Summary Information Format
Roundup Ready® sugar beet
According to Council Regulation 2002/812/EC

January 16, 2003

PART 2

SUMMARY INFORMATION FORMAT
FOR PRODUCTS CONTAINING GENETICALLY MODIFIED HIGHER PLANTS
(GMHPs)

A. GENERAL INFORMATION

This application refers to the sugar beet transformation event coded T9100152. The *cp4 epsps* gene, inserted in line T9100152, confers tolerance to glyphosate, the active ingredient in Roundup® herbicide.

The sugar beet event T9100152 is substantially equivalent to conventional beet, except for the expression of the glyphosate-tolerance trait.

Different varieties of glyphosate tolerant sugar beet are developed from event T9100152, using conventional breeding methods. With respect to this application, these different varieties are pooled under the designation Roundup Ready® varieties of sugar beet (abbreviated as RR sugar beet).

1. Details of notification

a) Member State of notification

Belgium.

b) Notification number

C/BE/99/01

c) Name of the product (commercial and other names)

The product consists of seeds and plants of Roundup Ready sugar beet expressing the CP4 EPSPS protein from Agrobacterium sp. strain CP4, which confers tolerance to glyphosate, the active ingredient of Roundup herbicide.

Commercial sugar beet varieties are developed from event T9100152, using conventional breeding methods. Each commercial variety, derived from event T9100152, will have a different commercial name.

d) Date of acknowledgement of notification:

December 1998.

2. Notifiers

® Roundup is a registered trademark of Monsanto Technology LLC
® Roundup Ready is a registered trademark of Monsanto Technology LLC

a) Name of notifiers i) Monsanto Company represented by Monsanto Europe S.A

ii) Syngenta Seeds SA

b) Address of notifiers i) Avenue de Tervuren 270-272, B-1150 Brussels, Belgium

ii) 12, Chemin de l´Hobit, F-31709 Saint Sauveur, France

c) Is the notifier domestic manufacturer:

Yes.

Importer:

No.

d) In the case of an import the name and address of the manufacturer shall be given

Not applicable.

3. General description of the product

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

The recipient plant is the sugar beet breeding line A1012. Breeding line A1012 was transformed using gene technology. The resulting transformation event is termed T9100152. Event T9100152 expresses two new genes, *cp4 epsps* and *gus*. *Cp4 epsps* renders the sugar beet plant tolerant to the herbicide glyphosate. *Gus* allows a simple and inexpensive screening tool for transgenic/non-transgenic plants.

The product that is subject of this application is seeds and plants of Roundup Ready sugar beet varieties (*Beta vulgaris*), and seeds and beet of any progeny derived from line T9100152 by conventional breeding. This application includes the cultivation and use of RR sugar beet in the European Union (EU) as any other sugar beet, including feed use.

These seed and progenies, derived from line T9100152, will be tolerant to glyphosate. The marketed seed may consist of inbred or hybrid lines developed using conventional breeding methods. Seed of RR beet will be marketed as new varieties of sugar beet, and the products obtained from these beets will be introduced into commerce in a manner consistent with other varieties of sugar beet.

b) Any specific form in which the product must not be placed on the market (seeds, cut-flowers, vegetative parts, etc.) as a proposed condition of the authorisation applied for

RR sugar beet has been demonstrated to be equivalent to conventional sugar beet, apart from its tolerance to glyphosate. RR sugar beet will therefore be used in the same manner as any other sugar beet product.

c) Intended use of the product and types of users

The users are breeders, farmers and processors (sugar factories). Sugar factories process sugar (sucrose) and other products like pulp, which will be used to feed livestock, principally dairy cattle. The users will notice no differences using and processing RR sugar beet, when compared to conventional beet.

