GenØk - Centre for Biosafety

Vår ref:2012/h90
Deres ref:2012/6286 ART-BI-DHT

Assessment of the technical dossier submitted under EFSA/GMO/BE/2011/90 for approval of transgenic maize, MON89034 from Bayer CropScience AG

Submitted to

Direktoratet for Naturforvaltning

by

David Quist
Centre for Biosafety – GenØk
July 2012

SUMMARY OF THE ASSESSMENT OF THE TECHNICAL DOSSIER RELATED TO EFSA/GMO/BE/2011/90

As a designated National Competence Center for Biosafety, our mission at GenØk in advice giving is to provide independent, holistic and useful analysis of technical and scientific information/reasoning in order to assist authorities in the safety evaluation of biotechnologies proposed for use in the public sphere.

The following information is respectfully submitted for consideration in the evaluation of product safety and corresponding impact assessment of MON89034, setting out the risk of adverse effects on the environment and health, including other consequences of proposed release under the pertinent Norwegian regulations.

This submission is structured to address specific provisions for an impact assessment required under the Norwegian Gene Technology Act of April 1993, focusing on the requirements in Appendix 2 - Principles for environmental risk assessment pursuant to sections 13-16 of the regulations, and Appendix 4 - Evaluation of ethical considerations, sustainability and benefit to society, cf section 17 of the "Regulations relating to impact assessment pursuant to the Gene Technology Act" of December 2005, pursuant to section 11 cf section 8. The information presented here may be applicable to more than one provision in different appendices.

We have targeted our critique to address the information needs under the relevant provisions that relate to our particular area of competence in biotechnology assessment as comprehensively as possible. Lack of commentary on our part towards any information under consideration should not be interpreted as specific endorsement of that information.

This submission was built in large part using the **Biosafety Assessment Tool** (https://bat.genok.org/bat/) produced by the University of Canterbury and GenØk – Centre for Biosafety. This is a free-to-the-public resource for hazard identification and risk assessment of genetically modified organisms.

All page numbers following quoted text that is not directly referenced refers to the technical dossier "Application for authorization of MON 89034 maize for cultivation in the European Union, according to Regulation (EC) No 1829/2003 on genetically modified food and feed", submitted by the Applicant.

Lastly, Codex Alimentarius guidelines allow Norway to ask for specific data of the type we identify and recommend obtaining. Norway therefore may request such information without concern of a challenge from the World Trade Organisation.

Specific recommendations

Based on our findings, we propose a few specific recommendations, summarized here and detailed in the critique below. This application is for cultivation of a GM event that has

already undergone an assessment for use as food and feed (please see dossier details and outcomes of regulatory assessment for case EFSA/GMO/NL/2007/37). The present application utilizes much of the information from this previous application. We wish to refer you to earlier assessments and critiques produced by member states, Bioteknologinemda, and the VKM (see attachments) on many aspects of the safety assessment.

Given these prior evaluations are readily available, we will focus our critique on supplementary information provided by the applicant to compliment the information submitted under EFSA/GMO/NL/2007/37 (see the technical dossier submitted with EFSA/GMO/NL/2007/37). Our evaluations includes new information from D.2.(b) - Molecular structure at the insertion site and in particular those that are related potential adverse effects that may follow from an environmental release, including sections D3 expression levels (exposure), and D11 "Environmental monitoring plan".

On the basis of our assessment, the Direktoratet for naturforvaltning is encouraged to request the following:

1. The Applicant should reperform the analysis using more sensitive methods (greater E-score sensitivities and smaller search sequence lengths) as well all relevant maize databases that maximize the likelihood of finding a accurate nucleotide match.

2. The Applicant should report the nucleotide sequences obtained from the endogenous maize DNA regions adjacent to the transgene insertion(s) so that independent analysis can be performed.

3. The Applicant should provide information demonstrating the genetic similarity of comparators used in compartive assessments in accordance with Regulation (EC) No 1829/2003 and Directive 2001/18/EC.

4. The Applicant should provide a case-specific monitoring plan to monitor potential unintended but anticipated exposure routes and levels, and to verify the assessment of exposure routes and levels into the environment.

5. The Applicant should provide greater details on the methods, locations and local considerations that should be identified for the establishment of baseline data.

