STATEMENT OF EFSA

Statement on a request from the European Commission related to an emergency measure notified by France under Article 34 of Regulation (EC) 1829/2003 to prohibit the cultivation of genetically modified maize MON 810[1]

European Food Safety Authority[2,3]

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ABSTRACT

Following a request from the European Commission, the European Food Safety Authority (EFSA) evaluated the documentation submitted by France under Article 34 of Regulation (EC) 1829/2003 in support of its request to prohibit the cultivation of genetically modified maize MON 810 in the EU. Neither the scientific publications cited in the documentation submitted by France with relevance to maize MON 810 nor the arguments put forward by France reveal any new information that would invalidate the previous risk assessment conclusions and risk management recommendations made by the EFSA GMO Panel. EFSA considers that the previous GMO Panel risk assessment conclusions and risk management recommendations on maize MON 810 remain valid and applicable. Therefore, EFSA concludes 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 adoption of an emergency measure on the cultivation of maize MON 810 under Article 34 of Regulation (EC) 1829/2003.

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KEY WORDS

GMO, maize (*Zea mays*), MON 810, France, emergency measure, environment, Directive 2002/53/EC

[1] On request from the European Commission, Question No EFSA-Q-2014-00203, approved on 31 July 2014.

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[3] Acknowledgement: EFSA wishes to thank EFSA staff: Yann Devos, Anna Lanzoni and Yi Liu for the preparatory work on this statement, and the members of the Working Groups on Food and Feed Risk Assessment and Environment Risk Assessment for the support provided to this statement.

Suggested citation: EFSA (European Food Safety Authority), 2014. Statement on a request from the European Commission related to an emergency measure notified by France under Article 34 of Regulation (EC) 1829/2003 to prohibit the cultivation of genetically modified maize MON 810. EFSA Journal 2014;12(8):3809, 18 pp., doi:10.2903/j.efsa.2014.3809.

Available online: www.efsa.europa.eu/efsajournal

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SUMMARY

On 25 March 2014, the European Commission requested the European Food Safety Authority (EFSA) to assess the supporting documentation submitted by France to notify an emergency measure under Article 34 of Regulation (EC) 1829/2003, prohibiting the cultivation of genetically modified maize MON 810 in the EU.

EFSA assessed the documentation supplied by the French Authorities and the scientific publications cited in the French Authorities' report. For each area of concern outlined in the French Authorities' report, EFSA assessed whether any of the scientific publications not previously addressed by EFSA and/or its GMO Panel, or any of the arguments put forward by France, would invalidate the previous GMO Panel conclusions on the safety of maize MON 810. Moreover, EFSA 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 French Authorities' report, EFSA noted that most of the cited scientific publications were addressed previously by EFSA and its GMO Panel in various scientific outputs. These publications were therefore not considered further. For the remaining scientific publications, EFSA has focused its assessment on those aspects that are relevant to maize MON 810.

Neither the scientific publications cited in the French Authorities' report with relevance to maize MON 810 nor the arguments put forward by France reveal any new information that would invalidate the previous risk assessment conclusions and risk management recommendations made by the EFSA GMO Panel. Therefore, EFSA considers that the previous GMO Panel risk assessment conclusions and risk management recommendations on maize MON 810 remain valid and applicable.

EFSA concludes 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 adoption of an emergency measure on the cultivation of maize MON 810 under Article 34 of Regulation (EC) 1829/2003.

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BACKGROUND AS PROVIDED BY THE EUROPEAN COMMISSION AND EFSA

The marketing of maize MON 810 (notification C/F/95/12-02) was authorised under Directive 90/220/EEC in the European Union (EU) for all, other than food, uses by the Commission Decision 98/294/EC of 22 April 1998 (EC, 1998). Consent was granted to the applicant (Monsanto Europe S.A.) by France on 3 August 1998. Food uses of maize derivatives were notified according to Article 5 of the Novel Food Regulation (EC) No 258/97 on 6 February 1998.

On 15 June 2009, the EFSA GMO Panel issued a scientific opinion on the renewal of the authorisation for the continued marketing of: (1) existing food and food ingredients produced from maize MON 810; (2) feed consisting of and/or containing maize MON 810, including the use of seed for cultivation; and (3) food and feed additives, and feed materials produced from maize MON 810. The EFSA GMO Panel concluded that "*maize MON 810 is as safe as its conventional counterpart with respect to potential effects on human and animal health*", and 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 nontarget 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*". In addition, 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*" (EFSA, 2009).

On 30 November 2011, the EFSA GMO Panel adopted a statement supplementing the environmental risk assessment conclusions and risk management recommendations on maize Bt11 cultivation (EFSA Panel on Genetically Modified Organisms, 2011c). In its statement, the EFSA GMO Panel concluded that "*subject to appropriate management measures, maize Bt11 cultivation is unlikely to raise additional safety concerns for the environment compared to conventional maize*" (EFSA Panel on Genetically Modified Organisms, 2011c). The EFSA GMO Panel considered that the environmental risk assessment conclusions and risk management recommendations on non-target Lepidoptera for maize Bt11 apply equally to maize MON 810 due to the similarities between both *Bt*-maize events (i.e., identity of amino acid sequence of the core of the Cry1Ab protein, similar biological activity against susceptible Lepidoptera, similar Cry1Ab protein expression level in pollen).

The EFSA GMO Panel further supplemented its previous risk management recommendations on maize Bt11 and MON 810 cultivation by reapplying the mathematical model developed by Perry et al. (2010, 2011, 2012), in order to consider additional hypothetical agricultural conditions, and to provide additional information on the factors affecting the insect resistance management (IRM) strategy (EFSA Panel on Genetically Modified Organisms, 2012d).

On 6 December 2012, following a request from the European Commission, the EFSA GMO Panel compiled its previous risk assessment conclusions and risk management recommendations on maize MON 810, and considered their validity in the light of new relevant scientific publications published from 2009 onwards (EFSA Panel on Genetically Modified Organisms, 2012e). Based on the performed literature search, the EFSA GMO Panel concluded that "*its previous risk assessment conclusions on maize MON 810 as well as its recommendations on risk management measures and monitoring remain valid and applicable.*"

Following requests of the European Commission to assess the annual post-market environmental monitoring reports on maize MON 810 cultivation submitted by the applicant, the EFSA GMO Panel issued scientific opinions on the 2009, 2010, 2011 and 2012 PMEM reports on maize MON 810 (EFSA Panel on Genetically Modified Organisms, 2011a, 2012a, 2013c, 2014). The EFSA GMO Panel noted shortcomings in the methodology for case-specific monitoring and general surveillance, and made recommendations to strengthen the annual PMEM activities on maize MON 810. So far, the

data submitted by the applicant in its PMEM reports did not indicate any adverse effects on human and animal health or the environment arising from the cultivation of maize MON 810.

