



**Review of Scientific Literature Relevant to the
Food/Feed and Environmental Risk Assessment of
Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 Maize**

Literature Review

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LIST OF ACRONYMS AND ABBREVIATIONS

CAB	Commonwealth Agricultural Bureaux
EFSA	European Food Safety Authority
EPSPS	5-enol pyruvylshikimate-3-phosphate synthase
ERA	Environmental Risk Assessment
EU	European Union
GMO	Genetically Modified Organism
ISAAA	International Service for the Acquisition of Agri-Biotech Applications
MEDLINE	MEDical Literature Analysis and Retrieval System (online version)
mEPSPS	double-mutated 5-enol pyruvylshikimate-3-phosphate synthase
NTO	Nontarget organisms
PAT	Phosphinothricin acetyltransferase
PICO/PECO	Population, Intervention/Exposure, Comparator, Outcomes
PMI	Phosphomannose isomerase

1.0 OBJECTIVE

The purpose of this systematic literature search is to identify literature and/or information on Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize, and all sub-combinations in scope that is relevant to the risk assessment of genetically modified organisms.

Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 is a combined trait maize line developed by Syngenta using conventional breeding techniques. The Bt11 component of this breeding stack includes the *cry1Ab* gene which encodes Cry1Ab, a truncated (615 aa) version of the native, full-length Cry1Ab protein produced by certain subspecies of *Bacillus thuringiensis*, a gram positive bacterium. Cry1Ab confers resistance to certain lepidopteran pests. Bt11 also includes the *pat* gene which encodes the enzyme phosphinothricin acetyltransferase, which confers tolerance to glufosinate herbicides. The *pat* gene in Bt11 maize was derived from *Streptomyces viridochromogenes*.

Maize plants derived from Event MIR162 contain the transgene *vip3Aa20*, which encodes the insecticidal protein Vip3Aa20, and the transgene *pmi*, which encodes the enzyme phosphomannose isomerase (PMI). Vip3Aa20 is a variant of the native Vip3Aa1 protein from the soil bacterium *B. thuringiensis strain AB88*, and is active against certain lepidopteran pests of maize, including *Spodoptera frugiperda* and *Helicoverpa zea*. The transgene *pmi* (also known as *manA*) was derived from *Escherichia coli strain K-12*. PMI enables transformed plant cells to utilize mannose as a primary carbon source; it was used as a selectable marker in the development of MIR162 maize.

Maize plants derived from the transformation Event MIR604 express a modified Cry3A (mCry3A) protein, for control of certain coleopteran pests, and a phosphomannose isomerase (MIR604 PMI) protein, which acts as a selectable marker enabling transformed plant cells to utilize mannose as a primary carbon source.

Maize event TC1507 (DAS-Ø15Ø7-1) was developed by insertion of a DNA fragment, PHI8999A, into the maize genome using microprojectile bombardment. TC1507 expresses the *B. thuringiensis var. aizawai* Cry1F insecticidal protein, which confers protection against certain lepidopteran pests such as European corn borer (*Ostrinia nubilalis*) and *Sesamia* spp. TC1507 also produces the phosphinothricin-N-acetyltransferase (PAT) protein from *S. viridochromogenes* which provides tolerance to glufosinate-ammonium herbicide.

Maize plants derived from transformation Event 5307 contain the gene *ecry3.1Ab* encoding an eCry3.1Ab protein and the gene *pmi* (also known as *manA*) encoding the enzyme PMI. The eCry3.1Ab protein is an engineered chimera of mCry3A and Cry1Ab proteins, and confers insecticidal activity against certain corn rootworm (*Diabrotica*) species. The gene *manA* was obtained from *E. coli strain K-12* and the protein it encodes was utilized as a plant selectable marker during development of 5307 maize.

Maize plants derived from the transformation Event GA21 produce a double-mutated 5-enolpyruvylshikimate-3-phosphate synthase (mEPSPS) protein that confers tolerance to herbicide products containing glyphosate.

Accordingly, this breeding stack hybrid expresses the introduced transgenes, producing the corresponding proteins present in Bt11, MIR162, MIR604, 1507, 5307, and GA21 maize plants. Records must be specific to Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize, or any of the in-scope sub-combinations to be considered relevant.

This report defines the 1) review question; 2) the search strategy; and 3) the explicit methods for selecting, and categorizing the records. The results of the selection process are reported including consideration of the implications of any findings. This report aims to comply with the European Food Safety Authority (EFSA) explanatory note on literature searching for GMO applications (EFSA 2019).

2.0 FORMULATING REVIEW QUESTIONS AND CLARIFYING THEIR PURPOSE

2.1 Review Question

The review question associated with this literature search is:

Do either food/feed products derived from Bt11 × MIR162 × MIR604 × TC1507 × 5307 × GA21 maize including all sub-combinations in scope or the combined intended traits have adverse effects on human and animal health, and/or the environment?

This review question follows the PICO/PECO structure with key elements “Population, Intervention/Exposure, Comparator, Outcomes” (Table 1).

TABLE 1 Review question in PICO/PECO structure

Element	Components of Review Question
<u>P</u> opulation	Human and animal health, and the environment
<u>I</u> ntervention/ <u>E</u> xposure	Bt11 × MIR162 × MIR604 × TC1507 × 5307 × GA21 maize and all sub-combinations in scope, derived food/feed products, and/or relevant combinations of Cry1Ab, Vip3Aa20, mCry3A, Cry1F, eCry3.1Ab, PAT, PMI, MIR604 PMI, and mEPSPS, and closely related variants
<u>C</u> omparator	conventional counterpart (if applicable)
<u>O</u> utcome	adverse effects

Because the search approach was not exclusionary (i.e., did not exclude the presence of any other events) searching the name of any of the single events comprising the Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize stack has the possibility to return literature relevant to the stack of interest and all sub-combinations. A binary and tertiary search term approach was developed to return literature on those titles related to all the possible stack combinations of this application. The sub-combinations relevant to the Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize market authorization in the European Union (EU) are listed in Table 2. A similar approach was used for the proteins associated with the events.

2.2 Eligibility/Inclusion Criteria

Tables 3 and 4 summarize the eligibility/inclusion criteria for establishing relevance of retrieved records. Table 3 provides high level key concepts for inclusion/exclusion and Table 4 provides more explicit information on the information/data requirements concept. The eligibility/inclusion criteria are provided in the order of importance or ease of finding information on the criteria within a publication. The first failed eligibility/inclusion criterion was used as the primary reason for exclusion, and the remaining criteria was not assessed. Internet pages results were screened by date to remove those published prior to 2019. Pages without dates were evaluated further using the criteria in Tables 3 and 4.

