Cotton GHB614 x T304-40 x GHB119

Organisation: The European GMO-free Citizens (De Gentechvrije Burgers)
Country: The Netherlands
Type: Others...

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a. Assessment:
b. Food Safety Assessment:
Toxicology

The EFSA has issued a scientific opinion in which it states: “The GMO Panel concludes that cotton GHB614 9 T304-40 9 GHB119, as described in this application, is as safe as its comparator and the tested non-GM reference varieties with respect to potential effects on human and animal health and the environment.”

How can they make such a claim? The toxicity of Roundup and glyphosinate-ammonium has been confirmed on several occasions by independent researchers. This cotton has been made resistant to it, and to a particular BT variety. BT isn't entirely innocent, either! One example: When the first GM soy beans which had been made resistant to the weedkiller Roundup arrived at the Port of Rotterdam in 1996, hardly any feeding tests and no food tests whatever were performed. The various accompanying reports are from Monsanto, which did not release them until 2000. And yet, as far back as 1995, a then Member of the British Parliament, Ms Angela Browning, had given Roundup the green light on behalf of the Ministry of Agriculture and Fisheries, which meant that the product could then be marketed throughout the EU. This is why we do not trust the reports issued by Monsanto and other multinationals. They are not independent. We now have sufficient knowledge of Roundup and glyphosate to say that they are carcinogenic and genotoxic, yet still you authorise GM crops which have been made resistant to them. Incomprehensible. See: https://www.sciencedirect.com/science/article/pii/S0278691518304800?via%3Dihub "The mechanism of DNA damage induced by Roundup 360 PLUS, glyphosate and AMPA in human peripheral blood mononuclear cells - genotoxic risk assessment panelEwelinaWoźniaka et al

https://doi.org/10.1016/j.fct.2018.07.035

"EPA emails show agency approved Monsanto herbicide label changes after consulting with company http://investigatemidwest.org/2018/07/25/lawsuit-epa-unlawfully-approved-monsantos-herbicide. Exposure to environmentally relevant doses of a glyphosate-based herbicide during pregnancy has been found not only to impair female fertility in rats, but to induce foetal growth retardation and malformations, including abnormally developed limbs, in their second-generation offspring. Perinatal exposure to a glyphosate-based herbicide impairs female reproductive outcomes and induces second-generation adverse effects in Wistar rats" • Authors and affiliations • María M. Milesi et al First Online: 09 June 2018 https://link.springer.com/article/10.1007/s00204-018-2236-6

Then there is the court case that is currently under way in San Francisco against Monsanto´s Roundup, in which the EFSA is cited: Quote: "Monsanto Lawyer Clashes With Cancer Expert in Roundup Trial July 13, 2018 HELEN CHRISTOPHI

Portier accused EFSA of failing to follow its own guidelines for evaluating herbicides, which he said state that if two positive animal tests are observed, the chemical in question must be classified as a possible carcinogen. https://www.courthousenews.com/monsanto-lawyer-clashes-with-cancer-expert-in-roundup-trial/ Day 4, July 13: Dr. Christopher Portier discusses flaws with U.S. and EU regulatory Evaluations for glyphosate. On day 4, Monsanto counsel, Kirby Griffis, tried to rattle Dr. Christopher Portier during cross-examination, confronting the expert witness with the EPA's conclusion that glyphosate is not likely to be carcinogenic to humans. During testimony, Dr. Portier slammed U.S. and European regulators over their methodology in evaluating glyphosate. For example, Dr. Portier testified that the EFSA missed 15 tumors in a series of rodent studies on glyphosate because the agency used the wrong methodology. During cross-examination, he said: "My entire career (has) been about using scientific evidence to make decisions, primarily about the carcinogenicity of compounds, and we've worked for years and years to do that appropriately. This was just so amazingly wrong in the way they were doing it."
https://www.organicconsumers.org/blog/kennedy-monsanto-roundup-trial-cancer-scientists
Van : Day 10: Secret Documents Reveal Monsanto's War on Cancer Scientists

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4. Conclusions and recommendations

Stichting Ekopark and the European GMO-free Citizens do not want this GM cotton to come onto the market in the EU. We will do everything in our power to stop it. This statement is issued jointly on behalf of Stichting Ekopark, Lelystad (NL).

