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COMMON FORMAT FOR Risk Assessment
(In accordance with Annex III of the Cartagena Protocol on Biosafety)

Risk assessment details	
1. Country Taking Decision:	South Africa
2. Title:	Application for Commodity Clearance of DAS-44406-6 in the Republic of South Africa.
3. Contact details:	Dow AgroSciences South Africa, P.O. Box 76129 Lynnwoodridge, Pretoria 0040, South Africa. Telephone: (+27) 12 361 8120; Fax (+27) 12 361 8126.
LMO information	
4. Name and identity of the living modified organism:	The LMO is DAS-44406-6. DAS-44406-6 soybean was developed using <i>Agrobacterium</i> -mediated transformation to stably incorporate the <i>aad-12</i> gene from <i>Delftia acidovorans</i> , the <i>2mepsps</i> from <i>Zea mays</i> and the <i>pat</i> gene from <i>Streptomyces viridochromogenes</i> , into soybean. The <i>aad-12</i> gene encodes the aryloxyalkanoate dioxygenase-12 (AAD-12) enzyme which, when expressed in plants, degrades 2,4-D into herbicidally-inactive 2,4-dichlorophenol (DCP). The <i>2mepsps</i> gene encodes a 5-enolpyruvylshikimate-3-phosphate synthase protein that is insensitive to glyphosate, thus providing tolerance to glyphosate in plants expressing the 2mEPSPS protein. The <i>pat</i> gene encodes the enzyme phosphinothricin acetyl transferase that inactivates glufosinate, and hence provides tolerance to glufosinate-ammonium herbicide.
5. Unique identification of the living modified organism:	DAS-44406-6
6. Transformation event:	DAS-44406-6
7. Introduced or Modified Traits:	Herbicide tolerance
8. Techniques used for modification:	Plasmid carried by <i>Agrobacterium tumefaciens</i>
9. Description of gene modification:	The disarmed <i>Agrobacterium tumefaciens</i> strain EHA101, carrying the binary vector with the <i>2mepsps aad-12</i> and <i>pat</i> genes within the T-DNA region, was used to initiate transformation. The <i>Agrobacterium tumefaciens</i> strain EHA101 carries the helper plasmid, pTiBo542. EHA101 was generated by inactivation of the T-DNA <i>onc</i> genes in strain A281.

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Characteristics of modification	
10. Vector characteristics (Annex III.9(c)):	The vector pDAB8264 is a binary one derived from <i>Agrobacterium tumefaciens</i> . <i>Agrobacterium tumefaciens</i> is a rod shaped Gram negative soil bacterium and is the causal agent of crown gall disease in over 140 species of dicot.
11. Insert or inserts (Annex III.9(d)):	<p>Three gene expression cassettes were present in the pDAB8264 vector for insertion into soybeans. The <i>aad-12</i> expression cassette is designed to express the plant-optimized aryloxyalkanoate dioxygenase (<i>aad-12</i>) gene that encodes the AAD-12 protein. The <i>aad-12</i> gene was isolated from <i>Delftia acidovorans</i> and the synthetic version of the gene was optimized to modify the G+C codon bias to a level more typical for plant expression. The <i>aad-12</i> gene encodes a protein of 293 amino acids that has a molecular weight of approximately 32 kDa. The insertion of <i>aad-12</i> into soybean plants confers tolerance to herbicides such as 2,4-D by production of the aryloxyalkanoate dioxygenase-12 enzyme (AAD-12).</p> <p>The <i>2mepsps</i> expression cassette is designed to express a double mutant maize 5-enolpyruvylshikimate-3-phosphate synthase gene that encodes the 2mEPSPS protein. The <i>2mepsps</i> gene was originally isolated from <i>Zea mays</i> and fused with an optimized chloroplast transit peptide, TPotp C, derived from maize and sunflower ribulose-1,5-bisphosphate carboxylase-oxygenase (RuBisCO) at its N-terminus. The <i>2mepsps</i> gene encodes a protein of 445 amino acids that has a molecular weight of approximately 47.5 kDa. The encoded protein is insensitive to glyphosate, thus providing tolerance to glyphosate in plants expressing the 2mEPSPS protein.</p> <p>The <i>pat</i> expression cassette is designed to express the plant-optimized phosphinothricin <i>N</i>-acetyl transferase (<i>pat</i>) gene that encodes the PAT protein. The <i>pat</i> gene was isolated from <i>Streptomyces viridochromogenes</i> and the synthetic version of the gene was optimized to modify the G+C codon bias to a level more typical for plant expression. The insertion of the <i>pat</i> gene into the soybean genome confers tolerance to glufosinate and was used as a selectable marker during the soybean transformation.</p>
Recipient organism or parental organisms (Annex III.9(a)):	
12. Taxonomic name/status of recipient organism or parental organisms:	Family; Leguminosae; Genus; <i>Glycine</i> ; Species; <i>Glycine max</i>

