Vår ref:2015/H_94
Deres ref: 2015/9649

Miljødirektoratet
Postboks 5672 Sluppen
7485 Trondheim
Dato: 13.10.15

Vedlagt er innspill fra GenØk – Senter for Biosikkerhet på offentlig høring under EU forordning 1829/2003 av oppsummert søknad for **EFSA/GMO/NL/2011/94**, bomull «event» GHB614xLLCotton25xMON15985, fra Bayer CropScience AG som gjelder mat, fòr, import og prosessering av genmodifisert bomull **GHB614xLLCotton25xMON15985.**

Vennligst ta kontakt hvis det er noen spørsmål.

Med vennlig hilsen,

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Vår ref:2015/H_94
Deres ref: 2015/9649

Assessment of the summary of the dossier under 1829/2003/EU of EFSA/GMO/NL/2011/94 cotton.

Sent to

Norwegian Environment Agency

by

GenØk- Centre for Biosafety
October 2015

KONKLUSJON PÅ NORSK

Vi trekker frem mangler i oppsummert søknad og data som ikke gir grunnlag for en konklusjon om sikker bruk, samfunnsnytte og bidrag til bærekraft av **GHB614xLLCotton25xMON15985 bomull**. Søker har ikke inkludert noe av den informasjonen omkring samfunnsnytte og bærekraft av **GHB614xLLCotton25xMON15985 bomull** som kreves i den norske genteknologiloven (Appendix 4) for godkjenning i Norge.

Hovedkonklusjon og anbefalinger:
Genøk–Senter for Biosikkerhet viser til brev fra Miljødirektoratet angående offentlig høring for **GHB614xLLCotton25xMON15985 bomull** i bruksområdet import og prosessering og til bruk i fòr og mat eller inneholdende ingredienser produsert fra **GHB614xLLCotton25xMON15985 bomull.**

Søker gir ikke opplysninger som adresserer vurderingskriteriene bærekraft, samfunnsnytte og etiske aspekter som forutsettes anvendt i den norske genteknologiloven. I denne sammenheng er det viktig å få dokumentert erfaringer med hensyn på effekter på miljø, helse og samfunnsaspekter. Denne type dokumentasjon er ikke tilstrekkelig i oppsummert søknad om omsetting av **GHB614xLLCotton25xMON15985 bomull** til import og prosessering og til bruk i fòr og mat eller inneholdende ingredienser produsert fra **GHB614xLLCotton25xMON15985 bomull.**

Vår konklusjon er at norske myndigheter ikke godkjenner bruk av **GHB614xLLCotton25xMON15985 bomull** til import og prosessering og til bruk i fòr og mat som det søkes godkjenning for.

ASSESSMENT OF THE SUMMARY OF THE TECHNICAL DOSSIER UNDER 1829/2003 OF EFSA/GMO/NL/2011/94 COTTON.

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 event **GHB614xLLCotton25xMON15985 cotton**, setting out the risk of adverse effects on the environment and health, including other consequences of proposed release under the pertinent Norwegian regulations.

*As we do not have access to the full technical dossier of **GHB614xLLCotton25xMON15985 cotton** we can not give a full assessment of this stacked event.*
Our assessment is therefore based on the information from the summary of the technical dossier and the data presented there.

We have previously commented on sub-combinations/single events of **GHB614xLLCotton25xMON15985 cotton** in the following:

- EFSA/GMO/NL/2010/77, GHB614xLLCotton25 (March 2011)
- EFSA/GMO/ES/2012/104 for 1GHB614 (March 2013).

Specific recommendations

Based on our findings, we propose some specific recommendations, summarized here and detailed in the go-through below.