Organisation: The European GMO-free Citizens (De Gentechvrije Burgers)
Country: The Netherlands
Type: Others...

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a. Assessment:
b. Food Safety Assessment:
Toxicology

Follow-up to our previous comments on the same GM cotton.

A new study raises new questions about the safety of GM Bt toxin.

The study performed in mice found that the GM Bt toxin Cry1Ac is immunogenic, allergenic, is able to induce anaphylaxis (a severe allergic response that can result in suffocation) and

causes pre-cancerous intestinal changes. The responses that Cry1Ac was found to produce in the mice included "mildly allergic manifestations" around the mouth, nose and ears, as well as wheezing, hair standing on end, and diarrhoea. Study of the allergenic potential of Bacillus thuringiensis Cry1Ac toxin following intra-gastric administration in a murine model of food-allergy Santos-Vigil, K. I., Ilhuicatzi-Alvarado, D., García-Hernández, A. L., Herrera-García, J.S., & Moreno-Fierros, L. (2018). International Immunopharmacology, 61, 185-196. https://www.sciencedirect.com/science/article/pii/S1567576918302467

In the words of the GM Bt crop developer Monsanto, the GM Bt toxins in GM crops were especially engineered to be "super toxins" because they have "broad spectrum activity". In contrast, natural Bt toxin affects only certain types of insect pests and degrades rapidly in daylight, so non-target organisms and human consumers are unlikely to be exposed. https://www.gmwatch.org/en/news/latest-news/18399

Ms Eijsten

I would now like to let Ms Eijsten have her say (by quoting a few fragments). She is an environmentally conscious citizen who has more than 80 complaints and appeals against GM market authorisations and/or GM field trials to her name, together with Mr J. van der Meulen, a chemical literature researcher. As long ago as 2001, she wrote: ..... "With regard to Bt. kurstaki, applicators suffer considerable misery if they get some of it on their face. The case of a scientist who accidentally injected himself with Bt. Israelensis and another kind of bacteria commonly found on human skin is interesting."

How considerate of Oregon Health Division to suggest, before a Bt.k. spraying programme, that "individuals with ... physician-diagnosed causes of severe immune disorders may consider leaving the area during the actual spraying."

"The 1991 Material Safety Data Sheet for Foray 48B states: "Repeated exposure via inhalation can result in sensitization and allergic response in hypersensitive individuals."

"Inert Ingredients. All Bt products contain ingredients other than Bt. These are identified only as "inert" ingredients and are called trade secrets by the manufacturers of the products. The "inert" ingredients are potentially the most toxic components of the formulations". Examples below.

Because "inerts" are called trade secrets, …. there is little public information …. available [that] indicates that they could cause health problems*" [meaning not entirely clear – translator]*. And then there are sodium hydroxide, sulfuric acid, phosphoric acid, methyl paraben and potassium phosphate as "inerts". These account for less than 10% of Foray 48B, but "they pose hazards". There follows a list of harmful effects: mild cases to irritation of the mucous membranes of the nose; damage of the upper respiratory tract; corrosive; severe deep skin burns, permanent loss of vision; severe bronchial constriction, and bronchitis; irritant to skin and mucous membranes; throat irritation; both methyl paraben and potassium phosphate were once registered by EPA as pesticide active ingredients. Sodium sulphite (inert) in Dipel 8AF: Up to ten per cent of asthmatics (about one million people in the United States) may react to sulphites, particularly those who are treated with steroids. Symptoms of exposure in those sensitive to sulphites usually involve the respiratory system, and can also include nausea, diarrhoea, lowered blood pressure, hives, shock, and loss of consciousness." The list goes on.

Enough of this doom and gloom. But just one more thing: formulations of Bt.i. are extremely unhealthy because the "inerts" in the product “deplete the dissolved oxygen in water”. The Bt.i formulation Teknar was “acutely toxic to brook trout fry, probably because of xylene used as "inert" in the product”.

