Extended Determination of Nonregulated Status for JR Simplot Company Innate™ V11 Potatoes

In response to a request from JR Simplot Company (hereinafter referred to as JR Simplot) to extend a determination of nonregulated status to JR Simplot’s V11 Snowden potatoes (hereinafter referred to as V11 potatoes) with low acrylamide potential and reduced black spot (petition number 15-140-01p), the Animal and Plant Health Inspection Service (APHIS) of the United States Department of Agriculture (USDA) has determined, based on similarity to its antecedent organisms, that V11 potatoes and progeny derived from them are unlikely to pose plant pest risks and are no longer to be considered regulated articles under APHIS’ Biotechnology Regulations (Title 7 of Code of Federal Regulations (CFR), part 340). This extension request is based upon APHIS’ determination of nonregulated status of its antecedent organisms: JR Simplot’s potato events, E12 Russet Burbank, F10 Ranger Russet and J3 Atlantic potatoes (hereinafter referred to as JR Simplot antecedent potato events), with low acrylamide potential and reduced black spot. JR Simplot antecedent potato events from petition number 13-022-01p were deregulated on November 10, 2014. APHIS-approved permits or acknowledged notifications that were previously required for environmental release, interstate movement, or importation under those regulations will no longer be required for V11 potatoes and their progeny. Importation of V11 potato seeds, other propagative material, and bulk or table stock, will still be subject to APHIS foreign quarantine notices at 7 CFR part 319 and the Federal Seed Act regulations at 7 CFR parts 201 and 361.

The same genetic construct pSIM1278, used to transform the JR Simplot antecedent potato events with low acrylamide potential and reduced black spot, was also used to transform and generate the V11 potato event. APHIS evaluated the plant pest risk of V11 potatoes by assessing its similarity to the deregulated JR Simplot antecedent potato events.

APHIS previously conducted a Plant Pest Risk Assessment on the antecedent organisms and found them unlikely to pose risks as plant pests. Based on a the plant pest similarity assessment (see Appendix A) of V11 potatoes to the antecedents, APHIS concludes that V11 potatoes are unlikely to pose a plant pest risk and should no longer be regulated under 7 CFR part 340. From the similarity assessment, APHIS concludes the following with respect to V11 potatoes and their progeny:

(1) No plant pest risk was identified from the transformation process, the insertion and/or expression of new genetic material, or from changes in metabolism in V11 potatoes.

(2) Disease and pest incidence and/or damage are not expected to be increased or atypical for V11 potatoes. No plant pest effects are expected on these or other agricultural products and no impacts are expected to APHIS pest control programs.
(3) Based on an evaluation of the gene products, and their similarity to the antecedents, V11 potatoes are unlikely to adversely impact nontarget organisms beneficial to agriculture.

(4) V11 potatoes are no more likely to become weedier or more difficult to control as a weed than the antecedents, which are not weedy.

(5) V11 potatoes are not likely to increase the weed risk potential of other species with which they can interbreed in the U.S. or its territories. Gene flow, hybridization and/or introgression of inserted genes from V11 potatoes to other sexually compatible relatives with which it can interbreed is not likely to occur.

(6) Significant changes to agricultural or cultivation practices (e.g. pesticide applications, tillage, irrigation, harvesting, etc.) from adoption of V11 potatoes are not expected.

(7) Horizontal gene transfer of the new genetic material inserted into the GE plant to other organisms is highly unlikely, and is not expected to lead directly or indirectly to disease, damage, injury or harm to plants, including the creation of new or more virulent pests, pathogens, or parasitic plants.

In addition to our finding that V11 potatoes are unlikely to pose a plant pest risk, APHIS reached a Finding of No Significant Impact (FONSI) for this action. V11 potatoes will have no significant impacts, individually or collectively, on the quality of the human environment and will have no effect on federally listed threatened or endangered species, species proposed for listing, or their designated or proposed critical habitats (http://www.aphis.usda.gov/biotechnology/not_reg.html).

Based on my review and consideration of all of the scientific and environmental data, analyses, information, and previous conclusions regarding the plant pest risk assessment for the antecedent organisms, the plant pest risk similarity assessment, the EA and FONSI, and my knowledge and experience as the APHIS Deputy Administrator for Biotechnology Regulatory Services, I have determined and decided that this determination of nonregulated status of V11 potatoes is the most scientifically sound and appropriate regulatory decision.