TABLE 2 List of sub-combinations in scope for Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize application

Highest order stack	Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21
Six sub-combinations of five events	Bt11 × MIR162 × MIR604 × 1507 × 5307
	Bt11 × MIR162 × MIR604 × 1507 × GA21
	Bt11 × MIR162 × MIR604 × 5307 × GA21
	Bt11 × MIR162 × 1507 × 5307 × GA21
	Bt11 × MIR604 × 1507 × 5307 × GA21
	MIR162 × MIR604 × 1507 × 5307 × GA21
Twelve sub-combinations of four events	Bt11 × MIR162 × MIR604 × 1507
	Bt11 × MIR162 × MIR604 × 5307
	Bt11 × MIR162 × 1507 × 5307
	Bt11 × MIR162 × 5307 × GA21
	Bt11 × MIR604 × 1507 × 5307
	Bt11 × MIR604 × 5307 × GA21
	Bt11 × 1507 × 5307 × GA21
	MIR162 × MIR604 × 1507 × 5307
	MIR162 × MIR604 × 1507 × GA21
	MIR162 × MIR604 × 5307 × GA21
	MIR162 × 1507 × 5307 × GA21
	MIR604 × 1507 × 5307 × GA21
Eleven sub-combinations of three events	Bt11 × MIR162 × 5307
	Bt11 × MIR604 × 5307
	Bt11 × 1507 × 5307
	Bt11 × 5307 × GA21
	MIR162 × MIR604 × 1507
	MIR162 × MIR604 × 5307
	MIR162 × 1507 × 5307
	MIR162 × 5307 × GA21
	MIR604 × 1507 × 5307
	MIR604 × 5307 × GA21
	1507 × 5307 × GA21
Five sub-combinations of two events	Bt11 × 5307
	MIR162 × 5307
	MIR604 × 5307
	1507 × 5307
	5307 × GA21

TABLE 3 Eligibility/inclusion criteria to establish relevance

Concepts	Criteria	Comment
Stacked events obtained by conventional crosses/subcombinations	This publication addresses the highest order breeding stack and/or a sub-combination or sub-combinations of the single events of the highest order breeding stack independently of its/their origin.	This permits the selection of publications on the higher order breeding stack and/or sub-combinations of the single events of the higher order that are in scope of the Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 application, independent of their origin. This permits the exclusion of publications on the single events of the higher stacked event, because the risk assessment of GMO applications for stacked events covers only products in the scope of the GMO application – i.e., the higher order and sub-combinations of the singles involved, independent of their origin.
Intervention/exposure	Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and all sub-combinations in scope, derived food/feed products, and/or the intended trait(s)	Intended traits include glufosinate herbicide tolerance, glyphosate herbicide tolerance, coleopteran insect resistance, lepidopteran insect resistance, and mannose metabolism. Herbicide tolerance is achieved by the expression of mEPSPS and PAT protein. Insect resistance traits are achieved by the expression of the Cry1Ab, Vip3Aa, and Cry1F proteins for protection against specific lepidopteran pests, and by the expression of the mCry3A, and eCry3.1Ab proteins for protection against specific coleopteran pests. Closely related variants are those that have the same tertiary level Crickmore nomenclature as Cry1Ab, Vip3Aa20, and eCry3.1Ab, and same secondary level Crickmore nomenclature for Cry1F and mCry3A. Any enzyme classified as a 5-enolpyruvylshikimate-3-phosphate synthase will be considered relevant if the EPSPS protein imparts the glyphosate tolerance trait. Any enzyme classified as a phosphinothricin acetyl transferase was considered relevant. Any enzyme classified as a phosphomannose isomerase will be considered relevant. Only unique protein combinations relevant to the stack and sub-combinations were in scope.

Concepts	Criteria	Comment
Information/data requirements	Data inform one or more information/data requirement(s) for the GMO and derived food/feed products under consideration, including the intended trait(s)	Publications that potentially contribute to the knowledge informing the risk assessment of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize (information/data requirements provided in Table 4) and all sub-combinations in scope will be considered relevant. Based on the scope of the application certain information/data requirements are excluded. These are also detailed in Table 4. Publications addressing issues such as benefits, socioeconomics, ethics, crop protection, detection methods, efficacy, public perception, and risk communication will be excluded using this criterion, as they are not relevant to the risk assessment as defined in this document.
Scope of GMO application	The pathways and level of exposure to the GMO, derived food/feed products, and the intended trait(s) addressed in the publication are relevant for the intended uses of the GMO and derived food/feed products under regulatory review	Publications must address pathways and levels of exposure relevant to the scope of the application: import and processing of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and all sub-combinations in scope for food/feed uses.
Reporting format	Original/primary data are presented in the publication or it is a risk assessment from a relevant key organization (such as regulatory agencies and risk assessment bodies involved in the risk assessment of GMOs)	Records that do not present original/primary data (e.g., editorials, reviews, position papers) were excluded. Risk assessments performed and reported by relevant key organizations were included as relevant if they address Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize, any of the relevant sub-combinations, and unique combinations of Cry1Ab, Vip3Aa20, mCry3A, Cry1F, eCry3.1Ab, PAT, MIR604 PMI, PMI, and mEPSPS and closely related variants.
Previously risk assessed publications	As indicated by EFSA, a publication should be included if it has not been previously risk assessed by EFSA and/or its GMO Panel, and is not cited/referenced in an EFSA/GMO Panel output	If a publication has previously been considered by EFSA it was excluded. Any cited/referenced publications contained within documents produced by EFSA and/or its GMO Panel were excluded.
Access	Full-text document is accessible	If potentially relevant full-text documents cannot be obtained, then they were listed in a table with a description of the (unsuccessful) methods used to try to obtain a copy.
Population	Human and animal health, and/or the environment are addressed as general protection goals	All of the information/data requirements categories described in Table 4 are thought to inform the risk assessment related to human and animal health, and/or the environment. Therefore, if a publication meets the inclusion criteria described in this Table and is relevant to the information/data requirements in Table 4 it was considered relevant.

Concepts	Criteria	Comment
Outcomes	Effects/impacts on human and animal health, and/or the environment are addressed	Publications that address Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize, or sub-combinations in scope also need to address effects/impacts on entities of concern, and potential determinants of exposure that place these entities at risk in order to be relevant to the risk assessment of Bt11 × MIR162 × MIR604 × GA21 maize, or sub-combinations in scope.
Comparator	If the publication is a comparative study that uses plant material as a test material, eligible publications must report a non-GM variety	Publications that address Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize or sub-combinations in scope, must also include a conventional counterpart as a comparator in those cases where comparative analysis is conducted and plant material is used as test material. Any uncertainties about the appropriateness of the comparator was addressed in the assessment of the publication.
Plant species	The publication may address the same plant species as the GMO under consideration, but could also address any plant species producing unique combinations of Cry1Ab, Vip3Aa20, mCry3A, Cry1F, eCry3.1Ab, PAT, PMI, MIR604 PMI, and mEPSPS proteins in the stack or sub-combinations in scope of the application.	The review question addresses the safe use of the intended trait(s) of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope. Therefore, GMOs that contain combinations of Cry1Ab, Vip3Aa20, mCry3A, Cry1F, eCry3.1Ab, PAT, MIR604 PMI, PMI, and mEPSPS or closely related variants unique to the product, and sub-combinations in scope, but are introduced into another plant species may be included. For certain types of data, the presence of the transgenic proteins in a different plant species does not impact the assessment of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope. Those types of data are identified as event-specific in Table 4.
Target pest/organisms	Target pests/organisms addressed in the study are established in the EU	Records related to the intervention/exposure and target pests/organisms were excluded because the scope of the application is import for food/feed uses, and this would be relevant for cultivation applications only.
Reporting format	A study should only be presented once, but if it is presented in more than one publication, all publications should be listed and grouped	Duplicate publications were excluded at the initial screening stage. Only one copy of a study was presented even if it is reported in different publications.

