PART II

SUMMARY OF THE APPLICATION FOR THE AUTHORIZATION OF GENETICALLY MODIFIED COTTON LLCOTTON25 AND DERIVED FOOD AND FEED IN ACCORDANCE WITH REGULATION (EC) NO. 1829.2003

A. GENERAL INFORMATION

1. Details of application

a) Member State of application: The Netherlands
b) Application number: Not available at the date of application
c) Name of the product (commercial and other names): Seed of genetically modified cotton (*Gossypium hirsutum*) with tolerance to glufosinate ammonium, derived by traditional breeding methods from crosses between GM cotton transformation event LLCotton25 (OECD code ACS-GHØØ1-3) and non-GM cotton cultivars.
d) Date of acknowledgement of valid application: Not available at the date of application

2. Applicant

a) Name of applicant: Bayer CropScience GmbH
b) Address of applicant: Bayer CropScience GmbH Industriepark Höchst, K 607 D-65926 Frankfurt a.M E-mail address: info@bayercropscience.com
c) Name and address of the person established in the Community who is responsible for the placing on the market, whether it be the manufacturer, the importer or the distributor, if different from the applicant (Commission Decision 2004/204/EC Art 3(a)(ii)): LLCotton25 will be imported and processed in the EU by the same groups who import, process and distribute commodity cottonseed today.

3. Scope of the application

☒ 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 Europe (Part C of Directive 2001/18/EC)

4. Is the product being simultaneously notified within the framework of another regulation (*e.g.* Seed legislation)?

Yes ☐	No ☒
If *yes*, specify	

5. Has the GM plant been notified under Part B of Directive 2001/18/EC and/or Directive 90/220/EEC?

Yes ☒	No ☐
If *no*, refer to risk analysis data on the basis of the elements of Part B of Directive 2001/18/EC	

6. Has the GM plant or derived products been previously notified for marketing in the Community under Part C of Directive 2001/18/EC or Regulation (EC) 258/97?

Yes ☒	No ☐
If *yes*, specify: 2001/18 notification No. C/ES/04/02.	

7. Has the product been notified in a third country either previously or simultaneously?

Yes ☒	No ☐
If *yes*, specify: Authorisation granted for cultivation and commercial use in USA, and requested for cultivation and commercial use in Australia, Brazil and Mexico. Authorisations granted or requested for import and commercial use in Canada, Japan and Korea.	

8. General description of the product

a) Name of the recipient or parental plant and the intended function of the genetic modification:

The recipient plant belongs to the species, *Gossypium hirsutum* L. The genetic modification intends to confer the tolerance to the herbicide glufosinate ammonium through the genetic locus defined as LLCotton25. LibertyLink® cotton varieties are developed by traditional breeding methods from crosses between LLCotton25 and non-GM cotton adapted for planting in the temperate cotton production regions of the Americas.

Herbicide tolerance is based upon the *bar* gene, a bialaphos resistance gene, isolated from the soil microorganism, *Streptomyces hygroscopicus*. The *bar* gene encodes the production of the enzyme, Phosphinothricin-Acetyl-Transferase (PAT). The specific enzymatic action of the PAT protein is tolerance to glufosinate ammonium herbicide.

Agricultural production of cotton requires weed control, and successful weed control depends upon combinations of management practices. For temperate cotton production, farmers use the planting of weed-free seed, crop rotation to break weed cycles, precision land levelling to aid irrigation, seed bed preparation, conservation tillage programs and the application of one or more herbicides.

Growing LibertyLink cotton allows; 1) more options to rotate herbicides for weed resistance management programs, 2) control of less sensitive weeds, *i.e.* amaranths, lambsquarters…, and 3) control of currently identified biotypes of herbicide resistant weeds, thus more options for crop management, lesser impact on cotton growing areas and potential implications for soil conservation through minimum tillage practices.

b) Types of products planned to be placed on the market according to the authorisation applied for:

LLCotton25 seed will be imported, processed and distributed in the European Union for all uses as any other cotton (food, feed and industrial uses) excluding cultivation.

Cottonseed products derived from event LLCotton25 (cottonseed meal and cottonseed oil) will be imported in the EU.

c) Intended use of the product and types of users:

In the EC, cotton seed and meal are used as high protein sources especially in the dairy industry around cotton growing regions. Cottonseed oil is an important vegetable oil source. Cottonseed products derived from event LLCotton25 will be imported in the EU from the major cotton growing areas as commodity and could be used for downstream purposes as food, feed and industrial products identically to non-GM cottons.

d) Specific instructions and/or recommendations for use, storage and handling, including mandatory restrictions proposed as a condition of the authorisation applied for:

No mandatory restrictions for use, storage and handling are proposed as a condition of the authorisation. All standard practices applicable to cotton today remain adequate for the handling of glufosinate ammonium-tolerant, LLCotton25 varieties.

When genetically modified cotton is placed on the EU market (including co-mingled with non-genetically modified cotton during use, storage and handling), the corresponding batch will be labelled and handled according to the legislation in application in the EU, in particular the Regulation No. 1830/2003 (EC).

e) Any proposed packaging requirements:

Seed containing LLCotton25 will be packaged as any other cotton.

f) A proposal for labelling in accordance with Articles 13 and Articles 25 of Regulation ((EC) 1829/2003. In the case of GMOs, food and/or feed containing or consisting of GMOs, a proposal for labelling has to be included complying with the requirements of Article 4, B(6) of Regulation (EC) 1830/2003 and Annex IV of Directive 2001/18/EC: LLCotton25 does not harbour characteristics that require specific labelling. Hence, no additional labelling is proposed on top of the GM labelling requirements foreseen in regulations (EC) 1829/2003 and 1830/2003.
g) Unique identifier for the GM plant (Regulation (EC) 65/2004; does not apply to applications concerning only food and feed produced from GM plants, or containing ingredients produced from GM plants): ACS-GHØØ1-3.
h) If applicable, geographical areas within the EU to which the product is intended to be confined under the terms of the authorisation applied for. Any type of environment to which the product is unsuited: No restrictions are necessary as LLCotton25 is suitable for food, feed and industrial uses in all regions of the European Union.

9. Measures suggested by the applicant to take in case of unintended release or misuse as well as measures for disposal and treatment

The majority of imported cotton commodities will be processed products from different levels of downstream processing without the ability for natural reproduction. Viable cottonseed will be imported in small quantities only. The safety profile in terms of human and animal health and environmental impact of seeds of LLCotton25 and conventional cottons are identical and do not constitute a hazard.

The case of accidental spillage of non-processed LLCotton25 seeds, in transit or at the processing facility, has been foreseen in the post market monitoring plan (see paragraph 11.4).