Michael J. Firko, Ph.D.
APHIS Deputy Administrator
Biotechnology Regulatory Services
Animal and Plant Health Inspection Service
U.S. Department of Agriculture

1/13/2014 Date
Appendix A

J.R. Simplot Company Request (15-140-01p) for Extension of Determination of Non-regulated Status of V11 Snowden Potatoes with Low Acrylamide Potential and Reduced Black Spot

OECD Unique Identifier: SPS-00V11-6

Plant Pest Risk Similarity Assessment

September 28, 2015

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This publication reports research involving pesticides. All uses of pesticides must be registered by appropriate State and/or Federal agencies before they can be recommended.
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A. Introduction

The Animal and Plant Health Inspection Service (APHIS) of the United States Department Agriculture (USDA) has received an extension request (petition number 15-140-01p) from the J.R. Simplot Company (hereafter referred to as JR Simplot).

USDA announced its determination of non-regulated status for ten potato events (petition number 13-022-01p) on November 10, 2014. These events included:

- E12 and E24—Russet Burbank varieties;
- F10 and F37—Ranger Russet varieties;
- J3, J55, and J78—Atlantic varieties; and
- G11, H37, and H50—proprietary varieties.

Presently, JR Simplot intends to pursue commercialization of the E12, F10 and J3 events. Therefore, the antecedent organisms identified in the extension request for the V11 Snowden potato event (hereafter referred to as the V11 event) are E12 Russet Burbank, F10 Ranger Russet and J3 Atlantic potatoes (hereafter referred to as JR Simplot antecedent potato events).

In accordance with §340.6(e)(2), JR Simplot requests that APHIS extend the non-regulated status for JR Simplot antecedent potato events with low acrylamide potential and reduced black spot potatoes to the genetically engineered (GE) V11 event (low acrylamide potential and reduced black spot potatoes) and any progeny derived from crosses of the V11 event with conventional potatoes, and any progeny derived from crosses of the V11 event with other GE potato varieties that have received a determination of nonregulated status, or are not considered regulated articles under regulations at 7 CFR Part 340.

APHIS regulations in 7 CFR part 340 regulate the introduction (importation, interstate movement, or release into the environment) of certain GE organisms and products. A GE organism is no longer subject to the regulatory requirements of Part 340 when APHIS determines that it is unlikely to pose a plant pest risk. A GE organism is considered a regulated article under Part 340 if the donor organism, recipient organism, or vector, or vector agent used in engineering the organism belongs to any genera or taxa designated in 7 CFR 340.2 and meets the definition of plant pest, or is an unclassified organism and/or an organism whose classification is unknown, or any product which contains such an organism, or any other organism or product altered or produced through genetic engineering which the Administrator determines is a plant pest or has reason to believe is a plant pest. The V11 event was produced by the Agrobacterium tumefaciens mediated transformation of potato internode sections (JR Simplot, 2015), and some of the introduced border sequences come from plant pest organisms listed in 7 CFR 340.2 (JR Simplot, 2015). Therefore, the V11 event is considered a regulated article under APHIS regulations at 7 CFR part 340.
Potential impacts in this Plant Pest Risk Similarity Assessment are those that pertain to plant pest risk associated with the V11 event and its progeny and their use in the absence of confinement relative to the JR Simplot antecedent potato events. APHIS utilizes data and information submitted by the applicant, in addition to current literature, to determine if the V11 event is any more likely than the JR Simplot antecedent potato events to pose a plant pest risk. APHIS specifies in 7 CFR 340.6(e) that an extension request for nonregulated status shall include information to establish the similarity of the antecedent organism to the regulated article in question.

APHIS may also consider information relevant to reviews conducted by other agencies that are part of the ‘Coordinated Framework for the Regulation of Biotechnology’(51 FR 23302, 1986; 57 FR 22984, 1992). Under the Coordinated Framework, the oversight of biotechnology-derived plants rests with APHIS, the Food and Drug Administration (FDA), and the Office of Pesticide Programs of the U.S. Environmental Protection Agency (EPA). Depending on its characteristics, certain biotechnology-derived products are subjected to review by one or more of these agencies.

