

Results of discussions at the Crops Subcommittee 5

Name: Herbicide glyphosate, glufosinate and dicamba tolerant sugar beet (modified cp4 epsps, pat, modified dmo, Beta vulgaris L. ssp. vulgaris var. altissima) (KWS20-1, OECD UI: KB-KWS201-6)

Contents of Type 1 Use, etc.: Cultivation, storage, transportation, and disposal in isolated fields, and acts incidental to these.

Applicant: Bayer CropScience Co., Ltd. The Agricultural Crops Subcommittee has decided to apply for Type 1 use based on the biodiversity impact assessment report submitted by the applicant.

We examined the details of the applicant's assessment regarding the impact on biodiversity when using Type 1 of this recombinant sugar beet in accordance with the regulations for use. The main items confirmed are as follows.

1 Regarding the results of biodiversity impact assessment This recombinant sugar beet contains pVS1 derived from *Pseudomonas aeruginosa* and *Escherichia*

It was created by introducing the T-DNA region of plasmid PV-BVHT527462, which was constructed based on the coli-derived plasmid pBR322, using the *Agrobacterium* method.

This recombinant sugar beet contains modified CP4 EPSPS protein derived from *Agrobacterium* CP4 strain.

One copy of an expression cassette containing the modified cp4 epsps gene encoding the PAT protein, the pat gene encoding the PAT protein derived from *Streptomyces viridochromogenes*, and the modified dmo gene encoding the KWS20-1 DMO protein derived from *Stenotrophomonas maltophilia* is integrated onto the chromosome. It has been confirmed by the segregation pattern of 25 genes and Southern blot analysis that it has been stably transmitted over multiple generations. Furthermore, it has been confirmed by Western blot analysis that the gene of interest is stably expressed over multiple generations.

(1) Competitive advantage Sugar beet was introduced to Japan around 1870, and has been cultivated in the southwestern warm region, the Tohoku region, and the North Sea.

Although it has been cultivated in the provinces, there are no reports of sugar beets growing wild in our country. This recombinant sugar beet contains modified CP4 EPSPS protein, PAT protein and modified KWS20-1.

Expression of the DMO protein confers resistance to the herbicides glyphosate, glufosinate, and dicamba. However, under natural conditions where the application of these herbicides is unlikely, it is difficult to imagine that herbicide tolerance will increase competitive advantage. Based on the above, this recombinant sugar beet has a competitive advantage within the scope of this application.

The applicant's conclusion that there is no risk of biodiversity impacts resulting from the project was judged to be appropriate.

(2) Productivity of harmful substances To date, there have been no reports that sugar beets produce harmful substances that may affect biodiversity.

This recombinant sugar beet contains a modified CP4 EPSPS protein that confers tolerance to the herbicide glyphosate, a PAT protein that confers tolerance to the herbicide glufosinate, and a modified KWS20-1 DMO that confers tolerance to the herbicide dicamba.

Although proteins are expressed, these proteins are not known to be harmful substances⁵. It was also confirmed that these proteins do not have sequences that are structurally similar to known allergens. The modified CP4 EPSPS protein is an enzyme that catalyzes the shikimate pathway for the biosynthesis of aromatic amino acids, but it is not the rate-limiting enzyme in this pathway, and even if the activity of the EPSPS protein increases, the final product of this pathway is It is thought that concentrations of certain aromatic amino acids will not increase 10 . Therefore, it is unlikely that harmful substances are produced in this recombinant sugar beet due to the modified CP4 EPSPS protein. Also,

The substrate specificity of the PAT protein and modified KWS20-1 DMO protein is extremely high, and because they do not use structurally similar plant endogenous substances as substrates, the PAT protein and modified KWS20-1 DMO protein are highly sensitive to the host. It is unlikely that it acts on the metabolic system and produces harmful substances. Based on the above, this recombinant sugar beet has a low production of harmful substances within the scope of this application.

The applicant's conclusion that there is no risk of causing any biological diversity impacts is considered to be appropriate.

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(3) Crossability Since there are no closely related wild species that can cross with this recombinant sugar beet growing in Japan,

No wild animals or plants were identified that could be affected by biodiversity due to hybridization.

Based on the above, we have determined that the applicant's conclusion that this recombinant sugar beet, within the scope of this application, is not likely to cause any impact on biodiversity due to hybridization is appropriate.

2 Conclusion of the Agricultural Products Subcommittee Based on the above, this recombinant sugar beet can be cultivated in isolated fields based on certain work procedures in a limited environment, and within the scope of activities associated with these, as well as cultivation, storage, transportation, and disposal in Japan. The conclusion of Biodiversity Impact Assessment Report 30 that there is no risk of causing any impacts on biodiversity was judged to be appropriate.