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SCIENTIFIC OPINION

Scientific Opinion on a request from the European Commission related to the emergency measure notified by France on genetically modified maize MON 810 according to Article 34 of Regulation (EC) No 1829/2003[1]

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

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ABSTRACT

Following a request of the European Commission, the European Food Safety Authority's Panel on Genetically Modified Organisms (EFSA GMO Panel) evaluated the documentation submitted by France in support of its request for the prohibition of the placing on the market of the genetically modified maize MON 810 according to Article 34 of Regulation (EC) No 1829/2003. The EFSA GMO Panel notes that some publications referred to by France were already part of the submission package by France for its safeguard clause and emergency measure on maize MON 810 in 2008. Those publications were addressed previously by the EFSA GMO Panel in its 2008 Scientific Opinion on the safeguard clause and emergency measure notified by France on maize MON 810. In the remaining documentation provided by France in support of the current emergency measure on maize MON 810, the EFSA GMO Panel could not identify any new science-based evidence indicating that maize MON 810 cultivation in the EU poses a significant and imminent risk to the human and animal health or the environment. With regard to issues related to management and monitoring of maize MON 810, the EFSA GMO Panel refers to its recent recommendations for management and monitoring measures of maize MON 810. In conclusion, the EFSA GMO Panel considers that, based on the documentation submitted by France, there is no specific scientific evidence, in terms of risk to human and animal health or the environment, that would support the notification of an emergency measure under Article 34 of Regulation (EC) No 1829/2003 and that would invalidate its previous risk assessments of maize MON 810.

KEY WORDS

GMO, maize (*Zea mays)*, MON 810, France, emergency measure, environment, Regulation (EC) No 1829/2003

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SUMMARY

On 20 February 2012, France provided to the European Commission a scientific argumentation in support of its request for the prohibition of the placing on the market of the genetically modified (GM) maize MON 810 according to Article 34 of Regulation (EC) No 1829/2003.

On 16 April 2012, the European Commission requested the European Food Safety Authority's Panel on Genetically Modified Organisms (EFSA GMO Panel) to assess the supporting documentation submitted by France.

The EFSA GMO Panel considered the relevance of concerns raised by France in the light of the most recent and relevant scientific data published in the scientific literature. During its evaluation of the supporting documentation, the EFSA GMO Panel has noted that some publications referred to by France were already part of the submission package by France for its safeguard clause and emergency measure on maize MON 810 in 2008. Those publications were addressed previously by the EFSA GMO Panel in its 2008 Scientific Opinion on the safeguard clause and emergency measure notified by France on maize MON 810, which concluded that no specific scientific evidence, in terms of risk to human and animal health or the environment, was provided that would justify the invocation of a safeguard clause under Article 23 of Directive 2001/18/EC and an emergency measure under Article 34 under Regulation (EC) No 1829/2003.

In the remaining documentation provided by France in support of the current emergency measure on maize MON 810, the EFSA GMO Panel could not identify any new science-based evidence indicating that maize MON 810 cultivation in the EU poses a significant and imminent risk to the human and animal health or the environment.

In relation to the management and monitoring of maize MON 810, the EFSA GMO Panel has previously assessed the Post-Market Environmental Monitoring (PMEM) plan and PMEM reports of maize MON 810 for the 2009 and 2010 growing seasons. Hence, the EFSA GMO Panel refers to its recent recommendations to improve the management and monitoring measures of maize MON 810 and other Bt-maize transformation events. The EFSA GMO Panel confirms from its evaluation of the PMEM results on maize MON 810 that no adverse effects on the environment, human and animal health due to maize MON 810 cultivation were identified during the 2009 and 2010 growing seasons.

In conclusion, the EFSA GMO Panel is of the opinion that, based on the documentation submitted by France, there is no specific scientific evidence, in terms of risk to human and animal health or the environment, that would support the notification of an emergency measure under Article 34 of Regulation (EC) No 1829/2003 and that would invalidate its previous risk assessments of maize MON 810.

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BACKGROUND AS PROVIDED BY THE EUROPEAN COMMISSION AND EFSA

The placing on the market for cultivation of the GM maize MON 810 in the European Union is authorized since 1998 through Commission Decision 98/294/EC of 22 April 1998 (EC, 1998) and the consent granted on 3 August 1998 by the Competent Authority of France.

On 9 February 2008, France notified to the European Commission a safeguard measure under Article 23 of Directive 2001/18/EC to provisionally prohibit the cultivation of the authorised maize MON 810 on its territory. On 13 February 2008, France also notified to the European Commission an emergency measure according to Article 34 of Regulation (EC) No 1829/2003. Consequently, on 27 February 2008, the Panel on Genetically Modified Organisms of the European Food Safety Authority (EFSA GMO Panel) was asked by the European Commission to assess the documentation provided by France in support of their measure requiring the prohibition of maize MON 810 cultivation. Having considered the overall information package submitted by France as well as a broad range of relevant scientific literature, the EFSA GMO Panel concluded, on 29 October 2008, that no specific scientific evidence had been provided by France that would justify the invocation of a safeguard clause under Article 23 of Directive 2001/18/EC and an emergency measure under Article 34 under Regulation (EC) No 1829/2003 (EFSA, 2008).