Several EU Member States invoked safeguard clause or emergency measures to provisionally restrict or prohibit the marketing of maize MON 810 on their territory. For all cases for which the EFSA GMO Panel or EFSA has been asked by the European Commission to evaluate whether the invocation was justifiable on the basis of the scientific information submitted in support of a safeguard clause or emergency measures, the EFSA GMO Panel or EFSA concluded that, in terms of risk to human and animal health and the environment, no new scientific evidence had been presented that would invalidate its previous risk assessment conclusions on maize MON 810 (EFSA, 2004, 2005, 2006a, b, 2008a, b, c, d, 2014a; EFSA Panel on Genetically Modified Organisms, 2012b, c, 2013a, b).

TERMS OF REFERENCE AS PROVIDED BY THE EUROPEAN COMMISSION

EFSA is requested in accordance with Article 31 of Regulation (EC) No 178/2002 to provide a statement:

- assessing if the French authorities submitted new scientific evidences in support to their request for a prohibition of GM maize MON 810 cultivation according to Article 34 of Regulation (EC) 1829/2003; and, where appropriate;
- indicating whether these new scientific evidences might lead the GMO Panel to reconsider its previous safety assessments of GM maize MON 810.

ASSESSMENT

At the request of the European Commission, EFSA assessed the documentation supplied by France (referred to hereafter as the French Authorities' report) and the scientific publications cited in the French Authorities' report. For each area of concern outlined in the French Authorities' report, EFSA assessed whether any of the scientific publications not previously addressed by EFSA and/or its GMO Panel, or any of the arguments put forward by France, would invalidate the previous GMO Panel conclusions on the safety of maize MON 810. Moreover, EFSA considered the relevance of concerns raised by the French Authorities in the light of the most recent and relevant scientific data published in the scientific literature.

During its evaluation of the French Authorities' report, EFSA noted that most of the cited scientific publications were addressed previously by EFSA and its GMO Panel in various scientific outputs (e.g., EFSA Panel on Genetically Modified Organisms, 2011b, c, 2012d, 2013c; EFSA, 2014b). These publications are therefore not considered further here, except for HCB (2013), which formed a key part of the basis of the scientific argumentation put forward in the French Authorities' report. In the case of the remaining scientific publications (Campagne et al., 2013; Mezzomo et al., 2013; Zhou et al., 2014), EFSA has focused its assessment on those aspects that are relevant to maize MON 810.

The EFSA assessment below is structured into the Sections used in the French Authorities' report.

1. EFSA assessment of Section I.1, "*Appearance of resistance on target pests*" as referred to in the French Authorities 'report

1.1. Cited scientific publications

Of the scientific publications cited in the French Authorities' report with relevance to maize MON 810, one publication (Campagne et al., 2013) was not previously considered by EFSA and/or its GMO Panel. Although the HCB (2013) report was recently considered by the EFSA GMO Panel (EFSA Panel on Genetically Modified Organisms, 2014), it is also addressed here for completeness. The possible relevance of these publications for the risk assessment of maize MON 810 was scrutinised.

- Campagne et al. (2013): The African stem borer (*Busseola fusca*) has evolved high levels of resistance to the Cry1Ab toxin expressed in *Bt*-maize in South Africa. Campagne et al. (2013) investigated the inheritance of Cry1Ab resistance in the African stem borer; they performed controlled crosses with *B. fusca* and evaluated larval survival on Cry1Ab-expressing *Bt*-maize and non-*Bt*-maize. The results showed that survival of resistant larvae and F_1 progeny from resistant × susceptible parents was not lower on Cry1Ab-expressing maize than on non-*Bt*-maize. The authors concluded that: (1) Cry1Ab resistance is inherited dominantly rather than recessively; and (2) insect resistance management (IRM) strategies for Cry1Ab-expressing *Bt*-maize must address the non-recessive inheritance of Cry1Ab resistance in *B. fusca* in South Africa.

- HCB (2013): In the HCB (2013) report, the French Haut Conseil des Biotechnologies (HCB) analysed the susceptibility of the European corn borer (ECB) and Mediterranean corn borer (MCB) (*Ostrinia nubilalis* and *Sesamia nonagrioides*, respectively) to the Cry1Ab toxin over time, using the resistance monitoring data supplied by the applicant (Monsanto, 2013). The HCB considered that the resistance monitoring data indicate that the susceptibility to the Cry1Ab toxin of ECB/MCB populations in Spain is decreased when compared with the reference laboratory strain. Therefore, the HCB recommended: "*(1) re-sampling in 2013 (i.e., prior to 2014) the ECB populations sampled in 2012 in South-West Iberia in order to determine whether there is any increase in their level of resistance to the Cry1Ab toxin; (2) making sure that susceptibility of laboratory reference strains is stable; and (3) improving refuge compliance by MON 810 maize growers in Europe*".

1.2. Relevance of the cited publications for the risk assessment of maize MON 810

1.2.1. Resistance management (Campagne et al., 2013)

As part of the GMO approval process, applicants submitting a market registration application for cultivation of *Bt*-crops proactively provide an IRM plan (Devos et al., 2013, 2014). IRM plans are designed to reduce the selection pressure associated with *Bt*-crops, in order to prevent or at least delay resistance evolution in the target insect pests (Bates et al., 2005; Alcalde et al., 2007; Andow, 2008; MacIntosh, 2010; Head and Greenplate, 2012). As currently implemented for several *Bt*-crops in several countries, IRM plans usually rely on the high dose/refuge (HDR) strategy (Gould, 1998; Glaser and Matten, 2003; MacIntosh, 2010).