6. The Applicant should describe how the monitoring report will review and evaluate the effectiveness, relevance, efficiency, and scientific quality of data derived from monitoring, including the continuity of the monitoring activities as it was described in the monitoring plan. Any unusal observations or identified adverse effects that is identified should be reported in a timely manner so that the appropriate response may be undertaken. These reports should also include a scientifically rigorous analysis of the results and conclusions, also considering site-specific conditions. The report should further highlight results that indicate adaptation of the monitoring plan, further research or review of risk management options or decisions.

7. The Applicant should also specficy how thereport will provide information on the practical experience from the monitoring and suggest the ways the plan may be revised as needed, as specified by the Competant Authority, and implemented by the Applicant. These may include adaptation of the monitoring plan, the establishment and/or adaptation of risk management measures, or the initation of new investigations or more in depth studies (in the case where followup studies are needed, how they should be designed and who should be responsible for their implementation should be decided by the Competent Authority, in accordance with the monitoring provisions adopted by the Party of Import).

8. The Applicant should indicate how monitoring reports could be made available on a central, openly accessible storage and presentation interface (e.g. a publically available website, housed by the Compentant Authority) so that it may be more broadly disseminated (including for public awareness and participation). Raw data should be stored by the Applicant and made available for independent review of the data, its interpretation, and conclusons drawn from the monitoring activities. Reporting should also be disseminated, as determined in the monitoring plan, via GMO registers establishd by the Competent Authority and other public databases.

9. The Applicant should submit required information on the social utility of MON89034 and its contribution to sustainable development, in accordance with the Norwegian Gene Technology Act.

Overall recommendation

From our analysis, we find that the deficiencies in the dossier do not support claims of safe use, social utility and contribution to sustainable development of MON89034. **Critically, the Applicant has not included any of the required information to assess social utility and sustainability as required in Appendix 4 of the Norwegian Gene Technology Act, which would be necessary for consideration of approval in Norway.** Hence at minimum, the dossier is deficient in information required under Norwegian law. A new application or reapplication should only be reconsidered with the delivery of the information requests recommended here, including any additional information deemed significant by the Norwegian authorities.

Therefore, in our assessment of MON89034, we conclude that based on the available data, including the safety data and monitoring plans supplied by the Applicant, the Applicant has not substantiated claims of environmental safety satisfactorily or provide the required information under Norwegian law to warrant approval in Norway at this time.

ASSESSMENT OF THE TECHNICAL DOSSIER RELATED TO EFSA/GMO/BE/2011/90

About the event

The transgenic maize event MON89034, developed by Monsanto Company, was developed via Agrobacterium-mediated transformation of two T-DNAs: One to confer insect tolerance through the expression of *Bt* insecticidal proteins Cry1A.105 and Cry2Ab2, lepidopteran insecticidal toxins derived from the soil bacterium, *Bacillus thuringiensis* and *Bacillus thuringiensis* subsp. Kurstaki, respectively, where both target sequences have undergone sequence modification optimized expression and do not exist in nature. And a second T-DNA containing the *nptII* gene encoding neomycin phosphotransferase II.

Assessment findings

As noted above, we will focus our critique on supplementary information provided by the applicant to compliment the information submitted under EFSA/GMO/NL/2007/37 (see the technical dossier submitted with EFSA/GMO/NL/2007/37). Our evaluations includes new information from D.2.(b) - Molecular structure at the insertion site and in particular those that are related potential adverse effects that may follow from an environmental release, including sections D3 expression levels (exposure), and D11 "Environmental monitoring plan" (D11).

1. New information contained in "Section D.2.(b) - Molecular structure at the insertion site"

The applicant reports a negative finding of homolgous sequences within the queried database using selected cutoff criteria. However, based on our assessment, the applicant has failed to produce a satisfactory analysis of the endogenous maize sequences bordering the insertion event(s) because of a conservative search methodology the minimizes the likelihood of finding actual and accurate matches their queries.

Details of their effort is reported confidentially as an amendment to the dossier p. 67 (please see "Responses to EFSA letter Ref. SR/SM/eb (2007)-2303901"), despite providing a sequence analysis that claims the were no matches to the endogenous maize border sequences adjacent to the insert, that applicant should perform searches using databases, methods, and reporting criteria that maximize the potential to identify and characterize junction sequences. For example, the Blast maize database (http://blast.maizegdb.org/home.php?a=BLAST_UI) or restricting the search to the maize genome would increase the likelihood of finding a match or improve the E-value of the hits (making finding a match more likely).