On 15 June 2009, following the request by the applicant for the renewal of the authorisation for placing maize MON 810 on the market, the EFSA GMO Panel adopted a Scientific Opinion[4] on the renewal under Regulation (EC) No 1829/2003 of maize MON 810 for import, processing for food & feed uses and cultivation (EFSA, 2009). The EFSA GMO Panel concluded that "*maize MON 810 is unlikely to have any adverse effect on the environment in the context of its intended uses, especially if appropriate management measures are put in place in order to mitigate possible exposure of non-target (NT) Lepidoptera"*. The EFSA GMO Panel recommended that, "*especially in areas of abundance of non-target Lepidoptera populations, the adoption of the cultivation of maize MON 810 be accompanied by management measures in order to mitigate the possible exposure of these species to maize MON 810 pollen*". Further, the EFSA GMO Panel advised that "*resistance management strategies continue to be employed and that the evolution of resistance in lepidopteran target pests continues to be monitored in order to detect potential changes in resistance levels in pest populations*".

On 30 November 2011, the EFSA GMO Panel adopted a Statement supplementing the evaluation of the environmental risk assessment (ERA) and risk management recommendations on the GM insect resistant maize Bt11 for cultivation (EFSA, 2011e). In its Statement on maize Bt11, the EFSA GMO Panel made recommendations for management measures and concluded that, "*subject to appropriate management measures, maize Bt11 cultivation is unlikely to raise additional safety concerns for the environment compared to conventional maize*". In light of the similarities between both GM Cry1Ab-expressing maize Bt11 and MON 810 (e.g., identity of amino acid sequence in core protein, similar biological activity against sensitive Lepidoptera, similar Cry1Ab protein expression level in pollen), the EFSA GMO Panel considered that the conclusions on the risk to non-target Lepidoptera from maize Bt11 apply equally to maize MON 810.

Furthermore, the EFSA GMO Panel was requested by the European Commission to assess the post-market environmental monitoring (PMEM) reports submitted by the applicant on the cultivation of maize MON 810 in 2009 and 2010. The EFSA GMO Panel therefore adopted a Scientific Opinion on the 2009 and 2010 PMEM reports on maize MON 810, on 7 September 2011 (EFSA, 2011c) and 7 March 2012 (EFSA, 2012) respectively. The EFSA GMO Panel noted shortcomings in the methodology for case-specific monitoring (CSM) and general surveillance (GS) and hence made recommendations for improvement of the PMEM of maize MON 810. However, these shortcomings identified in the methodology did not have any implications for conclusions on safety derived from the data submitted by the applicant in its PMEM reports. Hence, the EFSA GMO Panel did not identify

[4] This Scientific Opinion was published on the EFSA webpage on 30 June 2009.

adverse effects on the environment, human and animal health due to maize MON 810 cultivation during the 2009 and 2010 growing seasons.

On 20 February 2012, France notified to the European Commission its scientific argumentation in support of the prohibition of maize MON 810 cultivation in the EU, according to Article 34 of Regulation (EC) 1829/2003. The European Commission asked the EFSA GMO Panel to assess if new scientific evidence, that would indicate an environmental concern, was provided by France to support an emergency measure on maize MON 810. France endorsed its emergency measure on maize MON 810 through its decree[5] of 16 March 2012 suspending the cultivation of maize MON 810 varieties.

TERMS OF REFERENCE AS PROVIDED BY THE EUROPEAN COMMISSION

The EFSA GMO Panel is requested to provide a Scientific Opinion

(1) assessing if France submitted new scientific evidence in support of its prohibition of GM maize MON 810 cultivation according to Article 34 of Regulation (EC) 1829/2003; and, where appropriate,

(2) indicating whether this new scientific evidence might lead the EFSA GMO Panel to reconsider its previous safety assessments of GM maize MON 810.

In addition, the European Commission asked the EFSA GMO Panel to assess the 2012 study by Hilbeck *et al*. on the impact of Bt toxins on ladybird beetle (see Hilbeck *et al*., 2012a,b).

LEGAL CONTEXT OF THIS SCIENTIFIC OPINION

Article 34 of Regulation (EC) No 1829/2003, entitled 'Emergency measures', provides that "*where it is evident that products authorised by or in accordance with this Regulation are likely to constitute a serious risk to human health, animal health or the environment, …, measures shall be taken under the procedures provided for in Articles 53 and 54 of Regulation (EC) No 178/2002*".

Article 53 of Regulation (EC) No 178/2002 provides that "*where it is evident that food or feed originating in the Community or imported from a third country is likely to constitute a serious risk to human health, animal health or the environment, and that such risk cannot be contained satisfactorily by means of measures taken by the Member State(s) concerned, the Commission, acting … on its own initiative or at the request of a Member State, shall immediately adopt one or more of the following measures, depending on the gravity of the situation (…)*".

On 8 September 2011, the EU Court of Justice ruled that "*with a view to the adoption of emergency measures, Article 34 of Regulation No 1829/2003 requires Member States to establish, in addition to urgency, the existence of a situation which is likely to constitute a clear and serious risk to human health, animal health or the environment*". Furthermore, such measures can be envisaged only if they are supported by a comprehensive risk assessment indicating that such emergency measures are justified.

[5] http://legifrance.gouv.fr/affichTexte.do?cidTexte=JORFTEXT000025525099&categorieLien=id

ASSESSMENT

1. INTRODUCTION

The EFSA GMO Panel scrutinized the documentation[6] provided by France in support of its emergency measure on maize MON 810. According to the terms of reference set by the European Commission, the EFSA GMO Panel assessed whether the submitted documentation comprises new scientific information that would invalidate the conclusions of its previous risk assessments of maize MON 810.