TABLE 4 Overview of main categories of information/data requirements

Expert knowledge on data used in the risk assessment of the GMO is required but the list below provides some examples of relevant data/information. If certain data are considered stack/subcombination-specific or specific to unique combinations of the transgenic proteins expressed in the stack/subcombinations in scope then it is noted.

Information/data requirement	Non-exhaustive list of specific information/data requirements
Molecular characterization of the genetic modification of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in the scope	<ul style="list-style-type: none"> Information on the insert including: sequence, size, copy number, genetic element arrangement, deletions, location, sequence similarity searches, analysis of open reading frames (stack/sub-combination specific) Expression data of inserted/modified sequences (stack/sub-combination specific) Genetic stability (stack/sub-combination specific) Data on the equivalence between plant-produced and microbially-produced proteins (stack/sub-combination specific)
Agronomic, phenotypic, and compositional characterization of the Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize	<ul style="list-style-type: none"> Comparative assessment of agronomic and phenotypic characteristics under field or controlled conditions (stack/sub-combination specific) Comparative analysis of key nutritional constituents (stack/sub-combination specific)
Toxicological assessment of newly expressed protein(s), new constituents other than proteins, and the whole GM food/feed	<ul style="list-style-type: none"> Toxicity studies (protein combinations in scope) Feeding studies (stack/sub-combination specific)
Allergenicity assessment of the newly expressed protein and the GM food/feed, and adjuvanticity	<ul style="list-style-type: none"> Serum screening (protein combinations in scope) <i>In vivo</i> tests in animal models Expression data for endogenous allergens in maize (stack/sub-combination specific)
Nutritional assessment of the newly expressed protein(s), other new constituents, as well as potential alterations in the total diet of the consumer or the animal	<ul style="list-style-type: none"> Anticipated dietary intake of food/feed and the resulting nutritional impact (stack/sub-combination specific) Comparative growth performance studies with young rapidly growing animal species (stack/sub-combination specific)
Post-market monitoring	<ul style="list-style-type: none"> Description of mechanisms for determining actual changes to overall dietary intake patterns of the Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope, to what extent this has occurred, and whether or not the product induces known (side) effects or unexpected side effects Information on the reliability, sensitivity and specificity of the post market monitoring
Persistence and invasiveness assessment, including plant-to-plant gene transfer	<ul style="list-style-type: none"> Measurements of volunteer occurrence and establishment (stack/sub-combination specific) Replacement capacity (stack/sub-combination specific) Fitness of the Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 plant in various environmental conditions – if the relevant combinations or sub-combinations of newly expressed proteins are expressed in a different plant species then the publication may be considered relevant.
Assessment of plant to micro-organism gene transfer	<ul style="list-style-type: none"> This type of data is covered in the scope of the single event literature review.

Information/data requirement	Non-exhaustive list of specific information/data requirements
Assessment of interactions with target organisms	<ul style="list-style-type: none"> Excluded based on the scope of the application. The scope of this application covers the import, processing, and food and feed use of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in the scope in the EU. According to the EFSA ERA Guidance (EFSA 2010): “<i>resistance development is only relevant for applications with scope cultivation of GM plants and not for applications restricted to import and processing of GM plants, and their products</i>” (EFSA 2010). Therefore, an assessment of the potential resistance development in target organisms resulting from the import, processing, and food and feed use of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope is not relevant for this application.
Assessment of interactions with nontarget organisms	<ul style="list-style-type: none"> The EFSA ERA Guidance (EFSA 2010) states that: “in cases where the application does not include cultivation in the EU, direct environmental exposure of NTOs to the GM plant is via accidental release into the environment of seeds or propagules during transportation and processing. This may result in sporadic occurrence of feral plants and therefore exposure of NTO populations is likely to be negligible. The ERA will then focus on indirect exposure to products of the GM plant (e.g., through manure and faeces from animals fed the GM plant, and other by-products of industrial processes)”. Therefore, any publications that discuss direct exposure in test protein and laboratory studies or field survey data was considered not relevant based on scope of application.
Assessment of interactions with biogeochemical and abiotic processes	<ul style="list-style-type: none"> Excluded based on the scope of the application. The scope of this application covers the import, processing, and food and feed use of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope in the EU. According to the EFSA ERA Guidance (EFSA 2010): “<i>applications concerning food/feed uses and import, and processing do not require scientific information on possible environmental effects associated with the cultivation of the plant</i>” therefore, an assessment of the impacts of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope on biogeochemical processes resulting from specific cultivation, management and harvesting techniques is not relevant given the scope of this application.
Assessment of impact of specific cultivation, management and harvesting techniques	<ul style="list-style-type: none"> Excluded based on the scope of the application. The scope of this application covers the import, processing and food and feed use of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in the scope in the EU. Cultivation of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize in the EU is not included in the scope. According to the EFSA ERA guidance (EFSA 2010): “<i>for GM plants for import and processing that are not intended for cultivation in the EU, there is no need for an ERA for altered cultivation, management, and harvesting techniques</i>”. Therefore, an assessment of impact of specific cultivation, management, and harvesting techniques of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize is not relevant for this application.

Information/data requirement	Non-exhaustive list of specific information/data requirements
Risk mitigation	<ul style="list-style-type: none">Excluded based on the scope of the application. Risk mitigation measures such as high dose/refuge strategy, isolation distance from protected habitats hosting species of conservation concern that are at risk, integrated pest/weed management are only relevant to cultivation. The scope of this application covers the import, processing, and food and feed use of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope.
Post-market environmental monitoring	<ul style="list-style-type: none">Excluded based on the scope of the application. Monitoring such as insect resistance is relevant only to cultivation. The scope of this application covers the import, processing, and food and feed use of Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and sub-combinations in scope.

3.0 SEARCHING FOR/IDENTIFYING RELEVANT PUBLICATIONS

3.1 Electronic Bibliographic Databases

To search for different types of publications and unpublished work that could provide information on the review question, multidisciplinary citation databases which include grey literature (i.e., not peer reviewed) was used. Medline, Agricola, Commonwealth Agricultural Bureaux (CAB) Abstracts, and BIOSIS Previews (provided by Ovid Technologies) were searched. Each of the databases has a thesaurus. Searching these databases fulfills the requirement to search a minimum of at least two multi-disciplinary/large databases.

These databases were selected based on their coverage of scientific literature for relevant subjects including, but not limited to, biomedicine, plant diseases, agriculture, life sciences, pesticides, human health and nutrition, animal health, plant science, biotechnology, and environmental studies. Detailed information (e.g., list of subjects covered, coverage dates, update schedule, and sources for data) regarding each of the databases searched can be obtained upon request. The document types in these databases include journal articles, technical letters, notes, conference proceedings, book chapters, reports, and articles in press.

3.2 Internet Searches

3.2.1 Key organizations

The regulatory agencies that post their risk assessments on their websites and also regulate stacks (Table 5) were searched. The internet pages of these agencies were searched for documents related to Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and the sub-combinations in scope of the application.

