Decision Document

Evaluation of Event Bt11 x GA21 (Resistant to Lepidopteran insects and Tolerant to Glyphosate and Glufosinate Ammonium herbicides) for human and animal consumption

SUMMARY AND BACKGROUND

The process of food risk assessment of transformation events due to modern biotechnology is carried out by the National Service of Agrifood Health and Quality (SENASA), which is the regulatory agency depending on the Secretariat of Agriculture, Livestock, Fisheries and Food (SAGPyA).

The Agrifood Quality Directorate of SENASA is the area responsible for carrying out this activity. This office has a specific scientific team and the advice of a Technical Advisory Committee composed of experts from different scientific disciplines representing different sectors involved in production, industrialization, consumption, research and development of genetically modified organisms.

An application from Syngenta company was received on March 7, 2007, to carry out the human and animal food safety evaluation of the transformation event Bt11 x GA21, resistant maize to lepidopteran insects and tolerant to herbicides such as glyphosate and glufosinate ammonium.

The application was reviewed in order to confirm the compliance with all the criteria proposed in SENASA Resolution Nº 412/02, regulation that lays down the criteria and requirements for the human and animal food safety evaluation in genetically modified organisms.

The information submitted is analyzed, at a first instance, by the specific technical team and later subjected to evaluation to the Technical Advisory Committee and finally the Agrifood Quality Directorate evaluates it at a third instance and concludes in the present document.

EVALUATION

The Bt11 x GA21 maize was evaluated following the guidelines shown in SENASA Resolution Nº 412/02, on "Basis and Criteria for the Evaluation of Food Products derived from Genetically Modified Organisms", the "Requirements and Standards Procedure for the Evaluation of Human and Animal Safety of Food Products derived from Genetically Modified Organisms", and the "Requested

Information" for such evaluation. Such evaluation was carried out using the information submitted in the application, together with the additional information requested and consultations with experts to establish the safety for human and animal consumption.

1 – History of use and specification of the transformation event

Maize is the third most important cereal crop worldwide behind rice and wheat. It was domesticated in pre-Columbian America over 8000 years ago. It is commercially grown in several countries of the world.

Maize has a long history of safe use and no cases of intoxication or allergies have been reported due to its reasonable consumption.

Syngenta has developed Bt11 x GA21 maize, hybrid which contains such event stacking by conventional crossing (sexual) of the carrying lines of event Bt11 (OECD ID Nº SYN-BTØ11-1) and carrying lines of event GA21 (OECD ID Nº MON-ØØØ21-9).

The stacked event in Bt11 x GA21 maize confers tolerance to lepidopteran insects, especially to *Diatraea saccharalis*, and tolerance to herbicides which active principle is glyphosate and glufosinate ammonium. That is to say that in the same hybrid maize, the characteristics of resistance to lepidopteran insects and tolerance to the herbicide glufosinate ammonium coming from the parental line Bt11, and tolerance to glyphosate herbicide coming from the parental line GA21 is combined.

Both parental events, Bt11 and GA21, were duly evaluated and approved by SENASA for its human and animal consumption, after the completion of the same requirements and considerations used to evaluate this event. Therefore, it has been established its substantial equivalence when compared to its conventional counterpart, which are conventional maize materials traded in the market.

2 – Genetic stability and molecular characterization of the event

The company submits the comparative Southern blot analysis between a hybrid Bt11 x GA21 maize and the individual parental inbred events Bt11 and GA21, where it is demonstrated that the Bt11 x GA21 hybrid, obtained from conventional crossing of said parental lines, inherits in a stable way, the *cry1Ab* and *pat* genes of the parental event Bt11 and the *mepsps* gene of the parental event

GA21, maintaining the hybridization patterns in identical way to the parental events.

Moreover, the applicant submitted an inheritance study of the transgenes of F_2 of a Bt11 x GA21 hybrid maize, where the inheritance is verified independently from the *mepsps* gene of the GA21 event and the *cry1Ab* and *pat* genes of the Bt11 event, since it presents mendelian segregation (9:3:3:1).

At the same time, along the pre commercial step of the stacked event, the company has carried out morph agronomic evaluations, where it is demonstrated the gene stability and the effective levels of the protein expressed.