- The regulator is encouraged to make the full technical dossier of GHB614xLLCotton25xMON15985 cotton available to be able to perform a proper evaluation of the stacked event.
- The regulator is encouraged to ask the Applicant for an analysis of the genomic location of the inserted vectors.
- The regulator is encouraged to ask the Applicant to provide data from real control lines, isogenic lines, of cotton when it comes to comparison of protein levels.
- The regulator is encouraged to ask the applicant to address the potential of non-target effects of Bt toxins, especially in the context of their combined use in a stacked event.
- The regulator is encouraged to ask the Applicant to consider the possibility of cross resistance development to multiple Cry proteins due to the use of stacked events
- The regulator is encouraged to ask the Applicant to address what promoter is driving the expression of the ARM gene *aad*, as this is unclear from the summary.
- The regulator is encouraged to ask the Applicant to perform an analysis to verify the presence or absence of the *aad* protein.
- We find it ethically unacceptable to ban the use of glufosinate-ammonium based herbicides domestically due to health and environmental concerns, while supporting its use in other countries. This represents an unacceptable double standard for Norway, and we ask the regulators to reconsider the practice of separating health and environmental risk by national borders or regions.
- The applicant should include a full evaluation of the co-technology intended to be used with GHB614xLLCotton25xMON15985 cotton, namely glyphosate- and glufosinate-ammonium-based herbicides. Particular focus should be given to the level of accumulation of herbicides in the plants, particularly the parts used in food and feed production, and whether or not these levels of exposure could cause acute and/or chronic health issues. This needs to be tested in animal and feeding studies, separating the effects of the plant and the herbicide(s) by using both sprayed and unsprayed plant samples.
- In order to meet the requirements for the NGTA, the regulator is encouraged to ask the Applicant to submit information relevant for the assessment of the social utility of the GHB614xLLCotton25xMon15985 and its contribution to sustainable development. The information provided by the Applicant must be relevant for the agricultural context in the producing country. The information should include issues such as: Changes in pesticide use, emergence of herbicide resistant weeds, development of pest resistance in target populations, impacts on non-target organisms, potential for gene flow and possible impacts among farmers practicing different production forms for cotton cultivation in the producing country and share of the benefits among sectors of the society.

Overall recommendation

From our analysis, we find that the information provided in the summary of the technical dossier have deficiencies that do not support claims of safe use, social utility and contribution to sustainable development of **GHB614xLLCotton25xMON15985 cotton. 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.** A new application 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 **GHB614xLLCotton25xMON15985 cotton**, we conclude that based on the available data, the Applicant has not provided the required information under Norwegian law to warrant approval in Norway at this time.

ASSESSMENT OF THE SUMMARY OF THE TECHNICAL DOSSIER UNDER 1829/2003 OF EFSA/GMO/NL/2011/94 COTTON.

About the event

The event **GHB614xLLCotton25xMON15985** cotton was made by conventional crossing of lines containing the single events GHB614, LLCotton25 and MON15985.

The application of cotton event GHB614xLLCotton25xMON15985 is for food, feed, import and processing.

None of the parental lines is approved in Norway for any of the applications.

Cotton event GHB614xLLCotton25xMON15985 is not approved for food, feed, import or processing in EU. Parental lines and hybrid line GHB614xLLCotton25 are approved for this.

The cotton event GHB614xLLCotton25xMON15985 is cultivated in US. An authorisation for food, feed and industrial use is present in Australia, New Zealand, Canada, Japan, Korea and Mexico.

Assessment findings

The full technical dossier is not available for cotton event GHB614xLLCotton25xMON15985, thus we base our assessment on the summary provided by the Environmental Agency.

The event GHB614xLLCotton25xMON15985 (also referred to as *GTxLLXB2)* cotton is a stacked event that has tolerance to the herbicides glyphosate through the *2mEPSPS* gene and gluphosinate-ammonium through the *bar* gene. It also has resistance to certain lepidopteran insect pests through the Bt-toxins Cry1Ac and Cry2Ab2. In addition, it contains two antibiotic resistance marker (ARM) genes called *nptII* and *aad*. A visual marker called *uidA* (beta-D-glucuronidase) is also present.

Molecular characterization

Evaluation of the molecular characteristics of the GTxLLXB2 Cotton

The GTxLLxXB2 cotton plant is a crossbreed of three different GM plants. The GHB614 and LLCotton25 were produced by *Agrobacterium tumefaciens* mediated transformation. The MON15985 was produced particle acceleration transformation of MON 531 cotton which was previously modified via *Agrobacterium*-mediated transformation.

Concerns of the genomic location of the inserts

Both transformation procedures will insert the vector at random places in the plant genome. There has been performed Southern blot analysis, which confirms that the vectors are inserted in the genome, but there is no evidence in the summary of any investigation of where in the plant genome these inserts are. In addition, there are in total four independent transformation processes in the production of this plant, and this increases the need to establish where the inserts are in the plant genome, and whether they interfere with existing genes there.