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

Identical to the conditions for other beet used for the same purposes.

e) If applicable, geographical areas within the EU to which the product is intended to be confined under the terms of the authorisation applied for

Most of EU countries grow sugar beet. RR sugar beet is intended for the areas where sugar beet is grown (2087 Kha in the EU). The main production areas are Germany (519 Kha in 1996), France (430 Kha), Italy (285 Kha), Spain (173 Kha), and the United Kingdom (170 Kha).

f) Any type of environment to which the product is unsuited

The product is suited for areas where beet is currently cultivated. Beet is a common crop in arable agricultural systems in Europe, adapted to cool and moist growing conditions.

g) Any proposed packaging requirements

RR sugar beet has been shown to be substantially equivalent to other beet in growth, yield, survival, compositional and other characteristics. Hence, packaging of seed and processed products will be similar to that used for other beet varieties.

h) Any proposed labelling requirements in addition to those required by law

Information will be provided on seed packages and in accompanying documents in order for purchasers to be fully informed about the use of RR beet varieties.

i. Seed bags and packages.

In accordance with the requirements of Annex IV of Directive 2001/18, the product will be labeled with the following words “This product contains genetically modified organisms”. Packages and bags containing the seeds will be identified as Roundup Ready to allow farmers to know they are purchasing a RR sugar beet variety.

As for any other variety, all the usual pieces of information including variety name, seed quality, seed treatment, manufacturer's name and full address, will be given on the seed package.

ii. Accompanying documents.

The Roundup Ready trademark will also be a clear link to the technical guide provided to all purchasers of Roundup Ready branded sugar beet varieties. This material will contain information on the development, mode of action and use of Roundup Ready sugar beet seed, including the use of biotechnology in its development and the necessity to use the approved herbicide formulation containing glyphosate.
Plant materials derived from RR sugar beet will be marketed in the EU in accordance with the appropriate product legislation.

i) Estimated potential demand

(i) In the Community

This is understood as demand for GM seed. The success of the RR sugar beet in terms of increase in market share of seed of the RR sugar beet, will depend on the yield of the variety. Thus, it is expected that seed of a RR variety will follow traditional market forces.

(ii*) in export markets for EC supplies*

Seeds produced in EU are normally not exported outside EU. Varieties with RR sugar beet are expected follow this trend.

j) Unique identification code(s) of the GMO(s)

OECD has issued guidance on unique identifiers (OECD, 2002). According to these guidelines the unique identifier for T9100152 (also sometimes referred to as GTSB77) would be SY-GTSB77-8.

4. <u>Has the GMHP referred to in this product been notified under Part B of Directive 2001/18/EC and/or Directive 90/220/EEC?</u>

Yes ☒ No ☐

(i) If *no*, refer to risk analysis data on the basis of the elements of Part B of Directive 2001/18/EC.

5. <u>Is the product being simultaneously notified to another Member State</u> ?

Yes ☐ No ☒

(i) If *yes*, refer to risk analysis data on the basis of the elements of Part B of Directive 2001/18/EC

<u>Has the product been notified in a third country either previously or simultaneously</u>?

Yes ☒ No ☐

The product has been notified and approved in US.

6. Has the same GMHP been previously notified for marketing in the Community?

Yes ☒ No ☐

An application was submitted to France in 1996. This application was later withdrawn.

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

RR sugar beet varieties have been shown to be substantially equivalent to other beet except for their tolerance to glyphosate. Cultivated *B. vulgaris* varieties are not invasive, are weakly competitive outside cultivated areas, and possess few weedy characteristics. Furthermore, volunteer or bolting beet plants are readily managed using current agricultural practices including herbicides (other than glyphosate), hand weeding, and cultivation. Therefore, no need for specific measures is foreseen, if there is unintended release or misuse.
The measures for waste disposal and treatment for RR beet are identical to those for other beet.

B. NATURE OF THE GMHP CONTAINED IN THE PRODUCT

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

8. Complete name

a) Family name Chenopodiaceae

b) Genus *Beta*

c) Species *Vulgaris* (2n=18)

d) Subspecies *Vulgaris*

e) Cultivar/breeding line Event T9100152 and offspring thereof

f) Common name Beet

9. a) Information concerning reproduction

(i) Mode(s) of reproduction

Beet reproduces through seed only and is highly self-incompatible. The multiplication rate ranges from 350x in seed production fields to 10,000 for a single plant under optimal conditions.