There is so much literature on these Bt and other pesticides and formulations and their effects that it leaves me with a bad taste in my mouth." End of quotes. Amsterdam, 31 October 2001, Ms L. Eijsten; reproduced with permission.

https://www.gentechvrij.nl/dossiers/archief-lily-eijsten/wat-voor-smaak-heeft-een-allergeen-vraagt-het-voedingscentrum-zich-af/ Over Bt cotton:

An estimated 290,000 Indian farmers have committed suicide in 20 years. Small farms used to be the country’s economic backbone, but now owners struggle to make even a meagre profit and are drowning in debt. For some, the pressure is too much. Many blame GMO cotton for the failing farms. https://www.youtube.com/watch?v=XlYzd8bM9xg

Monsanto Case, 10 August 2018

After the verdict in the Monsanto´s Roundup case we said on 11 August in a tweet that was read more than 5,500 times: “And now the #EU must see to it that all imported GM crops are no longer wanted as food and feed.” The European GMO-free Citizens, Lelystad, The Netherlands. http://www.gentechvrij.nl

https://www.gentechvrij.nl/2018/08/11/monsanto-verliest-miljoenen-vanwege-roundupzaak/ More on the American jury's findings concerning Roundup and glyphosate:

On August 10, Monsanto was found guilty on all counts in the first of many cases connecting Roundup’s main ingredient, glyphosate, to cancer. The jury awarded Dewayne “Lee” Johnson, a Bay Area school district groundskeeper, $289 million. Quote from: Institute for Responsible Technology.

Monsanto loses millions in Roundup case

On 10 August, a jury in California came to the unanimous conclusion that Monsanto had failed to warn consumers about the dangers of Roundup. The chemicals giant, which was recently taken over by Bayer, was ordered to pay millions to a school groundskeeper, Mr Dewayne Johnson, whose tasks included spraying the liquid on the school playground. A burst pipe meant that he came into bodily contact with a large quantity of Roundup. Mr Johnson developed a rare form of cancer and does not have long to live. Despite this, he took up the fight with the help of the lawyers The Miller Firm, LLC, Baum, Hedlund, Aristei & Goldman PC, Audet & Partners, LLP and Kennedy & Madonna (in which Robert Kennedy Jr. is a partner). Monsanto is to appeal the decision.

See also: 11 August 2018. The Trouw newspaper: "Chemiereus Monsanto moet kankerpatiënt 289 miljoen betalen" ("Chemicals giant Monsanto ordered to pay cancer patient US$ 289 million"): https://www.trouw.nl/home/chemiereus-monsanto-moet-kankerpatient-289-miljoen-betalen-~ab0405ca/

Moms Across America say:

Johnson, a school pesticide applicator with non-Hodgkins Lymphoma, WINS his lawsuit against Monsanto! The jury (God bless them!!!) awarded Johnson 289.2 million dollars! 4,000 other plaintiffs are waiting to sue Monsanto and 100,000 are expected by the end of the year. Surely this decision has Bayer, who now owns Monsanto, reconsidering whether or not to continue to sell glyphosate-based products. https://twitter.com/yesmaam74

BBC News, 11 Aug 2018. Monsanto ordered to pay $289m damages in Roundup cancer trial. https://www.bbc.co.uk/news/world-us-canada-45152546

11 August 2018, 04:52. Süddeutsche Zeitung: Glyphosat. Monsanto muss 285 Millionen Dollar Schmerzensgeld zahlen ("Glyphosate: Monsanto ordered to pay US$ 285 million in compensatrion"). https://www.sueddeutsche.de/wirtschaft/glyphosat-monsanto-in-den-usa-zu-millionen-dollar-strafe-verurteilt-1.4089739

LE MONDE | 11.08.2018 à 00h56 • Updated on 11.08.2018 à 10h24 | By Stéphane Foucart : Glyphosate trial: Monsanto fined in historic judgment. The agrochemicals giant has been ordered to pay $ 289 million to Dewayne Johnson. The complaint lodged by the groundskeeper, who is a cancer sufferer, is the first to be examined by an American court. https://www.lemonde.fr/planete/article/2018/08/11/proces-du-glyphosate-monsanto-condamne-a-verser-289-millions-de-dollars-a-un-jardinier_5341423_3244.html

