Bioveiligheidsraad
Conseil de Biosécurité

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Secretariaat
Secrétariat

O./ref.: WIV-ISP/41/BAC/2016_0682

Title: Advice of the Belgian Biosafety Advisory Council on the application EFSA/GMO/NL/2007/47 from Pioneer under Regulation (EC) No. 1829/2003

Context

The application EFSA/GMO/NL/2007/47 was submitted by Pioneer on 24 September 2007 for the marketing of genetically modified (GM) soybean 305423 x 40-3-2 for food and feed uses, import and processing within the framework of Regulation (EC) No. 1829/2003[1].

Soybean 305423 x 40-3-2 is a two-event stack produced by conventional crossing. It expresses a high-oleic acid phenotype achieved by introducing a fragment of the soybean fad2-1 gene that results in the suppression of the expression of the endogenous omega-6 desaturase via RNA interference (RNAi). It also carries the genes encoding for ALS (GM-HRA) and CP4 EPSPS proteins, conferring tolerance to ALS-inhibiting herbicides and glyphosate respectively.

The application was officially acknowledged by EFSA on 19 February 2008. On the same date EFSA started the formal three-month consultation period of the Member States, in accordance with Articles 6.4 and 18.4 of Regulation (EC) No. 1829/2003 (consultation of national Competent Authorities within the meaning of Directive 2001/18/EC designated by each Member State in the case of genetically modified organisms being part of the products).

Within the framework of this consultation, the Belgian Biosafety Advisory Council (BAC), under the supervision of a coordinator and with the assistance of its Secretariat, contacted experts to evaluate the dossier, chosen from the common list of experts drawn up by the BAC and the Biosafety and Biotechnology Unit (SBB). Seven experts answered positively to this request, and formulated a number of comments to the dossier, which were edited by the coordinator. See Annex I for an overview of all the comments and for the list of comments actually placed on the EFSAnet on 19 May 2008.

The opinion of the EFSA Scientific Panel on GMOs was adopted on 14 July 2016 (EFSA Journal 2016;14(8):4566 [31 pp.][2]), and published together with the responses from the EFSA GMO Panel to comments submitted by the experts during the three-month consultation period.

The comments formulated by the experts together with the opinion of EFSA including the answers of the EFSA GMO Panel form the basis of the advice of the Biosafety Advisory Council given below.

[1] Regulation (EC) No 1829/2003 of the European Parliament and of the Council of 22 September 2003 on genetically modified food and feed (OJ L 268, 18.10.2003, p.1).
[2] See https://www.efsa.europa.eu/en/efsajournal/pub/4566

In addition, the scientific evaluations of the single events, namely soybean 305423 (EFSA/GMO/NL/2007/45), and soybean 40-3-2 (EFSA-GMO-RX-40-3-2) are taken into account in this advice. The Biosafety Council formulated a positive advice for both dossiers.
The two single soybean events are authorised in the EU for food and feed uses with the exception of GMO cultivation[3].

Scientific evaluation

1. Environmental risk assessment

According to the Biosafety Advisory Council no major risks were identified concerning the environment[4].

2. Molecular characterisation

With regard to the molecular characterisation, the Biosafety Advisory Council is of the opinion that the information provided is sufficient and does not raise safety concerns.

3. Assessment of food/feed safety and nutritional value

3.1. Assessment of compositional analysis

The composition of the GM soybean 305423 x 40-3-2 shows biologically relevant differences with conventional soybean in its fatty acid profile, as intended: an increase in oleic acid at the expense of polyunsaturated fatty acids.
As for the single-event soybean 305423, changes in the levels of other fatty acids are also observed. This unintended effect is likely to be related to the introduction of the GM-HRA enzyme, and is not expected to affect the conclusions on health and nutrition.
The Biosafety Advisory Council agrees with EFSA that the comparison with the two parental lines did not reveal any potential interaction that could be of concern for food and feed safety from the compositional viewpoint.

3.2. Assessment of toxicity

Soybean 305423 x 40-3-2 expresses two new proteins: GM-HRA and CP4 EPSPS. Based on previous assessments of these proteins and taking into account updated information provided by the applicant, the Biosafety Advisory Council is of the opinion that there are no indications that the newly expressed proteins in GM soybean 305423 x 40-3-2 may raise safety concerns regarding toxicity under the intended conditions of use.
It is also of the opinion that the simultaneous presence of these newly expressed proteins in the two-event stack soybean does not raise safety concerns regarding toxicity.

3.3. Assessment of allergenicity

In the context of previous applications assessed, the Biosafety Advisory Council did not identify concerns on allergenicity for the two newly expressed proteins GM-HRA and CP4 EPSPS. These conclusions remain valid based on the currently available information.

[3] See GMO register : <http://ec.europa.eu/food/dyna/gm_register/index_en.cfm>
[4] As the application doesn't imply a cultivation of the GM crop in the EU, a full environmental assessment is not required in EFSA procedure and was not performed.

The Biosafety Advisory Council is also of the opinion that the simultaneous presence of these newly expressed proteins in the two-event stack soybean does not raise safety concerns regarding allergenicity.
The Biosafety Advisory Council agrees with EFSA that there is no evidence that the genetic modification might significantly change the overall allergenicity of soybean 305423 x 40-3-2 when compared with that of its non-GM comparator and non-GM commercial soybean reference varieties.

3.4. Nutritional value

The Biosafety Advisory Council already assessed the nutritional consequences of the fatty acid profile modifications in the context of soybean 305423 (advice on application EFSA/GMO/NL/2007/45).
Taking into account the updated information provided by the applicant, the Biosafety Advisory Council is of the opinion that the conclusions drawn for soybean 305423 are also applicable to soybean 305423 x 40-3-2 and that this two-event stack soybean does not raise nutritional concerns in the context of the intended use.

4. Monitoring

With regard to monitoring, the Biosafety Advisory Council is of the opinion that the information provided is sufficient.

Conclusion

Based on the scientific assessment of the dossier done by the Belgian experts, taking into account the opinion of EFSA, the answers of the EFSA GMO Panel to the questions raised by the Belgian experts, the answers of the applicant to the EFSA GMO Panel questions and considering the data presently available, the Biosafety Advisory Council is of the opinion that in the context of its proposed uses, GM soybean 305423 x 40-3-2 is unlikely to pose any risk to human and animal health.

Given the scope of the application of this herbicide tolerant soybean with altered fatty-acid profile (no cultivation in EU) and the fact that the establishment of volunteer plants would be unlikely (soybean cannot survive without human assistance), the unintended environmental release of soybean 305423 x 40-3-2 is unlikely to pose any threat to the European environment.