TABLE 5 Key organization pages included in the search

Regulatory agency/risk assessment body	Web address
US Environmental Protection Agency	https://www.epa.gov/ingredients-used-pesticide-products/current-and-previously-registered-section-3-plant-incorporated
Health Canada ^a	https://www.canada.ca/en/health-canada/services/food-nutrition/genetically-modified-foods-other-novel-foods/approved-products.html
Office of the Gene Technology Regulator	http://www.ogtr.gov.au/
National Technical Commission on Biosafety ^b	http://ctnbio.mctic.gov.br/inicio
Ministry of Agriculture, Forestry and Fisheries	http://www.maff.go.jp/e/

^aAlso searches Environment and Climate Change Canada (<https://www.ec.gc.ca/cc/>), and Canadian Food Inspection Agency (<http://www.inspection.gc.ca/plants/plants-with-novel-traits/notices-of-submission/eng/1300143491851/1300143550790>).

^bPartial reports are excluded from review since there is no new information contained in these reports and they are not the official final opinion of the agency.

3.2.2 Web-based search engines and databases

General search engines such as GOOGLE Scholar and web-based databases known to contain information specifically on effects of GMOs were not searched. The search of the

databases and key organization websites is considered to provide an adequately comprehensive search of literature.

3.2.3 Manual searches

3.2.3.1 Checking reference lists

For any reviews, methodological publications, guidelines, and scientific opinions from regulatory agencies that were retrieved using the search strategy and classified as relevant to the review question, the reference list(s) from those records were manually searched for new records (2019 through the date the search was conducted) that met the eligibility/inclusion criteria.

3.2.3.2 Hand searching

Hand searching was not conducted. The search of the databases and key organization websites is considered to provide an adequately comprehensive search of literature.

3.2.3.3 Citation searching

Citation searching was not conducted. The search of the databases and key organization websites is considered to provide an adequately comprehensive search of literature.

3.3 Constructing the Search Strategy

3.3.1 Database searching

3.3.1.1 Approaches to develop searches

The “lumping” approach was utilized. A single search strategy was developed to capture all categories of information of interest in one search. This strategy was used because previous experience indicates that a manageable number of studies was returned.

3.3.1.2 Search terms

Identifying search terms

Search terms were identified by:

- Assessing subject indexing terms of relevant publications recorded in those electronic bibliographic databases that use thesauri
 - All publications returned from literature search reports that aim to comply with the EFSA explanatory note and deemed relevant to the review questions were examined to determine the subject indexing terms associated with it.

- Seeking suggestions from experts and stakeholders
 - The search terms were developed using a multi-disciplinary team (i.e., risk assessors, information specialists, regulatory affairs managers).

Free-text terms and subject indexing terms

The searches with the Ovid platform utilized the keyword search in the advanced search window. The keyword search uses a default set of fields designated .mp, which vary by database. Therefore, Ovid uses the term “keyword” to indicate that it is executing a multi-field search. In each database the specific fields searched are a different combination of free-text and controlled vocabulary fields, with Ovid switching automatically to the appropriate fields when a database is selected.¹

In Ovid, the fields used in the .mp keyword search are word searchable, therefore any search only has to find a single word in a controlled vocabulary field that contains phrases to return as search results all references indexed to that subject heading. Thus, a search strategy which includes “genetic*” will return the following (highlighted below):

- **Genetically modified foods** or **genetic engineering** in the Subject Headings field in Agricola,
- Zea mays: species, maize, common, **genetically modified**, strain-Bt10 [Gramineae] in the Organism field in BIOSIS Previews,
- **Genetically engineered organisms** in the Subject Headings field in CAB Abstracts,
- Plants, **Genetically Modified** / ge [Genetics] or **Genetic Engineering** in MeSH Subject Headings in Medline

Subsequent combining of terms, (genetic* AND (modif* OR engineer*)) (in bold), yields all references with these headings to be in the final results for that search set. Therefore, it is not necessary to search each exact controlled phrase in order to return all references for each of the specific headings.

¹In Agricola the .mp fields are: free-text—abstract; geographic area; identifier; meeting information; map information; note; original title; personal name as subject; title—and controlled vocabulary—category code; subject heading.

In BIOSIS Previews the .mp fields are: free-text—abstract; book title; gene name; miscellaneous descriptors; methods & equipment; original language book title; title—and controlled vocabulary—biosystematic codes; chemicals & biochemicals; concept codes; diseases; geopolitical locations; major concepts; organisms; parts, structure & systems of organisms; sequence data; super taxa; taxa notes; time.

In CAB Abstracts the .mp fields are: free-text—abstract; identifiers; original title; title—and controlled vocabulary—broad terms; geographic location; organism descriptors; subject headings.

In Medline the .mp fields are: free-text—abstract; keyword heading word; original title; synonyms; title; unique identifier—and controlled vocabulary—floating sub-heading word; name of substance word; organism supplementary concept word; protocol supplementary concept word; rare disease supplementary concept word; subject heading word.

Appendix A provides 1) the search history (including the full strategy used and fields searched as run in the database) and number of publications identified (line by line) for each bibliographic database prior to de-duplication, and 2) the subject indexing used by each database as shown within the brackets after each search term.

3.3.1.3 Free-text searching functions

The search terms were selected to incorporate a wide variety of synonymous and related terms. Truncation and wildcards were used where appropriate to capture different conventions in spelling and variation in the endings of terms.

3.3.1.4 Search strings

Search strings were combined with Boolean and proximity operators appropriate for the scope of the review.

3.3.1.5 Key elements of review questions to use for best result

A very large number of publications were returned using only the four key elements of Event, Intended trait, newly expressed protein(s), and Trade Name. To prevent a very large number of publications from being returned while still achieving sensitivity, additional key elements were added to the search strategy. Sensitivity was defined as the ability to return the previously deemed relevant articles with the new search string. ‘A very large number’ is not defined in the Explanatory Note (EFSA 2019); however, the number returned with other search strategies (e.g., (Event OR Intended Trait OR Newly Expressed Protein(s) OR Trade Name) or (Event OR Trade name OR ((Intended Trait OR Newly Expressed Protein(s)) AND (Plant Species or GMO)))) was so large that it could not be de-duplicated by the search platform.

Therefore, the search structure included the following search concepts/key elements; Event, Trade Name, Newly Expressed Protein(s), or Intended Trait in the same publications as terms describing plant species and/or GMO general terms. The search strategy employed was:

- Single event combinations OR Trade name OR Newly expressed protein combinations, OR (((Intended trait AND GMO general) OR GMO general × Intended trait) AND Plant species) OR GMO general × Intended trait - insecticidal

The search strategy employed captured literature relevant to Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize and is provided in Table 6. The same search string was used in all databases. Since the Ovid search platform simultaneously searches free-text and subject headings there is no disadvantage to using all search terms in all databases. For example, if ‘Genetically engineered organisms’ is a subject heading in CAB Abstracts but not in Agricola including this term in the search of the Agricola databases still allows for free-text searching of this term.

Because the binary and tertiary search strategy is not exclusionary it allows for the possibility of returning records on all of the subcombinations in scope of the application without searching all of the subcombinations separately (see Table 7 for an example matrix for the event terms).