Lavanguardia, Efe, Los Ángeles: Monsanto ordered to pay US$ 289 million to cancer-stricken groundskeeper

11/08/2018 01:37 Updated on 11/08/2018 12:17. The Superior Court of San Francisco rules that Monsanto failed to warn of the health risk associated with the use of the product, which contains glyphosate, a carcinogenic substance. https://www.lavanguardia.com/economia/20180811/451281329745/monsanto-pagara-289-millones-dolares-jardinero-dewayne-johnson-efectos-cancerigenos-glifosato-roundup.html?utm_source=Twitter&utm_medium=Social

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Allergenicity

Ms Eijsten:

Dangers to human health as a result of the modification are certainly present, and in no small way. The claim that “no harmful effects are expected” is old hat! The fact that substances from the formulation of pesticides via the mouth, skin, respiratory organs and enter the body (i.e. via food, among other things) is attested by a veritable mountain of literature and these substances in the formulation make the pesticide more of a threat to health than the so-called "active substance" alone.

Enclosed with this letter you will find a number of discussions of the problems associated with PAT, written by Mr H. v.d. Meulen.

* H. v.d. Meulen: ** Two studies which give rise to two diametrically opposed conclusions (Thompson versus Schulz); ** “Achtergrond”("Background") ** “Chemische reacties”

("Chemical reactions") ** “De 5 Substraten van PAT” ("The Five Substrates of PAT")** Commentary on a research article by A. Wehrmann ** Opmerkingen in verband met de ziekte v Crohn ("Comments on Crohn's disease").

Study performed by Hoechst (Dr Arno Schulz) into the substrates of phosphinothricin acetyltransferase (PAT). Amsterdam, 7 November 1999. Two studies which gave rise to diametrically opposed conclusions, i.e. Charles J. Thompson, 1987: Characterization of the herbicide-resistance gene bar from Streptomyces hygroscopicus: Dr Arno Schulz, 1993: L-Phosphinothricin N-Acetyl-transferase -Biochemical Characterization - a report included in Wehrmann 1996 (Schulz is the co-author). The subject is the characterization of the enzyme phosphinothricin acetyltransferase PAT, particularly the specificity of the substrates.

The first study concerns the reaction between phosphinothricin and acetyl co-enzyme A under the influence of the PAT enzyme, and compares it with a number of structural analogues of PPT phosphinothricin. One of the analogues was L-glutamate. The products of the reaction were identified using mass spectography, and the affinity was determined. In addition to phosphinothricin (PPT), a number of structural analogues were tested for the presence of an acetylerin reaction. L-Glutamine acid was one of the tested substances. With respect to PPT, the affinity of most of the substances was low: one substance did not react during the test, and a reaction occurred which gave rise to an identified product (the detection limit is not the subject of dispute) that can be reported on using hard figures; it would seem to be beyond doubt that glutamine acid is a substrate of PAT. The second study concerns the reaction of a large number of amino acids, including L-glutamine acid, which appeared in the first study, in a reaction mix with 100% excess PPT compared with the acetyl source acetyl co-enzyme A and PAT. Products of the reaction were identified using chromatography. Even with a very large excess of L-amino acid, no products of the reaction with the amino acids were detected. Only acetyl phosphinothricin was detected.

The authors concluded that the only substrate of PAT is PPT. The following contradicts this conclusion, which itself is incompatible with the results of the first study (incidentally, the first study is cited in the literature of the second study): no detection limit was determined for acetylized L-glutamine acid. The possibility of acetylized glutamine acid being an acetyl source for the acetylization of PPT was not considered.

This could have been done in the study by adding acetylized glutamine acid to the reaction mix in a quantity which was above the detection limit and determining whether the added quantity disappeared in the course of the reaction. Given the results of the first test, its disappearance is a foregone conclusion!! A reaction mix was used in which there was a large excess of a competing substrate, PPT. No observations were made with pure amino acids.

A discussion of the findings of the first study, focusing in particular on why the findings were different, is totally absent. Essentially, the authors of the second study accuse the authors of the first study of fabrication and fraud (the first study contains a treasure of numerical data; the second contains none). The second study does not take this aspect into sufficient account.