Prof. Maurice De Proft
President of the Belgian Biosafety Advisory Council

Annex I: Full comments of experts in charge of evaluating application EFSA/GMO/NL/2007/47 (BAC_2008_752) and comments submitted on the EFSAnet (ref. BAC_2008_753).

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May 2008

Compilation of comments of experts in charge of evaluating the application EFSA/GMO/NL/2007/47

Mandate for the Group of Experts: mandate of the Biosafety Advisory Council (BAC) of 4 March 2008
Coordinator: René Custers and Thierry Hance
Experts: Pascal Cadot (Consultant), Armand Christophe (UGent), Johan Claes (KH Kempen), Jean-Pierre Hernalsteens (VUB), André Huyghebaert (UGent), Peter Smet (Consultant), Nancy Terryn (UGent), Michel Van Koninckxloo (HEPHO)
Domains of expertise of experts involved: Genetic engineering, genome analysis, transgene expression, toxicology immunology, alimentary allergology, human nutrition, biochemistry of food/feed, analysis of food/feed, industrial processing, agronomy, crop protection management, agro-ecology, herbicide tolerance, soybean
Secretariat (SBB): Didier Breyer, Adinda De Schrijver, Martine Goossens, Philippe Herman

INTRODUCTION

Dossier **EFSA/GMO/NL/2007/47** concerns an application of the company **Pioneer Hi-Bred** for the marketing of the genetically modified **soybean 305423 x 40-3-2** for food and feed applications under Regulation (EC) 1829/2003.
The application has been officially acknowledged by EFSA on 19 February 2008.
The scope of the application is:
☒ GM plants for food use
☒ Food containing or consisting of GM plants
☒ Food produced from GM plants or containing ingredients produced from GM plants
☒ GM plants for feed use
☒ Feed containing or consisting of GM plants
☒ Feed produced from GM plants
☒ Import and processing (Part C of Directive 2001/18/EC)
☐ Seeds and plant propagating material for cultivation in European Union (Part C of Directive 2001/18/EC)

Depending on their expertise, the experts were asked to evaluate the genetically modified plant considered in the application on its 1) molecular, 2) environmental, 3) allergenicity, 4) toxicity and/or 5) food and feed aspects. It was expected that the expert should evaluate if the information provided

in the application is sufficient in order to state that the marketing of the genetically modified plant for its intended uses, will not raise any problems for the environment or human or animal health. If information is lacking, the expert was asked to indicate which information should be provided and what the scientifically reasoning is behind this demand.

The comments are structured as in the "Guidance document of the scientific panel on genetically modified organisms for the risk assessment of genetically modified plants and derived food and feed" (EFSA Journal (2004), 99, 1-94).

List of comments received from the experts

A. GENERAL INFORMATION

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

B. INFORMATION RELATING TO THE RECIPIENT OR (WHERE APPROPRIATE) PARENTAL PLANTS

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

C. INFORMATION RELATING TO THE GENETIC MODIFICATION

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

Comment 2

As described in this application, 305423x40-3-2 soybean has been obtained from traditional breeding methods between progeny of genetically modified 305423 soybean and 40-3-2 soybean. No new genetic modification has been introduced in 305423x40-3-2 soybean.
I would argue that this new variety can only be allowed on the market if the 2 parental lines are, so if both separate GM varieties are approved for the fields asked.
I understand from the dossier that 40-3-2 is already a few years approved, but the dossier on 305423 is still pending. It would have been nice to have some more information on what is the status of that dossier.
I have focused in my review on evidence on molecular basis that changes caused by the combination of the 2 parental lines have occurred.

Comment 3

The dossier involves plants obtained by classical sexual hybridisation between transgenic lines. According to the EFSA guidance document for the risk assessment of genetically modified plants containing stacked transformation events, the transgenes of both parents should be present without

changes. This is expected on the basis of the historic observation (Otten *et al.*, 1981) that transgenes behave in crosses as dominant Mendelian genetic markers and is confirmed by the information provided in this dossier.

D. INFORMATION RELATING TO THE GM PLANT

D.1 DESCRIPTION OF THE TRAITS AND CHARACTERISTICS WHICH HAVE BEEN INTRODUCED OR MODIFIED

Comments/Questions of the expert(s)

Comment 1

No others than the two parental lines.

Comment 2

No comments/questions

Comment 3

I agree with the information that is given in the dossier.

D.2. INFORMATION ON THE SEQUENCES ACTUALLY INSERTED OR DELETED

Comments/Questions of the expert(s)

Comment 1

No evidence for others than the 2 parental lines.
The results obtained from Southern blot analysis confirm the molecular equivalence and identical copy number of the inserted DNA present in 305423x40-3-2 soybean to that present in 305423 soybean and 40-3-2 soybean.

Comment 2

No comments/questions

Comment 3

The sequences present in both parental lines are detected intact in soybean 305423 x 40-3-2. As expected, there is no argument for any change of the inserted sequence.

D.3. INFORMATION ON THE EXPRESSION OF THE INSERT

Comments/Questions of the expert(s)

Comment 1

No comments/questions

Comment 2

The expression of the herbicide resistance genes of both parental can easily be confirmed phenotypically. The data given in the dossier show clearly that the down-regulation of the expression of the soybean omega-6 desaturase gene occurs with the same efficiency in soybean 305423 x 40-3-2 as in its parental line 305423.

D.4. INFORMATION ON HOW THE GM PLANT DIFFERS FROM THE RECIPIENT PLANT IN: REPRODUCTION, DISSEMINATION, SURVIVABILITY

Comments/Questions of the expert(s)

Comment 1

Statistically significant differences were observed for early population, final population and seedling vigor between 305423x40-3-2 soybean treated with glyphosate and ALS-inhibiting herbicides and non-GM control soybean in the across location analysis.
However, the range of individual values for early population, final population and seedling vigor for 305423x40-3-2 soybean and non-GM control soybean were within their respective tolerance interval.
Furthermore, this characteristic of the variety, useful for the growers (thus not in EU), will not raise any problems for the environment or human or animal health.

Comment 2

No comments/questions

Comment 3

I see no argument to accept that the reproduction, dissemination or survivability of soybean 305423 x 40-3-2 would be facilitated, compared to its parental transgenic parents or compared to non-transgenic soybeans, except when the plants are treated with the relevant herbicides.