Inadequate choice of control in the evaluation of molecular characteristics of the GTxLLXB2 Cotton plan

In the summary, the analyzed protein levels are compared to levels of protein in the plants that initially were used in the breeding of this stacked event. These are all GM plants, and therefore should not serve as a control of "normal" protein levels in the cotton plant.

In the investigation of protein levels and morphological features, the stacked plant should be compared to an isogenic cotton plant, and this should be the control for all risk assessments and analysis of this plant.

Recommendation:

- The regulator is encouraged to ask the Applicant for an analysis of the genomic location of the inserted vectors.
- The regulator is encouraged to ask the Applicant to provide data from real control lines, isogenic lines, of cotton when it comes to comparison of protein levels.
- The regulator is encouraged to ask the Applicant for the full dossier to be able to perform a full evaluation and comments on safety of the GTxLLXB2 Cotton plant.

Cry proteins
This stack contains two Bt-toxins, the Cry proteins Cry1Ac and Cry2Ab2, which have effects on certain Lepidopteran pests. They have also been analyzed for their potential non-target effects , as well as which organisms that has been used for testing and also their potential effects on biodiversity (Gilliand et al 2002, Crickmore 2005, Hilbeck and Schmidt 2006, Marvier et al 2007, Bøhn et al 2008, van Frankenhuyzen 2013).

Another issue is the potential acceleration in the development of resistance when multiple, targeted genes are used. This was the experience of Zhao et al (2005), who tested the effect of using broccoli plants containing Cry1Ac, Cry1C or both, on resistance development in a population of diamondback moths (*Plutella xylostella*).

Recommendation:

- The regulator is encouraged to ask the applicant address the potential of non-target effects of Bt toxins, especially in the context of their combined use in a stacked event.
- The regulator is encouraged to ask the Applicant to consider the possibility of cross resistance development to multiple Cry proteins due to the use of stacked events

Antiobiotic resistance marker (ARM) genes
The stacked event **GHB614xLLCotton25xMON15985 cotton** contains two ARM genes called *nptII* encoding neomycin phosphotransferase II (resistance to neomycin and kanamycin) and *aad* (resistance to spectinomycin and streptomycin) encoding 3''(9)-O-aminoglycoside adenylyltransferase.

In the summary of the technical application, only the expression of *nptII* is analysed in plant tissues and found expressed at a certain level. The protein level expressed from the other ARM gene *aad* is not mentioned and thus probably not analysed, but this remains unclear. Based on the summary of the dossier, we do not have an overview of whether the promoter driving its expression is active in plants or not or whether this is analysed.

NptII is often used as an ARM gene/selectable marker in GM plants and is considered to pose no risk to human or animal health or environment (EFSA, 2004, EFSA Journal 2009).This means that this gene has unlimited use.

The Norwegian Scientific Committee for Food Safety (VKM) has commented on health and environmental issues related to the use of ARM genes in GM plants (VKM, 2005) where they support the conclusion by EFSA (2009) but acknowledges that there is little information on the prevalence of such ARM genes in the environment and in Norway.

We support this view and strongly encourage the onset of ARM gene mapping in the environment for more knowledge on background level.

The Norwegian Genetechnology Act (NGTA) accounts for GMOs that are able to germinate, while the Food Act regulate import and production of processed, non-germinating products from GMOs for food and feed. Through NGTA, the principles of social utility, ethical considerations and sustainable development are evaluated for each GMO before approval, thus

many GMOs with ARM genes are prohibited because they are not considered as not contributing to these principles. The regulations in the Food Act prohibits food and feed that contains GMOs that are detectable in the end product.

By this, the stacked event **GHB614xLLCotton25xMON15985 cotton** is not allowed for food, feed, import or processing in Norway if it contains detectable levels of the GMs involved.

Many GM plants containing *nptII* have been approved for field trails and for marketing in several countries (Carrer et al 1993, Badosa et al 2004, Breyer et al 2014). The choice for using this marker gene has been driven by the fact that kanamycin is considered as "not important" in medical treatments and that kanamycin resistant bacteria are ubiquitous in nature (EFSA Journal 2009). However, kanamycin has recently been classified as a critically important antibiotic for human and animals (WHO, 2007). Literature survey indicates that only few data are available on the prevalence of *nptII* gene. The limited data demonstrates that there is a low level of presence of *nptII* in naturally occurring bacterial populations from agricultural soils (Smalla et al 1993, Gebhard and Smalla, 1999).