The EFSA GMO Panel looked for evidence for GMO-specific risks taking into consideration the EFSA Guidance Document for the ERA of GM plants (EFSA, 2010), as well as any related risk assessments on other Cry1Ab-expressing maize transformation events than maize MON 810 carried out previously (EFSA, 2009; 2011c,e; 2012). The EFSA GMO Panel considered the relevance of concerns raised by France in the light of the most recent and relevant scientific data published in the scientific literature. The EFSA GMO Panel also considered concerns expressed by some Member States supporting the current emergency measure notified by France on maize MON 810.

The EFSA GMO Panel considered the following concerns expressed by France on maize MON 810:

Concerns related to the ERA of maize MON 810:

- Fate, including dissemination, of the Cry1Ab protein in soil and water,
- Impacts on target pests & possible resistance evolution,
- Impacts on terrestrial and aquatic non-target organisms,
- Outbreaks of secondary pests.

Concerns related to management measures, including PMEM, for maize MON 810:

- Management strategy to delay possible resistance evolution in target pests,
- Management measures to limit exposure of non-target Lepidoptera,
- CSM and GS of maize MON 810.

The structure of the present Scientific Opinion follows the order of the above listed concerns. This Scientific Opinion is based on existing scientific outputs by the EFSA GMO Panel on maize MON 810 and related insect-resistant GM maize transformation events (e.g., maize Bt11). In these, the EFSA GMO Panel reviewed and assessed almost all of the publications referred to by France (EFSA, 2008; 2009; 2011a,b,c,d,e; 2012).

In relation to issues related to management and monitoring of GM plants, the EFSA GMO Panel assesses the scientific quality of the initial PMEM plans and subsequent PMEM reports submitted by applicants, whilst the final endorsement of both the PMEM plan and reports is the responsibility of risk managers.

[6] The documentation submitted by France is made publicly available on the webpage of the French Ministry of Agriculture, at http://agriculture.gouv.fr/consultation-mesure-d-urgence-MON810

2. CONCERNS RELATED TO THE ERA OF MAIZE MON 810

2.1. Fate of the Cry1Ab protein in soil and water

2.1.1. Fate of the Cry1Ab protein in soil

During and after growth of maize MON 810, Cry1Ab protein can enter soils e.g., by deposition of pollen, release from roots and, quantitatively most important, decaying plant material (EFSA, 2008, 2009, 2011b,d,e).

The EFSA GMO Panel notes that most of the publications (e.g., Donegan *et al*., 1995; Saxena and Stotzky, 2001; Sims and Holden, 1996; Tapp and Stotzky, 1998) referred to by France in its notification of an emergency measure to prohibit maize MON 810 cultivation were addressed previously in its 2008 Scientific Opinion on the French safeguard clause and emergency measure on the same GM maize transformation event (for further details, see EFSA, 2008). More publications referred to by France are dealt with here below.

Proteins can be a major source of energy, carbon and nitrogen for soil microorganisms. They are readily degradable by widely abundant extracellular microbial proteases (Jan *et al*., 2009) and there is no indication that Cry proteins would generally behave differently compared with other proteins (reviewed by Icoz and Stotzky, 2008). The fate and hence the persistence of Cry proteins in soil depends upon multiple factors, varying among environmental conditions (e.g., soil characteristics, microbial activity, temperature) (reviewed by Icoz and Stotzky, 2008). Even though Cry proteins are degraded or inactivated in soil within weeks (e.g., Gruber *et al*., 2011a,b), a residual fraction may persist longer under certain environmental conditions (EFSA, 2009, 2011b,d).

Laboratory studies have shown that, due to their chemical properties (e.g., surface charges), Cry proteins can be bound by sorption onto organo-mineral surfaces, i.e., those provided by clay particles or humic complexes, thereby reducing their accessibility for soil proteases (e.g., Tapp *et al*., 1994; Tapp and Stotzky, 1995, 1998; Crecchio and Stotzky, 2001; Pagel-Wieder *et al*., 2007; Madliger *et al*., 2011). Due to their relatively strong sorption to soil components, Cry1Ab, the most extensively studied Cry1 protein from GM crops in the literature, was found to be degraded more slowly in soil (under similar conditions) than e.g. Cry3Bb1 (see Baumgarte and Tebbe, 2005; Madliger *et al*., 2010, 2011; Miethling-Graff *et al*., 2010; Sander *et al*., 2010 referred to in EFSA, 2011d). In context of an ERA, the main question is whether the sorption of Cry1 protein would result in its accumulation in soil up to concentrations that would have an adverse effect on certain non-target soil organisms due to the repeated and large-scale cultivation of maize MON 810. The repeated cultivation of maize MON 810 has never indicated accumulation of Cry1Ab protein under field conditions (Hopkins and Gregorich, 2003, 2005; Baumgarte and Tebbe, 2005; Dubelman *et al*., 2005; Andersen *et al*., 2007; Hönemann *et al*., 2008; Icoz *et al*., 2008; Gruber *et al*., 2011a,b) suggesting that despite sorption, degradation rates were sufficiently high to avoid accumulation (see also EFSA, 2009).