The success of the HDR strategy is aided if the following conditions are met (reviewed by Tabashnik et al., 2013): (1) the *Bt*-toxin is expressed at appropriate levels in relevant plant parts at the appropriate times in relation to the target pest's life cycle; (2) initial resistance alleles are rare in the target insect pest population, so that nearly all resistance alleles will be in heterozygote individuals that cannot survive on the *Bt*-crop; (3) random mating occurs between resistant insects emerging in *Bt*-crops and susceptible insects preserved on refuges at sufficient levels; (4) resistance alleles are partially or fully recessive; and (5) fitness costs are associated with the resistance. Compliance with refuge requirements is an additional critical factor contributing to the success of IRM plans in delaying the rate at which resistance evolves.

When the conditions contributing to the success of the HDR strategy were not met, field-selected resistance to *Bt*-crops occurred, unless refuges were abundant or other (area-wide) integrated pest management (IPM) measures were implemented (reviewed by Tabashnik et al., 2013). Instances of field-selected resistance to *Bt*-maize active against lepidopteran maize pests have been reported in populations of *B. fusca* in South Africa (van Rensburg, 2007; Kruger et al., 2009, 2011; Van den Berg et al., 2013), and in populations of the fall armyworm (*Spodoptera frugiperda*) in Puerto Rico (Matten et al., 2008; Moar et al., 2008; Tabashnik, 2008; Tabashnik et al., 2008a; Storer et al., 2010, 2012), where larvae were able to survive on Cry1Ab-expressing *Bt*-maize MON 810 and Cry1F-expressing *Bt*-maize 1507, respectively.

Reasons for the above-mentioned instances of field-selected resistance range from the autosomally, non-recessive inheritance of resistance to specific agronomic/environmental factors, along with the insufficient planting of refuges of non-*Bt*-maize (Huang et al., 2011; Kruger et al., 2012; Tabashnik et al., 2013; Van den Berg et al., 2013).

EFSA reiterates that the instances of field resistance to lepidopteran-active *Bt*-maize have been reported outside Europe for two target pests in maize that are not present in the European fauna. Moreover, heterozygotes from resistant ECB strains obtained under laboratory conditions did not survive exposure to Cry1Ab-expressing *Bt*-maize plants, supporting the functional recessiveness of resistance to these plants (Siegfried and Hellmich, 2012; Siegfried et al., 2013). If the inheritance of

resistance is not recessive, then modelling results suggest that increasing refuge abundance can still substantially delay resistance (Tabashnik et al., 2008a, b).

At present, EFSA is not aware of early warning signs indicating increases in tolerance to Cry1Ab-expressing *Bt*-maize in field populations of ECB/MCB. Annual assessments of ECB/MCB susceptibility to Cry1Ab in the USA and EU have not revealed any significant change in susceptibility or identified populations that survive on Cry1Ab-expressing *Bt*-maize plants (after more than ten years of exposure to the Cry1Ab protein in the USA) (Farinós et al., 2004, 2011; Stodola et al., 2006; Andreadis et al., 2007; Siegfried et al., 2007; Crespo et al., 2009, 2010; EFSA Panel on Genetically Modified Organisms, 2011a, 2012a, 2013c, 2014; Siegfried and Hellmich, 2012). Moreover, no major resistance alleles have ever been recorded either through laboratory selection experiments (Chaufaux et al., 2001; Siqueira et al., 2004) or by F_2 screening of field populations (Andow et al., 2000; Bourguet et al., 2003; Stodola et al., 2006), strongly suggesting that the frequency of alleles conferring resistance to Cry1Ab-expressing *Bt*-maize plants is < 0.001 in all populations examined to date (Siegfried and Hellmich, 2012; Siegfried et al., 2013). The lack of resistance in ECB/MCB and other major insect pests targeted by *Bt*-crops attests that the HDR strategy is capable of preventing, or at least delaying, resistance under field conditions (Andow, 2008; Tabashnik et al., 2008a, b, 2009, 2013; Huang et al., 2011; Siegfried and Hellmich, 2012).

To ensure effective long-term ECB/MCB management and the sustainable use of *Bt*-maize, the EFSA GMO Panel advocated an IPM approach in which *Bt*-maize is only one of many management options. Moreover, resistance monitoring to detect early warning signs indicating resistance evolution in the field, compliance monitoring to assess farmers' compliance with IRM requirements and education (training) programmes aiding farmers to understand the importance of adhering to IRM requirements are essential to the success of the HDR strategy and should therefore continue to form an integral part of IRM plans for *Bt*-maize.

1.2.2. Resistance monitoring (HCB, 2013)

IRM plans for *Bt*-crops require routine monitoring for resistance evolution, so that early warning signs indicating increases in tolerance in the field may be detected (Siegfried and Spencer, 2012). A timely detection of such signs enables actions to limit the survival of resistant insects and to slow or prevent their spread should resistance have evolved among field populations (Siegfried et al., 2007).

Based on the 2012 resistance monitoring data for maize MON 810 (Monsanto, 2013), the applicant concluded that there is no indication of decreased Cry1Ab susceptibility in ECB/MCB populations in Spain. However, using the same dataset, the HCB considered that there is a decrease in susceptibility in the ECB/MCB populations in Spain. The EFSA GMO Panel previously assessed the above-mentioned dataset and the analysis reported by the HCB (2013) (see EFSA Panel on Genetically Modified Organisms, 2014). The EFSA GMO Panel concluded that the potentially increased Cry1Ab tolerance of the ECB/MCB populations, as hypothesised by the HCB, might be due to the declining performance of the reference laboratory strain (e.g., through the infection with pathogens or inbreeding depression). Even though the 2012 dataset showed a trend towards increased values of moulting inhibition concentration (MIC), no significant and consistent decrease in Cry1Ab susceptibility of the ECB and MCB field populations in Spain was observed over time (EFSA Panel on Genetically Modified Organisms, 2014).

In the 2012 PMEM report supplied by the applicant (Monsanto, 2013), the applicant acknowledged that the reference laboratory strain might have shown poor performance, yet the possible reasons were not discussed further. Therefore, the EFSA GMO Panel recommended that the applicant investigates the stability and quality of the reference laboratory strain. The recommendation of the EFSA GMO Panel is consistent with that of the HCB, which advocated "*making sure that susceptibility of laboratory reference strains is stable*".