The European Medical Agency (EMA) has also pointed out that little use of an antibiotic does not mean the same as that it is medicallyless important. They have also commented that the use of antiobiotics might change in the future as multiresistant bacteria occurs (EMEA 2007).

Based on this and the potential of horizontal gene transfer (HGT), the use of ARM genes in GM plants must be reconsidered and further restricted.

Recommendation:

- The regulator is encouraged to ask the Applicant to address what promoter is driving the expression of the ARM genes, as this is unclear from the summary.
- The regulator is encouraged to ask the Applicant to perform an analysis to verify the presence or absence of the *aad* protein.

Herbicide tolerance

This **GHB614xLLCotton25xMON15985 cotton** event contains two herbicidal tolerance genes, namely *2mepsps* and *bar* providing tolerance to the herbicides glyphosate and glufosinate-ammonium.

Glyphosate tolerance

In the recent years, glyphosate has received a lot of risk-related attention partly due to its increased use since the introduction of glyphosate-tolerant GM-plants (Dill et al., 2010, Cuhra et al., 2013). There have also been reports on negative effects in terrestrial and aquatic ecosystems (Blackburn and Boutin, 2003, Solomon and Thompson, 2003). Studies in animals and cell cultures have indicated that there could be health implications from exposure to glyphosate (Axelrad et al., 2003, Benachour et al., 2007, Cuhra et al., 2013). Among the health effects observed in animal models are histopathological changes in organs such as the liver, cell-division dysfunction in early embryos, negative impact on nerve-cell differentiation,

increased fetal mortality, growth reduction, and skeletal malformation. Additionally, the International Agency for Research on Cancer (IARC) recently released a report concluding that glyphosate is "probably carcinogenic to humans" (Fritschi et al., 2015).

Glufosinate ammonioum tolerance

Glufosinate-ammonium belongs to a class of herbicides that is banned in Norway and in EU (except for a limited use on apples) due to both acute and chronic effects on mammals including humans. Studies have shown that glufosinate-ammonium is harmful by inhalation, ingestion and skin contact and that serious health risks may result from exposure over time. Observations of patients poisoned by glufosinate-ammonium have found that acute exposure causes convulsions, circulatory and respiratory problems, amnesia and damages to the central nervous system (CNS) (Watanabe 1998). Chronic exposure in mice has been shown to cause spatial memory loss, changes to certain brain regions, and autism-like traits in offspring (Calas et al., 2008, Laugeray et al., 2014). According to EFSA, the use of glufosinate-ammonium will lead to farm workers being exposed to herbicide levels that exceed acceptable exposure levels during application.

Recommendations:

- We find it ethically unacceptable to ban the use of glufosinate-ammonium based herbicides domestically due to health and environmental concerns, while supporting its use in other countries. This represents an unacceptable double standard for Norway, and we ask the regulators to reconsider the practice of separating health and environmental risk by national borders or regions.

- The applicant should include a full evaluation of the co-technology intended to be used with **GHB614xLLCotton25xMON15985 cotton**, namely glyphosate- and glufosinate-ammonium-based herbicides. Particular focus should be given to the level of accumulation of herbicides in the plants, particularly the parts used in food and feed production, and whether or not these levels of exposure could cause acute and/or chronic health issues. This needs to be tested in animal and feeding studies, separating the effects of the plant and the herbicide(s) by using both sprayed and unsprayed plant samples.

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 (NGTA). In accordance with the aim of the NGTA, 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 elaborated in the regulations relating to impact assessment pursuant to the NGTA, section 17 and its annex 4. The NGTA, with its clauses on societal utility and sustainable development, comes into play with a view also to health, environmental and socio-economic impacts in other countries, such as where the GMOs are grown. In the following we identify areas that are relevant to consider in order to

assess social utility and sustainability aspects, and highlight the need for information to properly assess these issues.