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

1. Complete name

a) Family name:	*Malvaceae*
b) Genus:	*Gossypium*
c) Species:	*hirsutum*
d) Subspecies:	Not applicable.
e) Cultivar/breeding line or strain:	Coker312
f) Common name:	cotton

2 a. Information concerning reproduction

(i) Mode(s) of reproduction

Vegetative proliferation of cotton requires human intervention; therefore its mode of reproduction can be restricted to **sexual reproduction** only.

Cotton is mainly an **autogamous** species, however some degree of insect mediated **cross-pollination** may take place.

Gene flow can occur into an adjacent cotton crop, however, the rate is likely to be very low because there exists a combination of genetic, botanical, geographic and agricultural barriers to gene flow. Gene flow will not occur into wild *Gossypium* species, which are not present in Europe.

Further evidence for the limited amount of cross-pollination that occurs in cotton comes from the limited isolation distance required (30m) established for certification of hybrid plant materials by the AOSCA Handbook.

(ii) Specific factors affecting reproduction

The main abiotic environmental factors affecting cotton reproduction that also determine the areas of cotton production are of a **temperature profile**, such as a) active vegetative growth range: 15 - 38 °C, b) accumulated heat GD15.5°C need: 1,200 unit, c) number of frost free days: 200, d) rapid and consistent spring warming pattern, as well as high **light intensity.**

The **frequency of cross-pollination** varies with **the insect pollinator population** in particular with various wild bees, bumble bees (*Bombus* ssp.) and honey bees (*Apis mellifera*). All the factors reducing the density of pollinators such as the use of insecticides, or increased air humidity as the result of irrigation will essentially limit the extent of cross-pollination.

(iii) Generation time

Cotton in nature is a perennial shrub, which has been domesticated and converted to an annual crop. The generation time of cultivated cotton varies between 100 and 200 days.

2 b. Sexual compatibility with other cultivated or wild plant species

There are no identified non *G. hirsutum* plants that are sexually compatible with cultivated cotton in the EU.

Pre-zygotic, and ***post-zygotic*** **barriers** greatly limit the sexual compatibility of *G. hirsutum* with other plant species in the Gossypiae tribe. In addition plants of the *Gossypium* genus are not native to Europe. Several members of the genus are cultivated as ornamental plants (*e.g. Hibiscus rosa-sinensis*) or vegetables *(e.g. Abelmoschus esculentus*–okra), but hybridisation experiments of these species with *Gossypium sp*. failed or resulted in sterile seeds.

G. hirsutum, an allotetraploid species that combines the AADD genomes, will hybridise only with other tetraploid members of the *Gossypium* genus including *G. tomentosum, G. darwinii, G. mustelinum, G. hirsutum, G. lanceolatum, and G. barbadense*, which species are not known to have a habitat in Europe.

3. Survivability

a) Ability to form structures for survival or dormancy

Cotton is cultivated annually. Seeds are the only vegetative structure for survival. Some wild forms

may produce "hard seeds" that, upon drying, become impermeable to water and suffer delayed germination. However this trait is undesirable agronomically and has been largely eliminated from modern cultivars through breeding and selection.

Cultivated cotton does not produce seeds which can persist in the environment for long periods of time, furthermore cottonseed lacks the ability to develop dormancy.

b) Specific factors affecting survivability

The main factors affecting survivability of cotton are related to soil microclimate such as temperature and humidity. If planted in moist soil before the soil temperature reaches 15 °C, it is likely to rot.

4. Dissemination

a) Ways and extent of dissemination

The two differentiated reproductive structures suitable for dispersal of cotton genes in the environment are the seed and pollen.

- **Seed dispersal** could occur during transport, at sowing and essentially before and during harvest.
- **Pollen dispersal** studies conclude that when out-crossing occurs, it is localised around the pollen source and decreases significantly with distance. This is further exemplified by the isolation distance of 30 m which is used to assure genetic purity in breeding and seed production.

b) Specific factors affecting dissemination

Seed dispersal: Cotton seed has no structural modifications to facilitate transfer by animals. Dissemination is mainly the result of human activity.

Pollen dispersal in cotton shows a correlation with **insect prevalence**. Proximity of more attractive vegetation, climate and insect management will essentially limit the extent of cross-pollination.

5. Geographical distribution and cultivation of the plant, including the distribution in Europe of the compatible species

Plants of the tribe Gossypiae originated in the tropics and subtropics. Wild species of the tribe are extremely sensitive to photoperiod conditions and do not flower in long day-light regime, therefore they are essentially excluded from temperate climates. In spite of their origin, more than 50 % of cultivated cottons are produced in **temperate zone** above 30° Latitude N, but they also tend to be plants of the southern hemisphere.

Gossypium hirsutum in its wild form is distributed over the most arid areas of Central America and in the South and North of America, with wild populations that are rare and sporadic. Cultivated *G. hirsutum* (Upland or Mexican cotton) represents over 90 % of world-wide production besides one only "New Word" tetraploid species, *G. barbadense* (Pima or South American cotton) and two "Old Word" diploid species: *G. arboreum* and *G. herbaceum*. Main cotton producers are China, USA, India, Pakistan, Uzbekistan, Brazil and Turkey.

In Europe, the cultivated cotton is *G. hirsutum*. No wild relatives have been reported.

6. In the case of plant species not normally grown in the Member State(s), description of the natural habitat of the plant, including information on natural predators, parasites, competitors and symbionts

Cotton is commercially grown in **Greece**, **Spain** and **Portugal**.

7. Other potential interactions, relevant to the GM plant, of the plant with organisms in the ecosystem where it is usually grown, or used elsewhere, including information on toxic effects on humans, animals and other organisms

Cotton is known to interact with other organisms in the ecosystem including a range of **beneficial and pestiferous arthropods, bacteria, fungi, surrounding weed species, animals and humans**. The crop has been cultivated in Spain and Greece for decades and has a history of safe use.

The cotton crop was produced for fibre for thousands of years, in the 20th century it turned to food/feed channels. Cotton is not considered harmful or pathogenic to animals or humans, however the plant does produce a small amount of natural antinutritional factors such as **gossypol, cyclopropenoid fatty acids** and **phytic acid.**

With the exception of phytic acid, all the anti-nutritionals are subject to neutralisation during processing. Free gossypol bounds to lysine and other products, and then becomes unavailable to animals. Cyclopropenoid fatty acids are deactivated or removed from the oil by hydrogenation or during deodorization at 230-235°C.

C. INFORMATION RELATING TO THE GENETIC MODIFICATION

1. Description of the methods used for the genetic modification

The genetic modification was performed by *Agrobacterium* mediated introduction of the chimeric gene denoted as P*35S*::*bar*::3'*nos*.

2. Nature and source of the vector used

Plasmid pGSV71 is a derivative of pGSV1 (itself a derivative of the vector pBR322), which was constructed in *Escherichia coli*, and thereafter transferred to a suitable *Agrobacterium tumefaciens* strain.