To conclude, no new scientific evidence on the fate of Cry1Ab directly relating to maize MON 810 cultivation was reported in the documentation provided by France in support of the notified emergency measure. The EFSA GMO Panel reiterates that the persistence of Cry proteins can be variable, depending upon soil type and environmental conditions, and confirms its previous conclusions that this does not raise any safety concern.

2.1.2. Fate of the Cry1Ab protein in water

In agricultural landscapes where Bt-maize is cultivated, Cry1Ab protein may disseminate from its sites of cultivation into water bodies (Tank *et al*., 2010). In water bodies located up to 500 m from maize fields in Indiana (USA), detectable levels (0.56 ng/mL) of the Cry1Ab protein could be shown in surveyed water streams up to six months after harvest. Cry1Ab protein concentrations in water bodies were small compared with the amount known to cause adverse effects on sensitive target organisms and aquatic NTOs (Jensen *et al*., 2010).

Studies have demonstrated degradation of Cry1Ab protein from decaying plant material in aquatic environments (Wolt and Peterson, 2010; Carstens *et al*., 2011; EFSA, 2011b). It has also been shown that the maize tissue expressing Cry1Ab have comparable degradation rates to non-Bt-maize (Griffiths *et al*., 2009; Swan *et al*., 2009) (for further details, see EFSA, 2011d).

In their lower-tier study with the European corn borer, Jensen *et al*. (2010) confirmed no bioactivity of the Cry1Ab protein in senesced maize tissue exposed to aquatic environments for two weeks, supporting the proposal of Griffiths *et al*. (2009) of rapid degradation of the protein.

In the previous 2008 Scientific Opinion of the EFSA GMO Panel on the French safeguard clause and emergency measure on maize MON 810, the EFSA GMO Panel also addressed French concerns related to the presence of the *cry1Ab* gene and Bt-proteins in water (for further details, see EFSA, 2008).

The EFSA GMO Panel is of the opinion that no new scientific evidence on the fate of the Cry1Ab protein in water directly relating to maize MON 810 cultivation was reported in the documentation provided by France in support of the notified emergency measure.

2.2. Impacts on target pests & possible resistance evolution

The EFSA GMO Panel notes that some of the publications (i.e., Bourguet *et al*., 2003; Huang *et al*., 2007; Stodola *et al*., 2006; Van Rensburg, 2007) referred to by France in its notification of an emergency measure on maize MON 810, were previously addressed in its 2008 Scientific Opinion on the safeguard clause and emergency measure notified by France on the same GM maize transformation event (for further details, see EFSA, 2008). More publications referred to by France are dealt with here below.

The possible resistance evolution to the Cry1Ab protein in lepidopteran target pests continues to be a concern associated with the cultivation of maize MON 810, as resistance evolution may lead to altered pest control practices that may cause adverse environmental effects (EFSA, 2009; 2011b,d,e). In addition to target pests, other regionally important lepidopteran pests (e.g., *Sesamia cretica*, *Helicoverpa armigera*, *Mythimna unipuncta*) exposed to Lepidoptera-resistant maize events may also have the potential to evolve resistance to Cry1 proteins (EFSA, 2011b,c,e; 2012).

In its recent Scientific Opinion updating the ERA of maize 1507 (EFSA, 2011b) and Statement on maize Bt11 (EFSA, 2011e), the EFSA GMO Panel acknowledges instances of field resistance to Bt-maize outside Europe for two lepidopteran target pests in maize that are not present in the European fauna (Tabashnik *et al*., 2009; Huang *et al*., 2011): *Busseola fusca* in South Africa (Van Rensburg, 2007; Kruger *et al*., 2009, 2011b) and *Spodoptera frugiperda* in Puerto Rico, USA (Matten *et al*., 2008; Moar *et al*., 2008; Tabashnik, 2008; Tabashnik *et al*., 2008; Storer *et al*., 2010). The recent survey by Kruger *et al.* (2011a) revealed that, in South Africa, compliance with *refugia* requirements in the region was low especially during the initial 5-7 years after release and a large number of farmers applied conventional insecticides as preventative sprays on Bt-maize and *refugia* irrespective of stem borer infestation levels. Moreover, no Insect Resistance Management (IRM) measures were put in place at that time in Puerto Rico.

Consequently, the EFSA GMO Panel reiterates the need for farmers to comply with the non-Bt *refugia* implementation and refers to its Scientific Opinions on the annual PMEM reports on the cultivation of maize MON 810 in 2009 and 2010 (for further details, see EFSA, 2011c, 2012). In these Scientific Opinions, the EFSA GMO Panel reiterates its earlier recommendation that appropriate IRM strategies relying on the 'high dose/refuge' strategy continue to be employed, in order to delay the potential evolution of resistance to the Cry1Ab protein in lepidopteran target pests. Furthermore, in areas where other lepidopteran pests than the European and Mediterranean corn borer occur, they might also be subject to resistance evolution due to exposure to the Cry1Ab protein expressed in maize MON 810. Therefore, the EFSA GMO Panel recommended that these species are also considered by the applicant

in the context of IRM and CSM to monitor resistance evolution to the Cry1Ab protein in these species, as well as in GS through farmer questionnaires.

The EFSA GMO Panel found no evidence of resistance evolution in the European and Mediterranean corn borer in the PMEM reports of maize MON 810 for the 2009 and 2010 growing seasons, (for further details, see EFSA, 2011c, 2012).

The EFSA GMO Panel is of the opinion that no new scientific evidence directly relating to maize MON 810 cultivation and to possible resistance evolution by target pests was reported in the documentation provided by France in support of the notified emergency measure.