The use of antibiotic resistance genes as selectable markers

The GHB614xLLCotton25xMon15985 cotton is genetically modified to and contain two antibiotic resistance genes (*add* and *npt11*). The use of antibiotic resistance genes as selectable markers for transformation of GM plants is controversial, and measures are taken to limit this practice due to the dramatic increase in antibiotic resistant bacterial strains globally. Norwegian authorities are encouraging a very restrictive approach to the use of GM plants with antibiotic resistance genes and work for international prohibitions in this area. The Norwegian Food Act (2005) prohibits the use of GM plants with such genes, and GM plants that possess such genes have also previously been prohibited under the NGTA, partly because they are not considered to contribute to sustainable development globally. The EFSA panel concludes that the antibiotic resistance gene *add* should be restricted to field trial purposes and not be present in GM plants placed on the market, as these genes confer resistance to antibiotics which are used for therapy in defined areas of human and veterinary medicine (EFSA 2004). Consequently, we do not recommend an approval of the GHB614xLLCotton25xMon15985 as this GM cotton contain the antibiotic resistance genes *add* and *npt11* where at least one of them is expressed.

Environmental impacts of the Bt-toxin on target and non-target organisms

The GHB614xLLCotton25xMon15985 cotton does also confer resistance to certain lepidopteran and coleopteran pests. A growing number of studies and reviews indicate potential harm from cry toxins expressed by GM Bt plants to a range of non-target organisms (Holderbaum et al., 2015; Marvier et al. 2007; Rosi-Marshall et al. 2007; Bøhn et al. 2008; Bøhn et al. 2014). Resistance development among target pests has also been documented in Bt cotton fields (Tabashnik et al., 2013). Russel (2008) provides evidence from Australia where Bt cotton has been successfully adopted as part of a wider Intergrated Pest Management (IPM) strategy. This indicates the importance of introducing Bt cotton in a functional agricultural advisory context, and the possible gains if this is done. Development of resistance development in target pest populations in GM Bt cotton fields has been reported in the US (Tabashnik et al., 2013). Hence, Evaluation of resistance development within the target pest population and strategies suggested to halt this development, as well as impacts on non-target organisms is crucial in a sustainability assessment

Environmental and health impacts of the co-technology: glyphosate and glufosinat-ammonium

The evaluation of the co-technology, that is, secondary products that are intended to be used in conjunction with the GMO, is also considered important in the risk assessment of a GMO (Dolezel et al., 2009). Therefore, considerations of the co-products also warrant an evaluation of safe use. The GHB614xLLCotton25xMon15985 cotton confers tolerance to herbicides containing glyphosate and glufosinate-ammonium.

Glufosinate-ammonium is a class of herbicides that are banned in Norway and in the EU (except a limited use on apples) due to both acute and chronic effects on mammals including humans (see section on Herbicide tolerance for references and further elaboration on this issue)

Recent studies have also shown negative effects from glyphosate, both on species present in terrestrial and aquatic ecosystems and on animals and cell cultures (for further elaboration and references on this issue see section on Herbicide tolerance). Consequently, glyphosate is now increasing recognized as more toxic to the environment and human health than what it was initially considered to be. This is particularly a concern as the introduction of glyphosate tolerant GM plants has led to an increase in the use of glyphosate (Dill et al. 2010). Moreover, studies has shown increased levels of herbicide residues in herbicide tolerant GM crops (Bøhn et al. 2014), which could have health impacts on humans and animals consuming food/feed based on ingredients from this type of GM plants. In order to evaluate changes in the use of glyphosate/ glufosinate ammonium after the introduction of GHB614xLLCotton25xMon15985, the applicant should provide information about the use of these herbicides in cotton production in the US.

Glyphosate resistant weeds in cotton is vastly documented globally, including in the US[1]. The Applicant should provide information on the contribution of the GHB614xLLCotton25xMon15985 to the emergence of glyphosate/ glufosinate-ammonium resistance in weeds, management strategies to prevent herbicide resistance development in weeds, and if there are already cases of this in the areas intended for cultivation of the variety.

Socio-economic impacts

There is a clear lack of knowledge regarding social impacts of GM crop introduction for farmers in the Global North (Fisher et al. (2015), such as the US, which is currently the only country that have approved the cultivation of the GHB614xLLCotton25xMon15985 cotton. A literature review performed by Fisher et al. (2015) shows that Bt cotton was the most commonly addressed GM crop in studies reporting on social implications from cultivation of GM crops (from 2004- 2015). Still, most studies were undertaken in the Global South, and did primarily investigate economic impacts from cultivating GM crops. Hence, the authors conclude that there are very few studies that take a comprehensive view of social impacts associated with GM crops in agriculture.