3. Source of donor DNA, size and intended function of each constituent fragment of the region intended for insertion

The genetic elements to be transferred into the plant are described in Table 1.

Table 1. Size, source and intended function of each constituent fragment of the region intended for insertion

Source	Approximate Size (Kb)	Reference	Intended function
Right border repeat from the TL-DNA from pTiB6S3	0.03	Gielen *et al*., 1984	None, remaining part of the vector
Synthetic	0.03		Polylinker sequence
P*35S*: promoter region from the Cauliflower Mosaic Virus	1.39	Odell *et al*., 1985	Regulatory sequence for high level constitutive expression in the plant
bar: the coding sequence of the bialaphos resistance gene of *Streptomyces hygroscopicus.*	0.55	Thompson *et al*., 1987	Herbicide tolerance and selectable marker
Synthetic	0.02		Polylinker sequence
3'*nos*: a *Taq*I fragment from the 3' untranslated end of the nopaline synthase gene from the T-DNA of pTiT37	0.26	Depicker *et al*., 1982	Polyadenylation signal
Synthetic	0.05		Polylinker sequence
Left border repeat from the TL-DNA from pTiB6S3	0.03	Gielen *et al*., 1984	None; remaining part of the vector

Depicker A., Stachel S., Dhaese P., Zambryski P., Goodman H.M. 1982. Nopaline synthase: transcript mapping and DNA sequence. Journal of Molecular and Applied Genetics, 1, 561-573.

Gielen J., De Beuckeleer M., Seurinck J., Deboeck F., De Greve H., Lemmers M., Van Montagu M., Schell J. 1984. The complete nucleotide sequence of the TL-DNA of the *Agrobacterium tumefaciens* plasmid pTiAch5. The EMBO Journal 3, 835-846.

Odell J.T., Nagy F., Chua N.-H. 1985. Identification of DNA sequences required for activity of the Cauliflower Mosaic Virus 35S promoter. Nature 313, 810-812.

Thompson C.J., Rao Movva N., Tizard R., Crameri R., Davies J., Lauwereys M., Botterman J. 1987. Characterization of the herbicide resistance gene *bar* from *Streptomyces hygroscopicus*. The EMBO Journal 6, 2519-2523.

D. INFORMATION RELATING TO THE GM PLANT

1. Description of the trait(s) and characteristics which have been introduced or modified

All LibertyLink® crops are tolerant to commercial herbicides containing glufosinate ammonium (active form is L-glufosinate). Their herbicide tolerance is based upon the naturally occurring *bar* gene, isolated from soil microbes that produce L-phosphinothricin, a bacterial metabolite with antimicrobal and herbicidal activity. Glufosinate ammonium is the synthetic salt of this natural herbicide. Activity of the *bar* gene protects the microbe as it makes L-phosphinothricin. In a similar manner, expression of the *bar* gene in plants allows survival after a foliar spray with glufosinate ammonium herbicide. The *bar* gene codes for the enzyme phosphinothricin-acetyl-transferase that acetylates L-phosphinothricin (also known as L-glufosinate) to an inactive form. The PAT protein is a highly specific enzyme with only this one function. If left in its L-isomer form, phosphinothricin disrupts the normal process of amino acid synthesis and results in a lethal build-up of ammonium in the microbe or plant cell. In a manner not unlike an inadvertent over-fertilisation of a plant, glufosinate ammonium herbicides cause sensitive plants to release internal ammonia, leading to rapid plant death.

Cotton varieties with the genetic insertion LLCotton25 make the PAT protein mainly in their green leaf tissue. When sprayed with glufosinate ammonium herbicides, the LLCotton25 plants can continue to grow while surrounding weeds rapidly die.

Several formulations of glufosinate ammonium are commercially used in many regions of the world. Registered trade names include Liberty®, Ignite®, Finale® and Basta®. Registered uses in Europe include non-selective weed control in the floor of orchards and vineyards and desiccation of potatoes and oilseed rape prior to harvesting. LibertyLink® crops currently on the market in certain areas include varieties of corn, cotton, canola and soybean. None of them are cultivated in the European Union.

2. Information on the sequences actually inserted or deleted

a) The copy number of all detectable inserts, both complete and partial

Southern blot, PCR and sequence analysis demonstrated that the glufosinate ammonium-tolerant, cotton event LLCotton25 contains one copy of the *bar* gene.

b) In case of deletion(s), size and function of the deleted region(s)

Not relevant. No deletion occurred.

c) Chromosomal location(s) of insert(s) (nucleus, chloroplasts, mitochondria, or maintained in a non-integrated form), and methods for its determination

Based upon Southern blot and genetic segregation analysis, it was demonstrated that the DNA has integrated in a single genetic locus in the cotton nuclear genome (chromosome).

d) The organisation of the inserted genetic material at the insertion site

The characterisation of the inserted sequences in event LLCotton25 confirmed the presence of one copy of the *bar* gene cassette, and also the absence of vector backbone. There are no antibiotic resistance markers present in LLCotton25.

3. Information on the expression of the insert

a) Information on developmental expression of the insert during the life cycle of the plant

The amount of PAT protein in the leaves of LLCotton25 during the vegetative life cycle of the plant has an upper limit of approximately 130 µg/g fresh weight. The amount of PAT protein in seed is 70 µg/g fresh weight.

b) Parts of the plant where the insert is expressed

Linked to the plant promoter, *35S*, the expression of the *bar* gene is targeted to green tissue of the plant. Expression level was measured by PAT protein specific ELISA. It was found that PAT protein constituted 8 µg/g fresh weight of roots, 37 µg/g fresh weight of stems and 53 µg/g fresh weight of leaves. PAT protein comprises an average of 0.08, 0.23 and 0.19% of the total crude protein in roots, stems and leaves respectively, of cotton event LLCotton25. ELISA reactive PAT protein was not found in the non-transgenic control cotton organs. The limit of detection of the assay for the different matrices was 6.4 ng/g for roots, 14.7 ng/g for stems and 8.0 ng/g for leaves. Tissue samples were harvested from greenhouse grown cotton at the 2-4 leaf stage of growth.

From published experience with the 35S promoter in cotton, LLCotton25 plants were expected to show high levels of PAT protein in the leaves, and lesser amounts in the other organs. Indeed, we found the following order of PAT expression : leaf >> stem >>> roots >>> seed, pollen.