Additional comment from coordinator

Protocols are not fully identical, they are differences between sowing and treatment dates between trials (Table2, Annexe4)

A random bloc design was used to test the difference between variety including herbicide treatments using 6 sites in North America. This kind of design is powerful and commonly used in such study. However, I do not understand why a classical variance analyses (ANOVA3) including the analyses of interaction between factor and test of error based on the residual effect of these interactions was not done. Two kinds of statistical analyses were done 1) accross locations and individuals. The across location analyses results in an increase of the variation inside treatments, as particularity of each sites is not taken into account. In fact, the site factor should be used as an error term to test the actual treatment effect. As far as I can see in Annexe 4, it was not done or if done was not explained clearly. Moreover, when a difference is found significant, it is then compared to a tolerance level based on literature data obtained in a completely different context. Here, the question is not to know if the differences are in a large range of variation, but if they are significantly distant from the non gm plant. The tolerance intervals used are moreover very wide, and for several parameters, such as height, early and final population, start at zero! (Table 7, Annexe 4) At least, standard deviation must be used instead of this tolerance level! Finally, I think that the FDR adjustement used is not applicable here. See page 19 of Annexe 4 and following. So I do not agree with the statistical procedure used. Conclusion page 16 of 48 states that agronomic characteristics are comparable but they are in fact differing between GM and non GM plant, i.e. for seed vigor or height.

D5. GENETIC STABILITY OF THE INSERT AND PHENOTYPIC STABILITY OF THE GM PLANT

Comments/Questions of the expert(s)

Comment 1

No comments/questions

Comment 2

Genetic stability of the inserts is expected, on the basis of genetics, and confirmed by the information given in the dossier.

D.6. ANY CHANGE TO THE ABILITY OF THE GM PLANT TO TRANSFERR GENETIC MATERIAL TO OTHER ORGANISMS

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

Comment 2

No comments/questions

Comment 3

Sexual hybridisation with other soybeans would be the only mechanism for such exchange. This is very unlikely to occur, because soybean 305423 x 40-3-2 will be processed but not cultured in the European Union. In addition soybeans mainly self-pollinate and survive poorly in the environment, making such exchanges even more unlikely if transgenic material would be unintentionally released into the environment.

D.7. INFORMATION ON ANY TOXIC, ALLERGENIC OR OTHER HARMFUL EFFECTS ON HUMAN OR ANIMAL HEALTH ARISING FROM THE GM FOOD/FEED

Comment 1

The dossier is well established and discusses the safety (toxicological, allergenicity, food/feed nutrition) with own experiments and based on literature. The issues indicated in the Guidance Notes of the Biosafety Council (The safety assessment of genetically modified crops for food and feed use, April 2003) are well discussed. Furthermore, this soybean is obtained by traditional breeding methods between two GM soybeans, from which one was already authorised under previous EU legislation. Both of them are in the process of authorisation. This is indeed a strong indication, but not a proof, that soybean 305423x40-3-2 will not raise any additional problems for human or animal health as compared to control soybean. Most of the comments are related to this issue.

D.7.1 Comparative assessment

Comments/Questions of the expert(s)

Comment 1

The EFSA guidance document for the risk assessment of GM plants (EFSA 2006) states that "the comparison between GM plants and the most appropriate comparator **should cover more than one representative growing season**…" (page 23). Yet composition data were obtained from field trials during the 2005 growing season only (Annex 4). Are data of other growing seasons available?

Comment 2

The applicant refers several times to the traditional breeding method between two GM soybeans, and exploits this origin as an indication of the safety. The application would be more convincing if the safety is not only evaluated with respect to non-GM control soybean as a reference, but also with respect to the parent 305423 soybean and the 40-3-2 soybean. This yields information whether there is a synergistic effect of both parent species.

Comment 3

Soybean 305423 x 40-3-2 will be referred further as **submitted soybean.**

The submitted soybean was compared with a non-GMO soybean with comparable genetic background. In addition data from four commercial soybean varieties have also been used for comparison.
No particular questions.

D.7.2 Production of material for comparative assessment

Comments/Questions of the expert(s)

Comment 1

In part VI, page 139; 3. Other information is given than "Name and full address of the suppliers of control samples". Typing error?

Statistical methods. For several components, the calculated "tolerance interval" includes and is considerably larger than ranges found in literature (e.g palmitic acid in Appendix 4, Table 6 pp225: tolerance interval 2.93-19.6 vs literature range 7-15.8). If the determined value of the GM falls in either one, it is concluded that there is no compositional difference with regular soy. For the example given above, 6.48 % palmitic acid of total fatty acids is considered compositionally equivalent on basis of the tolerance interval, even though this is (slightly) outside the range reported in literature. Although this is not considered to be of biological relevance, it rises the question whether the tolerance interval should not be reduced from 99% to a lower value to be an acceptable indicator of compositional equivalency.

Additional comment from coordinator

I fully agree with that comment. Again, first as stated before, the statistical analyses is not appropriated to the data and the protocol used. See the above comments. Many differencse appeared between GM and non GM plant concerning composition in amino acid isoflavone and vitamins, those differences are minimized without actual argument. Table 7, differences in lectines in phytic acid are significant. So I could not agree with conclusion of page 213 of 563 of Annexe 4.
Page 306 of 563, Annexe 4: Consequences of the statement of GLP compliance are not discussed or taken into account in the conclusions.

Comment 2

No comments/questions

Comment 3

A field study, with a randomized block design, was conducted at six different locations in North America including the submitted and the non-GMO soybean. Composition data from commercial soybean varieties have been used as a baseline for expected and natural variations.
No particular questions.

D.7.3 Selection of material and compounds for analysis

Comments/Questions of the expert(s)

Comment 1

Saponins are present in soy in relatively high quantities (Berhow et al., 2006) and although poorly absorbed in humans (Hu et al., 2004) they can cause bloat in ruminants (Van Haver et al., 2003) and induce enteritis in salmon (Knudsen et al.)
Q: Would it not be indicated to determine the level of saponins in products derived from 305423x 40-3-2 soybeans and compare them with control soybeans?

Comment 2

The compositional analyses confirm that 305423x40-3-2 soybean grain is comparable to grain from non-GM control soybean with comparable genetic background and to commercial soybean grain except for the fatty acid composition.

Comment 3

The range of values for heptadecanoic acid, heptadecenoic acid and oleic acid for 305423x40-3-2 soybean were outside their respective tolerance intervals. All individual values for heptadecanoic acid, heptadecenoic acid and oleic acid for 305423x40-3-2 soybean were higher than the upper limit of their respective tolerance intervals and literature ranges.

It says in the dossier that heptadecanoic acid is present in vegetable oils, butter and meat, while heptadecenoic acid is present in beef, cheese and olive oil (USDA, 2006 and Pioneer data) but the reference USDA2006 does not show figures? Pioneer data also I did not find. I checked the USDA National Nutrient Database for Standard Reference Website but did not find data on heptadecenoic acid. As this is not my field of expertise I did not lose to much time, I guess other expert might know if the values of the GM are really out of scale or not.
Anyway this follows my general opinion on this GM variety, that it can only be allowed if the two parental lines are.