The susceptibility of the target insect pest has been shown to vary considerably depending upon the source of *Bt*-toxins used (Farinós et al., 2004; Saeglitz et al., 2006). Therefore, it is advisable to use the same *Bt*-toxin source throughout the duration of resistance monitoring. In its 2012 PMEM report, the applicant indicated that it used a new Cry1Ab toxin batch from 2012 onwards, which could have a different biological activity from the initial one used until 2010. The EFSA GMO Panel therefore assessed whether the hypothesised change in susceptibility could be attributed to the use of different Cry1Ab toxin batches. A study, performed by the applicant, indicated that the initial and new *Bt*-toxin batch have similar biological activity on ECB (Appendix 8 in Monsanto 2013). It is therefore considered unlikely that the hypothesised change in susceptibility can be attributed to the use of different Cry1Ab toxin batches.

To ensure an early detection of change in susceptibility of the ECB and MCB field populations, the EFSA GMO Panel strongly reiterated its previous recommendation for annual sampling of ECB/MCB in areas of high maize MON 810 adoption rate, especially in north-east Spain in 2014 (EFSA Panel on Genetically Modified Organisms, 2013d).

1.3. EFSA conclusion

Neither the results reported by Campagne et al. (2013) and HCB (2013) nor the arguments put forward by France in the French Authorities' report reveal any new information that would invalidate the previous risk assessment conclusions and risk management recommendations made by the EFSA GMO Panel.

2. EFSA assessment of Section I.3, "*Impact of MON810 maize on non-target invertebrates*" as referred to in the French Authorities' report

2.1. Cited scientific publications

From the scientific publications in the French Authorities' report with relevance to maize MON 810, two publications (Mezzomo et al., 2013; Zhou et al., 2014) were not previously considered by EFSA and/or its GMO Panel. The possible relevance of these publications for the risk assessment of maize MON 810 was scrutinised.

- Mezzomo et al. (2013): Mezzomo et al. (2013) studied the potential haematotoxicity (by conventional haematology) and genotoxicity (by *in vivo* micronucleus test) of spores of four *Bt*-strains expressing Cry toxins in Swiss albino mice. These *Bt*-strains were genetically modified to express Cry1Aa, Cry1Ab, Cry1Ac or Cry2Aa. The spores, re-suspended in distilled water, were given by oral gavage in a single administration of one of three doses (27, 136 or 270 mg of lyophilised spores/kg body weight (bw)). Binary combinations of lyophilised spores were also given at 270 mg/kg bw. The negative control group received distilled water (vehicle). As a positive control for the micronucleus test, mice received cyclophosphamide at 27 mg/kg bw. Group size was three male and three female mice. Blood and bone marrow were sampled after 24 hours in all groups. Blood samples were also taken after 72 hours or 7 days from additional groups receiving the high dose of the individual *Bt*-strains or 27 mg cyclophosphamide/kg bw. Based on the results, the authors concluded that administrations of *Bt*-spores provoked selective haematotoxicity, particularly for the erythroid lineage; a significant reduction in bone marrow cell proliferation demonstrated cytotoxic but not genotoxic effects.

- Zhou et al. (2014): Zhou et al. (2014) reported that the Cry1Ab toxin affects the metabolic enzymes acetylcholine esterase, glutathione peroxidase and superoxide dismutase in the predatory spiders *Ummeliata insecticeps* and *Pardosa pseudoannulata* when fed Cry1Ab-containing prey (fruit flies). The authors concluded that plant-produced *Bt*-proteins can affect non-target arthropods at the physiological and biochemical level, and reduce their fitness.

2.2. Relevance of the cited publications for the risk assessment of maize MON 810

2.2.1. Haematotoxicity (Mezzomo et al., 2013)

The test items were not purified Cry toxins, but viable *Bt*-spores expressing the Cry toxins. In addition, the *Bacillus* species used is known to be capable of producing enterotoxins (e.g., Gaviria Rivera et al., 2000) and a range of other toxic materials (Butko, 2003). Therefore, in the absence of an appropriate *Bt*-control strain, it is not possible to attribute any observed findings specifically to the Cry toxins, as was done by Mezzomo et al. (2013) in describing their results.

This acute study showed several key weaknesses which impede interpretation of the results. First, a study which addresses only peripheral blood findings of haematotoxicity has limited value in the absence of gross morphological and/or histopathological investigations. Haematological changes may be secondary to other pathological conditions (for example to haemorrhages) which were not monitored in this study. Second, vehicle control groups for the evaluation of peripheral blood analyses at 72 hours and 7 days after administration were missing. Finally, there is a lack of data indicating normal variation of the measured parameters under the study conditions, particularly considering the small number of animals per sex and per group and the use of a single vehicle control group (at 24 hours after administration). In addition, the micronucleus test was not performed in accordance with the current standards (OECD Guideline TG 474), in particular with respect to the number of animals used (only three animals per sex and group, instead of five), the dosing regime (sampling at only one time point, instead of at least two), as well as the presentation and interpretation of results (only group data but not individual data were considered).

Regarding the endpoints measured 24 hours after administration, the authors reported that four parameters of the erythrogram (mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), mean corpuscular volume (MCV) and red cell distribution width (RDW)) showed statistically significant differences compared with the vehicle control group, but no significant differences were identified in the leucogram. Overall, EFSA considers that the values of the erythrogram parameters for the four *Bt*-strains and for the three dose levels are very similar. The only notable difference remained between the test groups and vehicle control group for the MCV and its coefficient of variation (RDW). These differences were small in magnitude (around 10 %) and were not dose-related. Furthermore, these parameters are derived by calculation from the haematocrit, red blood cells (RBCs) and haemoglobin content of the blood, none of which showed a significant difference. Decreases in the percentage of polychromatic erythrocytes in the bone marrow of treated animals relative to the vehicle control group were also used as an argument to support effects on the erythroid lineage by the authors. However, owing to the limitations of the micronucleus test performed, this conclusion is not justified.

The authors also reported significant differences in platelet counts and related parameters (mean platelet volume (MPV), platelet large cell ratio (P-LCR) and platelet distribution width (PDW)) measured 24 hours after administration. Platelet counts are known to be highly variable, and the corresponding figures for the three dose levels showed no evidence of dose-effect relationship.

Based on these findings, EFSA concludes that the Mezzomo et al. (2013) publication does not support the conclusions on haematotoxicity associated with the Cry toxins.