4. Information on how the GM plant differs from the recipient plant in

a) Reproduction

The trait of herbicide tolerance had no effect as the mode and rate of reproduction are by seed production and are the same as for conventional cotton.

b) Dissemination

Two developmental stages in cotton are susceptible for dispersal: pollen and seed. No differences in dissemination capacity have been observed between genetically modified LLCotton25 and non-genetically modified cotton. Studies show that the genetic modification did not modify the characteristics of the cotton that could impact dissemination:

- no difference in pollen characteristics including viability by vital stain, fertility in crosses as either a male or female parent;
- no difference in pollen dispersal to cultivated cotton;
- no difference in seed morphology or fecundity measured as number of seed per boll, number of seed per plant and 100 seed weight;
- no difference in germination/stand count, seedling vigour or dormancy as measured by standard laboratory cotton seed physiology tests.

c) Survivability

For cultivated cotton, survival is most determined by seed characteristics. There is no indication of changes in the seed characteristics as a result of the genetic modification.

d) Other differences

The only biologically significant difference observed in field evaluations is that cotton varieties based upon transformation event LLCotton25 are tolerant to Liberty® herbicide, active ingredient glufosinate ammonium.

5. Genetic stability of the insert and phenotypic stability of the GM plant

The trait is inherited as a single dominant gene. To demonstrate the stability of the inserted DNA, Southern blot analysis was completed for different generations grown under different environmental conditions and for crosses into different genetic backgrounds.

The isolated DNA was digested with the *Nco*I restriction enzyme, which has two recognition sites in the transgene. Probing *Nco*I restricted genomic DNA with the *bar* gene cassette showed the two expected bands in all cotton event LLCotton25 samples. These bands represent an internal fragment and the junction between the transgenic sequences at the right border and upstream plant DNA sequences, and were identical in all samples.

The resulting Southern blots demonstrate the molecular stability of the cotton event LLCotton25 at the genetic level over multiple generations, different locations, and in 6 distinctive genetic backgrounds.

Phenotypic stability was demonstrated by Mendelian inheritance.

6. Any change to the ability of the GM plant to transfer genetic material to other organisms

a) Plant to bacteria gene transfer

No aspect of the nature of the genetic elements used gives any indication that a transfer from LLCotton25 to bacteria could occur.

b) Plant to plant gene transfer

Genetic transfer possible only to cotton. There is no evidence of genetic transfer and exchange under natural conditions with organisms other than those with which cotton is able to produce fertile crosses through sexual reproduction. There are no indications that the potential for successful exchange of genetic material has changed due to the genetic modification.

7. Information on any toxic, allergenic or other harmful effects on human or animal health arising from the GM food/feed

7.1 Comparative assessment

Choice of the comparator

Compositional analysis for seed compared LLCotton25 and its parent variety, Coker312.

7.2 Production of material for comparative assessment

a) Number of locations, growing seasons, geographical spread and replicates

The geographic range included the Southern United States cotton growing regions of Arkansas, Georgia, Mississippi, Missouri, North-Carolina, Texas. Seed samples were collected from two growing seasons (2000 and 2001), 15 locations, three treatments from almost every location, and a 3-fold replication per treatment. The three treatments consisted of: a) non-transgenic cotton grown using conventional herbicide weed control, b) transgenic cotton grown using conventional herbicide weed control, and c) transgenic cotton grown with Liberty® herbicide weed control.

b) The baseline used for consideration of natural variations

Published literature was consulted to establish a range of values to be expected for each nutritional component and ranges built from values of the non-transgenic, reference variety, Coker312.

7.3 Selection of material and compounds for analysis

Bayer CropScience undertook a systematic review of the composition of the seed derived from LLCotton25. The scope of the evaluation included the seed and selected processed seed products. The components selected for compositional and nutritional analyses comprise the important nutrients of cotton. These are proximates, amino acids, fatty acids, micronutrients such as vitamins and minerals, and anti-nutrients such as gossypol, cyclopropenoid fatty acids and phytic acid. The data demonstrate that cottonseed from LLCotton25 has the same nutritional composition as its non-transgenic counterpart, and values for nutritional components fall within the range of values reported for commodities in commerce.

Cottonseed oil is a high-quality cooking oil, due to its balance in unsaturated fatty acids, and high tocopherol (vitamin E) content. The lipid profile is preserved in LLCotton25. The fatty acid levels in the transgenic cottonseed oil samples are similar to the respective conventional cottonseed oil samples and within the range reported by the literature, and the tocopherol determinations show an excellent correspondence for crude and refined-deodorised cottonseed oil samples.

Antinutritional factors common to cotton were best measured in toasted in toasted cottonseed meal and are well below acceptable levels, and similar to levels in conventional cotton.

7.4 Agronomic traits

Throughout the field testing history of LLCotton25 there were no differences observed that could be attributed to pleitropic effects of the *bar* gene insertion. Neither did LLCotton25 differ from the parent variety in nutritional, agronomic or reproductive characters. The agronomic evaluations included a detailed phenotypic analysis based upon plant variety description, agronomic performance evaluations common to yield trials, pest resistance evaluations and agronomic practice evaluations. Field studies were conducted in Brazil in the 2001-2002 and 2002-2003 seasons. The variety development program performed replicated agronomic evaluations in 2000-2001 in Alabama, Arkansas, Georgia, Louisiana, Mississippi, Missouri, North Carolina, South Carolina, Tennessee and Texas. A summary of the comparisons between LLCotton25 and its parent cotton variety, Coker312, is provided in Table 2.

There is no indication in the data of agronomic performance that LLCotton25 is unlike cotton that is currently grown and consumed.

Table 2. Summary of parameters evaluated in the comparison of varieties containing LLCotton25 and the parent cotton variety, Coker312

Characteristic	Parameters	Finding
Plant morphology using PVP standards	Germination Seedling vigour Plant height Height to node ratio Sympodia length Disease susceptibility Leaf morphology Overall plant morphology	Same as recipient variety
Reproductive traits (PVP standards)	Days to first bloom Flower morphology Boll retention Days to 50% open bolls Fertility	Same as recipient variety
Fibre quality (PVP standards)	Micronaire Fibre elongation Fibre strength Fibre length Fibre length uniformity	Same as recipient variety
Field performance (PVP standards)	Emergence Stand establishment Vigour Height Yield Rate of growth (days to 50% open bolls)	Same as recipient variety
Pest and disease resistance	Severity rating for naturally occurring pathogens	Same as recipient variety
Fecundity (PVP standards)	Seed per boll Seed index (100 seed weight)	Same as recipient variety
Dormancy	Germination rate Overwintering	Same as recipient variety
Persistence	Census of volunteers in the subsequent season	Same as recipient variety
Nutritional composition of seed	Proximates, amino acids, minerals, vitamin E, fatty acids	Same as recipient variety
Anti-nutritional components	Gossypol, cyclopropenoid fatty acids, phytic acid	Same as recipient variety

7.5 Product specification

The derived food is cottonseed oil and cottonseed linters, and the derived feed the by-products of cottonseed processing (e.g. cottonseed meal).

Glufosinate ammonium-tolerant cotton transformation event LLCotton25 has been conventionally bred into an array of varieties with adaptation to the various zones of cotton cultivation (LLCotton25 varieties). LLCotton25 varieties belong to the species, *Gossypium hirsutum* L. and are distinguished from other cotton only by tolerance to the herbicide, glufosinate ammonium, the genetic locus defined as LLCotton25 and the presence of the PAT protein.