Comment 4

The soybean under study increases the amount of two odd chain fatty acids. The effect of this increase on possible health effects is well motivated for these components (including reference values from literature). However, it is not clear what might be the biochemical explanation for the increase of these unusual fatty acids. It might be possible that this increase is linked to an increase of other (possibly toxic) components (see, e.g., 2-ketobutyate in Kingsbury *et al*, 2006; LaRossa *et al.*, 1987). Another link is illustrated by Bjelk and Monaco (1992) who discussed the impact of the herbicide chlorimuron on the fatty acid biosynthesis.

It would make the dossier more convincing on this point if a possible biochemical pathway is discussed/hypothesized, based on a literature survey and/or experiments (see, e.g., van der Hoeven and Steffens, 2000).

In addition, a comparison of the compositional analysis with the results of each of the parent species will yield additional information about possible synergistic effects.

Comment 5

The OECD guidelines for the assessment of genetically modified soybean were followed; The compositional data include protein, fibre, carbohydrates, fat, ash, minerals fatty acids, amino acids, vitamins, secondary metabolites and anti-nutrients.
The applicant concludes that the submitted soybean is comparable to commercial soybean, with exception of the fatty acid profile, which reflects the intended modification: a high oleic phenotype.

A review of the data confirm these conclusion with some minor remarks.
Proximate analysis:
Significant differences identified are not consistent at each location and are within the tolerance levels and literature data.
As mentioned before, the applicant uses the crude fibre approach, traditionally used for animal feed and not for human food. I personally regret that the modern approaches of dietary fibre, generally used for human food are not followed.
This remark has been raised already several times in previous dossiers and also further substantiated during a hearing session at the SBB.
Minerals:
Important minerals are well covered in the analysis. No significant differences have been identified for most minerals. A significant difference in iron content was not confirmed at the across location level. Calcium values remain significantly different even at the across location level, but within literature range.
The observations with respect to iron, but particularly calcium, deserve attention.
Iron deficiency is very known in humans and is identified as a problem for particular vulnerable groups. As the value is within literature data the level is however not alarming.
Calcium is important in human nutrition and a mineral of concern for particular consumer groups. In case soybeans are used for the manufacture of soy drinks and other dairy analogs, calcium is generally a limiting mineral. Enrichment with calcium is however quite often applied. A further reduction of natural calcium levels has to be taken into account in the enrichment process. In case no calcium addition is performed the phenomenon deserves particular attention.
Fatty acids
In the comparative assessment the applicant concludes that there are not differences in the fatty acid profile with the exception of oleic acid, intended in the high oleic phenotype. All relevant fatty acids are included.
For particular fatty acids significant differences have been observed in line with the intended modification.
No further questions.
Amino acids
The whole range of important amino acids is covered.
No significant statistical differences were observed with the exception of aspartic acid at one location. The mean value of aspaxtic acid was however within the literature data for this amino acid;
No further questions.
Vitamins

Relevant vitamins for soybean have been studied, including vitamin B1, vitamin B2, folic acid and the whole range of tocopherols: α, β, γ and δ. In soybean, contrary to other vegetable oils, a range of tocopherols is naturally present. Tocopherols play an important role in human nutrition as natural antioxidants.
Some statistically significant differences were found but always within the range of literature data.
No further questions.

Isofavones

Isoflavones are important constituents in soybeans. Isoflavones are well studied, covering the range of constituents.
No statistically significant differences were found with the exception of glycitin and malonylglycitin. Differences are however within the range of literature data.
No further questions

Oligosaccharides

No statistically significant differences found. No further questions as the oligosaccharides are well studied.

Secondary metabolites and anti-nutrients

Constituents studied are lectins, phytic acid,and coumestrol. The applicant observed no statistically significant differences for these constituents.
The presence of trypsin inhibitor is an important trait in soybean. Some differences were found at particular locations. They are however within the range of literature data.

Conclusion:

The applicant presents convincing evidence, with some minor remarks, that the submitted soybean is comparable in terms of composition with the non-GMO soybean and with commercial varieties. Some differences are found in fatty acid composition in line with the intended modification. In my opinion differences found for some minor fatty acids are not of safety concern as they are also present in other natural fats and oils.

D.7.4 Agronomic traits

Comments/Questions of the expert(s)

Comment 1

The scope of the application does not include cultivation of 305423x40-3-2 soybean seed products in the EU.

Comment 2

No comments/questions

Comment 3

No comments

D.7.5 Product specification

Comments/Questions of the expert(s)

Comment 1

No comments/questions

Comment 2

No comments

D.7.6 Effect of processing

Comments/Questions of the expert(s)

Comment 1

No novel method of production and processing is envisaged.

Comment 2

No comments/questions

Comment 3

The applicant refers to the OECD, 2001 document where production and processing methods are reviewed. The submitted soybean will be processed according to existing methods. No novel method is envisaged. Unprocessed soybeans have no food use. All imported soybeans are crushed and further processed.
The processes are reviewed taking into account the particular composition. The GM-HRA and CP4 EPSPS are highly susceptible to proteolytic digestion and are unstable under heating conditions applied during processing.
I agree with this conclusion as soybeans, intended as a human food, have to be processed in order to inactivate the trypsin inhibitor. The expressed proteins are heat denatured.

D.7.7 Anticipated intake/extent of use

Comments/Questions of the expert(s)

Comment 1

It is to be expected that the high oleic acid type of soybean oil in 305423x40-3-2 will be used for other applications than soy oil also (that was one of the reasons to produce 305423). As a

consequence, the statement that the "total consumption of soybean products remaining unchanged" (part II, page 100) is uncertain.
If the processed products of 305423x40-3-2 will replace a portion of similar products from commercial soybean (part II, page 100), the intake of alphalinolenic acid is expected to decrease. This is not beneficial from a nutritional point of view. Indeed, dietary intake of this essential fatty acid is lower than recommended (e.g. Sioen 2007).

Comment 2

The information provided in the application is sufficient.

Comment 3

No comments/questions

Comment 4

The applicant concludes that the submitted soybean has equivalent food applications as the commercial varieties. Consumption data of soybean derivatives are cited according to GEMS/FOOD data, 2003 and other literature data like the EPIC study.
The applicant concludes that the composition of the submitted soybean is equivalent to commercial varieties. So no major shift in intake of nutrients due to soybeans is to be expected.
Comment:
As the submitted soybean has a high oleic fatty acid composition, this may affect the intake of particular fatty acids. In my opinion the modification in fatty acids is, from a human food perspective, in a positive direction as the high oleic oil will certainly show a better oxidation stability. In addition it is to be expected that in comparison to traditional soybean varieties the extracted oil has to be less modified, among others by hydrogenation. This may reduce the intake of (undesirable) trans fatty acids formed as by-products during hydrogenation.