2.3. Impacts on non-target organisms (NTOs)

2.3.1. Background

In its 2008 Scientific Opinion on the French safeguard clause and emergency measure on maize MON 810, the EFSA GMO Panel addressed the meta-analysis by Duan *et al*. (2008) referred to by France, which assessed direct effects on honeybee survival of Cry proteins from currently commercialised Bt-crops. The EFSA GMO Panel concluded that the low exposure level to Cry1Ab containing pollen combined with the low toxicity of the Cry1Ab protein is unlikely to result in any adverse effects on honeybees under normal apicultural conditions (EFSA, 2008).

In 2009, the possible adverse effects of maize MON 810 cultivation on NTOs were addressed in depth by the EFSA GMO Panel in its Scientific Opinion for the renewal of the placing on the market of maize MON 810 (EFSA, 2009). At that time, the EFSA GMO Panel considered a broad range of lower- and higher-tier data on NTOs (e.g., predators, parasitoids, pollinators, soil and aquatic non-target (NT) organisms) representative of relevant functional groups (for further details, see Section 6.1.4 of EFSA, 2009). The EFSA GMO Panel concluded that there was no evidence to indicate that the placing of maize MON 810 and derived products on the market is likely to cause adverse effects on NTOs in the context of its proposed uses. Concerning the NT Lepidoptera, the EFSA GMO Panel concluded that, on the basis of a modelling exercise (Perry *et al*., 2010), the amounts of maize MON 810 pollen grains found in and around maize fields are unlikely to adversely affect a significant proportion of non-target lepidopteran larvae. Nevertheless, considering the uncertainties inherent to all modelling exercises, it advised that, especially in areas of abundance of non-target Lepidoptera populations, the adoption of the cultivation of maize MON 810 be accompanied by management measures in order to mitigate the possible exposure of these species to maize MON 810 pollen.

2.3.2. Impacts on terrestrial[7] NTOs

In its Statement supplementing the ERA of maize Bt11 (EFSA, 2011e), the EFSA GMO Panel studied data on effects of Bt-maize pollen on Lepidoptera species provided by Darvas *et al*., (2004); Lang, (2004); Traxler *et al*., (2005); Lang and Otto, (2010) and the risk management proposals of Hofmann *et al*., (2010) and concluded that '*Bt-maize pollen might be hazardous to the larvae of lepidopteran species of conservation concern, and should therefore be the focus of specific risk management*' (EFSA, 2011e). The EFSA GMO Panel applied a very cautious approach and concluded that only locally exposed non-target Lepidoptera that are 'extremely sensitive' [representing hypothetical species not yet identified] to the Cry1Ab protein may be at risk if exposed to harmful amounts of maize Bt11 pollen (and by analogy of maize MON 810 pollen) (EFSA, 2011e).

Mortality is estimated in two phases: firstly locally, using the 'small-scale' parameters, and then globally, using the 'large-scale' parameters. The term 'locally' means spatially within the crop and its immediate margins, and temporally within the period of pollen shed and deposition. The term 'globally' means after averaging over an entire landscape or regional scale and over a whole growing season. The average expected global mortality is always reduced from the local expected mortality

[7] Terrestrial NTOs = plant- and ground-dwelling non-target organisms

because the latter represents an absolute 'worst-case' which would never occur in practice since it takes no account of large-scale processes (for further details, see EFSA, 2011b,e; Perry, 2011a,b; Perry *et al*., 2010, 2011a,b). The EFSA GMO Panel indicated the hypothetical risk for extremely sensitive NT Lepidoptera exposed to certain quantities of Cry1Ab-expressing maize pollen. No instances of particular species with such a sensitivity have been identified anywhere in the world. In its Statement supplementing the ERA of maize Bt11 (EFSA, 2011e), the EFSA GMO Panel also pointed out that, in a random sample of 500 lepidopteran species, theoretically only one species would be expected to be classified as 'extremely-sensitive'.

The known NT lepidopteran pest species *Plutella xylostella* is the most sensitive species so far identified and is classified as a highly sensitive species. The estimated percentage global mortality of *P. xylostella* never exceeds 1% for a maize Bt11/MON 810 crop surrounded with a 2 m margin of non-Bt maize crop and with a 0.01 host-plant/m² in the field. This global mortality percentage was estimated even with no mitigation measures and for conservative[8] values of the exposure level parameter *R = 0.08 (i.e.,* a maximum uptake of 80% of maize Bt11 (and/or maize MON 810) in a region where maize represents up to 80% of the arable land) (Perry *et al*., 2010, 2011a,b). With mitigation measures and with a more typical value of *R*, estimated global mortality would be considerably smaller than even this low estimate.

The EFSA GMO Panel reiterates that, in its Statement on maize Bt11 (EFSA, 2011e), a wide range of scenarios was explored, including worst-case assumptions for the exposure of European species of non-target Lepidoptera to the Cry1Ab protein from maize Bt11 (and by analogy MON 810) pollen to estimate mortality and to provide quantitative risk conclusions for these species. The EFSA GMO Panel focused on providing estimates of mortality at the local, small-scale level and giving information that will enable risk managers to translate these to global estimates of mortality appropriate to the region modelled (e.g., local protection goals (e.g., occurrence of Lepidoptera of conservational concern) and Cry1Ab-expressing maize in arable land). For further details, please consult EFSA, (2011b,e); Perry, (2011a,b) and Perry *et al*., (2010, 2011a,b).