For example, from the information given, it appears the applicant performed a "nBlast" search of 3012 bases and report no results. However, the criteria of E-score cut off of 1 would

minimize the likelihood of reporting a sequence match. Furthermore, by searching smaller fragments, a greater likelihood of sequence match with the database would be obtained.

Lastly, the nucleotide sequence of the adjacent endogenous border maize regions should be made available for independent analysis.

Recommendation: The applicant should reperform the analysis using more sensitive methods (greater E-score sensitivities and smaller search sequence lengths) as well all relevant maize databases that maximize the likelihood of finding a accurate nucleotide match.

Recommendation: The applicant should report the nucleotide sequences obtained from the endogenous maize DNA regions adjacent to the transgene insertion(s) so that independent analysis can be performed.

2. Information contained in ”D7.2. Production of material for comparative assessment” (p. 159)

In accordance with Regulation (EC) No 1829/2003 and Directive 2001/18/EC the use of (near) isogenic comparators in comparative assessments should be performed. However no information is provided by the applicant to demonstrate its compliance with these directives, made possible by presenting data on the level genetic or varietial similarity of comparators. This critique may be applied more generally within other comparative assessements that utlize reference lines, for example the nutritional comparative study using broiler chickens reported in 7.10.2. Nutritional assessment of GM feed on page 224.

Recommendation: The applicant should provide information demonstrating the genetic similarity of comparators used in compartive assessments in accordance with Regulation (EC) No 1829/2003 and Directive 2001/18/EC .

3. Information provided in “D11. Environmental monitoring plan”

In the development of a monitoring plan, an important aspect to keep into focus is what monitoring methodologies and plans will generated added value to the objective. Experience from environmental monitoring has indicated that unless all of the the details of the monitoring methdology, hypothesis formulation, data quality, and statistical power are well described from the start, the monitoring will not likely produce information that is meaningful and useful to the risk assessment. The liability here is that there will be the illusion that something useful has been done when in reality the monitoroing methodology implemented could not have actually identified meaningful changes in the first place. Perpetuation of faulty monitoring programs that fail on the basis of design rather than the potential for identifying changes give credence to the argument that monitoring does not produce meaningful results is not a efficient use of resources. Specific critiques and recommendations are given below.

11.3.Case-specific GM plant monitoring

The case-specific monitoring (CSM) plan focuses on strategies for instect-resistance management and evaluating their efficacy as an identified potential risk of insecticide resistance. However, CSM may also be utilized to verify the assumptions or conclusions regarding exposure of MON89034 or its products in the environment. This includes a) accidental spillage or exposure during transport, storage, or processing or other approved uses, b) persistence and accumulation of Cry1A.105 and Cry2Ab2 into the environment, and c) transgene expression under varying environments.

Recommendation: The applicant should provide a case-specific monitoring plan to monitor potential unintended but anticipated exposure routes and levels, and to verify the assessment of exposure routes and levels into the environment.

11.4.General surveillance for unanticipated adverse effects

In accordance Directive 2001/18/EC general surveillance is a compulsory. While the applicant gives some indications of their vision for a survellience plan that focuses on generation of qualitative data through farmer questionnaires. The applicant has not given sufficient specifications of parameter definitions, methods (including sampling), stastistical approach(es), baselines establishment, frequencies of observations, adaptability of monitoring plans to local conditions, external network use and integration to a level of detail to implement the monitoring activities and ensure the activity will produce meaningful results.

More specifically:

"The party placing the GM plant on the market will primarily consider general surveillance in the places where the GM plant is grown and monitor for any adverse effects of its cultivation at farm level." p. 288

Monitoring should be peformed in both the receiving environment and areas where MON89034 spread or exposure is likely. This includes protected areas and centres of crop origin and genetic diversity or ecologically sensistive regions with specific protection goals, including the use of buffer areas in order to detect unintended presence or unexpected effects.

"Statistical analysis has the task to identify effects within the monitoring characters and to establish whether the observed effect is due to the GM plant or to other

environmental and cultivation factors or random variation." p.289

It should be considered that other types of analysis or experiementation may be required to determine whether MON89034 would be the causal agent affecting an observed change. The applicant should describe processes to this end.