D.7.8 Toxicology

D. 7.8.1 Safety assessment of newly expressed proteins

Comments/Questions of the expert(s)

Comment 1

Mean concentrations of GM-HRA in 305423 soybean and 305423 x 40-3-2 soybean are indeed comparable.

a) GM-HRA protein measured in 305423 soybean

Growth stage/ Tissue	ng/mg Tissue Dry Weight		Standard deviation
	Mean	Range	
V2/Leaf	3.1	0.99 – 9.0	1.9
V5/Leaf	2.7	0 – 7.7	1.9
R3/Leaf	4.0	1.2 – 6.3	1.8
R3/Forage	5.7	0.78 – 51	12
R3/Root	0.18	0 – 0.63	0.22
R8/Grain	2.5	0 – 4.9	1.1

b) GM-HRA protein measured in 305423 x 40-3-2 soybean

Growth stage/ Tissue	ng/mg Tissue Dry Weight		Standard deviation
	Mean	Range	
V2/Leaf	2.1	0.66 – 4.5	1.1
V5/Leaf	2.3	0.91 – 4.6	1.0
R3/Leaf	4.4	1.7 – 9.9	1.9
R3/Forage	3.5	0.73 – 22	5.5
R3/Root	0.26	0 – 0.69	0.22
R8/Grain	3.1	1.9 – 4.6	0.85

Mean concentrations of CP4 EPSPS in 40-3-2 soybean and 305423 x 40-3-2 soybean are indeed comparable.

c) CP4 EPSPS protein measured in 40-3-2 soybean

Growth stage/ Tissue	ng/mg Tissue Dry Weight		Standard deviation
	Mean	Range	
V2/Leaf	5400	2900 – 9900	1900
V5/Leaf	5100	3300 – 7600	1300
R3/Leaf	1300	91 – 4100	1200
R3/Forage	2800	910 – 6300	1300
R3/Root	160	14 – 580	150
R8/Grain	320	220 – 430	50

d) CP4 EPSPS protein measured in 305423 x 40-3-2 soybean

Growth stage/ Tissue	ng/mg Tissue Dry Weight		Standard deviation
	Mean	Range	
V2/Leaf	5300	2800 – 10000	2100
V5/Leaf	5800	2400 – 10000	2300
R3/Leaf	2100	59 – 5100	1400
R3/Forage	3300	540 – 7500	2000
R3/Root	200	33 – 420	110
R8/Grain	410	320 – 520	70

Safety assessment of newly expressed proteins.

a) Degradation of the GM-HRA protein in simulated gastric fluid (author).

No data were provided in dossier EFSA/GMO/NL/2007/47.

b) Degradation of the GM-HRA protein in simulated intestinal fluid (author).

No data were provided in dossier EFSA/GMO/NL/2007/47.

c) Degradation of the CP4 EPSPS protein in simulated gastric fluid.

Rapid digestion was previously demonstrated.

d) Degradation of the CP4 EPSPS protein in simulated intestinal fluid.

Rapid digestion was previously demonstrated.

e) GM-HRA: Acute Oral Toxicity Study in Mice (Finlay, 2006).

Lack of acute toxicity was demonstrated in dossier EFSA/GMO/NL/2007/47. No further testing is needed.

f) CP4 EPSPS: Acute Oral Toxicity Study in Mice.

Lack of acute toxicity was demonstrated earlier. No further testing is needed.

Comment 2

In this section, the safety is discussed for the two proteins separately (GM-HRA and CP4 EPSPS), by using toxicity studies, sequence homology, presence in human food, … However, there is no discussion, nor reference to an acute toxicity study of the possible synergistic effect when combining both modifications in one species.

D.7.8.2 Testing of new constituents other than proteins

Comments/Questions of the expert(s)

Comment 1

No comments/questions

D.7.8.3 Information on natural food and feed constituents

Comments/Questions of the expert(s)

Comment 1

305423x40-3-2 soybean grain is comparable to grain from commercial soybean except for the fatty acid profile but without adverse effect.

Comment 2

No comments/questions

Comment 3

This item has been discussed before.

D.7.8.4 Testing of the whole GM food/feed

Comments/Questions of the expert(s)

Comment 1

The EFSA (2006) report states "if there are any indications for the **potential occurrence of unintended effects**…the whole GM food/feed should be tested." Although it is realized that the methodology of the paper of Ermakova (Ermakova 2006; Ermakova 2007) is highly criticized (e.g. Marshall 2007), the differences compared to controls in survival rates of rat pups of which the mothers were fed soy 40-3-2 before mating were of such magnitude that repeating this experiment with 305423x40-3-2 under the same conditions as in the paper and under optimalised conditions is considered to be prudent.

Comment 2

<u>a) 42-day poultry feeding study.</u>

No statistically significant differences were observed in mortality, weight gain, mortality adjusted feed efficiency, and carcass yields between broilers consuming diets produced with 305423x40-3-2 or

305423x40-3-2+Gly/SU soybean fractions and those consuming diets produced with near isoline control soybean fractions.

Based on the results from this study, it was concluded that 305423x40-3-2 soybean was nutritionally equivalent to non-transgenic control soybean with a comparable genetic background.

b) 90-Day rat feeding study.

Since the fatty acid composition in 305423x40-3-2 soybean is quite different from non-GM soybean and genes for two new proteins were introduced, **a 90-day rodent feeding study is recommended**, since synergistic effects cannot be excluded beforehand.

Comment 3

The information provided in the application is sufficient.

Comment 4

No comments/questions

D.7.9 Allergenicity

Comments/Questions of the expert(s)

Comment 1

Assessment of allergenicity of the introduced traits.
The reviewer agrees with the conclusion of the applicant when it is said that, according to current knowledge, GM-HRA and CP4 EPSPS are not likely to be allergenic.

Assessment of allergenicity of the whole GM plant.
The applicant has not evaluated the allergenicity of the whole GM plant. The fact that both parent plants were evaluated and the fact that both inserted proteins are not likely to be allergenic does not preclude any conclusion about allergenicity of the whole plant that must be considered as a new organism. It must also be emphasized that in the application for authorisation of genetically modified 305423 soybean and derived food and feed under Regulation (EC) N° 1829/2003 (application EFSA-GMO-NL-2007-45) to which it is referred in the present application, the number of soybean-sensitive sera used was considered as too limited. Given the high prevalence of soy allergy, it is essential to evaluate the allergenicity of the 305423x40-3-2 soybean, as the presence of the two new traits may alter the levels of expression of other proteins in the plants, and these differently regulated proteins might be allergens. It is important that the GM plant is not more allergenic than the natural counterpart. Sera of soy allergic patients can easily be found. It is recommended that at least 20 sera be used, in order to get a broader range of reactivity patterns. In addition, the sera must not be pooled, as some information (for example the visualisation of the emergence of an allergen for some patients) might be diluted and lost in a pool.
One cannot conclude, at the moment, about the allergenicity of 305423x40-3-2 soybean.