In response to the concerns expressed by France, the EFSA GMO Panel advises that its conservative assessment and conclusions on a possible risk for sensitive NT Lepidoptera should be put into local context and the risk assessed depending upon local cropping conditions and the presence of sensitive lepidopteran larvae. No supporting data concerning the sensitivity of any particular lepidopteran species present in France or elsewhere in the EU was provided. In summary, the EFSA GMO Panel considers that there is no significant and imminent risk for Lepidoptera.

In its supporting documentation, France is also concerned with possible sublethal effects of the Cry1Ab protein on natural enemies. Such effects were addressed by the EFSA GMO Panel in its 2009 Scientific Opinion on the renewal of maize MON 810 for import, processing for food & feed uses and cultivation (EFSA, 2009). The EFSA GMO Panel concluded that '*maize MON 810 will not cause reductions to natural enemies that are significantly greater from those caused by conventional farming where pesticides are used to control corn borers*'.

Contrasting conclusions are drawn in the existing literature from lower-tier feeding studies on the possible effects of Cry1Ab proteins on coccinellids.

Schmidt *et al.* (2009) and Hilbeck *et al.* (2012b) reported increased larval mortality in the coccinellid *Adalia bipunctata* in experimental feeding studies. Hilbeck *et al*. (2012a,b) suggested that this increased mortality was caused directly by the activated Cry1Ab protein and raised questions regarding whether this protein has an effect on taxa other than Lepidoptera, and concerning its mode of action in *A.bipunctata*. The authors reiterated that their lower tier laboratory studies including the Schmidt *et al*. (2009) studies "provide indications for possible hazards that require further

[8] leading to the greater mortality.

investigation (or possibly long-term field monitoring) to determine whether they pose a risk or translate into 'harm' in the field: no more and no less".

Alvarez-Alfageme *et al*. (2010) and Porcar *et al*. (2010) reported no effects on coccinelid larvae but very different experimental protocols were used in comparison to Schmidt *et al*. (2009) and Hilbeck *et al*. (2012b) which make the studies not directly comparable and may partly explain the different results obtained. However, Alvarez-Alfageme *et al.* (2010) also exposed coccinellid larvae to Cry1Ab protein through prey which accumulated Cry1Ab proteins in their body (spider mites) and no effects were detected.

As already assessed by EFSA (2009), neither a dose-response relationship, nor sublethal effects (on developmental time and adult body weight) on surviving specimens were observed by Schmidt *et al.* (2009). These parameters were not investigated by Hilbeck *et al*. (2012b). Both these features would be indicative of a typical sensitivity response to Cry proteins. Thus the EFSA GMO Panel is of the opinion that existing data are not sufficient to clearly identify a hazard or indicate a new mode of action of Cry proteins on the coccinellid species tested.

An important consideration is that it is unlikely that coccinellid larvae will be exposed to biologically relevant amounts of Cry1Ab protein from maize MON 810. The Cry1Ab protein content in maize MON 810 pollen (which is likely to be the most common source for possible Cry1Ab ingestion for *Adalia bipunctata*) is very low and ranges between 1-97 ηg/g fw (EFSA, 2009; Nguyen and Jehle, 2007). In addition, Bt-proteins are normally absent in aphids feeding on maize (Head *et al*., 2001; Raps *et al*., 2001), which constitute the main diet of many coccinelid larvae and therefore this alternative route of exposure to Cry1Ab protein from maize MON 810 can be considered negligible.

Moreover the EFSA GMO Panel considered higher-tier studies available in the literature (e.g., Pilcher *et al*., 1997; Jasinski *et al*., 2003; Dively and Rose, 2004; de la Poza *et al*., 2005; Lundgren and Wiedenmann, 2005; Eckert *et al*., 2006; Alvarez-Alfageme *et al*., 2008). In field studies, no adverse effects of Bt-maize (different events) were detected on a range of coccinellid species (as reviewed by EFSA 2009, 2011b,e). Therefore the EFSA GMO Panel considers the risk to ladybirds from maize MON 810 to be negligible.

At present, the EFSA GMO Panel is not aware of identified significant adverse effects of the Cry1Ab protein on non-target terrestrial arthropods. Lower- and higher-tier studies showed minimal to undetectable changes in non-target terrestrial arthropods (e.g., Marvier *et al*., 2007; Duan *et al*., 2008; Meissle and Romeis, 2008; Wolfenbarger *et al*., 2008; Malone and Burgess, 2009; Naranjo, 2009).

The EFSA GMO Panel is of the opinion that no new scientific evidence directly relating to maize MON 810 cultivation and to possible adverse effects on terrestrial (plant- and ground-dwelling) NTOs was reported in the documentation provided by France in support of the notified emergency measure.

2.3.3. Impacts on aquatic NTOs

Aquatic NTOs may be exposed to by-products of Bt-expressing maize entering into headwater streams. Based on exposure estimates, Carstens *et al*. (2011) identified shredders (according to Cummins *et al*., 1989) as the functional group within decomposers most likely to be exposed to Cry proteins.