(ii) Specific factors affecting reproduction, if any

To afford maximum seed yield beet requires: 1) a good establishment of the plantlet, 2) a vernalisation period of minimum 8 weeks with temperatures below 12 °C, 3) relatively long day during the bolting period, 4) sufficient nutrients and especially sufficient water supply during flowering, 5) sufficient pollen concentration during flowering, 6) warm and dry conditions during ripening period and harvest. A deficiency of one of these factors may affect the reproduction.

(iii) Generation time

Cultivated beet is normally biennial. The reproduction cycle for beet ranges from 9 to 16 months from seedling emergence to seed maturity.

9. b) Sexual compatibility with other cultivated or wild plant species

Beta vulgaris can be crossed under natural conditions with species in the genus *Beta. B. maritima and B. macrocarpa*ca hybridize with *B. vulgaris* under natural conditions. There is no evidence that *B.vulgaris* intercrosses with members of the Chenopodiaceae family other than the *Beta* genus.

10. Survivability

a) Ability to form structures for survival or dormancy

Seed is the only survival structure, and most seed left in the upper 5 centimeters of soil will germinate if the conditions are favorable. Seed that is ploughed deeper may remain dormant until the conditions are optimal for germination. It is known that seed may remain dormant for as long as 10 years or more.

b) Specific factors affecting survivability, if any

Beet plants rarely survive in subsequent crops, and are not considered as a weed problem. Numerous factors affect the ability of beet to survive. Beet is a biennial, highly sensitive to frost and poorly competitive. Importantly, beet is sensitive to tillage and to most broadleaf herbicides commonly used in rotational crops.

11. Dissemination

a) Ways and extent of dissemination

Beet seed have no special, morphological characteristics that facilitate dissemination. Beet seed will spread over short distances by several means such as threshing,

mechanical soil preparation, heavy rainfall, and animal intervention. The only significant means of dissemination of beet seed is in seed production process where large quantities are harvested, processed, and distributed.

b) Specific factors affecting dissemination, if any

Beet seed have no special, morphological characteristics that facilitate dissemination.

12. Geographical distribution of the plant

Cultivated beet is believed to have originated in the near Orient. It is now grown in temperate and Mediterranean climate zones.

13. 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

Sugar beet has been grown in Europe for the last 200 years.

14. Potentially significant interactions of the plant with other organisms in the ecosystem where it is usually grown, including information on toxic effects on humans, animals and other organisms

Certain interactions between beet and other organisms in the environment are known. Insects, nematodes and mammals feed on the leaves and roots. Beet is also susceptible to several fungal and viral diseases. Finally, beet competes poorly with weeds for light, nutrients and water. The presence of weeds in beet production fields is known to reduce the yield of beet significantly.

15. Phenotypic and genetic traits

Beet (*Beta vulgaris*) forms a rosette of leaves and a swollen root in the first year. Beetroot phenotypes cover a large range of colour (pink, red, orange, yellow and white) and shape. Cultivated beet is a biennial crop. The biennial nature of cultivated beet can be disturbed by a cold period of a minimum of 8 weeks (5-10° C), which may induce the reproductive phase with flowering and fructification in the first year.
Beet is normally a diploid with 2n=18. Most of the commercial beet varieties are diploid or triploid hybrids. Beet is generally highly self-incompatible. It is largely wind-pollinated, with insects playing a minor role.
Sugar beet, and mostly low dry matter content varieties, is very sensitive to frost.
Except for the introduced herbicide tolerance trait, RR sugar beet is equivalent to conventional sugar beet.

INFORMATION RELATING TO THE GENETIC MODIFICATION

16. Description of the methods used for the genetic modification

A disarmed *Agrobacterium tumefaciens* plant transformation system was used to produce line T9100152. This delivery system is well documented to transfer and stably integrate transferred DNA (T-DNA) into a plant nuclear chromosome (White, 1989;

Howard *et al.*, 1990). The particular vector used is pMON17204. Syngenta Seeds, using cytoplasmic male sterility in line A1012, developed the original transformation event.