Comment 2

No comments/questions

D.7.10 Nutritional assessment of GM food/feed

Comments/Questions of the expert(s)

Comment 1

Soy lecithin is one of the secondary products of processed soybean. Soy phospholipids can be used for the oxidative stabilisation of oils and fats (e.g. Murano Y et al., 2008). No data are given on the fatty acid composition of lecithin derived from 305423 soybeans which is possibly (and probably) also affected. If so, this may affect the possibility of soy phospholipids to improve the lipid profile favorably (Evans et al., 2007) especially considering that soy phospholipids containing 2 molecules of linoleic acid (this fatty acid is greatly reduced in soy 305423x40-3-2 oil; no data are given on phospholipids) are most active in promoting apoAI secretion. Pandy NR et al., 2008).
Q: Are data available on the fatty acid composition of phospholipids derived from 305423x40-3-2 soybeans?

Comment 2

The information provided in the application is sufficient.

Comment 3

No comments/questions

D.7.11 Post-market monitoring of GM food/feed

Comments/Questions of the expert(s)

Comment 1

Post-market monitoring of GM food and GM feed products containing, consisting of or derived from 305423x40-3-2 soybean is not necessary.

D.8. MECHANISM OF INTERACTION BETWEEN THE GM PLANT AND TARGET ORGANISMS (IF APPLICABLE)

Comments/Questions of the expert(s)

Comment 1

Not applicable

D.9. POTENTIAL CHANGES IN THE INTERACTIONS BETWEEN THE GM PLANT WITH THE BIOTIC ENVIRONMENT RESULTING FROM THE GENETIC MODIFICATION

D.9.1. Persistence and invasiveness

Comments/Questions of the expert(s)

Comment 1

Comment: The application does not include cultivation of 305423x40-3-2 soybean seed products in the EU. Therefore, any exposure to the environment will be limited to unintended release via spillage during transportation of the grain.
In Belgium, there is no likelihood for 305423x40-3-2 soybean to become environmentally persistent or invasive giving rise to any weediness. The marketing of this soybean will not raise any problems for the environment.

D.9.2 Selective advantage or disadvantage

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.3 Potential for gene transfer

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.4 Interactions between the GM plant and target organism

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.5 Interactions of the GM plant with non-target organism

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.6 Effects on human health

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.7 Effects on animal health

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.8 Effects on biogeochemical processes

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.9.9 Impacts of the specific cultivation, management and harvesting techniques

Comments/Questions of the expert(s)

Comment 1

The application does not include cultivation of 305423x40-3-2 soybean seed products in the EU.

D.10. POTENTIAL INTERACTIONS WITH THE ABIOTIC ENVIRONMENT

Comments/Questions of the expert(s)

Comment 1

The application does not include cultivation of 305423x40-3-2 soybean seed products in the EU.

D.11. ENVIRONMENTAL MONITORING PLAN

D.11.1 General

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.11.2 Interplay between environmental risk assessment and monitoring

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.11.3 Case-specific GM plant monitoring

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.11.4 General surveillance of the impact of the GM plant

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

D.11.5 Reporting the results of monitoring

Comments/Questions of the expert(s)

Comment 1

The information provided in the application is sufficient.

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Bioveiligheidsraad
Conseil de Biosécurité

19 May 2008

Secretariaat
Secrétariat

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Application EFSA/GMO/NL/2007/47
Comments submitted on the EFSAnet on mandate of the Biosafety Council

Mandate for the Group of Experts: mandate of the Biosafety Advisory Council (BAC) of 4 March 2008
Coordinator: René Custers and Thierry Hance
Experts: Pascal Cadot (Consultant), Armand Christophe (UGent), Johan Claes (KH Kempen), Jean-Pierre Hernalsteens (VUB), André Huyghebaert (UGent), Peter Smet (Consultant), Nancy Terryn (UGent), Michel Van Koninckxloo (HEPHO)
Domains of expertise of experts involved: Genetic engineering, genome analysis, transgene expression, toxicology immunology, alimentary allergology, human nutrition, biochemistry of food/feed, analysis of food/feed, industrial processing, agronomy, crop protection management, agro-ecology, herbicide tolerance, soybean
Secretariat (SBB): Didier Breyer, Adinda De Schrijver, Martine Goossens, Philippe Herman

INTRODUCTION

Dossier **EFSA/GMO/NL/2007/47** concerns an application of the company **Pioneer Hi-Bred** for the marketing of the genetically modified **soybean 305423 x 40-3-2** for food and feed applications under Regulation (EC) 1829/2003.
The application has been officially acknowledged by EFSA on 19 February 2008.
The scope of the application is:
☒ GM plants for food use
☒ Food containing or consisting of GM plants
☒ Food produced from GM plants or containing ingredients produced from GM plants
☒ GM plants for feed use
☒ Feed containing or consisting of GM plants
☒ Feed produced from GM plants
☒ Import and processing (Part C of Directive 2001/18/EC)
☐ Seeds and plant propagating material for cultivation in European Union (Part C of Directive 2001/18/EC)

Comments posted on the EFSAnet

The comments are structured according to the "Guidance document of the scientific panel on genetically modified organisms for the risk assessment of genetically modified plants and derived food and feed" (EFSA Journal (2004), 99, 1-94). The comments below are those that were posted on the EFSAnet. It should be noted that all the comments received from the experts are considered in the evaluation of this dossier and in formulating the final advice of the Biosafety Advisory Council. The compilation of all the comments that were received from the experts (including the references) is given in a separate document (ref. BAC_2008_752).

A. GENERAL INFORMATION

No comments

B. INFORMATION RELATING TO THE RECIPIENT OR (WHERE APPROPRIATE) PARENTAL PLANTS

No comments

C. INFORMATION RELATING TO THE GENETIC MODIFICATION

Comments/Questions of the expert(s)

As described in this application, 305423x40-3-2 soybean has been obtained from traditional breeding methods between progeny of genetically modified 305423 soybean and 40-3-2 soybean. No new genetic modification has been introduced in 305423x40-3-2 soybean.
We would argue that this new variety can only be allowed on the market if the 2 parental lines are, so if both separate GM varieties are approved for the fields asked.
We understand from the dossier that 40-3-2 is already a few years approved, but the dossier on 305423 is still pending. It would have been nice to have some more information on what is the status of that dossier.