TABLE 6 Search string strategy

Set	Field	Search string	Concepts/Key elements
1	Topic	Bt11 OR Bt 11 OR SYN-BT?11-1	Single event Bt11
2	Topic	MIR162 OR MIR 162 OR SYN-IR162-4	Single event MIR162
3	Topic	MIR604 OR MIR 604 OR SYN-IR6?4-5	Single event MIR604
4	Topic	DAS15?7 OR DAS 15?7 OR DAS15?7 OR DAS I5?7 OR 15?7 OR I5?7 OR TC 15?7 OR TC15?7 OR TC I5?7 OR TC15?7 OR DAS-?15?7-1 OR DAS?15?71	Single event TC1507
5	Topic	5307 OR SYN-?53?7-1	Single event 5307
6	Topic	GA21 OR GA 21 OR GA2I OR GA 2I OR MON ØØØ21* OR MON OOO21* OR MON 00021* OR MON empty setempty setempty set21* OR MONØØØ21* OR MONOOO21* OR MON00021* OR MONempty setempty setempty set21* OR M0N ØØØ21* OR M0N OOO21* OR M0N 00021* OR M0N empty setempty setempty set21* OR M0NØØØ21* OR M0N00021* OR M0N00021* OR M0Nempty setempty setempty set21*	Single event GA21
7		1 AND 5	Bt11 × 5307 – binary terms
8		2 AND 5	MIR162 × 5307 – binary terms
9		3 AND 5	MIR604 × 5307– binary terms
10		4 AND 5	1507 × 5307– binary terms
11		5 AND 6	5307 × GA21– binary terms
12		2 AND 3 AND 4	MIR162 × MIR604 × 1507 tertiary terms
13		7 OR 8 OR 9 OR 10 OR 11 OR 12	All Single event combinations OR Higher stacked event
14		Cry1Ab* OR Cry 1Ab* OR Cry1 Ab* OR Cry 1 Ab* OR CryIAb* OR Cry IAb* OR CryI Ab* OR Cry I Ab*	Newly expressed protein in Bt11
15		Phosphinothricin N acetyltransferase OR Phosphinothricin N acetyl transferase OR Phosphinothricin acetyltransferase OR Phosphinothricin acetyl transferase OR PPT acetyltransferase OR PPT acetyl transferase OR PT N acetyltransferase OR PT N acetyl transferase OR Glufosinate acetyltransferase OR Glufosinate acetyl transferase OR Gluphosinate acetyltransferase OR Gluphosinate acetyl transferase OR pat OR 111069-93-3 OR EC 2.3.1.183 OR E.C. 2.3.1.183	Newly expressed protein in Bt11 and TC1507
16		Vip3AA20* OR Vip3 AA20* OR Vip3 AA 20* OR Vip3A A 20*	Newly expressed protein in MIR162
17		Phosphomannoisomerase OR Mannose 6-phosphate isomerase OR Phosphomannoseisomerase OR Phosphomannose isomerase OR 9023-88-5 OR AAA24109 OR EC 5.3.1.8 OR E.C. 5.3.1.8	Newly expressed protein in MIR162, 5307 and MIR604
18		mCry3A* OR mCry 3A* OR mCry 3 A* OR Cry3A* OR Cry 3A* OR Cry 3 A*	Newly expressed protein in MIR604

Set	Field	Search string	Concepts/Key elements
19		5 enol pyruvyl shikimate 3 phosphate synthase OR 5 enolpyruvyl shikimate 3 phosphate synthase OR 5 enolpyruvylshikimate 3 phosphate synthase OR EPSP synthase OR MEPSP synthase OR EPSPS OR MEPSPS OR EC 2.5.1.19 OR E.C. 2.5.1.19	Newly expressed protein in GA21
20		CryIF* OR Cry 1F* OR Cry 1 F* OR Cry1 F* OR CryIF* OR Cry IF* OR Cry I F* OR CryI F*	Newly expressed protein in TC1507
21		(eCry3.1AB) OR (e-Cry3.1AB) OR (eCry3 1AB) OR (e-Cry3 1AB) OR (eCry3.1 AB) OR (e-Cry3.1 AB) OR (eCry3 1 AB) OR (e-Cry3 1 AB) OR (eCry 3.1AB) OR (e-Cry 3.1AB) OR (eCry 3 1AB) OR (e-Cry 3 1AB) OR (eCry 3.1 AB) OR (e-Cry 3.1 AB) OR (eCry 3 1 AB) OR (e-Cry 3 1 AB)	Newly expressed protein in 5307
22		(14 and 21) OR (14 and 17) OR (15 and 21) OR (15 AND 17)	Unique combinations of the newly expressed proteins in Bt11 × 5307
23		16 AND 21	Unique combinations of the newly expressed proteins in MIR162 × 5307
24		18 AND 21	Unique combinations of the newly expressed proteins in MIR604 × 5307
25		20 AND 21	Unique combinations of the newly expressed proteins in 1507 × 5307
26		(21 AND 19) OR (17 AND 19)	Unique combinations of the newly expressed proteins in 5307 × GA21
27		16 AND 18 AND 20	MIR162 × MIR604 × 1507 Combinations of the Newly expressed proteins
28		22 OR 23 OR 24 OR 25 OR 26 OR 27	All unique combinations of the newly expressed proteins
29		Agrisure ^{*a}	Trade name MIR162, MIR604, Bt11
30		Herculex ^{*a}	Trade name TC1507
31		Duracade ^{*a}	Trade name 5307
32		29 OR 31 OR (29 AND 30)	Relevant trade name combinations and commercial trade names

33	((Insect OR Insects OR coleoptera* OR lepidoptera* OR pest OR pests OR stalkborer* OR stalk borer* OR borer* OR cornborer* OR corn borer* OR noctuidae OR Crambidae OR Chrysomelidae OR earworm* OR ear worm* OR armyworm* OR army worm* OR cutworm* OR cut worm* OR rootworm* OR root worm* OR Ostrinia OR O nubilalis OR Diatraea OR D grandiosella OR D crambidoides OR Helicoverpa OR H zea OR Spodoptera OR S frugiperda OR S exigua OR Papaipema OR P nebris OR Elasmopalpus OR E lignosellus OR D saccharalis OR Striacosta OR S albicosta OR Agrotis OR A ipsilon OR Feltia OR F jaculifera OR Pseudaletia OR P unipuncta OR Diabrotica OR D virgifera OR D barberi OR ECB OR SWCB OR SCSB OR CEW OR FAW OR SCB OR WBC OR WCRW OR WCR OR NCRW OR MCR OR MCRW) ADJ2 (toleran* OR resistan* OR protect* OR control*) OR B thuringiensis OR Bacillus thuringiensis OR ((glufosinate* OR gluphosinate* OR Basta* OR Liberty* OR Ignite* OR Rely* OR Finale* OR Challenge* OR gl?phosate OR gl?fosate OR roundup* OR round up* OR herbicide* OR pesticide*) ADJ2 (toleran* OR resistan* OR protect*))	Intended traits
34	GMO* OR LMO* OR GM OR GE OR transgen* OR ((genetic* OR living OR biotech*) ADJ3 (modif* OR transform* OR manipul* OR improv* OR engineer* OR deriv*)) OR stack*	GMO general
35	33 AND 34	(Intended trait AND GMO general)
36	GMHT OR GEHT OR GMHR OR GEHR OR GMHTs OR GEHTs OR GMHRs OR GEHRs	GMO general × intended traits – herbicidal
37	35 OR 36	(Intended trait AND GMO general) OR GMO general × Intended trait - herbicidal
38	Maize* OR corn* OR Zea mays OR Z mays	Plant species
39	37 AND 38	((Intended trait AND GMO general) OR GMO general × Intended trait)) AND Plant species
40	((Bt OR Bacillus thuringiensis OR B thuringiensis) ADJ5 (maize* OR corn* OR mays)) OR Btmaize* OR Btcorn*	GMO general × Intended trait - insecticidal
41	13 OR 32 OR 28 OR 39 OR 40	Single event combinations OR Trade name OR Newly expressed protein combinations, OR (((Intended trait AND GMO general) OR GMO general × Intended trait) AND Plant species) OR GMO general × Intended trait - insecticidal

*Trade names for all the single events, the highest order stack and commercial sub-combinations all have Agrisure in their names. Therefore, by simply searching Agrisure it is possible to capture all relevant literature. Searching Duracade will pick up commercialized stacks that are listed in this table as well as single event combinations based on the binary search strategy.