17. Nature and source of the vector used

The plant transformation vector pMON17204 is a disarmed *A. tumefaciens* binary vector containing four genes (*cp4 epsps, uidA, gox, nptII*) between the right and left borders, and a bacterial selectable marker gene (*aad*) located outside the borders.

18. Size, source [name of donor organism(s)] and intended function of each constituent fragment of the region intended for insertion

The table below describes the DNA segments intended for insertion.

Genetic Element	**Size (Kb)**	**Function**
Right Border	0.025	A 25-nucleotide sequence that acts as the initial point of DNA transfer into plant cells originally isolated from pTiT37 (Depicker *et al.*, 1982).
P-FMV	0.672	The 35S promoter from a modified figwort mosaic virus (CoMVb) used to drive expression of CP4 EPSPS and *gox* genes (Shepard *et al.*, 1987; Richins *et al.*, 1987; Gowda *et al.*, 1989; Sanger *et al.*, 1993).
AEPSPS/CTP2	0.31	The N-terminal chloroplast transit peptide sequence from the *Arabidopsis thaliana* EPSPS gene (Timko *et al.*, 1988).
Cp4 epsps	1.363	The 5-enolpyruvylshikimate-3-phosphate synthase (CP4 EPSPS) gene from *Agrobacterium* sp. strain CP4 (Padgette *et al.*, 1996).
E9 3'	0.63	The 3' end of the pea rbcS E9 gene that provides the polyadenylation sites for the CP4 EPSPS and GUS genes (Coruzzi *et al.*, 1984; Morelli *et al.*, 1985).
P-e35S	0.615	The cauliflower mosaic virus (CaMV) promoter (Odell *et al.*, 1985) with the duplicated enhancer region (Kay *et al.*, 1985) used to drive expression of the GUS and *nptII* genes.
Genetic Element	**Size (Kb)**	**Function**
UidA	1.809	The *uidA* gene from *E. coli* encoding a ß-D-glucuronidase (GUS) protein (Jefferson *et al.*, 1986).

CTP1	0.165	The N-terminus encoding the chloroplast transit peptide of the small subunit 1A of rubisco from *A. thaliana* (Timko *et al.*, 1988).
GOX	1.295	The coding sequence of the glyphosate oxidoreductase (*gox*) gene isolated from *Ochronobactrum anthropi* (Barry *et al.*, 1992).
NOS 3'	0.256	The 3' nontranslated region of the nopaline synthase gene which terminates transcription and directs polyadenylation (Fraley *et al.*, 1983).
NptII	0.795	The *nptII* gene encoding the enzyme neomycin phosphotransferase II (Beck *et al.*, 1982).
Left Border	0.025	A 25-nucleotide sequence that delimits the T-DNA transfers and acts as the endpoint of DNA transfer into plant cells. It was originally isolated from pTiA6 (Barker *et al.*, 1983).

INFORMATION RELATING TO THE GMHP

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

The introduced trait that is tolerance to the herbicide glyphosate. Event 77 also expresses the *gus* gene, which allows for the detection of the event by a colorimetric assay.

20. Information on the sequences actually inserted/deleted/modified

a) Size and structure of the insert and methods used for its characterisation, including information on any parts of the vector introduced in the GMHP or any carrier or foreign DNA remaining in the GMHP

The size and structure of the insert has been determined. The insert contains the *cp4 epsps*, *uidA*, and part of the *gox* gene; the *nptII* gene, the bacterial marker and ori's were not incorporated into the sugar beet genome. In total 8 kb was transferred to the sugar beet genome.

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

Not relevant.

c) Location of the insert in the plant cells (integrated in the chromosome, chloroplast, mitochondrion, or maintained in a non-integrated form), and methods for its determination

The trait follows the standard Mendelian segregation for single insertions. This indicates that the insert is located in the nuclear chromosomes.

d) Copy number and genetic stability of the insert

Copy number of the inserted DNA was determined using a Southern blot analysis. The Southern blot analysis revealed that event 77 carry one copy of the inserted DNA. Stability over generations was studied using Southern blot techniques. This study shows that the insert is stable over four generations.

e) In case of modifications other than insertion or deletion, describe function of the modified genetic material before and after the modification as well as direct changes in expression of genes as a result of the modification

Not relevant.