Both parental lines must be authorized in accordance with regulation EC/1829/2003 before this dossier should be analysed. It is too early to take a decision now.

D. INFORMATION RELATING TO THE GM PLANT

D.1 DESCRIPTION OF THE TRAITS AND CHARACTERISTICS WHICH HAVE BEEN INTRODUCED OR MODIFIED

No comments

D.2. INFORMATION ON THE SEQUENCES ACTUALLY INSERTED OR DELETED

No comments

D.3. INFORMATION ON THE EXPRESSION OF THE INSERT

No comments

D.4. INFORMATION ON HOW THE GM PLANT DIFFERS FROM THE RECIPIENT PLANT IN: REPRODUCTION, DISSEMINATION, SURVIVABILITY

Comments/Questions of the expert(s)

Protocols are not fully identical, they are differences between sowing and treatment dates between trials (Table2, Annexe4)

A random bloc design was used to test the difference between variety including herbicide treatments using 6 sites in North America. This kind of design is powerful and commonly used in such study. However, I do not understand why a classical variance analyses (ANOVA3) including the analyses of interaction between factor and test of error based on the residual effect of these interactions was not done. Two kinds of statistical analyses were done 1) across locations and individuals. The across location analyses results in an increase of the variation inside treatments, as particularity of each sites is not taken into account. In fact, the site factor should be used as an error term to test the actual treatment effect. As far as I can see in Annex 4, it was not done or if done was not explained clearly.
Moreover, when a difference is found significant, it is then compared to a tolerance level based on literature data obtained in a completely different context. Here, the question is not to know if the differences are in a large range of variation, but if they are significantly distant from the non gm plant. The tolerance intervals used are moreover very wide, and for several parameters, such as height, early and final population, start at zero! (Table 7, Annex 4) At least, standard deviation must be used instead of this tolerance level! Finally, I think that the FDR adjustment used is not applicable here. See page 19 of Annex 4 and following. So I do not agree with the statistical procedure used. Conclusion page 16 of 48 states that agronomic characteristics are comparable but they are in fact differing between GM and non GM plant, i.e. for seed vigor or height.

D5. GENETIC STABILITY OF THE INSERT AND PHENOTYPIC STABILITY OF THE GM PLANT

No comments

D.6. ANY CHANGE TO THE ABILITY OF THE GM PLANT TO TRANSFER GENETIC MATERIAL TO OTHER ORGANISMS

No comments

D.7. INFORMATION ON ANY TOXIC, ALLERGENIC OR OTHER HARMFUL EFFECTS ON HUMAN OR ANIMAL HEALTH ARISING FROM THE GM FOOD/FEED

D.7.1 Comparative assessment

Comments/Questions of the expert(s)

The EFSA guidance document for the risk assessment of GM plants (EFSA 2006) states that "the comparison between GM plants and the most appropriate comparator **should cover more than one representative growing season**…" (page 23). Yet composition data were obtained from field trials during the 2005 growing season only (Annex 4). Are data of other growing seasons available?

The applicant refers several times to the traditional breeding method between two GM soybeans, and exploits this origin as an indication of the safety. The application would be more convincing if the safety is not only evaluated with respect to non-GM control soybean as a reference, but also with respect to the parent 305423 soybean and the 40-3-2 soybean. This yields information whether there is a synergistic effect of both parent species.

D.7.2 Production of material for comparative assessment

Comments/Questions of the expert(s)

Comment 1

Statistical methods. For several components, the calculated "tolerance interval" includes and is considerably larger than ranges found in literature (e.g palmitic acid in Appendix 4, Table 6 pp225: tolerance interval 2.93-19.6 vs literature range 7-15.8). If the determined value of the GM falls in either one, it is concluded that there is no compositional difference with regular soy. For the example given above, 6.48 % palmitic acid of total fatty acids is considered compositionally equivalent on basis of the tolerance interval, even though this is (slightly) outside the range reported in literature. Although this is not considered to be of biological relevance, it rises the question whether the tolerance interval should not be reduced from 99% to a lower value to be an acceptable indicator of compositional equivalency.

Comment 2

The statistical analyses are not appropriated to the data and the protocol used. See the above comments. Many difference appeared between GM and non GM plant concerning composition in amino acid isoflavone and vitamins, those differences are minimized without actual argument. Table 7, differences in lectines in phytic acid are significant. So we could not agree with conclusion of page 213 of 563 of Annexe 4.
Page 306 of 563, Annexe 4: Consequences of the statement of GLP compliance are not discussed or taken into account in the conclusions.

D.7.3 Selection of material and compounds for analysis

Comments/Questions of the expert(s)

Comment 1

Although the OECD consensus document on "Compositional considerations for new varieties of soybean: key food and feed nutrients and anti-nutrients" does not prescribe the analysis of saponins, one expert has suggested to include saponins in the compositional analysis as they are present in soy in relatively high quantities [Berhow et al., 2006] and although poorly absorbed in humans [Hu et al. , 2004], they can cause bloat in ruminants [Van Haver et al.,2003] and induce enteritis in salmon [Knudsen et al., 2007).

Comment 2

It says in the dossier that heptadecanoic acid is present in vegetable oils, butter and meat, while heptadecenoic acid is present in beef, cheese and olive oil (USDA, 2006 and Pioneer data) but the reference USDA 2006 does not show figures? Pioneer data also I did not find. I checked the USDA National Nutrient Database for Standard Reference Website but did not find data on heptadecenoic acid. As this is not my field of expertise I did not lose to much time, I guess other expert might know if the values of the GM are really out of scale or not.
Anyway this follows my general opinion on this GM variety, that it can only be allowed if the two parental lines are.

Comment 3

The soybean under study increases the amount of two odd chain fatty acids. The effect of this increase on possible health effects is well motivated for these components (including reference values from literature). However, it is not clear what might be the biochemical explanation for the increase of these unusual fatty acids. It might be possible that this increase is linked to an increase of other (possibly toxic) components (see, e.g., 2-ketobutyate in Kingsbury *et al*, 2006; LaRossa *et al.*, 1987). Another link is illustrated by Bjelk and Monaco (1992) who discussed the impact of the herbicide chlorimuron on the fatty acid biosynthesis.

It would make the dossier more convincing on this point if a possible biochemical pathway is discussed/hypothesized, based on a literature survey and/or experiments (see, e.g., van der Hoeven and Steffens, 2000).

In addition, a comparison of the compositional analysis with the results of each of the parent species will yield additional information about possible synergistic effects.

Comment 4

The OECD guidelines for the assessment of genetically modified soybean were followed; The compositional data include protein, fibre, carbohydrates, fat, ash, minerals fatty acids, amino acids, vitamins, secondary metabolites and anti-nutrients.

