Import and processing of genetically modified cotton GHB614

COGEM Advice CGM/080509-01

Summary

COGEM has been asked to advice on an application concerning the import and processing for use in food and feed of the genetically modified cotton line GHB614. Cultivation is not part of this application.

The cotton line GHB614 was produced by Agrobacterium-mediated transformation and contains the 2mepsps gene. As a result, this line is tolerant to glyphosate-containing herbicides.

In Europe, no wild relatives of cotton are present and modern cotton cultivars do not possess any of the attributes commonly associated with problematic weeds. There are no reasons to assume that the gene inserted will increase the potential of cotton to establish feral populations. Moreover, cotton cannot survive the climatological conditions in Northwest Europe. Without irrigation, cotton volunteers cannot survive and establish themselves in the wild. Therefore, COGEM is of the opinion that incidental spillage of seeds of this cotton line will not pose a risk to the environment in the Netherlands nor in Northwest Europe.

Furthermore, in the opinion of COGEM the molecular characterisation is complete and performed adequately.

In view of these considerations, COGEM concludes that the import and processing of cotton line GHB614 poses a negligible risk to the environment in the Netherlands.

Introduction

The present application by Bayer Cropscience concerns the import and processing of the genetically modified cotton line GHB614 for use in food and feed. Cotton is mainly cultivated for the use of cotton lint. Cottonseeds are harvested as rest products and used as feed, or for the production of cottonseed oil for human consumption. Genetically modified cotton line GHB614 expresses the *2mepsps* gene which confers tolerance to glyphosate-containing herbicides.

Aspects of the crop

Cotton is a member of the genus *Gossypium* and belongs to the *Malvacea* family (1). More than 95% of commercial cotton is upland cotton, *G. hirsutum*, while long staple cotton, *G. barbadense*, occupies a small area of less then 5% (2).

Major producers of seed cotton and lint are China, the United States of America, India, Pakistan, Brazil and Turkey. Together, these countries are responsible for 80% of the total cotton production (3). Within the European Union, cotton is mainly grown in

Greece and on a smaller scale in Spain and Bulgaria (4). It should be noted that only non-genetically modified (gm) cotton is grown in Europe.

Depending on cultivar and climate, the growth period can range from 160 to 220 days. The crop will flower about eight weeks after planting. In the following two months, a cottonboll will develop and will finally open. About eight weeks later, the cotton fibers have reached full length and cellulose content and the cotton can be harvested (5).

Cotton is highly sensitive to temperature. It does not start its vegetative activity until the temperature reaches 15°C and the activity is delayed when the temperature rises above 38°C. For normal development, cotton needs an average of 150 days with temperatures between these values (6). The optimum temperature for germination is 34°C, for growth of seedlings 24-29°C and for later continuous growth 34°C. When the crop is grown at lower temperatures, the production of the vegetative branches increases and the cropping period will be extended. Reduced light intensity will retard flowering and fruiting. Because cotton is susceptible to frost, the whole growth period of six months has to be free of frost (2,6).

In areas where the rainfall is less then 500 mm a year, irrigation should be applied (7). In places where cotton is grown as a rain-fed crop, the average rainfall is 800-1200 mm (5).

Cultivated cotton is predominantly a self-pollinating species. But the prevalence of insects strongly influences outcrossing rates for cotton. Many field-based assessments estimate out-crossing rates at 10% or less, although rates up to 80% have been found. The pollen remains viable up to a period of twelve hours (7).

Cotton has no wild-relatives in Europe (2). Cotton is a domesticated crop and modern cotton cultivars do not possess any of the attributes commonly associated with problematic weeds, such as dormancy, persistence in soil banks, germination under adverse environmental conditions, rapid vegetative growth, a short life cycle, very high seed output, high seed dispersal and long-distance dispersal of seeds (7). Cotton volunteers occur in cotton growing areas and are relatively common where cotton seed is used as lifestock feed. There is no indication, however, that these volunteers establish feral populations. Seeds that do not germinate are likely to be removed by seed predators or rot, rather than become incorporated into a persistent soil seed bank (7).

