* genes in soils or other environments, a low-level gene transfer to *Agrobacterium* species, including *tumefaciens*, is thought not to confer a new trait and selective advantage. Considering the intended uses of the GM cotton products and the above assessment, the EFSA GMO Panel has therefore not identified a concern associated with a potential horizontal gene transfer from cotton MON 88913 to bacteria.

(b) Plant-to-plant gene transfer

Considering the intended uses of cotton MON 88913 and the physical characteristics of cotton seeds, a possible pathway of dispersal is from cottonseed accidental spillage during transportation and/or processing and possible pollen dispersal from occasional feral GM cotton plants.

The genus *Gossypium* consists of at least four species: *Gossypium arboreum*, *Gossypium barbadense*, *Gossypium herbaceum* and *Gossypium hirsutum*. *Gossypium herbaceum* is reported (Zohary and Hopf, 2000) to be a traditional fibre crop in the eastern Mediterranean area already in the pre-Columbus period (before 1500 AD). In southern Europe, *G. herbaceum* and *G. hirsutum* have been grown since the 19th century, giving rise to occasional feral plants in the same area (Davis, 1967; Tutin et al., 1992) but no sexually compatible wild relatives of *G. hirsutum* have been reported in Europe. Therefore, the plant-to-plant gene transfer from this GM cotton is restricted to cultivated and occasional feral populations.

Survival of cotton outside cultivation in Europe is mainly limited by a combination of low competitiveness, absence of a dormancy phase, susceptibility to diseases and cold climate conditions.

Since these general characteristics of this GM cotton are unchanged, the inserted traits are not likely to provide a selective advantage outside cultivation in Europe (see Section 6.1.1.1).

The EFSA GMO Panel also takes into account the fact that this application does not include cultivation of the GM cotton within the EU so that the likelihood of cross-pollination between the imported GM cotton and cotton crops and occasional feral cotton plants is considered to be extremely low. Even if feral populations of cotton MON 88913 were established or transgene flow occurred to cultivated and feral cotton, a selective advantage would occur only if the complementary glyphosate-based herbicides were applied.

6.1.1.3. Interactions of the GM plant with target organisms[36]

Considering the intended uses of cotton MON 88913, which exclude cultivation, and the absence of target organisms, potential interactions of the GM plant with target organisms were not considered an issue by the EFSA GMO Panel.

6.1.1.4. Interactions of the GM plant with non-target organisms[37]

Owing to the intended uses of cotton MON 88913, which exclude cultivation, potential interactions of the GM cotton with non-target organisms were not considered an issue by the EFSA GMO Panel.

6.1.1.5. Interaction with the abiotic environment and biogeochemical cycles[38]

Owing to the intended uses of cotton MON 88913, which exclude cultivation, potential interactions with the abiotic environment and biogeochemical cycles were not considered an issue by the EFSA GMO Panel.

6.2. Post-market environmental monitoring[39]

The objectives of a post-market environmental monitoring (PMEM) plan according to Annex VII of Directive 2001/18/EC are (1) to confirm that any assumption regarding the occurrence and impact of potential adverse effects of the GMO, or its use, in the environmental risk assessment are correct and (2) to identify the occurrence of adverse effects of the GMO, or its use, on human health or the environment which were not anticipated in the environmental risk assessment.

Monitoring is related to risk management, and thus a final adoption of the PMEM plan falls outside the mandate of EFSA. However, the EFSA GMO Panel gives its opinion on the scientific content of the PMEM plan provided by the applicant (EFSA, 2011b). The potential exposure to the environment of cotton MON 88913 would be through ingestion by animals and their manure and faeces, leading to exposure of gastrointestinal tract and soil microbial populations to recombinant DNA, and through accidental release into the environment of GM cotton seeds during transport and/or processing. The scope of the PMEM plan provided by the applicant is in line with the intended uses. As the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects, no case-specific monitoring is necessary.