B. Development of the V11 Snowden potato

In producing the antecedents, JR Simplot used genetic engineering to introduce into the background of commercial potato cultivars two traits that are of interest to potato consumers, producers and processors: reduced acrylamide potential and reduced black spot bruise (petition number 13-022-01p). V11 was produced by introducing the same genes and traits into the Snowden parent variety which is the 2nd most popular potato variety used for making potato chips. Multiple traits would be difficult to achieve through conventional breeding. Potato is tetraploid, highly heterozygous and sensitive to inbreeding depression. Each potato parent variety must be independently transformed to achieve the desired phenotype in that variety. The same genetic construct pSIM1278 used to transform the JR Simplot antecedent potato events was used to transform and generate the V11 event. The intended purpose of the JR Simplot antecedent potato events and the V11 event is to provide the potato processing industry with new potato lines with low acrylamide potential and reduced black spot bruise. Both of these changes are intended to benefit potato consumers, producers, and processors. For example, California’s Proposition 65 requires that food manufacturers warn consumers about the dangers of acrylamide in their products. The low acrylamide potential is intended to benefit consumers because of concerns about the health effects of ingesting acrylamide, and to benefit the industry relative to Proposition 65. The reduced black spot bruise is intended to benefit consumers by providing a higher quality product, to benefit producers by reducing culls at delivery, and to benefit processors by reducing pick-outs.

APHIS BRS completed a detailed plant pest risk assessment (PPRA) and environmental assessment (EA) for the JR Simplot antecedent potato events (i.e., the ten events, three of which are the antecedents associated with this PPRSA) (http://www.aphis.usda.gov/biotechnology/petitions_table_pending.shtml). The EA fully addressed all resource areas of potential concern. In the antecedent petition, 13-022-01p, APHIS concluded on the basis of the EA that the impacts would not be
significant. The agency issued Findings of No Significant Impacts (FONSI) and made
determinations of non-regulated status for each event. As of November 2014, APHIS has
deregulated ten JR Simplot GE potato events with low acrylamide potential and reduced
black spot.

C. Description of Inserted Genetic Material, Its Inheritance and Expression, Gene Products, and Changes to Plant Metabolism

To inform APHIS of the potential hazards resulting from the genetic modification and
potential routes of exposure related to the inserted DNA and its expression products,
APHIS assessed data and information presented in the extension request related to the
similarity of the V11 event to the JR Simplot antecedent potato events and : the
transformation process; the source of the inserted genetic material and its function in both
the donor organism and the GE crop event; and the integrity, stability and mode of
inheritance of the inserted genetic material through sexual or asexual reproduction and
the number of loci inserted.

APHIS also assessed data presented in the extension request on whether the genetic
modification results in expression of new genes, proteins, or enzymes, suppression of
existing genes and their products, or changes in plant metabolism or composition in the
V11 event. The assessment encompasses a consideration of the expressed double
stranded RNA (dsRNA) and any observed or anticipated effects on plant metabolism
including, e.g. any relevant changes in levels of metabolites, anti-nutrients, or nutrients in
harvested potatoes derived from the V11 event compared to the JR Simplot antecedent
potato events or those in the conventional counterparts and other comparators.

Description of the genetic modification and inheritance of inserted DNA

Transformation of the V11 event (Solanum tuberosum subsp. tuberosum) was
accomplished through the Agrobacterium tumefaciens mediated transformation of potato
internode segments from Snowden variety as described in JR Simplot’s extension request
(JR Simplot, 2015, p. 26).

The exact genetic construct pSIM1278 (JR Simplot, 2015, p. 11), used to transform the
JR Simplot antecedent potato events, was used to transform and generate the V11 event.
The T-DNA of pSIM1278 contains DNA sequence intended to down-regulate four genes
through the mechanism of RNA interference (RNAi):

- The first cassette down-regulates expression of the potato asparagine synthetase-1
gene (Asn1) and the potato polyphenol oxidase-5 gene (Ppo5). This cassette is
comprised of two 404-bp inverted repeat fragments of Asn1 and two 143-bp
inverted repeat fragments of Ppo5. The Asn1 and Ppo5 fragments are arranged
between the two convergent native potato promoters—the potato Agp promoter of
the ADP glucose pyrophosphorylase gene (Agp) and the potato Gbss promoter of
the granule-bound starch synthase gene (Gbss) that are primarily active in tubers.
These promoters drive expression of the inverted repeats to generate double-
stranded RNA targeting the native potato genes *Asn1* and *Ppo5* for down-regulation, also known as gene silencing.