2.2.2. Potential adverse effects on spiders (Zhou et al., 2014)

The findings reported by Zhou et al. (2014) suggest that the Cry1Ab toxin can affect enzyme activity in predatory spiders when fed Cry1Ab-containing prey. Even though the spiders have been shown to take up the Cry1Ab toxin when feeding on the Cry1Ab-containing prey, no correlation can be made between the Cry1Ab toxin content measured in the spiders and the observed differences in enzyme activity in the spiders. Some limitations of the study (such as undefined number of organisms tested

and replications; lack of details on how the protoxin was quantified in the diet; ill-characterised and -described purity of the Cry1Ab toxin; lack of a positive control) make it difficult to explain the mechanism leading to the observed differences. Therefore, EFSA considers that the dataset reported by Zhou et al. (2014) is preliminary and is not sufficient to demonstrate a new mode of action of the Cry1Ab protein on spiders.

In addition, the biological relevance of the observations made remains difficult to assess, as it is not clear how the observed differences in enzyme activity are related to fitness parameters of the spiders; the authors did not report on life-table parameters of the spiders. Moreover, the reported response was transient.

Finally, exposure has not been sufficiently characterised. The test model used by Zhou et al. (2014) does not reflect realistic exposure conditions; spiders are generalist predators, and the level of exposure to the Cry1Ab toxin will depend on the prey spectrum of individual species. In the experiments performed by Zhou et al. (2014), the variability of food uptake was not controlled, and this may have influenced the observed variability of the Cry1Ab toxin content in the spiders. Therefore, it remains challenging to extrapolate the results of the observations made under controlled conditions to spider activity under field conditions.

At present, EFSA is not aware of identified significant adverse effects of the Cry1Ab toxin on spiders. Laboratory studies have indicated that plant-produced Cry proteins have no direct effects on life-table parameters of spiders after ingestion, whereas field studies confirmed that population densities of spiders are not adversely affected (reviewed by Peterson et al., 2011). Those findings are in line with the outcomes of meta-analyses, in which a broad range of beneficial arthropods including spiders were addressed (Marvier et al., 2007; Wolfenbarger et al., 2008; Naranjo, 2009; Peterson et al., 2011; Albajes et al., 2013; Comas et al., 2014).

2.3. EFSA conclusion

Neither the results reported by Mezzomo et al. (2013) and Zhou et al. (2014) nor the arguments put forward by France in the French Authorities' report reveal any new information that would invalidate the previous risk assessment conclusions and risk management recommendations made by the EFSA GMO Panel.

3. EFSA assessment of Section II, "*Management measures needed to protect the environment*" as referred to in the French Authorities' report

Section II of the French Authorities' report lists all the recommendations for risk management measures made by the EFSA GMO Panel in of its various scientific outputs (e.g., EFSA, 2009; EFSA Panel on Genetically Modified Organisms, 2011a, c, 2012a, d, e, 2013c, 2014), and does not present any new scientific information.

4. EFSA assessment of Section III, "*Insufficient implementation of management measures*" as referred to in the French Authorities' report

Section III of the French Authorities' report focuses on the legal enforcement of EFSA's recommendations. This issue is not in EFSA's remit and therefore not considered further.

5. Conclusions

Neither the scientific publications cited in the French Authorities' report with relevance to maize MON 810 nor the arguments put forward by France reveal any new information that would invalidate the previous risk assessment conclusions and risk management recommendations made by the EFSA GMO Panel. Therefore, EFSA considers that the previous GMO Panel risk assessment conclusions and risk management recommendations on maize MON 810 remain valid and applicable.

EFSA concludes 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 adoption of an emergency measure on the cultivation of maize MON 810 under Article 34 of Regulation (EC) 1829/2003.

DOCUMENTATION PROVIDED TO EFSA

1. Letter from the European Commission, dated 25 March 2014, 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 16 April 2014, from the EFSA Executive Director to the European Commission.

REFERENCES

Alcalde E, Amijee F, Blache G, Bremer C, Fernandez S, Garcia-Alonso M, Holt K, Legris G, Novillo C, Schlotter P, Storer N and Tinland B, 2007. Insect resistance monitoring for Bt maize cultivation in the EU: proposal from the industry IRM working group. Journal of Consumer Protection and Food Safety, 2(S1), 47–49.

Albajes R, Lumbierres B, Pons X and Comas J, 2013. Representative taxa in field trials for environmental risk assessment of genetically modified maize. Bulletin of Entomological Research, 103, 724–733.

Andreadis SS, Alvarez-Alfageme FA, Sánchez-Ramos I, Stodola TJ, Andow DA, Milonas PG, Savopoulou-Soultani M and Castañera P, 2007. Frequency of resistance to *Bacillus thuringiensis* toxin Cry1Ab in Greek and Spanish population of *Sesamia nonagrioides* (Lepidoptera: Noctuidae). Journal of Economic Entomology, 100, 195–201.

Andow DA, 2008. The risk of resistance evolution in insects to transgenic insecticidal crops. Collection of Biosafety Reviews, 4, 142–199.

Andow DA, Olson DM, Hellmich RL, Alstad DN and Hutchinson WD, 2000. Frequency of resistance alleles to *Bacillus thuringiensis* toxin in an Iowa population of European corn borer. Journal of Economic Entomology, 93, 26–30.

Bates SL, Zhao J-Z, Roush RT, Shelton AM, 2005. Insect resistance management in GM crops: past, present and future. Nature Biotechnology, 25, 57–62.

Bourguet D, Chaufaux J, Séguin M, Buisson C, Hinton JL, Stodola TJ, Porter P, Cronholm G, Buschman LL and Andow DA, 2003. Frequency of alleles conferring resistance to Bt maize in French and US corn belt populations of the European corn borer, *Ostrinia nubilalis*. Theoretical and Applied Genetics, 106, 1225–1233.

Butko P, 2003. Cytolytic toxin Cyt1A and its mechanism of membrane damage: Data and hypotheses. Applied and Environmental Microbiology, 69, 2415–2422.

Campagne P, Kruger M, Pasquet R, Le Ru B and Van den Berg J, 2013. Dominant inheritance of field-evolved resistance to Bt corn in *Busseola fusca*. PloS ONE, 8, e69675.