7.6 Effect of processing

The LLCotton25 varieties are grown using the agronomic practices of the region of production, and the seed is harvested, transported, stored and processed using the same processes as cotton currently in commerce. The genetic modification was not aimed at changing the processing method.

Upon chemical analysis, the nutritional composition of whole seed and processed seed (delinted seed, lint, untoasted and toasted cottonseed meal, crude and refined cottonseed oil) were found to be equivalent to other cotton.

Processing using heat, for example cooking, high pressure steam, plus solvents, alkali treatments, degrades the PAT protein, but not the DNA. In crude cottonseed oil, the PAT protein is not detected, while DNA can be detected. In refined cottonseed oil, neither DNA nor the PAT protein can be detected.

7.7 Anticipated intake/extent of use

The intake of cottonseed oil and linters in the diet of the European Union is not anticipated to change with the introduction of LLCotton25 varieties. Cottonseed and cottonseed products derived from LLCotton25 varieties are not different in quality or nutritional composition from the cottonseed products now consumed. No change in the use patterns for cotton is anticipated. No potential dietary and nutritional impacts have been identified for cottonseed and cottonseed products derived from LLCotton25 varieties.

The *per capita* consumption of cottonseed oil for the European diet is 0.1 kg/year. The extremes of cottonseed oil consumption in the member States include 0.5 kg/person/year in Spain and 1 kg/person/year in Greece. Austria, Luxembourg, Germany and Italy do not consume any. The *per capita* consumption in Turkey is 2.9 kg/year

7.8 Toxicology

7.8.1 Safety assessment of newly expressed proteins

The PAT protein is not toxic for mammals and does not possess any of the characteristics associated with food allergens. Findings to support this conclusion include:

- The coding sequence of the *bar* gene is derived from a common soil microbe not known to be a pathogen.
- The PAT protein is quickly degraded and denatured in gastric and intestinal fluids of domestic animals and humans.
- The PAT enzyme is highly substrate specific. It acts on its target, glufosinate ammonium but it does not act on glutamate, the closest structural analogue of L-glufosinate.
- There were no adverse effects found in mice, even at a high dose level of the PAT protein, after intravenous administration.

7.8.2 Testing of new constituents other than proteins

No other constituent than the PAT protein is novel and no changes in composition of cotton were discovered by chemical analysis.

7.8.3 Information on natural food and feed constituents

Natural constituents of cotton have not been changed in LLCotton25. Extensive compositional analysis was undertaken, taking into consideration the OECD consensus document on “compositional considerations for new varieties of cotton: key food and feed nutrients and anti-nutrients”. Equivalence in the whole, linted seed was demonstrated for all proximates, fibre compounds, and the total amino acids. Good agreement between the findings for LLCotton25, the comparator and the baseline support the conclusion of compositional equivalence to cotton currently in commerce.

7.8.4 Testing of the whole GM food/feed

An animal feeding study was conducted to supplement the safety evaluation: this feeding study was performed with male broiler chickens. Poultry were selected to evaluate the effects of a feed component over an entire life span and under conditions of rapid growth, thus the assay is highly sensitive for nutritional deficiencies or toxic effects.

The broiler chicken is an economically significant and widely distributed food animal. The species used is based upon commercial practice and is very sensitive to detect differences in nutrient quality because of its rapid growth (30-fold increase in body weight over four weeks). This study showed no indications that neither the event LLCotton25 nor the transformation process itself, has adverse effects on feeding, growth or general health. Moreover, no negative impacts of the nutritional quality of the event LLCotton25 were observed on poultry.

7.9 Allergenicity

7.9.1 Assessment of allergenicity of the newly expressed protein

The PAT protein does not possess any of the characteristics associated with food allergens.

The PAT protein has no homology with any known allergens, toxins or antinutrients.

The PAT protein has no glycosylation sites present on certain food allergens.

The PAT protein forms only an extremely minor part of the crude protein fraction in LLCotton25, making it unlikely to become a food allergen, which tend to be major proteins.

7.9.2 Assessment of allergenicity of the whole GM plant or crop

Cotton (*Gossypium hirsutum* L.) is not considered an allergenic food.

Plants are known to naturally produce toxins and allergens that often serve the plant as natural defense compounds against pests and pathogens. The inclusion of cottonseed products in human food or animal feed is limited due to the presence of some anti-nutrients in cottonseed that could act as toxic compounds. These anti-nutritional and toxic factors are gossypol, cyclopropenoid fatty acids (CPFA) and phytic acid. Gossypol and phytic acid are present in the meal and the seed. Thus, the cottonseed is processed to reduce the content of gossypol and CPFA to acceptable levels as well as to minimise the toxicological properties of these two compounds

Cottonseed oil intended for human consumption is highly purified: the purification process substantially reduces the content of CPFA and gossypol. Therefore, cottonseed oil and meal are currently considered not to contain common food toxins or anti-nutritional components of concern for human and animal health, because either the product only has minor amounts of these active compounds or their levels decrease (or they even disappear) during processing.

A consideration of specific food safety issues did not identify food allergenic potential as one outcome that would cause concern for human consumption. Edible oils that are refined, bleached and deodorised do not appear to pose a risk to allergic individuals, as they contain virtually no proteins. Literature to date on cottonseed oil validates this theory: the absence of water-soluble allergens in cottonseed oil is correlated with no clinical allergy observations after consumption of cottonseed oil. Therefore, no allergic reaction is expected from its current use pattern.

7.10 Nutritional assessment of GM food/feed

7.10.1 Nutritional assessment of GM food

The introduced trait in LLCotton25 is intended for agronomic benefits. Extensive compositional analysis was undertaken, taking into consideration the OECD consensus document on "compositional considerations for new varieties of cotton: key food and feed nutrients and anti-nutrients". No change in the nutritional composition was intended and upon extensive analysis, none was found.

The primary use of cotton is for the textile industry. However the by-products of cotton ginning find many uses in human and animal diets. Compositional equivalence was demonstrated for the food proprieties of the cottonseed oil and linters. The key nutrients, fatty acids and vitamin E (tocopherol), which are the principal components of cottonseed oil, were investigated. The lipid profile is preserved in LLCotton25, and the fatty acid levels in the transgenic cottonseed oil samples are similar to the respective conventional cottonseed oil samples and within the range reported by the literature.

Cottonseed oil from LLCotton25 has the same nutritional composition as its non-transgenic counterpart, and values for nutritional components fall within the range of values reported for commodities in commerce.