The background to the conclusion that PAT has only one substrate - PTT - is as follows: PAT, a GM product, occurs in herbicide (PPT)-resistant crops. In order for this GM product to be given market authorisation, its toxicity must first be determined. Could this GM product react with the contents of our gut, e.g. with the important amino acid L-glutamine acid? It would

take a tonne of research money to downplay it. Total denial seems to be HOECHST's preferred strategy!

We believe that the conclusion drawn from the second study is totally unfounded and that the study itself does not deserve the name. It is incompetent, and the people who cite it need to have that incompetence pointed out to them. J. van der Meulen, L. Eijsten. https://www.gentechvrij.nl/dossiers/archief-lily-eijsten/onderzoek-van-hoechst-dr-arno-schulz-betreffende-de-substraten-van-phosphinothricinacetyltransferasepat/ (Reproduced with permission).

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5. Others

This GM cotton must not be allowed on the market! GM cotton which is already on the market must be labelled to the effect that it is genetically modified. In this way we will know which cotton wool, cotton buds, bandages, tampons, materials, clothing, etc. to leave in the shops. Edible products must also be labelled! Moreover, GLA will be banned and Roundup will soon follow!

Organisation: Testbiotech
Country: Germany
Type: Non Profit Organisation

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a. Assessment:
Molecular characterisation

Besides resistance to glyphosate, the plants are doubled in the genetic condition that confers resistance to glufosinate. This causes a higher amount of the PAT enzyme to be produced in the plants.

The cotton produces two truncated and chimeric versions of Bt toxins that do not exist naturally. No detailed consideration was undertaken regarding the extent to which the truncation of the Bt proteins will change its biological characteristics. The DNA sequences used for the expression of these proteins have not been made public, although this information is very relevant for the risk assessment of the genetically engineered cotton.

Further, the insertion of the constructs creates several new open reading frames. EFSA did not assess unintended gene products, such as miRNA, that can emerge from the insertion of the transgenes.

In order to enable further independent risk assessment, the full DNA sequence inserted into the plants should be made available, including all open reading frames.

EFSA (2018a) did not request any detailed analysis based on so-called -omics (transcriptomics, metabolomics, proteomics) to investigate changes in the overall metabolism in the plants. EFSA assumed that the data from phenotypic characteristics and compositional analysis would not indicate any need for further investigations. However, these data did show many significant changes (see below). In general, data on phenotypic characteristics and compositional analysis can be used as complementary data, but these are not as sensitive as -omics data and cannot replace them.

It is known (Christ et al. 2017) that the PAT/bar acts upon plant endogenous amino acids leading to ectopic accumulation of two metabolites. This effect was overlooked for more than 20 years despite many relevant plants being risk assessed by EFSA. As EFSA (2018b) states, the GMO panel was not aware of the finding at the time of previous risk assessments. However, it is now at least aware of these findings and that metabolomic studies should have been requested. Such studies would be especially relevant in this case because the stacked cotton shows an increased expression of the bar gene.

Expression data provided on the newly produced proteins indicate higher rates for application of the complementary herbicides (EFSA 2018c). According to the expert opinion of Member States (EFSA 2018c), this pattern of gene expression indicates an effect of herbicide application in combination with the stacked event. Therefore, the EFSA conclusion that no indications for combinatorial effects were observed in the plants is not correct.

Furthermore, it is known that the Bt content in the plants depends on environmental impact. For example, environmental stress can cause unexpected patterns of expression in the newly introduced DNA (see, for example, Trtikova et al., 2015; Adamczyk & Meredith 2004). Therefore, the plants should have been subjected to a much broader range of defined environmental conditions and stressors in order to gather reliable data on gene expression and functional genetic stability.

Further, the method used to determine the amount of Bt toxins (ELISA) is known to be dependent on the specific protocols used. The data are not sufficiently reliable without further evaluation by independent labs. For example, Shu et al. (2018) highlight difficulties in measuring the correct concentration of Bt toxins produced by the genetically engineered plants (see also Székács et al., 2011). Without fully evaluated test methods to measure the expression and the concentration of the Bt toxins, risk assessment will suffer from substantial methodological gaps.