Chaufaux J, Séguin M, Swanson JJ, Bourguet D and Siegfried BD, 2001. Chronic exposure of the European maize borer (Lepidoptera: Crambidae) to Cry1Ab *Bacillus thuringiensis* toxin. Journal of Economic Entomology, 94, 1564–1570.

Comas C, Lumbierres B, Pons X and Albajes R, 2014. No effects of *Bacillus thuringiensis* maize on nontarget organisms in the field in southern Europe: a meta-analysis of 26 arthropod taxa. Transgenic Research, 23, 135–143.

Crespo ALB, Spencer TA, Alves AP, Hellmich RL, Blankenship EE, Magalhães LC and Siegfried BD, 2009. On-plant survival and inheritance of resistance to Cry1Ab toxin from *Bacillus thuringiensis* in a field-derived strain of European corn borer, *Ostrinia nubilalis*. Pest Management Science, 65, 1071–1081.

Crespo ALB, Spencer TA, Tan SY and Siegfried BD, 2010. Fitness costs of Cry1Ab resistance in a field-derived strain of *Ostrinia nubilalis* (Lepidoptera: Crambidae). Journal of Economic Entomology, 103, 1386–1393.

Devos Y, Meihls LN, Kiss J and Hibbard BN, 2013. Resistance evolution to the first generation of genetically modified *Diabrotica*-active *Bt*-maize events by western corn rootworm: management and monitoring considerations. Transgenic Research, 22, 269–299.

Devos Y, Aguilera J, Diveki Z, Gomes A, Liu Y, Paoletti C, du Jardin P, Herman L, Perry JN and Waigmann E, 2014. EFSA's scientific activities and achievements on the risk assessment of genetically modified organisms (GMOs) during its first decade of existence—looking back and ahead. Transgenic Research, 23, 1–25.

EC (European Commission), 1998. Commission Decision of 22 April 1998 concerning the placing on the market of genetically modified maize (*Zea mays* L. line MON 810), pursuant to Council Directive 90/220/EEC (98/294/EC). Official Journal, L131, 32–33.

EFSA (European Food Safety Authority), 2004. Opinion of the Scientific Panel on Genetically Modified Organisms on a request from the Commission related to the Austrian invoke of Article 23 of Directive 2001/18/EC. The EFSA Journal, 78, 1–13.

EFSA (European Food Safety Authority), 2005. Opinion of the Scientific Panel on Genetically Modified Organisms on a request from the Commission related to the safeguard clause invoked by Hungary according to Article 23 of Directive 2001/18/EC. The EFSA Journal, 228, 1–14.

EFSA (European Food Safety Authority), 2006a. Opinion of the Scientific Panel on Genetically Modified Organisms on a request from the Commission related to genetically modified crops (Bt176 maize, MON 810 maize, T25 maize, Topas 19/2 oilseed rape and Ms1xRf1 oilseed rape) subject to safeguard clauses invoked according to Article 16 of Directive 90/220/EEC. The EFSA Journal, 338, 1–15.

EFSA (European Food Safety Authority), 2006b. Opinion of the Scientific Panel on Genetically Modified Organisms on a request from the Commission related to the safeguard clause invoked by Greece according to Article 23 of Directive 2001/18/EC and to Article 18 of Directive 2002/53/EC. The EFSA Journal, 411, 1–26.

EFSA (European Food Safety Authority), 2008a. Request from the European Commission related to the safeguard clause invoked by Hungary on maize MON 810 according to Article 23 of Directive 2001/18/EC. The EFSA Journal, 756, 1–18.

EFSA (European Food Safety Authority), 2008b. Request from the European Commission related to the safeguard clause invoked by Greece on maize MON 810 according to Article 23 of Directive 2001/18/EC. The EFSA Journal, 757, 1–12.

EFSA (European Food Safety Authority), 2008c. Scientific Opinion of the Panel on Genetically Modified Organisms related to a request from the European Commission related to the safeguard clause invoked by France on maize MON 810 according to Article 23 of Directive 2001/18/EC and the emergency measure according to Article 34 of Regulation (EC) No 1829/2003. The EFSA Journal, 850, 1–46.

EFSA (European Food Safety Authority), 2008d. Scientific Opinion of the Panel on Genetically Modified Organisms on a request from the European Commission related to the safeguard clause invoked by Austria on maize MON 810 and T25 according to Article 23 of Directive 2001/18/EC. The EFSA Journal, 891, 1–64.

EFSA (European Food Safety Authority), 2009. Scientific Opinion of the Panel on Genetically Modified Organisms on applications (EFSA-GMO-RX-MON 810) for the renewal of authorisation for the continued marketing of (1) existing food and food ingredients produced from genetically modified insect resistant maize MON 810; (2) feed consisting of and/or containing maize MON 810, including the use of seed for cultivation; and of (3) food and feed additives, and feed materials produced from maize MON 810, all under Regulation (EC) No 1829/2003 from Monsanto. The EFSA Journal, 1149, 1–84.

EFSA (European Food Safety Authority), 2014a. Statement on a request from the European Commission related to the emergency measure notified by Greece on genetically modified maize MON 810 according to Article 18 of Directive 2002/53/EC. EFSA Journal 2014:12(6), 7 pp. doi:10.2903/j.efsa.2014.3732.

EFSA (European Food Safety Authority), 2014b. Request to review the scientific basis of two Testbiotech reports on maize 1507. Available online: http://registerofquestions.efsa.europa.eu/roqFrontend/questionLoader?question=EFSA-Q-2014-00192