7.10.2 Nutritional assessment of GM feed

Extensive compositional analysis was undertaken, taking into consideration the OECD consensus document on "compositional considerations for new varieties of cotton: key food and feed nutrients and anti-nutrients". The by-products of cottonseed processing (cottonseed meal and cottonseed hulls) can be used in animal feed. Cotton contains some anti-nutritional factors, most of which are concentrated in the meal fraction. The anti-nutritional factors include gossypol, cyclopropenoid fatty acids, and phytic acid. With the exception of phytic acid, all of the anti-nutritional factors are subject to heat denaturation. Cottonseed meal is typically subjected to a moist heat treatment to facilitate oil removal. This treatment denatures proteins and detoxifies the gossypol that otherwise would cause the cottonseed meal to be improper as animal feed. Anti-nutritional factors common to cotton were best measured in toasted cottonseed meal and are well below acceptable levels, and similar to levels in conventional cotton.

7.11 Post-market monitoring of GM food/feed

No post-market monitoring plan is required for GM food/feed produced from LLCotton25. A traditional comparator, the cotton variety, Coker312l, was used in the comparative analysis (D.7.1-3). The intent of the genetic modification was for agronomic benefit (D.7.4), no change in the nutritional composition or value was intended and no change was identified (D.7.6, D.10). No health claims are intended and LLCotton25 will not be marketed as an alternative to or replacement for traditional cotton (D7.5). LLCotton25 has no specific properties that might increase the dietary intake compared to traditional cotton (D.7.7). There is no evidence that the long term nutritional and health status of the European population could be impacted by the marketing of LLCotton25 (D.7.8-10).

8. Mechanism of interaction between the GM plant and target organisms (if applicable)

Not applicable. There are no target organisms.

9. Potential changes in the interactions of the GM plant with the biotic environment resulting from the genetic modification

9.1 Persistence and invasiveness

A review of the reproductive and vegetative fitness finds that LLCotton25 compares to its parent variety Coker312 in all aspects except the tolerance to glufosinate ammonium herbicide. Subsequent season monitoring for volunteers has found no indication of increased persistence or invasiveness of LLCotton25.

9.2 Selective advantage or disadvantage

None. USDA concluded that the PAT protein does not confer a selective advantage. Agronomic performance shows no disadvantage. The only circumstance in which a selective advantage could happen would be if some plants from escaped seed would be sprayed with glufosinate ammonium. The likelihood that some escaped seed would germinate is very low because most of the imported seed is non-viable. In addition the herbicide glufosinate ammonium is not likely to be used in the vicinity of seed storage facilities, processing plants or roadways, areas where such an escape might occur.

9.3 Potential for gene transfer

Plant to bacteria gene flow. In order for any horizontal gene transfer to lead to a new type of micro-organism and therefore to introduce a significant impact, some of the following conditions will have to be fulfilled:
- the uptake should result in the incorporation of complete undegraded DNA
- the plant targeted genes should result in significant expression in a prokaryotic background
- the expression should represent a significant increase over the background level
- the traits should convey a competitive advantage to the strain in which they are incorporated.

Sequence analysis of elite event LLCotton25 confirmed (Section D.2), the insertion of one copy of the *bar* gene cassette only and also the absence of vector backbone sequences. LLCotton25 does not contain either an origin of replication from plasmid pGSV71 or any sequences responsible for an enhanced frequency of recombination. Furthermore the introduced *bar* gene is under the control of the 35S promoter, which is not functional in bacteria. Considered altogether, these facts make the possibility of gene transfer from plants of LLCotton25 to bacteria to be unlikely.

Plant to plant gene flow. Gene flow to other crop cotton is possible in cotton producing areas of Europe. Studies find the potential to be small. Impacts of outcrossing to other cultivated cotton can be managed with modest isolation distances in commercial production.

Likelihood of gene flow. Gene flow can occur into an adjacent cotton crop, however, the rate is likely to be very low because there exists a combination of genetic, botanical, geographic and agricultural barriers to gene flow. Gene flow will not occur into wild *Gossypium* species, which are not present in Europe. Measurement of natural cross pollination from LLCotton25 to cultivated cotton found the rate of outcrossing to be the same as other cotton, about 0.3% between plants at distances between 1 and 12 meters.

The only foreseeable chance for LLCotton25 to outcross to cotton in Europe would be the unlikely case of imported seed spilled in transit, if plants established within 12 meters of cultivated cotton.

Consequence of gene flow. The transfer of the *bar* gene into cultivated cotton will not exacerbate problems of weed control or adversely impact agriculture. Glufosinate ammonium is used mainly in agricultural areas in Europe, and the weed management of roadsides and the yards of processing facilities based on the use of glufosinate ammonium is not in practice.

Nevertheless LLCotton25 is not intended to be grown in Europe and thus, this risk is only hypothetical. No adverse impact to biodiversity was identified.

9.4 Interactions between the GM plant and target organisms

The introduced trait is not a pesticidal trait. There are no target organisms.

9.5 Interactions of the GM plant with non-target organisms

Three possible interactions with other organisms were examined. The genetic modification, tolerance to the herbicide, glufosinate ammonium, did not change the interaction of GM cotton varieties with other organisms in the absence of herbicide application. Under agricultural conditions when the herbicide is used: i.) some advantage may be gained in plant population dynamics; ii.) in habitats outside agriculture, the interaction with other plant communities is like any other cotton; iii.) no changes could be identified in interactions with non-target organisms in the environments under which glufosinate ammonium tolerant cotton will be cultivated (USA and Brazil). Under agricultural conditions, with direct comparisons of herbicide application, insect population diversity and measures of sensitivity to natural pathogens of cotton found no advantage for the transgenic event LLCotton25.

a) Effects on biodiversity in the area of cultivation
Under pressure of selection in an area treated with glufosinate ammonium, LLCotton25 may establish in the environment and, thereby, modify the biodiversity. Furthermore it might transfer the trait via pollen flow to other cultivated cotton (wild relatives of cotton are not found in Europe) in the vicinity and contribute to their establishment and modification of the biodiversity too. However extensive environmental risk assessment has been carried out with LLCotton25 in various countries and approvals have been granted/are anticipated in the USA, Brazil, Australia and Mexico. Moreover the scope of the present application does not include cultivation in Europe and is limited to “import and processing” in EU of LLCotton25.

b) Effects on biodiversity in other habitats
LLCotton25 will be imported as mostly non-viable seed. Therefore the likelihood that some imported seed could escape from silos or lorries and germinate is very low. The very rare LLCotton25 plants that would germinate only have a selective advantage in those cases where the herbicide glufosinate ammonium is used. In all other cases, the likelihood to establish a feral population of LLCotton25 is not higher than conventional cotton.

c) Effects on non-target organisms
There are no non-target organisms specific to LLCotton25 compared to non-genetically modified cotton. There are no observed effects of the herbicide-tolerant cotton on non-target organisms. Field observations found no differences in insect populations, or reactions to natural infestation of cotton pathogens.