Agrisure Designation	Product
Agrisure Duracade 5122, Agrisure Duracade 5122A, Agrisure Duracade 5122 E-Z Refuge, Agrisure Duracade 5122A E-Z Refuge	Bt11 × MIR604 × TC1507 × 5307 × GA21
Agrisure Duracade 5222, Agrisure Duracade 5222A, Agrisure Duracade 5222 E-Z Refuge, Agrisure Duracade 5222A E-Z Refuge	Bt11 × MIR162 × MIR604 × TC1507 × 5307 × GA21

TABLE 7 Matrix for the event terms in scope for Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21 maize

Stacks in scope of the application	Bt11 × 5307	MIR162 × 5307	MIR604 × 5307	1507 × 5307	5307 × GA21	MIR162 × MIR604 × 1507
Bt11 × MIR162 × MIR604 × 1507 × 5307 × GA21	X	X	X	X	X	X
Bt11 × MIR162 × MIR604 × 1507 × 5307	X	X	X	X		X
Bt11 × MIR162 × MIR604 × 5307 × GA21	X	X	X		X	
Bt11 × MIR162 × MIR604 × 1507 × GA21						X
Bt11 × MIR162 × 1507 × 5307 × GA21	X	X		X	X	
Bt11 × MIR604 × 1507 × 5307 × GA21	X		X	X	X	
Bt11 × MIR162 × MIR604 × 5307	X	X	X			
Bt11 × MIR162 × 1507 × 5307	X	X		X		
Bt11 × MIR162 × 5307 × GA21	X	X			X	
Bt11 × MIR604 × 1507 × 5307	X		X	X		
Bt11 × MIR604 × 5307 × GA21	X		X		X	
Bt11 × 1507 × 5307 × GA21	X			X	X	
MIR162 × MIR604 × 1507 × 5307 × GA21		X	X	X	X	
Bt11 × MIR162 × MIR604 × 1507						X
MIR162 × MIR604 × 1507 × 5307		X	X	X		X
MIR162 × MIR604 × 5307 × GA21		X	X		X	
MIR162 × MIR604 × 1507 × GA21						X
MIR162 × 1507 × 5307 × GA21		X		X	X	
MIR604 × 1507 × 5307 × GA21			X	X	X	
Bt11 × MIR162 × 5307	X	X				
Bt11 × MIR604 × 5307	X		X			
Bt11 × 1507 × 5307	X			X		
Bt11 × 5307 × GA21	X				X	
MIR162 × MIR604 × 5307		X	X			
MIR162 × MIR604 × 1507						X
MIR162 × 1507 × 5307		X		X		
MIR162 × 5307 × GA21		X			X	
MIR604 × 1507 × 5307			X	X		

Stacks in scope of the application	Bt11 × 5307	MIR162 × 5307	MIR604 × 5307	1507 × 5307	5307 × GA21	MIR162 × MIR604 × 1507
MIR604 × 5307 × GA21			X		X	
1507 × 5307 × GA21				X	X	
Bt11 × 5307	X					
MIR162 × 5307		X				
MIR604 × 5307			X			
1507 × 5307				X		
5307 × GA21					X	

3.3.1.6 Use of multiple languages

The search terms used were in the English language or utilized the Roman alphabet. For the event name and trade names that do not use words in the English language, translations are unlikely to exist.

3.3.1.7 Time period

Due to the use of multiple (i.e., 4) multi-disciplinary databases and redundancy in coverage it is unlikely that late addition of a publication would be missed. Therefore, the returned literature was limited to that which was published between January 1, 2019 and the date of the last database update prior to the search (see Table 9).

The records returned from the search of the regulatory agency webpages were manually excluded if they were dated prior to 2019. If a date could not be determined for the record, then the record was reviewed for relevance using the criteria in Tables 3 and 4.

3.3.1.8 Internet searching of regulatory agency webpages

The search terms selected are the event and protein names from the International Service for the Acquisition of Agri-Biotech Applications (ISAAA) (Table 8). The descriptions and information for the top 50 hits or 10% of the total hits (whichever is greater) for each search term/web page was collected.

TABLE 8 Nomenclature for the single event and newly expressed proteins from the ISAAA database for use in searching regulatory agency web pages

Event	Search term	Concepts/Key Elements
MIR604	MIR604	Event name
MIR604	mCry3A	Newly expressed protein
MIR604	Phosphomannose isomerase	Newly expressed protein
5307	5307	Event name
5307	eCry3.1Ab	Newly expressed protein
5307	Phosphomannose isomerase	Newly expressed protein

3.4 Reference Publications

Previously conducted literature reviews have returned literature relevant to the risk assessment of the single events Bt11, MIR162, MIR604, 1507, 5307, and GA21 events or Cry1Ab, Vip3Aa20, mCry3A, Cry1F, eCry3.1Ab, PAT, PMI, MIR604 PMI, and mEPSPS proteins ([REDACTED]). Therefore, the previously conducted literature reviews serve as a validation of the search terms.

4.0 SUMMARIZING AND REPORTING THE DATA

4.1 Selecting Publications

4.1.1 Database records

The process for selecting relevant publications was conducted in two stages. The first stage required a preliminary assessment of titles and abstracts. Those records that are clearly not relevant from reviewing the title only were excluded from further review. For those records that appeared relevant or have unclear relevance the abstract was reviewed. Those records that are clearly not relevant from reviewing the abstract were excluded from further review, while records that are relevant or have unclear relevance were reviewed in Stage 2.

Full-length articles were reviewed in Stage 2. An explanation of exclusion is provided for any full-length records that are deemed not relevant in Stage 2. Any relevant records identified in Stage 2 were subjected to reliability assessment and evaluation of the implications of the record on the food and feed or environmental risk assessments.

Two independent reviewers examined the records for inclusion/exclusion for each eligible information/data requirement at all stages of review. Reviews and selections were conducted independently. During the rapid assessment process (Stage 1), only records that were deemed clearly not relevant by all reviewers were excluded from further review. This conservative approach ensures that all potentially relevant records are evaluated until they are deemed to be either relevant or clearly not relevant in Stage 2. Following the Stage 1 reviews, reviewers scored the records as either 1) relevant or unclear relevance, 2) clearly not relevant.

A kappa test was performed after the Stage 1 review and before any discussion of abstracts over which there was disagreement by the reviewers. Of the 331 records reviewed from the databases at Stage 1 there were 323 agreements to exclude, 3 records where both reviewers agreed to include the records for Stage 2 review, and 5 disagreements: reviewer A selected to include 3 records while reviewer B selected to exclude those records, and reviewer B selected to include 2 records while reviewer A selected to exclude them. This yielded a kappa test score of 0.54.