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2011a. Scientific Opinion on the annual Post-Market Environmental Monitoring (PMEM) report from Monsanto Europe S.A. on the cultivation of genetically modified maize MON 810 in 2009. EFSA Journal 2011:9(10), 66 pp. doi:10.2903/j.efsa.2011.2376.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2011b. Scientific Opinion updating the evaluation of the environmental risk assessment and risk management recommendations on insect resistant genetically modified maize 1507 for cultivation. EFSA Journal 9(12), 73 pp. doi:10.2903/j.efsa.2011.2429.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2011c. Statement supplementing the evaluation of the environmental risk assessment and risk management recommendations on insect resistant genetically modified maize Bt11 for cultivation. EFSA Journal 9(12), 45 pp. doi:10.2903/j.efsa.2011.2478.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2012a. Scientific Opinion on the annual Post-Market Environmental Monitoring (PMEM) report from Monsanto Europe S.A. on the cultivation of genetically modified maize MON 810 in 2010. EFSA Journal 10(3), 35 pp. doi:10.2903/j.efsa.2012.2610.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2012b. 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. EFSA Journal 10(5), 21 pp. doi:10.2903/j.efsa.2012.2705.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2012c. Scientific Opinion on a request from the European Commission related to the safeguard clause notified by Greece on genetically modified maize MON 810 according to Article 23 of Directive 2001/18/EC. EFSA Journal 10(9), 20 pp. doi:10.2903/j.efsa.2012.2877.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2012d. Scientific Opinion supplementing the conclusions of the environmental risk assessment and risk management recommendations for the cultivation of the genetically modified insect resistant maize Bt11 and MON 810. EFSA Journal 10(12), 32 pp. doi:10.2903/j.efsa.2012.3016.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2012e. Scientific Opinion updating the risk assessment conclusions and risk management recommendations on the genetically modified insect resistant maize MON 810. EFSA Journal 10(12), 98 pp. doi:10.2903/j.efsa.2012.3017.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2013a. Scientific Opinion on a request from the European Commission related to the emergency measure notified by Italy on genetically modified maize MON 810 according to Article 34 of Regulation (EC) No 1829/2003. EFSA Journal 11(9), 7 pp. doi:10.2903/j.efsa.2013.3371.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2013b. Scientific Opinion on a request from the European Commission related to the emergency measure notified by Luxembourg on genetically modified maize MON 810 according to Article 34 of Regulation (EC) No 1829/2003. EFSA Journal 11(9), 7 pp. doi:10.2903/j.efsa.2013.3372.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2013c. Scientific Opinion on the annual Post-Market Environmental Monitoring (PMEM) report from Monsanto Europe S.A. on the cultivation of genetically modified maize MON 810 in 2011. EFSA Journal 11(12), 38 pp. doi:10.2903/j.efsa.2013.3500.

EFSA Panel on Genetically Modified Organisms (GMO Panel), 2014. Scientific Opinion on the annual post-market environmental monitoring (PMEM) report from Monsanto Europe S.A. on the cultivation of genetically modified maize MON 810 in 2012. EFSA Journal 12(6), 29 pp. doi:10.2903/j.efsa.2014.3704.

Farinós GP, de la Poza M, Hernández-Crespo P, Ortego F and Castañera P, 2004. Resistance monitoring of field populations of the corn borers *Sesamia nonagrioides* and *Ostrinia nubilalis* after 5 years of Bt maize cultivation in Spain. Entomologia Experimentalis et Applicata, 110, 23–30.

Farinós GP, Andreadis SS, de la Poza M, Mironidis GK, Ortego F, Savopoulou-Soultani M and Castañera P, 2011. Comparative assessment of the field-susceptibility of *Sesamia nonagrioides* to the Cry1Ab toxin in areas with different adoption rates of Bt maize and in Bt-free areas. Crop Protection, 30, 902–906.

Gaviria Rivera AM, Granum PE and Priest FG, 2000. Common occurrence of enterotoxin genes and enterotoxicity in *Bacillus thuringiensis*. FEMS Microbiology Letters, 190, 151–155.

Glaser JA and Matten SR, 2003. Sustainability of insect resistance management strategies for transgenic Bt corn. Biotechnology Advances, 22, 45–69.

Gould F, 1998. Sustainability of transgenic insecticidal cultivars: integrating pest genetics and ecology. Annual Review of Entomology, 43, 701–726.

HCB (Haut Conseil des Biotechnologies), 2013. Commentaires sur le rapport de surveillance de culture du MON 810 en 2012. Available online: http://www.hautconseildesbiotechnologies.fr/IMG/pdf/131108_Surveillance_mais_MON810_2012_Commentaires_CS_HCB.pdf

Head GP and Greenplate J, 2012. The design and implementation of insect resistance management programs for Bt crops. GM Crops & Food: Biotechnology in Agriculture and the Food Chain, 3, 144–153.

Huang F, Andow AA and Buschman LL, 2011. Success of the high-dose/refuge resistance management strategy after 15 years of Bt crop use in North America. Entomologia Experimentalis et Applicata, 140, 1–16.

Kruger M, Van Rensburg JBJ and Van den Berg J, 2009. Perspective on the development of stem borer resistance to Bt maize and refuge compliance at the Vaalharts irrigation scheme in South Africa. Crop Protection, 28, 684–689.

Kruger M, Van Rensburg JBJ and Van den Berg J, 2011. Resistance to Bt maize in *Busseola fusca* (Lepidoptera: Noctuidae) from Vaalharts, South Africa. Environmental Entomology, 40, 477–483.

Kruger M, Van Rensburg JBJ and Van den Berg J, 2012. Transgenic Bt maize: farmers' perceptions, refuge compliance and reports of stem borer resistance in South Africa. Journal of Applied Entomology, 136, 38–50.

MacIntosh SC, 2010. Managing the risk of insect resistance to transgenic insect control traits: practical approaches in local environments. Pest Management Science, 66, 100–106.

Marvier M, McCreedy C, Regetz J and Kareiva P, 2007. A meta-analysis of effects of Bt cotton and maize on nontarget invertebrates. Science, 316, 1475–1477.

Matten SR, Head GP and Quemada HD, 2008. How governmental regulation can help or hinder the integration of Bt crops into IPM programs? In: Integration of insect-resistant genetically modified crops within IPM programs. Eds Romeis J, Shelton AM, Kennedy GG. Springer Science + Business Media BV, Amsterdam, The Netherlands, 27–39.

Mezzomo BP, Miranda-Vilela AL, Freire IdS, Barbosa LCP, Portilho FA, Lacava ZGM and Grisolia CK, 2013. Hematotoxicity of *Bacillus thuringiensis* as spore-crystal strains Cry1Aa, Cry1Ab, Cry1Ac or Cry2Aa in Swiss albino mice. Journal of Hematology & Thromboembolic Diseases, 1, 104.

Moar W, Roush R, Shelton A, Ferré J, MacIntosh S, Leonard BR and Abel C, 2008. Field-evolved resistance to *Bt* toxins. Nature Biotechnology, 26, 1072–1074.