Consequently, the risk assessment of molecular characteristics is not conclusive and is not sufficient to show food and feed safety.

Adamczyk Jr, J.J., & Meredith Jr, W. R. (2004) Genetic basis for variability of Cry1Ac expression among commercial transgenic Bacillus thuringiensis (Bt) cotton cultivars in the United States. Journal of Cotton Science, 8(1): 433-440.

Christ, B., Weng, J. K., Guyer, L., Hochstrasser, R., Francisco, R., Hörtensteiner, S., & Aubry, S. (2017). Non-specific activities of the major herbicide-resistance gene BAR. Nature plants, 1. https://www.nature.com/articles/s41477-017-0061-1

EFSA GMO Panel (2018a). Scientific Opinion on the assessment of genetically modified cotton GHB614 9 T304-40 9 GHB119 for food and feed uses, import and processing under

Regulation (EC) No 1829/2003 (application EFSA-GMO-NL-2014-122). EFSA Journal 2018;16(7):5349, 32 pp. https://doi.org/10.2903/j.efsa.2018.5349

EFSA (2018b) Public comments on genetically modified oilseed rape Ms8, Rf3 and Ms8 x RF3 (….) Ref BU/GdS/EW/FA/SM/cb – OC-2018-19139196, accessed via the register of EFSA

EFSA GMO Panel (2018c). Comments form the experts of Member States on Scientific Opinion on the assessment of genetically modified cotton GHB614 9 T304-40 9 GHB119 for food and feed uses, import and processing under Regulation (EC) No 1829/2003 (application EFSA-GMO-NL-2014-122). EFSA Journal 2018;16(7):5349, 32 pp. accessed via the register of EFSA.

Shu, Y., Romeis, J., Meissle, M. (2018) No interactions of stacked Bt maize with the non-target aphid Rhopalosiphum padi and the spider mite Tetranychus urticae. Front. Plant Sci. 9: 39. doi: 10.3389/fpls.2018.00039

Székács, A., Weiss, G., Quist, D., Takács, E., Darvas, B., Meier, M., Swain, T., Hilbeck, A., (2011) Interlaboratory comparison of Cry1Ab toxin quantification in MON 810 maize by ezyme-immunoassay. Food and Agricultural Immunology, 23(2): 99-121.

Trtikova, M., Wikmark, O.G., Zemp, N., Widmer, A., Hilbeck, A. (2015) Transgene expression and Bt protein content in transgenic Bt maize (MON810) under optimal and stressful environmental conditions. PloS One, 10(4): e0123011.

Comparative analysis (for compositional analysis and agronomic traits and GM phenotype)

Field trials were only performed in the US, at eight registered sites for only one growing season (2012), and not in any other cotton producing regions. The parental plants were not grown in parallel, thus no direct comparison can be made that would allow assessment of effects due to the process of stacking.

Around one third of the 27 parameters measured for the phenotype were found to be significantly different in comparison to the conventional plants. Some of them belong to categories III/IV which indicate major differences.

In plant composition, more than half of the 53 parameters measured were significantly different (more than 30) in comparison to the conventional plants. Again, some of them belong to categories III/IV which indicate major differences.

Taken as isolated data these differences might not directly raise safety concerns, nevertheless, the large number of effects should have led to further investigations.

Therefore, EFSA should have requested further studies e.g. > data from omics (proteomics, transcriptomics, metabolomics), > data representing more extreme environmental conditions such as those caused by climate change, > data representing more areas of commercial cotton

cultivation, > more data on stress reactions under controlled conditions > and the impact of the dosage of the complementary herbicide that was sprayed, as well as the number of times it was sprayed onto the plants under practical conditions.

Instead, EFSA (2018a) has relied solely on the newly introduced statistical method known as the "test of equivalence". This method can be helpful to make some assumptions on the relevance of the significant findings. However, it cannot replace a detailed assessment of the high number of significant differences.

Based on the available data, no final conclusions can be drawn on the safety of the plants.