Reviews on social and economic impacts from GM crop cultivation (e.g. economic gains, distribution of benefits, access to seeds and improved wellbeing) relevant for a sustainability assessment indicate that these effects have been very complex, mixed and dependent on the agronomic, socio-economic and institutional settings where the technology has been introduced (Glover, 2010). Similarly, Fisher et al. (2015), and point to factors such as different political and regulatory contexts when explaining differences reported in distribution of economic gains and farmers' access to seeds in studies included in the review. This underlines that it cannot be expected that the same effects will apply between different social and environmental contexts. In order to meet the requirements in the NGTA, further investigations of social and economic implications from cultivating insect resistant and herbicide tolerant GM cotton, particularly in the US, is needed.

[1] http://weedscience.org/summary/crop.aspx

Social and economic impacts from gene flow and co-existence management
The cultivation of GM plants in general is causing problems with regard to co-existence. An evaluation of the occurrence of volunteer plants in the producing countries and suggested control strategies is important for a sustainability assessment. Information about the strategies adopted to ensure co-existence with conventional and organic cotton production and potential consequences for these production forms in the US is required for an assessment of social and economic impacts in the producer country.

Assessment of alternatives
It is also important to evaluate whether alternative options (e.g. the parental non-GM version of the GHB614xLLCotton25xMon15985) may achieve the same outcomes in a safer and ethically justified way. Furthermore, in order to evaluate whether the GHB614xLLCotton25xMon15985 contributes to social utility, it is important to consider current and future demand for this GM cotton product for food, feed and processing purposes in Norway and to what extent this demand is/can be satisfied by existing sources.

Ethical considerations
While it is understood that the Applicant has not applied for deliberate release of the GHB614xLLCotton25xMon15985 in Norway, the acceptance of a product in which the intended use involves the use of a product banned in Norway, as the glufosinate-ammonium, would violate basic ethical and social utility criteria, as laid out in the NGTA. Therefore we find that it would be ethically incongruous to support a double standard of safety for Norway on one hand, and safety for countries from which Norway may import its food and feed on the other. This line of reasoning is consistent with the provisions under the NGTA to assess ethical, social utility and sustainable development criteria not only for Norway, but for countries from which Norway imports food and feed. Specifically, this issue is relevant particularly in the revised guidelines for impact assessment pursuant to the Act of 2005 Section 17, "*Other consequences of the production and use of genetically modified organisms"* points 2 and 3, "*ethical considerations that may arise in connection with the use of the genetically modified organism(s)*», and "*any favorable or unfavorable social consequences that may arise from the use of the genetically modified organism(s)*", respectively.

Recommendation:

- In order to meet the requirements for the NGTA, the regulator is encouraged to ask the Applicant to submit information relevant for the assessment of the social utility of the GHB614xLLCotton25xMon15985 and its contribution to sustainable development. The information provided by the Applicant must be relevant for the agricultural context in the producing country. The information should include issues such as: Changes in pesticide use, emergence of herbicide resistant weeds, development of pest resistance in target populations, impacts on non-target organisms, potential for gene flow and possible impacts among farmers practicing different production forms for cotton cultivation in the producing country and share of the benefits among sectors of the society
- We do not recommend an approval of the GHB614xLLCotton25xMon15985 as it contain the two antibiotic resistance genes *aad* and *npt11* whereof at least one of them is expressed.

Conclusion

The GHB614xLLCotton25xMon15985 expresses the antibiotic resistance gene *npt11* while the expression of *aad* is not known. The GHB614xLLCotton25xMon15985 is tolerant to glufosinate-ammonium, which is banned for use in Norway. Banning the use of glufosinate-ammonium based herbicides domestically due to health and environmental concerns, while supporting its use in other countries would be ethically unacceptable. The applicant does not attempt to identify socio-economic implications, nor demonstrate a benefit to the community and a contribution to sustainable development from the use of the GHB614xLLCotton25xMon15985 and does therefore not provide sufficient information as required by the NGTA.

References

Axelrad JC., Howard CV and MClean, WG (2003). The effects of acute pesticide exposure on neuroblastoma cells chronically exposed to diazinon. Toxicology, 185**,** pp67-78.

Benachour N., Sipahutar H., Moslemi S., Gasnier C., Travert C and Seralini GE (2007). Time- and dose-dependent effects of roundup on human embryonic and placental cells. Arch Environ Contam Toxicol, 53**,** pp.126-33.

Badosa E., Moreno C and Montesinos E (2004). Lack of detection of ampicillin resistance gene transfer from Bt176 transgenic corn to culturable bacteria under field conditions. FEMS Microbiol. Ecol, 48, pp. 169-178.