9.6 Effects on human health

No effects on human health are indicated for people working with, coming into contact with or in the vicinity of an environmental release of LLCotton25. Cotton seed of LLCotton25 has the same nutritional quality as cotton in commerce. The plants of LLCotton25 have the same qualities as other cotton. No toxic or allergic effect from handling LLCotton25 has been observed on workers in the field since 1999, year of its first field release.

9.7 Effects on animal health

The primary use of cotton is for its lint; however cotton seed and the by-products of cotton processing are often included in animal diets. The nutritional composition of the seed was demonstrated to be equivalent to other cotton by chemical analysis.

To support the finding of nutritional equivalence and to demonstrate bioavailability, poultry were fed diets containing cotton under study conditions designed to evaluate growth and health parameters. Poultry were selected to evaluate the effects of a feed component over an entire life span and under conditions of very rapid growth, thus the assay is highly sensitive for nutritional deficiencies or toxic effects. No differences were identified for nutritive value of the seed and no indications of toxic or adverse effects were associated with any of the sources of cotton in the tested animal species.

Cottonseed of transformation event LLCotton25 is not anti-nutritional or toxic for animals and no effects on animal health are expected.

9.8 Effects on biogeochemical processes

Potential effects on biogeochemistry were assessed indirectly in agronomic studies designed to identify best agronomic practices for growing glufosinate ammonium-tolerant cotton. For example, studies to evaluate the fitness of the event found cotton varieties containing the transformation event, LLCotton25 are not different in seed or lint yield response to soil composition than comparable cotton varieties.

Chemical analysis of the components seed and lint found no differences in the mineral composition and thus no reason to consider mineral utilisation from the soil to be different than for conventional cotton.

Moreover the scope of the present application does not include cultivation in Europe and is limited to "import and processing" in the EU of LLCotton25.

9.9 Impacts of the specific cultivation, management and harvesting techniques

LLCotton25 varieties will be grown in principally the United States of America (USA), Brazil, Mexico and Australia. Cottonseed produced in the USA enters the European Union (EU) by import as commodity cotton seed. Crushing, processing and consumer packaging are accomplished in the EU. No new crushing or processing activities are required for LLCotton25.

Cotton in agricultural production requires weed control and successful weed control depends upon combinations of management practices. For cotton production, farmers use the planting of weed-free seed, crop rotation to break weed cycles, precision land levelling to aid irrigation, seed bed preparation, conservation tillage programs, irrigation and the application of one or more herbicides.

Advantages for farmers provided by the Liberty® cotton system include: 1) more options to rotate herbicides for resistance management programs; 2) control of less sensitive weeds (*i.e.* amaranths, lambsquarters, sedge, barnyardgrass, foxtail); and 3) removal of difficult to control weeds (*i.e.* morning glories); thus more options for crop management, lesser impact on cotton growing areas and potential implications for soil conservation through minimum tillage practices.

Moreover the scope of the present application does not include cultivation in Europe and is limited to "import and processing" in the EU of LLCotton25.

10. Potential interactions with the abiotic environment

No interaction with the abiotic environment is foreseen that would differ from cotton now in cultivation and in commerce. Lesser soil erosion may be a benefit of the cultivation of LLCotton25 as farmers growing it will be able to practice minimum tillage and conservation tillage systems.

Moreover the scope of the present application does not include cultivation in Europe and is limited to "import and processing" in the EU of LLCotton25.

11. Environmental monitoring plan (not if application concerns only food and feed produced from GM plants, or containing ingredients produced from GM plants and if the applicant has clearly shown that environmental exposure is absent or will be at levels or in a form that does not present a risk to other living organisms or the abiotic environment)

11.1 General (risk assessment, background information)

The scope of this application is the import of seed derived from LLCotton25 for food, feed and industrial uses. No authorisation for growing is requested in the Member States of the European Union.

Environmental risk assessment for the import of LLCotton25 into the European Union identified no risk, however a potential adverse effect could be anticipated if pollen from LLCotton25 were to fertilise commercial cotton in European cotton production. The only foreseeable chance for LLCotton25 to outcross to cotton in Europe would be if imported seed spilled in transit, if that seed was viable and plants established within a short distance of cultivated cotton. The likelihood that viable seed could be released unintentionally into the cotton growing environment of the EU community is very small, as :

- the amount of cotton imported as viable seed is small,
- the seed imported for crushing is loaded into **sealed cargo containers** which serve as overland transport via tractor trailer or railcar to the processing facility,
- outcrossing happens only at a low frequency because of the highly self pollinating nature of cotton,
- cotton pollen have a short life time (no more than a few hours).

11.2 Interplay between environmental risk assessment and monitoring

Because there are no adverse effects identified relating to import of herbicide-tolerant LLCotton25, the resulting monitoring to perform is limited to a general surveillance of potential adverse effects, immediate or delayed, direct or indirect, of the GMO on human health and/or the environment which are not covered in the e.r.a.

11.3 Case-specific GM plant monitoring (approach, strategy, method and analysis)

Since no risk has been identified, there is no need for a case-specific monitoring plan.

11.4 General surveillance of the impact of the GM plant (approach, strategy, method and analysis)

The scope of this application is the import of seed derived from LLCotton25 for food, feed and industrial uses, no authorization for growing is requested at the moment in the Member States of the European Union. The general surveillance will be focused on those domains involved from import to crushing facilities. The identification of possible unanticipated adverse effects of the GMO on human or livestock health and/or the environment, which were not anticipated in the e.r.a., can be addressed under the general surveillance. The people and their networks participating in the surveillance plan would tend, although not exclusively, to be best suited to identify possible unanticipated adverse effects of the GMO to the receiving environment and/or human or livestock health.

Background data.

Of the current and future EU Member States that cultivate cotton; Greece, Spain and, to a low extent, Portugal and Bulgaria, only Spain and Greece have imported commodity cottonseed for crushing (potentially viable seed) in the last five years in a reasonable amount (although never higher than 1% of the national production). The potential parameters determining a possible gene transfer are: a) seed spillage in ports, processing facilities, and along transit routes. However these sites probably do not provide an opportunity for feral cotton populations to establish and are generally far removed from cotton cultivation, and seed spillage is practically impossible due to the use of sealed containers b) the application of glufosinate ammonium herbicide, which could give

LLCotton25 plants an advantage, c) outcrossing happens only at a low frequency because of the self pollinating nature of cotton and d) cotton pollen have a short life time.

Thus, if cotton were to spill at a port or along a roadside or at a crushing facility, it is very unlikely it would establish a feral population, or that it would outcross to commercial cotton. If LLCotton25 were to spill in one of these environments, the result would be the same as for any other cotton. The introduced trait of tolerance to the herbicide glufosinate ammonium would not provide a survival advantage, as long as glufosinate ammonium is not applied in these environments.