"The central tool is an annual farm questionnaire addressed to a subset of farmers cultivating MON 89034." p.231

Questionnaires may be useful to provide supplementary information to monitoring but not the central activity, as it lacks sufficient analytical rigor to support robust monitoring effort. When questionnaires are used, in some cases farm or land managers, and not the individual "in the field" workers themselves have the best overview or integrative knowledge on the parameters of interest, particularly for large farms and the first to identify changes. The applicant should consider the appropriaprite targets of questionnaires.

"It has to be noted that established routine surveillance networks (providing monitoring of agriculture, variety registration monitoring, plant protection, plant health and soil surveys and ecological/environmental monitoring) might provide useful data on background or baseline characters on a landscape or national level (e.g. climatic conditions, cultivation practices) or on monitoring characters (plant diseases or pests, invasiveness, weeds). The party placing the GM plant on the market may therefore consider to use information from this type of networks on an ad hoc basis (e.g. if a potential adverse effect is reported in a subset of questionnaires in a certain region) to assess whether this effect is associated with the GM plant or with another influencing factor (Figure 54). Networks for the agricultural and the non-agricultural environment as well as for human andlivestock health might be consulted, as appropriate." p. 294

We disagree. This kind of information should be considered criticial for establishing baselines data.

Determining a causal link between an adverse effect and one or more GMOs requires the relevant baseline be established or identified. This is only possible if the observed effect can be compared to the occurrence of the effect in receiving environments or indicators that have not been previously exposed to the GMO. While such baseline data may be readily available, it may also need to be generated before the introduction of the GMO or in parallell with a similar receiving enviornment that does not contain the GMO. This requires a description of scientifically robust methods for constructing baselines. This must specify the appropriate spatial scale and account for effects of spatial heterogeneity on the representativeness of the baseline in the areas to be compared.

"In addition to the above-mentioned general surveillance actions directed to MON 89034 growers, international traders, grain processors, users of maize grain, an other stakeholders, the party placing MON 89034 on the market will actively monitor existing information sources such as official websites, scientific publications and expert reports on GM crops in order to identify, collate and follow-up on potentially adverse observations made for this

maize or any other relevant information, in particular with respect to occupational health, animal feed safety or putative ecological effects of the release of this maize." p. 297

The details of how this would become harmonized and integrated into the GS survellience plan needs to be specified by the applicant.

Recommendation: The applicant should provide greater details on the methods, locations and local considerations that should be identified for the establishment of baseline data, including specifics on how existing monitoriung networks would be utilized to generate data.

11.4.3.3.2 Number of farm questionnaires to be sampled

The use quantitative data and tranformation for statistical analysis as suggested by the applicant will not support a precautionary-based approach to analyzing potential adverse effects. For example, the applicant proposes to use sample sizes based on statistical power that is "high as possible" on the basis of Type I and Type II error both set at 0.01 for hypothesis testing. This seems to be difficult to achieve. The sample sizes necessary to support sufficient statistical power at the given thresholds appears insufficient. More realistic sampling is recommended under lower thresholds (greater acceptance) of posible Type II error to acheive greater statistical power.

11.5.Reporting the results of monitoring

The applicant suggests that reporting would come in the form of synthesis report to competant authorities. However, reporting of the data obtained and made available for independent analysis would strengthen the confidence and robustness, and transparency of the analysis.

Reporting is essential to provide not just results, but critical feedback its efficiency and efficacy towards meeting the stated objectives in the monitoring plan. Second, it can help make sure that results will support further assessment, changes to risk management or decisionmaking.

Recommendation:

The applicant should describe how the monitoring report will review and evaluate the effectiveness, relevance, efficiency, and scientific quality of data derived from monitoring, including the continuity of the monitoring activities as it was described in the monitoring plan. Any unusal observations or identified adverse effects that is identified should be reported in a timely manner so that the appropriate response may be undertaken. These reports should also include a scientifically rigorous analysis of the results and conclusions, also considering site-specific conditions. The report should further highlight results that indicate adaptation of the monitoring plan, further research or review of risk management options or decisions.

The applicant should also specficy how thereport will provide information on the practical experience from the monitoring and suggest the ways the plan may be revised as needed, as specified by the Competant Authority, and implemented by the Applicant. These may include adaptation of the monitoring plan, the establishment and/or adaptation of risk management measures, or the initation of new investigations or more in depth studies (in the case where followup studies are needed, how they should be designed and who should be responsible for their implementation should be decided by the Competent Authority, in accordance with the monitoring provisions adopted by the Party of Import).