21. Information on the expression of the insert

a) Information on the expression of the insert and methods used for its characterisation

The promoters used to drive the expression of the genes are considered to be constitutive, i.e. the genes are expressed in all cells in the plant. Immunological techniques were used to measure expression.

b) Parts of the plant where the insert is expressed (e.g. roots, stem, pollen, etc.)

The promoters used to drive the expression of the genes are considered to be constitutive, i.e. the genes are expressed in all cells in the plant.

22. Information on how the GMHP differs from the recipient plant in

a) Mode(s) and/or rate of reproduction

No observation has been made that would indicate any difference in the mode or rate of reproduction.

b) Dissemination

No observation has been made that would indicate any difference in the level of dissemination.

c) Survivability

No observation has been made that would indicate any difference in the rate of survivability.

d) **Other differences**

Not relevant.

23. Potential for transfer of genetic material from the GMHP to other organisms

Beta vulgaris can be crossed under natural conditions with species in the genus *Beta. B. maritima and B. macrocarpa* can hybridise with *B. vulgaris* under natural conditions. Studies has been conducted that shows that RR sugar beet also can hybridise with these wild beets. The frequency of transfer was equal to that of conventional sugar beet. There is no evidence that *B. vulgaris* intercrosses with members of the Chenopodiaceae family other than the *Beta* genus.

24. Information on any harmful effects on human health and the environment, arising from the genetic modification

No toxic, allergenic or other harmful effects on animal health are expected from the genetic modification.

25. Information on the safety of the GMHP to animal health, where the GMHP is intended to be used in animal feedstuffs, if different from that of the recipient/parental organism(s)

The overall safety to the environment and to the animal and human health from marketing of RR sugar beet is judged to be equal to that of other beet marketed for the same purposes.

26. Mechanism of interaction between the GMHP and target organisms (if applicable), if different from that of the recipient/parental organism(s)

The glyphosate tolerance trait is intended to provide protection to the crop when Roundup herbicide is applied to control competing weeds. There is therefore no target organism.

27. Potentially significant interactions with non-target organisms, if different from the recipient or parental organism(s)

On the basis of the characterization of the introduced proteins and the compositional analyses, no specific interactions of RR sugar beet with non-target organisms are to be expected, beyond those that occur with other sugar beet varieties. Extensive observations in the field have also confirmed that there are no differences between RR beet and the non transgenic counterpart in their phenotype, susceptibility to diseases and predators, and yield, indicating that there is no alteration in the interactions with predatory or beneficial non-target organisms.

28. Description of detection and identification techniques for the GMHP, to distinguish it from the recipient or parental organism(s)

Southern blot or PCR techniques could be employed for the detection and identification of the inserted nucleotide sequences. A specific ELISA has been developed and could be used to detect the CP4 EPSPS or GUS proteins in individual plants.
An event specific detection method, based on PCR, has been developed. This detection method was submitted as part of the C/BE/99/01 dossier.

C. INFORMATION ON THE POTENTIAL ENVIRONMENTAL IMPACT FROM THE RELEASE OF THE GMHP

29. Potential environmental impact from the release or the placing on the market of GMOs (Annex II, D2 of Directive 2001/18/EC), if different from a similar release or placing on the market of the recipient or parental organism(s)

Analysis of the characteristics of RR sugar beet has shown that the likelihood of potential adverse effects on human health and the environment in the European Union, resulting from its cultivation and use as any other sugar beet, including use in animal feed but is negligible. Therefore, the overall environmental risk posed by the GMHP is also negligible, and strategies for risk management for RR sugar beet would be the same as for traditional sugar beet.

30. Potential environmental impact of the interaction between the GMHP and target organisms (if applicable), if different from that of the recipient or parental organism(s)

RR sugar beet has no target organism.