Monsanto, 2013. Annual monitoring report on the cultivation of MON 810 in 2012; Czech Republic, Portugal, Romania, Slovakia, and Spain. Available online: http://ec.europa.eu/food/plant/gmo/reports_studies/docs/report_2012_mon_810/report_2012_mon_810_en.pdf

Naranjo SE, 2009. Impacts of *Bt* crops on non-target invertebrates and insecticide use patterns. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources, 4, 1–23.

Peterson JA, Lundgren JG, Harwood JD, 2011. Interactions of transgenic *Bacillus thuringiensis* insecticidal crops with spiders (Araneae). Journal of Arachnology, 39, 1–21.

Perry JN, Devos Y, Arpaia S, Bartsch D, Gathmann A, Hails RS, Kiss J, Lheureux K, Manachini B, Mestdagh S, Neemann G, Ortego F, Schiemann J and Sweet JB, 2010. A mathematical model of exposure of non-target Lepidoptera to Bt-maize pollen expressing Cry1Ab within Europe. Proceedings of the Royal Society B: Biological Sciences, 277, 1417–1425.

Perry JN, Devos Y, Arpaia S, Bartsch D, Gathmann A, Hails RS, Kiss J, Lheureux K, Manachini B, Mestdagh S, Neemann G, Ortego F, Schiemann J and Sweet JB, 2011. The usefulness of a mathematical model of exposure for environmental risk assessment. Proceedings of the Royal Society B: Biological Sciences, 278, 982–984.

Perry JN, Devos Y, Arpaia S, Bartsch B, Ehlert C, Gathmann A, Hails RS, Hendriksen NB, Kiss J, Messéan A, Mestdagh S, Neemann G, Nuti M, Sweet JB, Tebbe CC, 2012. Estimating the effects of Cry1F *Bt*-maize pollen on non-target Lepidoptera using a mathematical model of exposure. Journal of Applied Ecology, 49, 29–37.

Saeglitz C, Bartsch D, Eber S, Gathmann A, Priesnitz KU and Schuphan I, 2006. Monitoring the Cry1Ab susceptibility of European corn borer in Germany. Journal of Economic Entomology, 99, 1768–1773.

Siegfried BD and Hellmich RL, 2012. Understanding successful resistance management: The European corn borer and Bt corn in the United States. GM Crops & Food: Biotechnology in Agriculture and the Food Chain, 3, 184–193.

Siegfried BD and Spencer T, 2012. Bt resistance monitoring in European corn borer and western corn rootworms. In: Gene containment. Eds Oliver M and Li Y. John Wiley & Sons, New York, NY, USA, 43–55.

Siegfried BD, Spencer T, Crespo AL, Storer NP, Head GP, Owens ED and Guyer D, 2007. Ten years of Bt resistance monitoring in the European corn borer: what we know, what we don't know, and what we can do better. American Entomologist, 53, 208–214.

Siegfried BD, Rangasamy M, Wang H, Spencer T, Haridas CV, Tenhumberg B, Sumerford DV and Storer NP, 2013. Estimating the frequency of Cry1f resistance in field populations of the European corn borer (Lepidoptera: Crambidae). Pest Management Science, 70, 725–733.

Siqueira HAA, Moellenbeck D, Spencer T and Siegfried BD, 2004. Cross-resistance of Cry1Ab-selected *Ostrinia nubilalis* (Lepidoptera: Crambidae) to *Bacillus thuringiensis* δ-endotoxins. Journal of Economic Entomology, 97, 1049–1057.

Stodola TJ, Andow DA, Hyden AR, Hinton JL, Roark JJ, Buschman LL, Porter P and Cronholm GB, 2006. Frequency of resistance to *Bacillus thuringiensis* toxin Cry1Ab in southern United States corn belt population of European corn borer (Lepidoptera: Crambidae). Journal of Economic Entomology, 99, 502–507.

Storer NP, Babcock JM, Schlenz M, Meade T, Thompson GD, Bing JW and Huckaba RM, 2010. Discovery and characterization of field resistance to Bt maize: *Spodoptera frugiperla* (Lepidoptera: Noctuidae) in Puerto Rico. Journal of Economic Entomology, 103, 1031–1038.

Storer NP, Kubiszak ME, King EJ, Thompson GD and Santos AC, 2012. Status of resistance to Bt maize in *Spodoptera frugiperda*: Lessons from Puerto Rico. Journal of Invertebrate Pathology, 110, 294–300.

Tabashnik BE, 2008. Delaying insect resistance to transgenic crops. Proceedings of the National Academy of Sciences of the USA, 105, 19029–19030.

Tabashnik BE, Gassmann AJ, Crowder DW and Carrière Y, 2008a. Insect resistance to *Bt* crops: evidence versus theory? Nature Biotechnology, 26, 199–202.

Tabashnik BE, Gassmann AJ, Crowder DW and Carrière Y, 2008b. Reply to Field-evolved resistance to *Bt* toxins. Nature Biotechnology, 26, 1074–1076.

Tabashnik BE, Van Rensburg JBJ and Carrière Y, 2009. Field-evolved insect resistance to *Bt* crops: definition, theory and data. Journal of Economic Entomology, 102, 2011–2025.

Tabashnik BE, Brévault T and Carrière Y, 2013. Insect resistance to Bt crops: lessons from the first billion acres. Nature Biotechnology, 31, 510–521.

Van den Berg J, Hilbeck A and Bøhn T, 2013. Pest resistance to Cry1Ab Bt maize: field resistance, contributing factors and lessons from South Africa. Crop Protection, 54, 154–160.

van Rensburg JBJ, 2007. First report of field resistance by the stem borer, *Busseola fusca* (Fuller) to Bt-transgenic maize. South African Journal of Plant and Soil, 24, 147–151.

Wolfenbarger LL, Naranjo SE, Lundgren JG, Bitzer RJ and Watrud LS, 2008. Bt crop effects on functional guilds of non-target arthropods: a meta-analysis. PLoS ONE, 3, e2118.

Zhou J, Xiao K, Wei B, Wang Z, Tian Y, Tian Y and Song Q, 2014. Bioaccumulation of Cry1Ab protein from an herbivore reduces anti-oxidant enzyme activities in two spider species. PLoS ONE, 9, e84724.