Rosi-Marshall *et al*. (2007) reported that by-products of Bt-expressing maize entered headwater streams in the USA and claimed on the basis of experimental data obtained under lower-tier conditions that this would reduce growth and increase mortality of some non-target aquatic arthropods, especially trichopteran species (see also Chambers *et al*., 2010). Since important background information on levels of exposure and sensitivity of caddisflies to Bt-proteins are missing in the paper by Rosi-Marshall *et al*. (2007), it is widely concluded by others that the conclusions about risk made by the authors are not supported by the data presented in the paper (ACRE, 2007; Beachy *et al*., 2008; Parrott, 2008). Nonetheless, it could be concluded that a potential hazard for trichopterans has been

identified under laboratory conditions when exposed to high doses of Bt-proteins (EFSA, 2009, 2011b).

Recent lower-tier bioassays with four different non-target aquatic leaf-chewing arthropod species (two caddisflies, a crane fly and an isopod) showed no effect on the larvae of caddisflies when fed senesced leaf tissues of Cry1Ab-expressing maize *ad libitum* for 30 days, whereas the negative effects observed on the crane fly and isopod were attributed to tissue-mediated differences among the isogenic line treatments (Jensen *et al*., 2010; Lamp, 2010). The authors attributed the lack of observable toxic effects in their study to the reduction of bioactivity of the Cry1Ab protein, as maize tissues used were previously exposed for two weeks to environmental conditions (terrestrial or aquatic environments). Moreover, no adverse effects on the abundance and biomass of Trichoptera have been reported in natural conditions in Tier 3 studies so far (Chambers *et al*., 2010).

Although there is indication of a potential hazard for trichopterans under laboratory conditions when exposed to high doses of Cry proteins, no substantial aquatic exposure to the Cry1Ab protein contained within maize plant tissue is expected. Carstens *et al*. (2011) calculated that, even under worst-case conditions, the exposure of shredders to Bt-maize is low (for further details, see EFSA, 2011b).

Furthermore, the EFSA GMO Panel considered the paper by Bøhn *et al*. (2010) as referred to by France in its supporting documentation. As stated in the EFSA GMO Panel Scientific Opinion on GM insect-resistant maize MON88017 (for further details, see EFSA, 2011d), Bøhn *et al*. (2008, 2010) revealed that *Daphnia magna*, a filter-feeder, fed a 100 % suspension of maize MON 810 flour under lower-tier conditions had a higher mortality and reduced fitness performance, as compared with the non-Bt-maize treatment, suggesting toxic effects of the Cry1Ab protein. However, it remains unclear whether the unusual delays in development of *D. magna* fed non-Bt-maize have been caused by nutritional deficiencies related to the maize-based diet or the presence of the Cry1Ab protein (EFSA, 2009d; Ricroch *et al*., 2010).

The EFSA GMO Panel is of the opinion that no new scientific evidence directly relating to maize MON 810 cultivation and to possible adverse effects on aquatic NTOs was reported in the documentation provided by France in support of the notified emergency measure.

2.4. Outbreaks of secondary pests

Cultivation of Bt-crops, such as maize MON 810, may result in reduction of the use of insecticides and may cause changes in crop rotations in response to reduced pest pressure (Gómez-Barbero *et al.*, 2008; Brookes and Barfoot, 2010). However, this reduction in pesticide use and the narrow spectrum of activity of Cry proteins may provide an opportunity for secondary pests, previously controlled by insecticides used against key target pests, to reach damaging levels (Wang *et al*., 2008; Lu *et al*., 2010). Natural enemies failing to fully control secondary pests, and reduced competition with target pests might also play a role in secondary pest outbreaks (Catangui and Berg, 2006; Sanvido *et al*., 2007; Eichenseer *et al*. 2008; Romeis *et al*., 2008; Fitt, 2008; Kennedy, 2008; Naranjo *et al*., 2008; Dorhout and Rice, 2010; Lu *et al*., 2010; Virla *et al*., 2010). During the last decade *Striacosta albicosta* (the western bean cutworm) expanded across the corn belt in the USA due to the decrease of competition from other lepidopteran target pests as a consequence of Bt-maize cultivation (Michel *et al*., 2010). The western bean cutworm is not affected by the Cry1Ab protein expressed in Bt-maize, and was therefore able to exploit the ecological niche of the more susceptible *Helicoverpa zea* (corn earworm) and European corn borer (Catangui and Berg, 2006; Dorhout and Rice, 2010, Hutchison *et al*., 2011). However, *S. albicosta* is not present in European maize ecosystems (for further details, see EFSA, 2011e).

It should also be noted that the emergence of secondary pests is not specific to Bt-crop cultivations only or maize MON 810 in particular. Arthropod assemblages in agricultural fields are in a continuous fluctuation in terms of their species number, composition and individual densities over time and space.

Human interventions, including pest control, influence these parameters. Whenever pest management of crops changes, the abundance of some pest species may decline and other pest species may increase.

If secondary pests reach damaging levels, additional pest control measures might be necessary and some changes in management could result in adverse environmental effects. In general, it is recommended to adhere to integrated pest management (IPM) principles to manage pests and secondary pests and minimise environmental impacts (Meissle *et al.*, 2011). Predicting the incidence of secondary pests and the environmental consequences of changes in management measures is highly dependent upon cultivation practices, farming systems and regional environmental factors.

The EFSA GMO Panel is of the opinion that no new scientific evidence directly relating to maize MON 810 cultivation and to possible outbreaks of secondary pests was reported in the documentation provided by France in support of the notified emergency measure.