- The second cassette lowers reducing sugars by down-regulating the potato phosphorylase-L (*PhL*) gene and the potato starch-associated (*R1*) gene by targeting the down-regulation of their promoters. This cassette is comprised of an inverted repeat fragment of the 508-bp promoter region *PhL* and a 531-bp inverted repeat fragment of *R1*. Similar to the first cassette, the *PhL* and *R1* inverted repeat fragments are transcribed by the potato *Agp* and *Gbss* promoters.

**Expression of inserted DNA and changes in gene expression, new proteins or metabolism**

As with the previously deregulated events, no novel proteins are expressed in the V11 event. The JR Simplot antecedent potato events and the V11 event contain no marker genes.

APHIS reviewed the information provided by JR Simplot in the extension request and determined the following:

- The V11 event contains a single, intact copy of the pSIM1278 T-DNA with a 14-bp deletion of the left border and 3-bp deletion of the right border. (JR Simplot, 2015).
- The T-DNA is stably inherited from generation to generation.
- The V11 event does not contain any back bone sequence of extraneous DNA fragments from the transformation plasmid.
- Similar to the JR Simplot antecedent potato events, the V11 event contains a stable, well-characterized insert.
- There were no changes in gene expression, metabolism or additional proteins between the V11 event and JR Simplot antecedent potato events.

**D. Potential Plant Pest and Disease Impacts**

APHIS assessed data and information presented in the extension request related to the similarity of the V11 event to JR Simplot antecedent potato events to determine whether potential plant pest or disease impacts are likely to result from the transformation process, from DNA sequences from plant pests, or from any other expression products, new enzymes, proteins or changes in plant metabolism or composition in the V11 event that are known or anticipated to cause disease symptoms, or to affect plant pests or diseases or plant defense responses. APHIS also assessed whether V11 event is more likely to have significantly increased disease and pest susceptibility as compared to JR Simplot antecedent potato events. Impacts or changes in similarity to the JR Simplot antecedent potato events to the V11 event were assessed to determine if they would (1) affect and/or result in significant introduction or spread of a damaging pest or disease to other plants; (2) result in the introduction, spread, and/or creation of a new disease; and/or (3) result in a significant exacerbation of a pest or disease for which APHIS has a control program.
Plant Protection and Quarantine (PPQ) is an APHIS program that safeguards agriculture and natural resources from the entry, establishment, and spread of animal and plant pests and noxious weeds into the United States; and supports trade and exports of U.S. agricultural products. PPQ responds to new introductions of plant pests to eradicate, suppress, or contain them through various programs in cooperation with state departments of agriculture and other government agencies. These may be an emergency or longer term domestic programs that target a specific pest. A variety of insect, plant disease, mollusk, nematode or weed programs exist (USDA-APHIS-BRS, 2015), however, none specifically target pests of the V11 event.

Because the genetic makeup and transformation of the V11 event are identical to previously deregulated JR Simplot antecedent potato events, no significant changes in composition are expected from the expression of genes in the V11 event. Similarly, the V11 event is not expected to differ from the antecedents in its ability to harbor or transmit plant pathogens or pests and cause indirect plant pest effects on other agricultural products.