Parameters to evaluate

Different parameters influence the possible occurrence and/or establishment of feral cotton populations. Outcrossing possibilities and pollen viability are already discussed in detail in other papers and do not need to be repeated here. Remaining parameters to be used are: a) accidental spillage in ports, along transit routes and around crushing facilities, b) occurrence and/or establishment of feral LibertyLink populations of cultivated cotton, and c) usage frequency of glufosinate ammonium in harbours, along transport routes and around crushing facilities.

Implementing general surveillance

Upon approval of LLCotton25 in the EU, Bayer CropScience will ensure that awareness of the GM crop is made widely available through stakeholders by providing key information, and will invite these stakeholders to participate in general surveillance:

- Inform European operators, especially traders and processors of bulk mixtures of cotton seed (grain), that LLCotton25 has been authorized in the EU for import and use thereof as any other cotton, excluding cultivation in the EU;
- Supply European operators, especially traders and processors of bulk mixtures of cotton seed, with information about LLCotton25 products and their safety in accordance with the requirements of Directive 2001/18/EC, relating to the Placing on the Market of the GM crop;
- Inform European operators involved in the import of cotton seed that labeling of products for the European market must be executed in accordance with articles 4 and 5 of Regulation (EC) 1830/2003 of the European parliament and of the Council of 22 September 2003 concerning traceability and labelling of GMO's and traceability of food and feed products produced from GMOs and amending Directive 2001/18/EC;
- Supply European operators involved in the import of cotton seed with the OECD Unique Identifier Code: [LLCotton25: ACS-GHØØ1-3]
- Review with European operators involved in the import and processing of cotton seed the existing measures to minimize grain spillage and clean-up practices in the frame of good manufacturing practices and environmental management systems already in place for crushing facilities and ports in the EU;
- Invite European operators involved in the import and processing of cotton seed, to provide regular feedback on the general surveillance;
- Request from European operators involved in the import and processing of cotton seed, to report in timely fashion any unanticipated adverse effects associated with the use of the product, so that decisive (if necessary remediating) action can be taken, including risk-reducing measures;
- In addition, company experts will actively screen for information on the product which could be publicly available on the web or published in literature and to inform CA and Commission in case adverse effects will be reported, in particular with respect to human, animal and/or environmental safety;
- Provide to European operators involved in the import and processing of cotton seed, the reference to the Register: http://europa.eu.int/comm/food/food/biotechnology/authorisation/commun_register_en.htm.

Further information on the product and relevant legislation will be available from a number of sources, including industry and government websites, official registers and government publications.

11.5 Reporting the results of monitoring

Bayer CropScience propose to submit general surveillance reports on an annual basis, following the initial placing on the market (first import). A final report will be made at the end of the consent.

Indirect effects refer to a causal chain of events with an effect on human health and the environment. Observations of indirect effects might, in some cases, be delayed. Since surveillance will also include the observation of potential indirect and/or delayed effects, we propose to include a report covering potential indirect or delayed effects at the stage of re-evaluation or at the end of a given consent in the case where Bayer CropScience does not apply for a renewal. An evaluation of the need for additional, post-consent surveillance will be included in such a report.

If information that confirms an adverse effect which alters the existing risk assessment becomes available to the notifier from users or other sources, Bayer CropScience is required immediately to inform the Competent Authority which gave consent for marketing of the GM crop, and in collaboration with the Competent Authority, to evaluate the information and, if necessary, to take proportional measures necessary to protect human or livestock health and/or the environment. Bayer CropScience will submit a Report, consisting of a scientific evaluation of the potential adverse effect and a conclusion on the safety of the product. The report will also include, where appropriate, the measures that were taken to ensure the safety of human or livestock health and/or the environment.

12. Detection and event-specific identification techniques for the GM plant

A discriminating PCR (dPCR) method and control materials have been provided to the DG Joint Research Centre – Community Reference Laboratory – as defined by EU Regulation 1829/2003.

E. INFORMATION RELATING TO PREVIOUS RELEASES OF THE GM PLANT AND/OR DERIVED PRODUCTS

1. History of previous releases of the GM plant notified under Part B of the Directive 2001/18/EC and under Part B of Directive 90/220/EEC by the same notifier

a) Notification number No releases of LLCotton25 have been made under Part B until 2004; first experimental releases in the EU are expected in 2005. Notification number B/ES/04/14
b) Conclusions of post-release monitoring No release in Europe, however in the USA, no persistent volunteers that could not be managed by current agricultural practice were observed.
c) Results of the release in respect to any risk to human health and the environment (submitted to the Competent Authority according to Article 10 of Directive 2001/18/EC) No release in Europe, however in the USA, no human health or environmental risks were observed.

2. History of previous releases of the GM plant carried out outside the Community by the same notifier

a) Release country LLCotton25 has been commercially released in the USA in 2004, and grown commercially on more than 130,000 hectares. **USA (field release since 1999, no longer regulated since 2004)** Authority overseeing the releases: United States Department of Agriculture (USDA) Information on the releases at www.aphis.usda.gov/ **Brazil (field release since 2001)** Authority overseeing the releases: Comissão Técnica Nacional de Biossegurança (CTNBio) Information on the releases at http://www.mct.gov.br/ctnbio **Australia (field release since 2002)** Authority overseeing the releases: OGTR Information on the releases at http://www.ogtr.gov.au/gmorec/ir.htm
b) Authority overseeing the release See E.2.a.
c) Release site See. E.2.a
d) Aim of the release See E.2.a., Field releases for breeding and variety development, technical developments for best agronomic practices and cotton integrated pest management systems have been conducted.
e) Duration of the release The generation time for cotton from planting to harvest is 100 to 200 days.

f) Aim of post-releases monitoring Volunteer LLCotton25 plants in subsequent season.
g) Duration of post-releases monitoring One or two seasons, until no volunteers observed.
h) Conclusions of post-release monitoring Occurrence of volunteers is very infrequent and dependent upon mild conditions in the winter season.
i) Results of the release in respect to any risk to human health and the environment No risk to human health or the environment has been indicated by the field release experience.

3. Links (some of these links may be accessible only to the competent authorities of the Member States, to the Commission and to EFSA):

a) Status/process of approval The JRC websites http://gmoinfo.jrc.it /gmc_browse.asp and http://gmocrl.jrc.it/statusofdoss.htm provide publicly accessible links to up-to-date databases on the regulatory progress of notifications under Directive 2001/18/EC and Regulation (EC) No 1829/2003.
b) Assessment Report of the Competent Authority (Directive 2001/18/EC) Not yet available
c) EFSA opinion Not yet available
d) Commission Register (Commission Decision 2004/204/EC) Not yet available
e) Molecular Register of the Community Reference Laboratory/Joint Research Centre Information on detection protocols will likely be posted at www.gmo-crl.jrc.it/
f) Biosafety Clearing-House (Council Decision 2002/628/EC www.bch.biodiv.org/
g) Summary Notification Information Format (SNIF) (Council Decision 2002/812/EC) www.gmoinfo.jrc Reference notification C/ES/04/02