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13. Common name of recipient organism or parental organisms:	Soybean
14. Point of collection or acquisition of recipient or parental organisms:	Privately owned germplasm.
15. Characteristics of recipient organism or parental organisms related to biosafety:	Soybean is one of the oldest cultivated crops and a well-known crop plant worldwide. Soybean is a common source of food and feed with a centuries-long history of safe use and consumption around the world. Soybean is widely grown in South Africa and has a history of safe use. Soybean seed is however known to contain a number of natural anti-nutritional components, which are completely or partially inactivated during processing. Trypsin (proteinase) inhibitors are known to have anti-nutritive properties in animals fed unprocessed soybeans. Other anti-nutrients include lectins, stachyose and raffinose, phytoestrogens and phytate. Some of these anti-nutrients relate to their impact on human nutrition, while others relate to animal nutrition in general including livestock.
16. Centre(s) of origin of recipient organism or parental organisms:	North and Central China. Specific geographic coordinates are unknown.
17. Centres of genetic diversity, if known, of recipient organism or parental organisms:	Centers of genetic diversity of soybean are the same as its centre of origin.
18. Habitats where the recipient organism or parental organisms may persist or proliferate:	Soybean does not persist or proliferate outside of agriculture in South Africa. There are no known populations in any natural habitat in the country.

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Donor organism or organisms (Annex III.9(b)):	
19. Taxonomic name/status of donor organism(s)	<p>1. <i>Nicotiana tobacum</i>: donor of the Matrix Attachment Region RB7: Magnoliopsida; Solanales; Solanaceae; <i>Nicotiana</i></p> <p>2. <i>Arabidopsis thaliana</i>: donor of the AtUbi10 promoter and 5' untranslated region, Histone H4A748 3' UTR and Histone H4A748 promoter Rosids; Brassicales; Brassicaceae; <i>Arabidopsis</i></p> <p>3. <i>Delftia acidovorans</i>: donor of the <i>aad-12</i> gene Betaproteobacteria; Comamonadaceae; <i>Delftia</i></p> <p>4. <i>Agrobacterium tumefasciens</i>: donor of the 3' UTR of the AtuORF23 and ATUORF1 and T-DNA Border A and T-DNA Border B Alfa Proteobacteria; Rhizobiales; Rhizobiaceae; <i>Agrobacterium</i></p> <p>5. <i>Cassava vein mosaic virus</i>: donor of the CsVMV promoter <i>Caulimoviridae</i>; <i>Cavemovirus</i>; Species: Cassava vein mosaic virus</p> <p>6. <i>Streptomyces viridochromogenes</i>: donor of the <i>pat</i> gene Betaproteobacteria; Actinobacteridae; Actinomycetales; Streptomycineae; Streptomycetaceae; <i>Streptomyces</i></p> <p>7. <i>Zea mays</i>: donor of the <i>2mepsps</i> gene and TPotp C: Liliopsida; Poales; Poaceae; <i>Zea</i></p> <p>8. <i>Helianthus annuus</i>: donor of TPotp C; Asterids; Asterales; Asteraceae; <i>Helianthus</i></p>
20. Common name of donor organism(s):	Tobacco; Arabidopsis; Bacteria; Virus; Maize; Sunflower
21. Point of collection or acquisition of donor organism(s):	None of the donor organisms was collected from a specific geographical location.