E. Potential Impacts on Non-target Organisms Beneficial to Agriculture

APHIS has previously evaluated the potential impacts on non-target organisms beneficial to agriculture that could result from the deregulation of Simplot antecedent potato events. The JR Simplot antecedent potato events were determined by APHIS to be unlikely to have an adverse effect on non-target organisms in the environment. The genetic construct pSIM1278 used to transform the deregulated varieties contains DNA sequence intended to down-regulate four genes through the mechanism of RNA interference (RNAi):

- **Asn1** (asparagine synthetase) for reduced free asparagine contributing to low acrylamide potential
- **R1** (water dikinase) and **PhL 9** (phosphorylase-L 9) for lower reducing sugars contributing to low acrylamide potential
- **PhL** (phosphorylase-L) for lower reducing sugars contributing to low acrylamide potential; and
- **Ppo5** (polyphenol oxidase-5) for reduced black spot

The exact same genetic construct, pSIM1278, used to transform the previously deregulated JR Simplot antecedent potato events, was used to transform the V11 event. Therefore, based on the high similarity of the V11 event to the JR Simplot antecedent potato events, the compositional similarity of the V11 event to its parent variety, the unlikely impacts of non-target effects due to RNAi, and on the finding that the JR Simplot antecedent potato events were unlikely to harm non-target organisms, APHIS concludes that it is unlikely that V11 event will have an adverse effect on non-target organisms, including those beneficial to agriculture.
F. Potential for Enhanced Weediness of the V11 Snowden Potato

The biology of potato is well studied and understood. As documented in the PPRA of the JR Simplot antecedent potato events, potatoes are not known to be weedy or persistent; they are incapable of survival outside of cultivation (Holm et al., 1979; Muenscher, 1980; Love, 1994; OECD, 1997).

In addition to considerations of the known biology of potato, APHIS analyzed information submitted in the petition on the antecedent organisms on a suite of agronomic characteristics and plant-disease and plant-insect interactions. This agronomic data from the field showed that the antecedents were not different than their non-transgenic comparator. The assessments concluded that the antecedents were unlikely to become weeds. Based on the high similarity of the V11 event to the JR Simplot antecedent potato events expressing similar proteins, and on the finding that the antecedent organisms were unlikely to become weeds, APHIS concludes that it is unlikely that V11 event will become a weed.

APHIS has previously assessed the potential of the JR Simplot antecedent potato events to become weeds. In addition, APHIS has also assessed the potential weediness of many other GE potato events representing a variety or traits. For both the antecedents and the other GE potato events, it was concluded that the new traits would not make the potato events any more likely to become a weed. Therefore, because of the similarity of the V11 event to the JR Simplot antecedent potato events, APHIS has determined that the V11 event is no more likely to be a weed.

G. Potential Impacts on the Weediness of Any Other Plants with which the V11 Snowden Potato Can Interbreed

APHIS evaluated the potential for gene introgression to occur from the JR Simplot antecedent potato events to sexually compatible wild relatives and considered whether such introgression would result in increased weediness. Those assessments found that among native Solanum spp. in the U.S., cultivated potato is potentially sexually-compatible only with the two tuber-bearing species, S. jamesii and S. stoloniferum (previously S. fendleri (Spooner et al., 2004)). Neither of these species is listed on U.S. or State weed lists (USDA-NRCS, 2013a). Gene flow, hybridization and/or introgression of the introduced genetic material from JR Simplot potatoes to the wild relatives, S. stoloniferum and S. jamesii, is unlikely to occur. In the case of S. stoloniferum, where there is a remote possibility of gene introgression, APHIS concluded that even if such introgression were to occur, this species is not considered a weed.

APHIS concluded that the gene silencing cassettes originating from the JR Simplot antecedent potato events were unlikely to impact the weediness of this wild species since the JR Simplot antecedent potato events do not exhibit characteristics that cause them to be any weedier than other cultivated potatoes. Therefore, the V11 event is not expected to increase the weed risk potential of other species with which they can interbreed in the
U.S. and its territories based on their similarity to the JR Simplot antecedent potato events.

**H. Potential Changes to Agriculture or Cultivation Practices**

APHIS assessed whether significant changes to agricultural or cultivation practices from the JR Simplot antecedent potato events is likely to impact plant diseases or pests or their management, including any APHIS control programs. This includes consideration of any changes in pesticide applications, tillage, irrigation, harvesting, etc. as they relate to plant pests and diseases.

APHIS did not identify any significant changes to agricultural or cultivation practices (e.g. pesticide applications, tillage, irrigation, harvesting, rotations, management of volunteers, etc.) from the JR Simplot antecedent potato events and concluded that no impact on plant diseases or pests or their management is likely to occur. Based on the similarity of the V11 event to the JR Simplot antecedent potato events expressing the same proteins, APHIS concludes that it is unlikely that any significant changes to agriculture or cultivation practices would be associated with the V11 event and therefore no impact on plant diseases or pests of their management is likely to occur.