Molecular characterisation

Cotton line GHB614 was produced by *Agrobacterium*-mediated transformation using the vector pTEM2. This vector contains the following elements:

- Ph4a748At, constitutive promoter derived from Arabidopsis thaliana
- TPotpC, chloroplast transit peptide (CTP), based on CTP sequences from sunflower (*Helianthus annuus*) and maize (*Zea mays*)
- Intron1 h3At, intron from A. thaliana

- *2mepsps* gene, modified 5-enolpyruvylshikimate-3-phosphate synthase gene originally derived from *Z. mays*
- 3'histon At, terminator derived from A. thaliana

Properties of the introduced gene conferring herbicide tolerance

The cotton line GHB614 was produced by Agrobacterium-mediated transformation and contains the *2mepsps* gene, which encodes a modified 5-enolpyruvylshikimate-3-phosphate synthase 2mEPSPS protein. The *epsps* gene was originally isolated from maize (*Zea mays L.*). The modified 2mEPSPS protein differs from the wild type EPSPS enzyme by two amino acid substitutions. EPSPS is a naturally occurring enzyme involved in the biosynthesis of aromatic amino acids. In non transgenic cotton lines, glyphosate acts by binding to and inhibiting the function of naturally occurring EPSPS. Consequently, aromatic amino acids are no longer formed, leading to plant death. In contrast, 2mEPSPS is not affected by glyphosate because of a reduced binding affinity. Cotton GHB614 expresses 2mEPSPS, aquiring a high tolerance to glyphosate (9).

EPSPS proteins are active in the chloroplasts of a plant cell. The sequence encoding the chloroplast transit peptide (TPotpC) is fused to the *2mepsps* gene, resulting in the transport of the transgenic 2mEPSPS protein to the chloroplast (10).

Molecular characterisation

The applicant provided studies on the molecular analysis of genetically modified cotton line GHB614. Southern blot analysis confirms that a single copy of the insert cassette is integrated and no backbone sequences are present. The insert cassette and flanking regions have been sequenced and the applicant determined that 17 bp were deleted at the target site upon integration of the T-DNA in the genomic DNA. PCR and Southern Blot analysis demonstrate that the left and right border flanking sequences are of cotton plant origin. COGEM is of the opinion that the molecular analysis was adequate and complete.

General surveillance plan

A general surveillance plan is supplied by the applicant. COGEM is of the opinion that the applicant should describe in more detail how the general surveillance will be organized and should indicate which organizations will be involved. However, in the opinion of COGEM the surveillance plan is adequate for the Netherlands, because cotton cannot survive the North western European climate. In the specific case of the Netherlands, a more detailed general surveillance plan is therefore of less importance.

Advice

The present application concerns the import and processing for feed and food purposes of a gm cotton line. Cultivation is not part of the application. Therefore, the risk assessment focuses on the accidental spillage of cottonseeds. As stated above, cotton

plants are very sensitive to temperature. A reasonably high temperature (an average of 150 days with a temperature between 15 and 38°C) is required in all stages of development. The Dutch climate is far from ideal for growing cotton. During the warmest months (April to October), the average temperature is around 14°C. The average rainfall for spring and summer is 375 mm and is below the required 500 mm. Moreover, the frost periods during the winter make it impossible for cotton to survive and establish itself in the Netherlands.

Climate conditions in other parts of the European Union are more suitable for growing cotton. At the moment cotton is grown in Greece, Spain and Bulgaria. However, it is not to be expected that the spillage of cottonseeds in these countries leads to the establishment of feral populations because no self sustaining feral populations have been observed in Europe. All European cotton is irrigated due to shortage of rainfall in the growing season. Furthermore as stated above, modern cotton cultivars do not possess any of the attributes commonly associated with problematic weeds and there are no reasons to assume that the inserted genes will increase the potential of the cotton to run wild.

In view of the above, COGEM is of the opinion that there is no risk that incidental spillage of cottonseeds will lead to the spread of cotton within Northwest Europe. Therefore, COGEM is of the opinion that the proposed import and processing of cotton line GHB614 does poses a negligible risk for the environment in the Netherlands.

References

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