Blackburn L and Boutin, C (2003). Subtle Effects of Herbicide Use in the Context of Genetically Modified Crops: A Case Study with Glyphosate (Roundup®). Ecotoxicology, 12**,** pp. 271-285.

Breyer D., Kopertekh L and Reheul D (2014). Alternatives to Antibiotic Resistance Marker Genes for In Vitro Selection of Genetically Modified Plants – Scientific Developments, Current Use, Operational Access and Biosafety Considerations. Critical Reviews in Plant Sciences, 33, pp.286-330.

Bøhn T., Cuhra M., Traavik T., Sanden M., Fagan J. and Primicerio R (2014). Compositional differences in soybeans on the market: Glyphosate accumulates in Roundup Ready GM soybeans. Food Chemistry, 153**,** pp.207-215.

Bøhn, T., Primicerio, R., Hessen, D. O. Traavik. T (2008). Reduced fitness of *Daphnia magna* fed a Bt-transgenic maize variety. Archives of Environmental Contamination and Toxicology, 55, pp.584-592.

Calas, AG., Richard, O., Même, S., Beloeil, JC., Doan, BT., Gefflaut, T., Même, W., Crusio, WE., Pichon J and Montecot, C. (2008). Chronic exposure to glufosinate-ammonium induces spatial memory impairments, hippocampal MRI modifications and glutamine synthetase activation in mice. NeuroToxicology, 29**,** pp. 740-747.

Carrer H., Hockenberry TN, Svab Z and Maliga P (1993). Kanamycin resistance as a selectable marker for plastid transformation in tobacco. Mol Gen Genet, 241, pp.49-56.

Committee for Medicinal Products for Veterinary Use and Committee for Medicinal Products for Human Use. Presence of the antibiotic resistance marker gene nptII in GM plants for food and feed uses. 22.02.2007.
www.emea.europa.eu/docs/en_GB/document_library/Other/2010/01/WC500054091.pdf

Crickmore N (2005). Using worms to better understand how Bacillus thuringiensis kills insects. Trends in Microbiology, 13, pp.347-350.

Cuhra M., Traavik T and Bøhn T (2013). Clone- and age-dependent toxicity of a glyphosate commercial formulation and its active ingredient in Daphnia magna. Ecotoxicology, 22**,** pp. 251-262.

Dill GM., Sammons RD., Feng PC., Kohn F., Kretzmer K., Mehrsheikh A., Bleeke M., Honegger JL., Farmer D and Wright, D (2010). Glyphosate: discovery, development, applications, and properties. Glyphosate Resistance in Crops and Weeds: History, Development, and Management, John Wiley and Sons, Inc., Hoboken, pp.1-33.

Dolezel M, Miklau M, Eckerstorfer M, Hilbeck A, Heissenberger A and Gaugitsch H (2009). Standardising the Environmental Risk Assessment of Genetically Modified Plants in the EU / Standardisierung der Umweltrisikoabschätzung gentechnisch veränderter Pflanzen in der EU. BfN – pp.259.

EFSA (2004). Opinion of the Scientific Panel on Genetically Modified Organisms on the use of antibiotic resistance genes as marker genes in genetically modified plants. www.efsa.europa.eu/fr/press/news/gmo040419a.htm

EFSA Journal (2009). Consolidated presentation of the joint Scientific Opinion of the GMO and BIOHAZ Panels on the "Use of Antibiotic Resistance Genes as Marker Genes in Genetically Modified Plants" and the Scientific Opinion of the GMO Panel on "Consequences of the Opinion on the Use of Antibiotic Resistance Genes as Marker Genes in Genetically Modified Plants on Previous EFSA Assessments of Individual GM Plants". 1108: 1-178

EMEA (2007). COMMITTEE FOR MEDICINAL PRODUCTS FOR VETERINARY USEAND COMMITTEE FOR MEDICINAL PRODUCTS FOR HUMAN USE Presence of the antibiotic resistance marker gene nptII in GM plants for food and feed uses. EMEA/CVMP/56937/2007-Final.

Fisher K., Ekener-Petersen E., Rydhmer L and Bjornberg EK (2015). Social impacts of GM crops in agriculture: A Systematic literature review. Sustainability, 7, 8598 – 8620.