The applicant concludes that the submitted soybean is comparable to commercial soybean, with exception of the fatty acid profile, which reflects the intended modification: a high oleic phenotype.

A review of the data confirm these conclusion with some remarks concerning the fatty acid composition.

Proximate analysis:
In the proximate analysis the applicant has used the crude fibre approach, in line with the OECD guidelines. It has been remarked that modern approaches for human food studies use the dietary fibre approach, and that it may be recommendable to amend the OECD guidelines on this point.

Minerals
Important minerals are well covered in the analysis. No significant differences have been identified for most minerals. A significant difference in iron content was not confirmed at the across location level. Calcium values remain significantly different even at the across location level, but within literature range.
The observations with respect to iron, but particularly calcium, deserve attention.
Iron deficiency is very known in humans and is identified as a problem for particular vulnerable groups. As the value is within literature data the level is however not alarming.
Calcium is important in human nutrition and a mineral of concern for particular consumer groups. In case soybeans are used for the manufacture of soy drinks and other dairy analogues, calcium is generally a limiting mineral. Enrichment with calcium is however quite often applied. A further reduction of natural calcium levels has to be taken into account in the enrichment process. In case no calcium addition is performed the phenomenon deserves particular attention.

Fatty acids
The fatty acid analysis has confirmed the expected changes in the fatty acid profile: high in oleic acid and low in linoleic and linolenic acid. On top of that other changes in the fatty acid profile have been shown: some decrease in palmetic acid and some increases in for instance heptadecanoic acid and heptadecenoic acid.

The dossier is well motivated with regard to the possible health effects of these changes in the fatty acid profile (including reference values from literature). However, it is not clear what might be the biochemical explanation for the increase of these unusual fatty acids. It might be possible that this increase is linked to an increase of other (possibly toxic) components (see, e.g., 2-ketobutyate in Kingsbury *et al*, 2006; LaRossa *et al.*, 1987). Another link is illustrated by Bjelk and Monaco (1992) who discussed the impact of the herbicide chlorimuron on the fatty acid biosynthesis.

It would make the dossier more convincing on this point if a possible biochemical pathway is discussed/hypothesized, based on a literature survey and/or experiments (see, e.g., van der Hoeven and Steffens, 2000).

Amino acids, vitamins, oligosaccharides, secondary metabolites and anti-nutrients are well covered and do not raise any further questions.

D.7.4 Agronomic traits

No comment

D.7.5 Product specification

No comment

D.7.6 Effect of processing

No comment

D.7.7 Anticipated intake/extent of use

Comments/Questions of the expert(s)

It is to be expected that the high oleic acid type of soybean oil in 305423x40-3-2 will be used for other applications than soy oil also (that was one of the reasons to produce 305423). As a consequence, the statement that the "total consumption of soybean products remaining unchanged" (part II, page 100) is uncertain.
If the processed products of 305423x40-3-2 will replace a portion of similar products from commercial soybean (part II, page 100), the intake of alphalinolenic acid is expected to decrease. This is not beneficial from a nutritional point of view. Indeed, dietary intake of this essential fatty acid is lower than recommended (e.g. Sioen 2007).

D.7.8 Toxicology

D. 7.8.1 Safety assessment of newly expressed proteins

Comments/Questions of the expert(s)

In this section, the safety is discussed for the two proteins separately (GM-HRA and CP4 EPSPS), by using toxicity studies, sequence homology, presence in human food, … However, there is no discussion, of the possible synergistic effect when combining both modifications in one species.

D.7.8.2 Testing of new constituents other than proteins

No comment

D.7.8.3 Information on natural food and feed constituents

No comment

D.7.8.4 Testing of the whole GM food/feed

No comment

D.7.9 Allergenicity

Comments/Questions of the expert(s)

Assessment of allergenicity of the whole GM plant.
It must be emphasized that in the application for authorisation of genetically modified 305423 soybean and derived food and feed under Regulation (EC) N° 1829/2003 (application EFSA-GMO-NL-2007-45) to which it is referred in the present application, the number of soybean-sensitive sera used was considered as too limited. It is important that the GM plant is not more allergenic than the natural counterpart. Sera of soy allergic patients can easily be found. It is recommended that at least 20 sera be used, in order to get a broader range of reactivity patterns. In addition, the sera must not be pooled, as some information (for example the visualisation of the emergence of an allergen for some patients) might be diluted and lost in a pool.
One cannot conclude, at the moment, about the allergenicity of 305423x40-3-2 soybean.

D.7.10 Nutritional assessment of GM food/feed

Comments/Questions of the expert(s)

Soy lecithin is one of the secondary products of processed soybean. Soy phospholipids can be used for the oxidative stabilisation of oils and fats (e.g. Murano Y et al., 2008). No data are given on the fatty acid composition of lecithin derived from 305423 soybeans which is possibly (and probably) also affected. If so, this may affect the possibility of soy phospholipids to improve the lipid profile favourably (Evans et al., 2007) especially considering that soy phospholipids containing 2 molecules of linoleic acid (this fatty acid is greatly reduced in soy 305423x40-3-2 oil; no data are given on phospholipids) are most active in promoting apoAI secretion (Pandy NR et al., 2008).
Are data available on the fatty acid composition of phospholipids derived from 305423x40-3-2 soybeans?

D.7.11 Post-market monitoring of GM food/feed

No comment

D.8. MECHANISM OF INTERACTION BETWEEN THE GM PLANT AND TARGET ORGANISMS (IF APPLICABLE)

Not applicable

D.9. POTENTIAL CHANGES IN THE INTERACTIONS BETWEEN THE GM PLANT WITH THE BIOTIC ENVIRONMENT RESULTING FROM THE GENETIC MODIFICATION

D.9.1. Persistence and invasiveness

No comment

D.9.2 Selective advantage or disadvantage

No comment

D.9.3 Potential for gene transfer

No comment

D.9.4 Interactions between the GM plant and target organism

No comment

D.9.5 Interactions of the GM plant with non-target organism

No comment

D.9.6 Effects on human health

No comment

D.9.7 Effects on animal health

No comment

D.9.8 Effects on biogeochemical processes

No comment

D.9.9 Impacts of the specific cultivation, management and harvesting techniques

Not applicable.

D.10. POTENTIAL INTERACTIONS WITH THE ABIOTIC ENVIRONMENT

Not applicable.

D.11. ENVIRONMENTAL MONITORING PLAN

D.11.1 General

No comment

D.11.2 Interplay between environmental risk assessment and monitoring

No comment

D.11.3 Case-specific GM plant monitoring

No comment

D.11.4 General surveillance of the impact of the GM plant

No comment

D.11.5 Reporting the results of monitoring

No comment

References

see document BAC_2008_752 in annex