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b. Food Safety Assessment:
Toxicology

Bayer presented data from 90-day feeding trials undertaken solely with the three parental plants. These studies suffer from methodological weaknesses. Further, in each case, a considerable number of significant effects were shown to occur in the rats. Taking into account the uncertainties from the molecular assessment and the data from composition analysis and phenotypical characteristics, it is obvious that further studies with the stacked plants should have been requested. These additional feeding studies are also necessary to assess potential combinatorial effects between stressors produced in the plants (such as the Bt proteins) and the residues from spraying the complementary herbicides (see also Then & Bauer-Panskus, 2017).

Furthermore, because truncated and synthetic versions of the Bt proteins are produced in the plants, food safety of a combination of these toxins would require a detailed investigation.

There are further relevant issues e.g. the potential impact on the intestinal microbiome also needs to be considered. Such effects might be caused by the residues from spraying since glyphosate has been shown to have negative effects on the composition of the intestinal flora of cattle (Reuter et al., 2007) poultry (Shehata et al., 2013) and rats (Mao et al., 2018). Further, Bremmer and Leist (1997) examined the possible conversion of NAG to glufosinate in rats. In general, antibiotic effects and other adverse health effects might occur from exposure to a diet containing these plants that were not assessed under pesticide regulation. However, these adverse effects on health might be triggered by the residues from spraying with the complementary herbicide (see also van Bruggen et al., 2017).

In addition, as far as the exposure of the food chain to Bt toxins is concerned, EFSA should have requested data on the overall combined exposure to Bt toxins caused by the introduction of Bt plants into the EU. Currently, there are already 30 events that produce Bt toxins authorised for import. The exposure stemming from these imports should have been added to that of the stacked cotton.

Consequently, the toxicological assessment carried out by EFSA is not sufficient to show food and feed safety.

Bremmer, J.N. and Leist, K.-H. (1997) Disodium-N-acetyl-L-glufosinate; AE F099730 – Hazard evaluation of Lglufosinate produced intestinally from N-acetyl-L-glufosinate. Hoechst Schering AgrEvo GmbH, Safety Evaluation Frankfurt. TOX97/014. A58659. Unpublished.

Mao, Q., Manservisi, F., Panzacchi, S., Mandrioli, D., Menghetti, I., Vornoli, A., Bua, L., Falcioni, L., Lesseur, C., Chen, J., Belpoggi, F., Hu, J. (2018) The Ramazzini Institute 13-week pilot study on glyphosate and Roundup administered at human-equivalent dose to Sprague Dawley rats: effects on the microbiome, Environmental Health, 17:50. https://doi.org/10.1186/s12940-018-0394-x

Reuter, T, Alexander, T.W., Martinez, T.F., McAllister, T.A. (2007) The effect of glyphosate on digestion and horizontal gene transfer during in vitro ruminal fermentation of genetically modified canola. Journal of the Science of Food and Agriculture, 87(15), 2837-2843. https://onlinelibrary.wiley.com/doi/abs/10.1002/jsfa.3038

Shehata, A.A., Schrödl, W., Aldin, A.A., Hafez, H.M., Krüger, M. (2012) The effect of glyphosate on potential pathogens and beneficial members of poultry microbiota in vitro. Curr Microbiol, 6(4): 350-358. https://link.springer.com/article/10.1007/s00284-012-0277-2

Then, C., & Bauer-Panskus, A. (2017) Possible health impacts of Bt toxins and residues from spraying with complementary herbicides in genetically engineered soybeans and risk assessment as performed by the European Food Safety Authority EFSA. Environmental Sciences Europe, 29(1): 1. https://enveurope.springeropen.com/articles/10.1186/s12302-016-0099-0

Van Bruggen, A.H.C., He, M.M., Shin, K., Mai, V., Jeong, K. C., Finckh, M.R., Morris, J.G. (2018) Environmental and health effects of the herbicide glyphosate. Science of The Total Environment, 616: 255-268. https://www.sciencedirect.com/science/article/pii/S0048969717330279

Allergenicity

Bt toxins are known to be immunogenic. They seem to act as allergens and adjuvant effects are likely to occur. In regard to immunogenicity (non-IgE-mediated immune adverse reactions), it is generally acknowledged that Bt toxins are immunogenic (Rubio-Infante & Moreno-Fierros, 2016; Adel-Patient et.al., 2011; Andreassen et.al., 2015a,b; Andreassen et.al., 2016; see also Then & Bauer-Panskus, 2017). Thus, there are some substantial reasons for concern that reactions to allergens can be enhanced (see also EFSA 2018d, minority opinion). This is relevant since in food/feed the Bt toxins can be mixed with allergens from soybeans, amongst others. Mixing with soybeans can also substantially prolong the degradation of the Bt toxins in the gastric system (Pardo-López et al., 2009).