The PMEM plan proposed by the applicant includes (1) the description of an approach involving operators (federations involved in cotton import and processing) reporting to the applicant via a centralised system any observed adverse effect(s) of GMOs on human health and the environment; (2) a coordinating system established by EuropaBio for the collection of the information recorded by the various operators; and (3) the use of networks of existing surveillance systems (Lecoq et al., 2007; Windels et al., 2008). The applicant proposes to submit a PMEM report on an annual basis and a final report at the end of the consent.

[36] Technical Dossier/Sections D8 and D9.4.
[37] Technical Dossier/Section D9.5.
[38] Technical Dossier/Sections D9.8 and D10.
[39] Technical Dossier/Section D11 and Additional information, January 2008.

The EFSA GMO Panel is of the opinion that the scope of the PMEM plan provided by the applicant is in line with the intended uses of cotton MON 88913 as the environmental risk assessment did not cover cultivation and identified no potential adverse environmental effects. In addition, the EFSA GMO Panel acknowledges the approach proposed by the applicant to put in place appropriate management systems to restrict environmental exposure in the case of accidental release of viable seeds of cotton MON 88913. The EFSA GMO Panel agrees with the reporting intervals proposed by the applicant in the PMEM plan.

6.3. Conclusion

The scope of application EFSA-GMO-UK-2007-41 is for food and feed uses, import and processing and does not include cultivation. Considering the intended uses of cotton MON 88913, the environmental risk assessment is concerned with indirect exposure, mainly through ingestion by animals, and their manure and faeces leading to exposure of gastrointestinal tract and soil microorganisms, and with the accidental release into the environment of viable cotton MON 88913 seeds (e.g. during transport and/or processing). In the case of accidental release into the environment of viable cotton MON 88913 seeds during transport and/or processing, there are no indications of an increased likelihood of spread and establishment of feral cotton plants. Considering its intended uses as food and feed, interactions with the biotic and abiotic environment were not considered to be an issue. Risks associated with an unlikely but theoretically possible horizontal gene transfer from cotton MON 88913 to bacteria have not been identified. The scope of the PMEM plan provided by the applicant is in line with the intended uses of cotton MON 88913 and the guidance document of the EFSA GMO Panel on PMEM of GM plants (EFSA, 2011b). In addition, the EFSA GMO Panel acknowledges the approach proposed by the applicant to put in place appropriate management systems to restrict environmental exposure in cases of accidental release of viable cotton MON 88913 seeds. The EFSA GMO Panel agrees with the reporting intervals proposed by the applicant in the PMEM plan.

CONCLUSIONS AND RECOMMENDATIONS

The molecular characterisation data establish that the GM cotton MON 88913 contains a single insert consisting of a single copy of the CP4 EPSPS expression cassette. No other parts of the plasmid used for transformation are present in cotton MON 88913. Hazard identification based on the bioinformatic analyses could not be completed because the version of the database used by the applicant for similarity searches of the ORFs spanning the inserted DNA–genomic DNA junctions to known toxins was outdated (from 2001). Therefore, the EFSA GMO Panel cannot exclude that one of these ORFs might resemble a known toxin not included in this version of the database. Consequently, the EFSA GMO Panel cannot conclude on the safety of these ORFs based on updated information. Genetic stability studies did not raise a safety issue. The levels of the CP4 EPSPS protein in cotton MON 88913 have been sufficiently analysed.

Based on the information available, the EFSA GMO Panel concludes that no biologically relevant differences which would raise safety concerns were identified in the composition or agronomic and phenotypic characteristics of plants and seeds obtained from cotton MON 88913.

The EFSA GMO Panel considered that there are no indications that the CP4 EPSPS protein expressed in cotton MON 88913 may be allergenic or toxic. No biologically relevant differences were identified in the nutritional characteristics of cotton MON 88913 compared with its conventional counterparts as indicated by compositional data. The EFSA GMO Panel concludes that cotton MON 88913 is as nutritious as non-GM reference varieties and that it is unlikely that the overall allergenicity of the whole plant is changed.

Considering its intended uses as food and feed, interactions with the biotic and abiotic environment were not considered to be an issue by the EFSA GMO Panel. In the case of accidental release into the environment of viable cotton MON 88913 seeds during transport and/or processing, there are no

indications of an increased likelihood of spread and establishment of feral cotton plants. Risks associated with an unlikely but theoretically possible horizontal gene transfer from cotton MON 88913 to bacteria have not been identified.