31. Possible environmental impact resulting from potential interactions with non-target organisms, if different from that of the recipient or parental organism(s)

a) Effects on biodiversity in the area of cultivation

As the trait in T9100152 is herbicide tolerance, no interaction between the GMHP and non-target organisms are identified in the area of cultivation.

b) Effects on biodiversity in other habitats

Observations during field-testing showed RR sugar beet has not become more invasive or persistent than traditional beet, and will therefore affect biodiversity like traditional sugar beet.
The shift from traditional sugar beet herbicides to glyphosate will result in increased flexibility in application and potentially more efficient weed control. What impact this may have on the biodiversity - insects and flora - was examined in a large scale experiment conducted in UK (Coghlan, 2002). The experiments suggest that the careful use of GM

technology can encourage wildlife diversity in GM crops that has been lost in conventional crops.

c) Effects on pollinators

Traditional sugar beet is not known to have effect on pollinators. On the basis of the characterization of the introduced proteins and the compositional analyses, no specific interactions of RR sugar beet with non-target organisms are to be expected, beyond those that occur with other sugar beet varieties.

d) Effects on endangered species

Traditional sugar beet is not known to have effect on endangered species. On the basis of the characterization of the introduced proteins and the compositional analyses, no specific interactions of RR sugar beet with non-target organisms are to be expected, beyond those that occur with other sugar beet varieties.

D. INFORMATION RELATING TO PREVIOUS RELEASES

32. History of previous releases 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

See question 6.

b) Conclusions of post-release monitoring

Compared to other beet fields, no unexpected event has been observed in the RR sugar beet fields.

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)

The monitoring of the trial sites did not reveal any impact of RR beet on the environment or on human health. Thus, no impact on human health and the environment has been recorded in any of the field trials with event T9100152.

33. History of previous releases carried out inside or outside the Community by the same notifier

a) Release country

Sweden, Finland, Denmark, Germany, France, Belgium, the Netherlands, UK, Ireland, Italy, Spain, USA, Canada, Czech Republic, Slovakia, Romania, Ukraine, Russia, Belarus, Moldova, Lithuania, Chile and Poland.

b) Authority overseeing the release

Ministry of Agriculture, Ministry of Health or Ministry of Environment.

c) Release site

Various.

d) Aim of the release

The aim of the releases was many-fold, including efficiency, yield, hybrid registration and impact on biodiversity.

e) Duration of the release

Most trials were one-year trials, but some were part of studies that lasted several years (e.g. SCIMAC).

f) Aim of post-releases monitoring

To find any beet plant that might establish itself from dormant seed or beet debris.

g) Duration of post-releases monitoring

One or three years, depending on the conditions in the permits.

h) Conclusions of post-release monitoring

Compared to other sugar beet fields, no unexpected event has ever been observed in the RR sugar beet trials.

i) Results of the release in respect to any risk to human health and the environment

No beet fraction, either processed or unprocessed, has entered the food/feed chain. No trial has ever resulted in any negative impact on human health or the environment.

E. INFORMATION RELATING TO THE MONITORING PLAN - IDENTIFIED TRAITS, CHARACTERISTICS AND UNCERTAINTIES RELATED TO THE GMO OR ITS INTERACTION WITH THE ENVIRONMENT THAT SHOULD BE ADDRESSED IN THE POST COMMERCIALISATION MONITORING PLAN

In part C of the dossier C/BE/99/01 (Predicted behaviour of the product) an assessment on the environmental impact of the product was performed and concluded that the introduction of RR sugar beet will have no adverse effect on the environment. In the same section, the effect of RR sugar beet on human and animal health was evaluated: compositional analyses demonstrated that RR sugar beet are substantially equivalent to other sugar beet and the expressed proteins are safe for consumption by humans and animals. It was concluded that RR sugar beet are equivalent to other sugar beet regarding human and animal health. In addition, no negative impact on human health and the environment was ever recorded in any of the field trials.

Due to the fact that the environmental and human health safety assessment for RR sugar beet has not identified any specific risks related to its placing on the market, the monitoring plan for RR sugar beet will be focused on general surveillance for unanticipated, adverse effects.

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