New findings (Santos-Vigil et al., 2018) indicate allergenic potential of Cry toxins after intra-gastric administration in a murine model. Thus, the EFSA assumption that a detailed assessment of the allergenic potential of Cry toxins is not necessary, is simply wrong.

Consequently, the assessment on allergenicity cannot be regarded as conclusive.

Adel-Patient, K., Guimaraes, V.D., Paris, A., Drumare, M.F., Ah-Leung, S., Lamourette, P., ... & Créminon, C. (2011) Immunological and metabolomic impacts of administration of Cry1Ab protein and MON 810 maize in mouse. PloS one, 6(1): e16346. http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0016346

Andreassen, M., Rocca, E., Bøhn, T., Wikmark, O.G., van den Berg, J., Løvik, M., ... & Nygaard, U. C. (2015a) Humoral and cellular immune responses in mice after airway administration of Bacillus thuringiensis Cry1Ab and MON810 cry1Ab-transgenic maize. Food and agricultural immunology, 26(4): 521-537. http://www.tandfonline.com/doi/abs/10.1080/09540105.2014.988128

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Others

According to Regulation (EU) No 503/2013, the applicant has to ensure that post-market monitoring is developed to collect reliable information with respect to the detection of indications of whether any (adverse) effects on health may be related to genetically modified food or feed consumption. Some experts from Member States (EFSA 2018b) have made appropriate demands regarding the implementation this obligation. Accordingly, the monitoring report should deliver detailed information on: i) actual volumes of the cotton imported into the EU, ii) the ports and silos where shipments of the cotton being unloaded, iii) the processing plants where the cotton was transferred to, iv) the amount of the stacked cotton as used on farms for feed, and v) transport routes of the stacked cotton.

The applicant is further requested to explain how the PMM of the stacked cotton in mixed GMO commodities imported, processed or used for food/feed would be put into practice. Since traders may co-mingle the stacked cotton with other imported commercial genetically engineered cotton that is processed or used for food/feed, the applicant is requested to explain how the monitoring will be designed to distinguish between potential adverse effects caused by stacked cotton and those caused by other genetically engineered cotton, such as parental plants.

The monitoring should be run in regions where the stacked cotton will be transported, stored, packaged, processed or used for food/feed. In case of substantial losses and spread of the stacked cotton, all receiving environments need to be monitored.

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3. Environmental risk assessment

EFSA (2018a) acknowledges that the stacked cotton seeds can give rise to volunteer plants that might persist for some time in the environment, especially in the Mediterranean region. To assess the environmental risks conferred by these genetically engineered offspring plants, experimental data are necessary. It is known that next generation effects can emerge in genetically engineered plants that are not present in the original plants (see, for example, Lu and Yang, 2009; Zhang et al., 2018). Furthermore, plants with additional expression of the EPSPS enzyme are known to show a higher fitness even if no glyphosate is applied (Fang et al., 2018).

Lu, B.-R., Yang, C., 2009. Gene flow from genetically modified rice to its wild relatives: Assessing potential ecological consequences. Biotechnol. Adv., Biotechnology for the Sustainability of Human SocietyInvited Papers from IBS 2008 27, 1083–1091. https://doi.org/10.1016/j.biotechadv.2009.05.018

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Consequently, environmental risk assessment carried out by EFSA is not acceptable.

4. Conclusions and recommendations

The EFSA risk assessment should be rejected.

When making his decision the risk manager should also take into account issues that are related to pesticide regulation. In this case, glufosinate-ammonium is about to be prohibited in the European Union.