Therefore, it can be concluded that the stacked event Bt11 x GA21 inherits the stability of Bt11 and GA21 parental events.

3 – Products and levels of expression

The products of the new expression are Cry1Ab protein and PAT protein, derived from event Bt11, and the modified enzyme mEPSPS derived from event GA21.

The modified enzyme mEPSPS (5-enolpyruvylshikimate-3-phosphate-synthase) encoded for *mepsps* gene is expressed in all plat tissues, producing a tolerance to herbicides that contain glyphosate. The *cry1Ab* gene encodes for the expression of Cry1Ab protein, which is also expressed in the whole plant and confers exclusive resistance to certain lepideroptean insects. Such resistance gene to insects is linked to the *pat* gene, which is encoded for the PAT protein and confers tolerance to the post emergent herbicide glufosinate ammonium and acts as a gene marker in the Bt11 parental event, being expressed mainly in leaves and ears.

Products as well as levels of protein in the new expressions are similar to the parental events.

4 – Compositional analysis

The results of compositional analyses of hybrid maize with stacked events submitted by the applicant, did not show significant biological differences in the 65 analytes measured in grain and forage, having into account their compositional nutrient profiles, compared to conventional maize traded in the market and the parental lines.

Based on this, it is concluded that Bt11 x GA21 maize is equivalent to commercial maize and the parental lines.

5 – Additional studies

The company submitted additional studies on animal food, specifically in a food research with broiler chickens during 44 days and another one with rats for 90 days. None of them showed adverse effects regarding control diets, reaching the conclusion that maize with stacked events does not produce negative effects.

6 – Metabolic interactions

Having evaluated the genetic stability, molecular characterization, products and levels of expression, compositional analyses and morph agronomic studies, it is concluded that no metabolic interaction is expected between single events when they are combined or stacked in a conventional way that might impact on the food safety of the stacked event as it is the case of Bt11 x GA21 hybrid maize.

7 – Allergenicity

No substances or allergenic proteins have been detected in the donor organism or in the recipient organism.

The mutated enzyme mEPSPS as well as Cry1Ab and PAT proteins, demonstrated not to share any relevant structural or immunological similarity, when compared with allergens, toxins or pharmacological active proteins in the framework of bioinformatics studies and database of known allergens.

Therefore, it is concluded that the possibility of a stacked event of Bt11 x GA21 maize to express allergenic substances for humans and/or animals is negligible.

8 – Toxicity

Regarding the study with rats during 90 days using Bt11 x GA21 grain maize in their diets, it is also demonstrated, after analyzing the observations in relation to corporal weight, feed consumption, clinical condition (including a set of functional and oftalmological observations), pathology clinic, weight of organs and histopathology, that there was not any toxicological effect related to the consumption of the event.

As a consequence, it is concluded that the event in Bt11 x GA21 maize do not pose any toxicological risk to be consumed by humans and animals.

9- Conclusion

After performing a complete food risk assessment to the material submitted by Syngenta company, and having into consideration that:

- The studies on genetic stability, molecular characterization, products and levels of expression, compositional analyses and morph agronomic studies, show no metabolic interaction between single events when they are combined or stacked in a conventional way in the Bt11 x GA21 hybrid maize,

- It is substantially and nutritionally equivalent to its non transgenic counterpart,

- Food safety of proteins in the new expression presented in Bt11 x GA21 maize was duly analyzed on a case by case basis. This means that parental lines as well as stacked event were evaluated, concluding that there are no relevant biological similarities in the structure or sequence homology when compared with allergens, toxins or pharmacological active proteins.

It is concluded that stacked event of Bt11 x GA21 maize is similar to its conventional counterpart; therefore, it is as safe and wholesome as conventional hybrid maize.

According to what has been previously exposed and having into account the current scientific knowledge available as well as the requirements and criteria internationally accepted, no objections can be found to approve Bt11 x GA21 maize for human and animal consumption.

10- Regulations and recommendations:

- SENASA Resolution Nº 1265/99
- SENASA Resolution Nº 412/02
- Principles for the risk analysis of food obtained by modern biotechnological means (CAC/GL 44-2003)
- Guidelines for the safety evaluation of food derived from plants of recombinant DNA (CAC/GL 45-2003)

- Consensus Documents for the work on the Safety of Novel Foods and Feeds (OECD)