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Testbiotech comment on EFSA Scientific Opinion on an application by Dow AgroSciences (EFSA-GMO-NL-2013-116) for placing on the market of genetically modified insect-resistant soybean DAS-81419-2 for food and feed uses, import and processing under Regulation (EC) No 1829/2003

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Introduction

The genetically engineered soybean DAS-81419-2 produces two synthetically derived Bt toxins (Cry1Ac (synpro) and Cry1Fa2). In addition, the soybeans are engineered to be resistant to the herbicide glufosinate.

1. Molecular characterisation

There are unintended structural changes in the genome: One of the gene constructs responsible for the production of the Bt toxin shows rearrangements. An additional short fragment is inserted and the genome of the soybean shows a deletion of 59 bp. There are 9 new open reading frames (ORF) in the flanking regions and several hundred ORFs within the gene constructs inserted (FSANZ 2014). Gene products from the additional open reading frames were assessed in regard to translation into potential proteins. But no assessment was made of any other gene products such as miRNA. Thus, uncertainties remain about other biologically active substances emerging from the method of genetic engineering.

The additional DNA added to the sequences of the Cry1Ac and Cry1F DNA is not meant to change its toxicity, but nevertheless the biological functions of the proteins might be changed. Further, compared to their native templates, the toxins in the plant are truncated and activated. But the protein assessment carried out by EFSA (2016a) does not address these details.

The expression of the toxin was only measured under field conditions in the US. It is unclear to which extent specific environmental conditions can influence the overall concentration of the toxins in the plants. The plants should have been subjected to a much broader range of environmental conditions to obtain reliable data on gene expression and functional genetic stability. Environmental stress can also cause unexpected patterns of expression in the newly introduced DNA (see Trtikova et al., 2015).

In addition, more varieties should have been included into the field trials since it is known that the genetic background of the varieties can influence the level of gene expression (see Trtikova et al., 2015).

Further, all parts of the plants should be taken into account for risk assessment. Expression data have to be considered as one of the starting points in the risk assessment of the plant, so the assessment of the data cannot be reduced to those parts of the plants entering the food chain.

2. Comparative analysis (for compositional analysis and agronomic traits and GM phenotype)
The significant changes observed were set aside without more detailed investigations of underlying mechanisms, evaluation of tendencies in the data and more targeted experiments (see EFSA, 2016b). No data from Omics (proteomics, transcriptomics, metabolomics) were used to assist the compositional analysis and the assessment of the phenotypical changes. The additional reference varieties used for the statistical assessment were chosen without sufficient reasoning. Consequently, it cannot be ruled out that data noise may be masking biologically relevant effects. As a result, the comparative analysis suffers from many uncertainties and remains inconclusive.

There are further flaws in the generation of the data:

- Despite South America being one of the most important regions for the production of soybeans, no data have been requested from environments representing these regions.
- No data representing more extreme environmental conditions, such as those caused by climate change, were generated.
- In addition, more varieties should have been included into the field trials to see how the gene constructs interact with the genetic background of the plants.
- Furthermore, data from soybeans sprayed with the complementary herbicide should have been requested. While DowAgro Sciences claims that they will not encourage farmers to spray glufosinate during cultivation, there is no reason why farmers in North and South America should not apply glufosinate in response to aggravated pressure from glyphosate-resistant weeds. Thus, it has to be expected that the imported soybeans will to a great extent contain residues from spraying, and might show changes in composition due to the application of the herbicide.
- Finally, the choice of the components used for the assessment followed an outdated version of OECD Guidelines from 2001 instead of those from 2012.

Based on the available data, no final conclusions can be drawn on changes in the composition of the plants.

Toxicology
There are several gaps in the risk assessment:

- Despite it being known that Bt toxins can cause effects in several different ways, only one mode of action was considered (for overview: Hilbeck & Otto, 2015).
- Despite it being known that Bt toxins can show synergies with each other and as well as with other compounds, no detailed investigation of combinatorial effects were conducted (for overview: Then, 2010).
- There are no reliable data to assess the exposure of the food chain to Bt toxins. Soybeans can be processed in a broad range of products by using various methods for heating, germinating etc.
- Interaction with plant components (such as protease inhibitors) that can delay the degradation of the Bt toxins, were not taken into account (Pardo-López et al., 2009).
- No testing of the whole plant (feeding studies) was requested even though there were still several uncertainties after the comparative assessment and the molecular analysis.

As a result, the toxicological assessment carried out by EFSA is not acceptable.

Allergenicity

There are several relevant issues regarding allergenicity and the immune system that were left aside in EFSA risk assessment.

- A range of studies found indications of adjuvant effects triggered by Bt toxins. EFSA simply referred to its earlier opinion on genetically engineered cotton 281-24-236 x 3006-201-23. This opinion from 2010 only mentions two older studies and does not take more recent findings into account (for overview see: Rubio-Infante N. & Moreno-Fierros L., 2015). Further, compared to cotton, soybeans show a much higher content of potentially allergenic proteins. Therefore, adjuvant effects have to be considered much more carefully.
- The sera samples used for assessment are very low in number and there are substantial uncertainties about the outcome (see, for example, the comment made by a Belgian expert, EFSA 2016 b).
- The assessment did not take into account the risk to more vulnerable groups of people, such as infants.

Overall, the assessment is insufficient to rule out impacts on the immune system.

Others

Monitoring should be case specific. Exact data on exposure to the soybean should be made available. Possible health impacts have to be monitored in detail. Controls regarding residues from spraying with glufosinate have to be established. Accumulated effects that might stem from mixtures with other genetically engineered plants have to be taken into account in the monitoring plan.

Conclusions and recommendations

The risk assessment undertaken by EFSA should not be accepted. It does not identify knowledge gaps or uncertainties and fails to assess toxicity, impact on the immune system and the reproductive system. The monitoring plan has to be rejected because it will not make the necessary data available.

References:

EFSA (2016a) Scientific Opinion on an application by Dow AgroSciences (EFSA-GMO-NL-2013-116) for placing on the market of genetically modified insect-resistant soybean DAS-81419-2 for food and feed uses, import and processing under Regulation (EC) No 1829/2003. EFSA Journal 2016;14(12):4642, 23 pp.

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