Subsequently, the reviewers met to discuss the abstracts in which they disagreed and moved the 3 records over which there was disagreement forward to full-length review for a total of 6 records that were reviewed in Stage 2. There was no disagreement among the reviewers after Stage 2; therefore, no tie breaker review was needed.

Because of the format of documents retrieved from internet searching of key organizations (i.e., title and abstract is not often provided) the kappa test was conducted only on the output of the database search.

4.1.2 Records from key organizations

The records returned from searching the websites of key organizations were considered relevant if they are risk assessments, scientific opinions/reports concerning the commercial release of GMO being examined or documents on the biology of the crop of interest. The

regulatory agency webpages that are searched do not post primary data; therefore, all other document types were not considered relevant.

The format of records returned from regulatory agency websites does not often meet the format required to assess them using the 2-stage process followed for the database records. Those websites at which the records are published in English were assessed by two independent reviewers. Due to format full-text documents were assessed to determine relevance. For those websites where the records are not published in English, the results were reviewed by a native speaker. If the document was deemed to be a relevant document type then it was translated into English and two independent reviewers determined if it met the criteria for inclusion. The rationale for record exclusion is provided only for those records classified as one of the relevant document types and if excluded based on other eligibility criteria.

The Intersecretarial Commission on Biosafety of GMOs and National Advisory Commission on Agricultural Biotechnology do not post the relevant document types on their websites; therefore those agency websites were not searched.

For the purposes of generating the statistics related to the records returned from the search of the regulatory agency websites certain assumptions were made. A unique internet record was defined as a unique URL. If the URLs for two documents were identical except for the file format (e.g., pdf versus .doc or .docx), one of the documents was considered a duplicate and excluded from statistical accounting. Documents that were classified as relevant were manually examined to determine if there were any duplicates among them. If a duplicate was identified then it was excluded.

Documents that are clearly labeled as draft or with a line for a signature that is blank were not reviewed.

4.2 Results of the Publication Selection Process

For electronic bibliographic databases, the date on which the search was conducted, the date of the most recent update of the database, the service provider used, date span of the search, any limits applied to the search (e.g., study types, dates, languages) and the total number of records retrieved before and after removing duplicates was recorded (Table 9).

Additionally, the line-by-line strategy with the number of publications identified per line is presented. See Appendix A.

TABLE 9 **Electronic bibliographic database search results**

Database	Search Date dd/mm/yyyy	Service provider	Date span of the search dd/mm/yyyy^a	Any limits applied to the search	Total number of records retrieved after removing duplicates
Agricola	11/10/2020	Ovid Technologies	01/01/2019 – 30/09/2020	Dates	38
BIOSIS Previews	11/10/2020	Ovid Technologies	01/01/2019 – 05/10/2020	Dates	127
CAB Abstracts	11/10/2020	Ovid Technologies	01/01/2019 – 05/10/2020	Dates	40
Medline	11/10/2020	Ovid Technologies	01/01/2019 – 10/10/2020	Dates	126

^aOvid only allows results to be limited by year. The results were de-duplicated across databases and then de-duplicated against the prior year's returned records. The frequency of database update varies. Ovid has provided us with the following update information: Agricola updated monthly on the 1st of the month, BIOSIS Previews updated weekly on Mondays, CAB Abstracts updated weekly on Mondays and Medline updated daily.

For records from websites, the following were recorded (if available): the website name and service publisher used, justification for choosing the source, the URL, the date on which the search was conducted, the date of the most recent website update at the time it was searched, the date span of the search, the search terms used, any limits to the search, and the number of relevant records retrieved (Table 10).

There were no relevant publications returned from searching regulatory agency websites so no manual references were searched as presented in the flow chart of the publication selection process (Figure 1).

TABLE 10 Regulatory agency webpage search details

Regulatory agency name	URL	Date of search dd/mm/yyyy	Date of most recent website update dd/mm/yyyy	Date span of search ^a	Total number of records retrieved after removing duplicates	Number of relevant records
Health Canada ^b	https://www.canada.ca/en/health-canada/services/food-nutrition/genetically-modified-foods-other-novel-foods/approved-products.html	29/07/2020	05/28/2020	No limitations	41	0
Ministry of Agriculture, Forestry and Fisheries	http://www.maff.go.jp/e/	20/07/2020	No update information provided	No limitations	3	0
National Technical Commission on Biosafety ^c	http://ctnbio.mcti.gov.br/	15/07/2020	No update information provided	No limitations	1	0
Office of the Gene Technology Regulator	http://www.ogtr.gov.au/	23/07/2020	23/07/2020	No limitations	1	0
US Environmental Protection Agency	https://www.epa.gov/ingredients-used-pesticide-products/current-and-previously-registered-section-3-plant-incorporated	05/08/2020	14/07/2020	No limitations	0	0

^aRecords published prior to 2019 were manually excluded (if any).

^bAlso searches Environment and Climate Change Canada (<https://www.ec.gc.ca/cc/>) and

Canadian Food Inspection Agency (<http://www.inspection.gc.ca/plants/plants-with-novel-traits/notices-of-submission/eng/1300143491851/1300143550790>).

^cPartial reports are excluded from review and statistics since there is no new information contained in these reports and they are not the official final opinion of the agency.

The results of the selection process are recorded in Table 11.

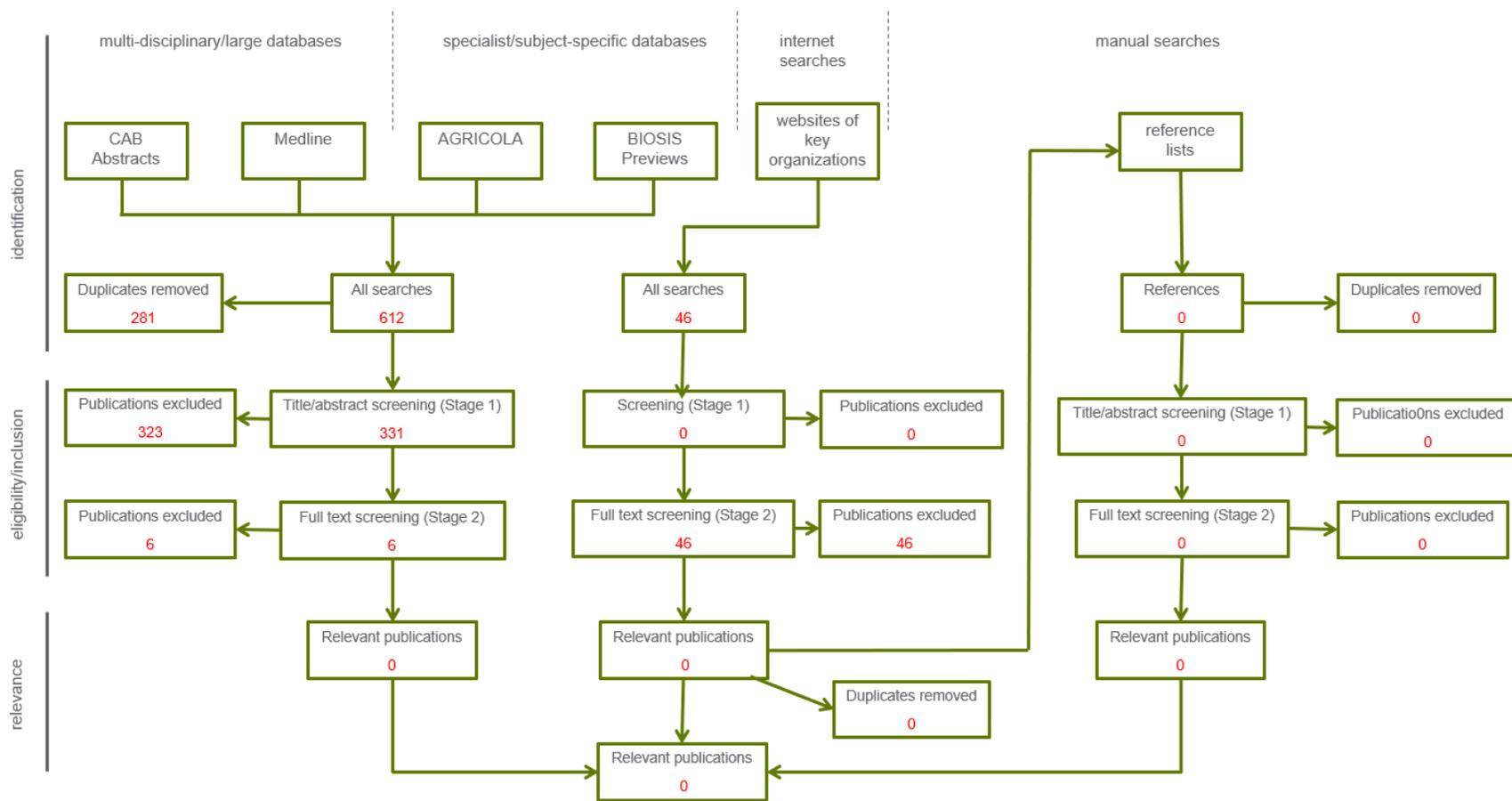
TABLE 11 Results of the publication selection process, for each review question and or group of information/data requirements searched