Fritschi L., MClaughlin J., Sergi C., Calaf G., Le Cureieux F., Forastiere F., Kromhout H and Egephy P (2015). Carcinogenicity of tetrachlorvinphos, parathion, malathion, diazinon, and glyphosate. Red, pp.114.

Gebhard F and Smalla K (1999). Monitoring field releases of genetically modi¢ed sugar beets for persistence of transgenic plant DNA and horizontal gene transfer. FEMS Microbiology Ecology, 28, pp. 161-272.

Gilliand A, Chambers CE, Bone EJ and Ellar DJ (2002). Role of *Bacillus thuringiensis* Cry1 delta endotoxin binding in determining potency during lepidopteran larval development. Applied and Environmental Microbiology, 68, pp.1509-1515.

Glover D (2010). Exploring the Resilience of Bt Cotton's 'Pro-Poor Success Story'. Development and Change, 41, pp. 955–981.

Hilbeck A and Schmidt JEU (2006). Another view on Bt proteins - how specific are they and what else might they do? Biopesticides International, 2, pp.1-50.

Holderbaum DF., Cuhra M., Wickson F., Orth AI., Nodari OR and Bøhn T (2015). Chronic responses of daphnia magna under dietary exposure to leaves of a transgenic (event mon810) bt–maize hybrid and its conventional near-isoline. Journal of Toxicology and Environmental Health, Part A, 78, pp. 993–1007.

Laugeray A., Herzine A., Perche O., Hebert B., Aguillon-Naury M., Richard O., Menuet A., Mazaud-Guittot S., Lesne L., Briault S., Jegou B., Pichon J., Montecot-Duborg C and Mortaud S (2014). Pre- and Postnatal Exposure to Low Dose Glufosinate Ammonium Induces Autism-Like Phenotypes in Mice. Frontiers in Behavioral Neuroscience, 8**,** pp.390.

Marvier M., McCreedy C., Regetz J and Kareiva P (2007). A Meta-analysis of effects of BT cotton and maize on non-target invertebrates. Science, 316, pp. 1475-77.

Norwegian Gene Technology Act (2005). Act of 2 April 1993 No. 38 Relating to the Production and Use of Genetically Modified Organisms, etc.

Rosi-Marshall E.J., Tank JL Royer TV., Whiles MR., Evans-White M., Chambers C., Griffiths NA., Pokelsek J and Stephen ML (2007). Toxins in transgenic crop byproducts may affect headwater stream ecosystems. Proceedings of the National Academy of Sciences of the United States of America 104, pp. 16204-16208.

Russel AW (2008). GMOs and their contexts: A comparison of potential and actual performance of GM croms in a local agricultural setting. Geoforum 39, pp. 213 – 222.

Smalla K., van Overbeek LS., Pukall R and van Elsas JD (1993). Prevalence of nptII and Tn5 in kanamycin-resistant bacteria from different environments. FEMS Microbiology Ecology, 13, pp. 47-58.

Solomon K. and Thompson D. (2003). Ecological Risk Assessment for Aquatic Organisms from Over-Water Uses of Glyphosate. Journal of Toxicology and Environmental Health, Part B, 6**,** pp. 289-324.

Tabashnik B E., Brévault T and Carrière Y (2013). Insect resistance to Bt crops: lessons from the first billion acres. Nature Biotechnology, 39, pp 510- 521

van Frankenhuyzen K (2013). Cross-order and cross-phylum activity of *Bacillus thuringiensis* pesticidal proteins. Journal of Invertebrate Pathology, 114, pp. 76-85.

VKM (2005). Hovedkomitéen: Helse-og miljøvurdering – antibiotikaresistensmarkører i genmodifiserte planter. Doknr 05/312-5.

Watanabe T and Sano T (1998). Neurological effects of glufosinate poisoning with a brief review. Human & Experimental Toxicology 17:35-39.

WHO (2007). Critically important antimicrobials for human medicine: Categorization for the development of risk management strategies to contain antimicrobial resistance due to nonhuman antimicrobial use. Report of the second WHO expert meeting, Copenhagen, 29-31 May 2007.

Zhao JZ, Cao J, Collins H, Bates SL, Roush RT, Earle ED and Shelton A (2005). Concurrent use of transgenic plants expressing a single and two Bacillus thuringiensis genes speeds insect adaptation to pyramided plants. Proceeding of the National Academy of Sciences, 102(24), pp. 8426-8430.