**I. Potential Impacts from Transfer of Genetic Information to Organisms with which the V11 Snowden Potato Cannot Interbreed**

APHIS has previously examined the potential for the JR Simplot antecedent potato events, expressing RNA interference (RNAi) of the \textit{Asn1}, \textit{R1}, \textit{PhL} and \textit{Ppo5} genes to be horizontally transferred without sexual reproduction to other organisms and whether such an event could lead directly or indirectly to disease, damage, injury or harm to plants, including the creation of new or more virulent pests, pathogens, or parasitic plants. The horizontal gene transfer between unrelated organisms is one of the most intensively studied fields in the biosciences since 1940, and the issue gained extra attention with the release of transgenic plants into the environment (Dröge et al., 1998). Potential risks from stable horizontal gene transfer (HGT) from genetically engineered organisms to another organism without reproduction or human intervention were recently reviewed (Keese, 2008). Mechanisms of HGT include conjugation, transformation and transduction, and other diverse mechanisms of DNA and RNA uptake and recombination and rearrangement, most notably through viruses and mobile genetic elements. APHIS has previously reviewed the potential for horizontal gene transfer from GE potato to bacteria, fungi, invertebrates, viruses, and parasitic plants (USDA, 2014).

APHIS previously concluded that HGT of the inserted genetic material from the JR Simplot antecedent potato events to other organisms is highly unlikely, and is not expected to lead directly or indirectly to disease, damage, injury or harm to plants, including the creation of new or more virulent pests, pathogens, or parasitic plants. Therefore, APHIS concludes that HGT from the V11 event to other organisms is also highly unlikely.
J. Conclusion

APHIS has reviewed the information submitted in the extension request, supporting documents, and other relevant information to assess the similarity of plant pest risk of the V11 event compared to the JR Simplot antecedent potato events. APHIS concludes that the V11 event is **no more likely** to pose a plant pest risk than the previously deregulated JR Simplot antecedent potato events.

K. References


USDA (2015) 15-140-01p Extension Request


### Appendix A

<table>
<thead>
<tr>
<th>Description</th>
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<th>Antecedent</th>
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<td><em>Ppo5</em> gene from <em>S. verrucosum</em> (RNAi) 10-Kb spacer from <em>S. tuberosum</em> to create hairpin <em>Gbss</em> granule-bound starch synthase promoter from <em>S. tuberosum</em></td>
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<td>fragment of the <em>PhL</em> promoter from <em>S. tuberosum</em> (RNAi) Spacer-2, a 257 bp fragment from <em>S. tuberosum</em> to create hairpin</td>
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Petition 15-140-01p | Antecedent 1  
JRS01 – E12, F10, J3  
Petition 13-022-01p | Comments |
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| Mechanism of Action | $Asn1$: reduces free asparagine  
$R1$: lowers reducing sugars  
$PhL$: lowers reducing sugars  
$Ppo5$: reduces enzymatic browning | $Asn1$: reduces free asparagine  
$R1$: lowers reducing sugars  
$PhL$: lowers reducing sugars  
$Ppo5$: reduces enzymatic browning | Same as Antecedent |
<p>| Date of antecedent EA/ EIS | N/A | July 2014 | |</p>
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<tr>
<td><strong>Disease and pest susceptibilities</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to change disease and pest susceptibilities</td>
<td></td>
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<tr>
<td><strong>Impacts on beneficial non-targets</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to impact beneficial non-target organisms</td>
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<tr>
<td><strong>Enhanced weediness</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to enhance weediness</td>
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</tr>
<tr>
<td><strong>Enhanced weediness of relatives</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to enhance weediness of relatives</td>
<td></td>
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<tr>
<td><strong>Changes to agriculture or cultivation practices</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to change agriculture or cultivation practices</td>
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<tr>
<td><strong>Horizontal Gene Transfer</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to affect the probability of horizontal gene transfer</td>
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<tr>
<td><strong>Plant Pest Risk</strong></td>
<td>Similar as antecedent</td>
<td>Unlikely to pose a plant pest risk</td>
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