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22. Characteristics of donor organism(s) related to biosafety:	<p>1. <i>Nicotiana tabacum</i>: <i>N. tabacum</i> is a native of tropical and subtropical America but it is now commercially cultivated worldwide (its leaves are commercially grown in many countries to be processed into tobacco).</p> <p>2. <i>Arabidopsis thaliana</i>: is native to Europe, Asia, and northwestern Africa. It is an annual (rarely biennial) plant usually growing to 20–25 cm tall. The small size of its genome makes <i>Arabidopsis thaliana</i> useful as a model plant for plant biology study and has been widely used safely for studies including genetic mapping and sequencing, etc.</p> <p>3. <i>Delftia acidovorans</i>: is a non glucose-fermenting, gram-negative, non spore-forming rod prevalent in soil and fresh water. Some species have also been isolated from activated sludge and clinical specimens. <i>Delftia acidovorans</i> can be used to transform ferulic acid into vanillin and related flavor metabolites. This utility has led to a history of safe use for <i>D. acidovorans</i> in the food processing industry. This strain also produces polyhydroxyalkanoates that are being developed as biomaterials for medical applications. There are limited reports of <i>D. acidovorans</i> causing infections in compromised patients. There are no reports of this strain producing any allergens.</p> <p>4. <i>Agrobacterium tumefaciens</i>: is a bacterium that is found in soils worldwide. It infects plants but does not infect humans or animals.</p> <p>5. <i>Cassava vein mosaic virus</i>: is a plant pathogenic virus that infects many crops and is therefore a commonly ingested virus which however does not infect humans or animals, or even plants outside its host range.</p> <p>6. <i>Streptomyces viridochromogenes</i>: is a common soil bacterium that produces the tripeptide L-phosphinothricyl-L-alanyl-alanine (L-PPT), which was developed as a non-selective herbicide by Hoechst Ag. It is ubiquitous in nature with no known adverse effects on human and animal health.</p> <p>7. <i>Zea mays</i>, is a major cereal crop grown for food and feed.</p> <p>8. <i>Helianthus annuus</i> has sunflower has a long history of safe use for production of edible oils and livestock feed.</p>
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Intended use and receiving environment

23. Intended use of the LMO (Annex III 9(g)):	For all uses as for any other soybean, excluding cultivation.
24. Receiving environment (Annex III.9(h)):	This is not an application for release of DAS-44406-6 into the environment of South Africa.

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Risk assessment summary

25. Detection/Identification method of the LMO (Annex III.9(f)):	PCR detection methods to confirm the molecular identity of DAS-44406-6 have been developed and are currently undergoing scientific assessment at the European Commission Joint Research Centre (EU JRC) (see http://gmo-crl.jrc.ec.europa.eu/statusofdoss.htm).
	The Institute for Reference Materials and Measurements (IRMM) is collaborating with Dow AgroSciences to develop certified reference materials for DAS-44406-6 soybean.
26. Evaluation of the likelihood of adverse effects (Annex III.8(b)):	The scope of this application does not include cultivation of DAS-44406-6 soybean in South Africa. Any exposure to the environment will be limited to any unintended release of DAS-44406-6 soybean, which could occur via accidental spillage during loading/unloading of the vessels, trains and trucks carrying the load of commodity grain, including DAS-44406-6 soybean, destined for processing into animal feed or human food products. However, this limited exposure is highly unlikely to give rise to any adverse effect and, if necessary, any spillage could be easily controlled by the application of current agricultural practices used for the control of volunteer soybean plants. Furthermore, soybean is known to be a weak competitor in the wild, which cannot survive outside cultivation without human intervention. Environmental conditions at the sites of handling are unlikely to be conducive to germination, growth and reproduction of soybean grain that is incidentally released.
27. Evaluation of the consequences (Annex III.8(c)):	In the unlikely event that some grain containing DAS-44406-6 soybean (from grain imports) may end up in the environment of South Africa before being crushed, the impact would be negligible as data supported a conclusion that DAS-44406-6 soybean is substantially equivalent to conventional soybean; soybean plants cannot survive without human intervention.
	Furthermore, South Africa is not the centre of origin for <i>G. max</i> and there are no wild relatives in South Africa with which soybean can outcross.
28. Overall risk (Annex III.8(d)):	The overall risk posed by this GMO is negligible.
29. Recommendation (Annex III.8(e)):	The overall risk is negligible. No recommendations other than procedures that may apply to conventional soybean are applicable.

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30. Actions to address uncertainty regarding the level of risk (Annex III.8(f)):	There is no uncertainty regarding the risk profile.
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Additional information	
31. Availability of detailed risk assessment information:	All data relating to this risk assessment have been presented in this dossier.
32. Any other relevant information:	To the best of our knowledge, all relevant information has been supplied in this dossier.
33. Attach document:	<i>Not applicable to applicant</i> <Specific types of entry: option to choose a file from the local source and 'upload' a copy to the BCH server>
34. Notes:	<Text entry>