The applicant should indicate how monitoring reports could be made available on a central, openly accessible storage and presentation interface (e.g. a publically available website, housed by the Compentant Authority) so that it may be more broadly disseminated (including for public awareness and participation). Raw data should be stored by the Applicant and made available for independent review of the data, its interpretation, and conclusons drawn from the monitoring activities. Reporting should also be disseminated, as determined in the monitoring plan, via GMO registers establishd by the Competent Authority and other public databases.

4. Social utility and sustainability aspects

In addition to the EU regulatory framework for GMO assessment, an impact assessment in Norway follows the Norwegian Gene Technology Act. In accordance with the aim of the Norwegian Gene Technology Act, production and use of the GMO shall take place in an ethically and socially justifiable way, under the principle of sustainable development. This is further elaborated in section 10 of the Act (approval), where it is stated that

> *"significant emphasis shall also be placed on whether the deliberate release represent a benefit to the community and a contribution to sustainable development"*.

These issues are further detailed in the regulation on consequence assessment section 17 and its annex 4. The Applicant has not provided relevant information that allows an evaluation of the issues laid down in the aim of the Act, regarding ethical values, social justification of the GMO within a sustainable development. Given this lack of necessary information for such an evaluation, the Applicant has not demonstrated a benefit to the community and a contribution to sustainable development from the use of MON89034. The Applicant should thereby provide the necessary data in order to conduct a thorough assessment on these issues, or the application should be refused.

It is also important to evaluate whether alternative options, (e.g. the parental non-GM version of MON89034) may achieve the same outcomes in a safer and ethically justified way.

Further, the Norwegian Gene Technology Act, with its clauses on societal utility and sustainable development, comes into play with a view also to health and environmental effects in other countries, such as where GMOs are grown. For instance, it is difficult to extrapolate on hazards or risks taken from data generated under different ecological,

biological, and genetic contexts as regional growing environments, scales of farm fields, crop management practices, genetic background, interactions between cultivated crops, and surrounding biodiversity are all likely to affect the outcomes. Hence it cannot be expected that the same effects will apply between different environments and across continents.

Recommendation: The Applicant should submit required information on the social utility of MON89034 and its contribution to sustainable development, in accordance with the Norwegian Gene Technology Act.

Conclusion

Available information for risk assessment evaluation

This evaluation is based on the Applicant's own submitted information, along with our own expertise in related fields. The relevant scientific literature is very limited in some cases, yet we have tried to extract information from the peer-reviewed literature that may inform the scientific validity of the information under consideration. In situations where lack of knowledge, complexity and uncertainty are high, particularly in relation to unknown adverse effects that may arise as a result of approval for release of a living modified organism into the environment or food supply, the available information may not be sufficient to warrant approval. Further information may address some of these issues, however an accurate description of uncertainties provided by the applicant would provide a more useful basis for assessing the level of risk that may come with regulatory approval of the GMO, taken on a case-by-case basis.

In all cases, product-related safety testing should have an independent and unbiased character. This goes both for the production of data for risk assessment, and for the evaluation of the data.

The lack of compelling or complete scientific information to support the claims of the Applicant documented here highlights the need for independent evaluation of the dossier as performed here, including the raw data produced by the Applicant. We therefore support better transparency and independent review of information to ensure high standards within the regulatory process. This would include any information provided by the Applicant used to justify confidentiality claims on any scientific data. We encourage the authorities to insist on this level of transparency and accessibility to all scientific data (including raw data) to ensure the scientific validity of the information presented.

Overall recommendation

Above we highlight a number of issues in relation to the questionable safe use of MON89034 that do not justify a conclusion of safe use, social utility and contribution to sustainable development. Critically, the Applicant's environmental monitoring plan lacks sufficient

details and descriptions to support the required monitoring activities, and has not included any of the required information to assess social utility and sustainability as required in Appendix 4 of the Norwegian Gene Technology Act, which would be necessary for consideration of approval in Norway. Taken together, these deficiencies fail to address the necessary safety regulations under Norwegian Law, and thus the application is incomplete and should not be approved. A new application or reapplication should only be reconsidered with the delivery of the information requests recommended here, including any additional information deemed significant by the Norwegian authorities.

Therefore, in our assessment of MON89034 we conclude that based on the available data, including the safety data supplied, the Applicant has not substantiated claims of safety satisfactorily to warrant approval in Norway at this time.