The scope of the PMEM plan provided by the applicant is in line with the intended uses of cotton MON 88913 and the guidance document of the EFSA GMO Panel on PMEM of GM plants (EFSA, 2011b). In addition, the EFSA GMO Panel acknowledges the approach proposed by the applicant to put in place appropriate management systems to restrict environmental exposure in cases of accidental release of viable cotton MON 88913 seeds. The EFSA GMO Panel agrees with the reporting intervals proposed by the applicant in the PMEM plan.

The EFSA GMO Panel considers that the information available for cotton MON 88913 is not sufficient to reach a final overall conclusion due to partially outdated bioinformatic analyses.

DOCUMENTATION PROVIDED TO EFSA

1. Letter from the Competent Authority of the United Kingdom received on 11 April 2007 concerning a request for placing on the market of cotton MON 88913 for food and feed uses, import and processing in accordance with Regulation (EC) No 1829/2003.

2. Acknowledgement letter dated 17 April 2007 from EFSA to the Competent Authority of the United Kingdom.

3. Letter from EFSA to applicant dated 10 September 2007 requesting additional information under completeness check.

4. Letters from applicant to EFSA received on 3 October 2007 and on 8 October 2007 providing additional information under completeness check.

5. Letter from EFSA to applicant dated 19 October 2007 delivering the 'Statement of Validity' for application EFSA-GMO-UK-2007-41, cotton MON 88913 submitted by Monsanto Europe S.A. under Regulation (EC) No 1829/2003.

6. Letter from EFSA/DJ JRC to applicant dated 30 October 2007 requesting additional information and stopping the clock.

7. Letter from EFSA to applicant dated 20 December 2007 requesting additional information and maintaining the clock stopped.

8. Letter from applicant to EFSA received on 21 January 2008 providing additional information.

9. Letter from EFSA to applicant dated 30 January 2008 accepting the additional information requested by EFSA on 20 December 2007 (received on 21 January 2008) but maintaining the clock stopped for the DG/JRC.

10. Letter from EFSA to applicant dated 27 February 2008 requesting new additional information and maintaining the clock stopped for EFSA and for the DG/JRC.

11. Letter from applicant to EFSA received on 3 June 2008 providing additional information.

12. Letter from EFSA to applicant dated 24 June 2008 requesting clarifications on additional information and maintaining the clock stopped for EFSA and for the DG/JRC.

13. Letter from EFSA to applicant dated 7 August 2008 re-starting the clock for DG/JRC and maintaining the clock stopped for EFSA.

14. Letter from applicant to EFSA received on 1 September 2008 providing clarifications on additional information.

15. Letter from EFSA to applicant dated 21 October 2008 re-starting the clock.

16. Letter from EFSA to applicant dated 2 February 2009 requesting additional information and stopping the clock.

17. Letter from applicant to EFSA received on 15 September 2010 providing clarifications on the scope of the application.

18. Letter from applicant to EFSA received on 1 December 2010 providing additional information.

19. Letter from EFSA to applicant dated 11 July 2012 regarding the finalisation of the EFSA assessment.

20. Letter from applicant to EFSA received on 20 August 2012 replying to EFSA letter dated 11 July 2012 and providing a timeline for submission of responses.

21. Letter from EFSA to applicant dated 9 October 2012 regarding the finalisation of the EFSA assessment.

22. Letter from applicant to EFSA received on 3 December 2012 providing additional information.

23. Letter from EFSA to applicant dated 20 February 2013 providing further clarifications regarding the finalisation of the EFSA assessment.

24. Letter from EFSA to applicant dated 11 April 2013 re-starting the clock EFSA.

25. Letter from applicant to the Competent Authority of the United Kingdom dated 1 July 2013, requesting the extension of the scope of application EFSA-GMO-UK-2007-41 to cover (1) cotton MON 88913 for food and feed use; 2) food and feed containing or consisting of cotton MON 88913; and (3) food produced from or containing ingredients produced from cotton MON 88913 and feed produced from cotton MON 88913.

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