3. CONCERNS RELATED TO MANAGEMENT MEASURES, INCLUDING PMEM, FOR MAIZE MON 810

In its supporting document, France asked the European Commission to consider appropriate management measures associated to the cultivation of maize MON 810 in the EU. In this respect, the document provided by France lists the EFSA GMO Panel's recommendations for management and monitoring measures as detailed in the EFSA GMO Panel Scientific Opinions on the annual PMEM reports of maize MON 810 (EFSA, 2011c, 2012) and the Statement on maize Bt11 (EFSA, 2011e).

Against this background, in July 2011, the EFSA GMO Panel adopted an updated Guidance Document on the PMEM of GM plants (EFSA, 2011a) which provides applicants and risk managers with guidance on the strategy, methodology and reporting of PMEM of GM plants. Detailed recommendations were also given to the applicant for the improvement of its IRM/CSM and GS of maize MON 810 (for further details, see EFSA, 2011c, 2012). The applicant was provided with specific recommendations to improve its IRM plan (e.g., non-Bt *refugia*, sampling over time in 'hotspot areas' with high uptake of maize MON 810 and multivoltine target pests) as well as the methodology of the GS of maize MON 810 (see Appendix 1 to EFSA, 2011c, 2012 providing a methodological guidance for the assessment of the farmer questionnaires).

In addition, in 2009, the EFSA GMO Panel already recommended (EFSA, 2009) that, especially in areas of abundance of non-target Lepidoptera populations, the adoption of the cultivation of maize MON 810 be accompanied by appropriate management measures in order to mitigate the possible exposure of these species to maize MON 810 pollen. The implications of these management measures should be considered in the PMEM plan. Further details on the framework to implement appropriate risk mitigation measures, wherever it is necessary, are given in the recent Statement of the EFSA GMO Panel on the similar Cry1Ab-expressing GM maize Bt11 (for further details, see EFSA, 2011e). The EFSA GMO Panel reiterates that, through its Statement on maize Bt11, risk managers are provided with guidance to: (i) estimate the mortality of exposed non-target Lepidoptera with a range of various sensitivities to maize Bt11 (and by analogy to maize MON 810) pollen and (ii) choose risk mitigation measures proportionate to the level of identified risk and to the protection goals pertaining to their region.

The EFSA GMO Panel confirms from its evaluation of the PMEM results on maize MON 810 that no adverse effects on the environment, human and animal health due to maize MON 810 cultivation were identified during the 2009 and 2010 growing seasons.

CONCLUSIONS

The EFSA GMO Panel has scrutinized the documentation provided by France in support of its emergency measure on GM maize MON 810. The EFSA GMO Panel considered the relevance of concerns raised by France in the light of the most recent and relevant scientific data published in the scientific literature.

During its evaluation of the supporting documentation, the EFSA GMO Panel has noted that some publications referred to by France were already part of the submission package by France for its safeguard clause and emergency measure on maize MON 810 in 2008. Those publications were addressed previously by the EFSA GMO Panel in its 2008 Scientific Opinion on the safeguard clause and emergency measure notified by France on maize MON 810 (EFSA, 2008), which concluded that no specific scientific evidence, in terms of risk to human and animal health or the environment, was provided that would justify the invocation of a safeguard clause under Article 23 of Directive 2001/18/EC (EC, 2001) and an emergency measure under Article 34 under Regulation (EC) No 1829/2003 (EC, 2003).

In the remaining documentation provided by France in support of the current emergency measure on maize MON 810, the EFSA GMO Panel could not identify any new science-based evidence indicating that maize MON 810 cultivation in the EU poses a significant and imminent risk to the human and animal health or the environment.

In relation to the management and monitoring of maize MON 810, the EFSA GMO Panel has previously assessed the PMEM plan and PMEM reports of maize MON 810 for the 2009 and 2010 growing seasons. Hence, the EFSA GMO Panel refers to its recent recommendations to improve the management and monitoring measures of maize MON 810 and other Bt maize transformation events. The EFSA GMO Panel confirms from its evaluation of the PMEM results on maize MON 810 that no adverse effects on the environment, human and animal health due to maize MON 810 cultivation were identified during the 2009 and 2010 growing seasons.

In conclusion, the EFSA GMO Panel is of the opinion that, based on the documentation submitted by France, there is no specific scientific evidence, in terms of risk to human and animal health or the environment, that would support the notification of an emergency measure under Article 34 of Regulation (EC) No 1829/2003 and that would invalidate its previous risk assessments of maize MON 810.

DOCUMENTATION PROVIDED TO EFSA

1. Letter from the European Commission, dated 22 February 2012, to the EFSA Executive Director requesting the assessment by EFSA of the scientific elements supporting the French request for a prohibition of the placing on the market of GM maize MON 810 for cultivation purposes in the EU.

2. Acknowledgement letter, dated 13 March 2012, from the EFSA Executive Director to the European Commission.

3. Letter from the Austrian Federal Ministry of Health, dated 21 March 2012, to the Head of the DG SANCO unit E1 concerning the French emergency measure on maize MON 810.

4. Letter from the Hungarian Ministry of rural Development, dated 22 March 2012, to the Head of the DG SANCO unit E1 concerning the French emergency measure on maize MON 810.

5. Letter from the European Commission, dated 16 April 2012, to the EFSA Executive Director requesting the assessment by the EFSA GMO Panel of the scientific elements supporting the French request for a prohibition of the placing on the market of GM maize MON 810 for cultivation purposes in the EU.

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