Review question and/or category of information/data requirement(s) captured in the search	Number of publications
Publications identified after all ^a searches of the scientific literature (excluding duplicates)	377
Database results identified	331
Internet results identified	46
Manual searching – checking reference lists ^b	0
Publications excluded from the search results after screening of title and abstracts*** (stage 1)	323
Database results excluded	323
Internet results excluded	0
Manual searching - reference results excluded	0
Publications screened using full-text (stage 2)	52
Database results screened	6
Internet results screened	46
Manual searching - reference results screened	0
Publications excluded after full-text screening	51
Database results full-text excluded	6
Internet results full-text excluded	46
Manual search – references excluded	0
Unobtainable publications	0
Unclear publications	0
Publications considered relevant	0
Database results relevant	0
Internet results relevant	0
References from relevant internet documents	0

^aBoth from electronic bibliographic databases and other sources of scientific literature.

^bNo relevant records were returned from the searching of regulatory agency websites therefore there are no records reported as a result of manual searching.

FIGURE 1 Flow chart of the publication selection process



4.3 Relevant Publications

No publications were considered relevant after detailed assessment of full-text documents.

No publications were considered relevant after detailed assessment of internet documents.

4.4 Excluded Publications After Detailed Assessment of Full-Text Documents

A list of the full bibliographic references for all excluded studies retrieved from database searching after detailed assessment of full-text documents for relevance (i.e., stage 2), with justification for their exclusion, is recorded in Table 12.

TABLE 12 Report of all publications excluded after detailed assessment of full-text documents

List of bibliographic references for all relevant publications, classified by category of information/data requirements*			
Study author(s) and year	Title	Source	Reason(s) for exclusion based on eligibility/inclusion criteria table
Alves <i>et.al.</i> 2020	Dung beetles and the conservation of diversity in an agricultural landscape with maize fields and Atlantic Forest remnants	Acta Oecologica	Intervention/exposure - The publication examined dung beetle populations adjacent to cultivated fields (i.e., the fields were not planted by the researchers). The authors asked the farmers to qualify their inputs in a survey where one of the options was “transgenic maize” and the results were compiled based on survey results. Therefore, while it is not know whether any of the maize products planted are those within the scope of this literature review because the data were combined it is not relevant.
Dively <i>et al.</i> 2020	Evaluation of gene flow in structured and seed blend refuge systems of non-Bt and Bt corn	Journal of Pest Science	Intervention/exposure - This study was performed on Agrisure 3000GT which is not a stack or subcombination associated with this review.
Fast <i>et.al.</i> 2020	Transgene expression in sprayed and non-sprayed herbicide-tolerant genetically engineered crops is equivalent	Regulatory Toxicology and Pharmacology	Intervention/exposure – This study was not performed on the stack or sub-combinations in scope for this review.
Hart <i>et al.</i> 2019	Insecticidal proteins	Official Gazette of the United States Patent & Trademark Office	Intervention/exposure – The sequences would be relevant to the single events and not this stack or subcombinations.
Liu <i>et.al.</i> 2020	Development of a sensitive monoclonal antibody-based sandwich ELISA to detect Vip3Aa in genetically modified crops	Biotechnology Letters	Intervention/exposure – This study was not performed on the stack or sub-combinations in scope for this review
Walters <i>et al.</i> 2020	Meeting technical challenges for protein characterization and surrogate equivalence studies that resulted from insecticidal protein co-expression in maize event MZIR098	Transgenic Research	Information/data requirements – While the protein combination is relevant to the unique protein combinations associated with the stack, the endpoints measured in the experiment are not relevant to the data/information requirements for the stack.

Report of all publications excluded after assessment of internet documents

Forty-six records excluded are not presented in a table because they did not fulfill the eligibility criteria for report format.

4.5 Unobtainable Publications

No publications were considered unobtainable.

4.6 Unclear Publications

No publications were considered unclear.

4.7 Full-Text Documents

No publications were considered relevant after detailed assessment of full-text documents therefore no relevant documents accompany this final report.

4.8 Narrative Synthesis/Summary of Relevant Publications

A narrative synthesis/summary of the relevant studies describing their overall volume, strength, and direction per main category of information/data requirements was not reported because this literature review was conducted for annual PMEM reports on GMOs authorized in the EU market and therefore it is not required.

4.9 Implications of Relevant Publications on Risk Assessment

There were no relevant publications.

5.0 RECORDS TO BE MAINTAINED

Records maintained include, but are not limited to, documentation of database search dates, database update dates, resolution of differences of opinion on records, the report, and any amendments or deviations.

6.0 ARCHIVING OF RECORDS

The protocol, protocol amendments or deviations, raw data, related documentation, and the final report are archived at Syngenta in Research Triangle Park NC, USA.

Walters FS, Young S, Graser G. 2020. Meeting technical challenges for protein characterization and surrogate equivalence studies that resulted from insecticidal protein co-expression in maize event MZIR098. *Transgenic Res.* 29:109-124.

APPENDIX SECTION

APPENDIX A Search history and subject indexing



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<input type="checkbox"/>	14 (Cry1Ab* or Cry 1Ab* or Cry1 Ab* or Cry 1 Ab* or Cry1Ab* or Cry 1Ab* or Cry1 Ab* or Cry 1 Ab*).mp. [mp=meeting information, title, original title, map information, note, abstract, heading words]	940	Advanced	Display Results More	
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<input type="checkbox"/>	42	limit 41 to yr="2019 -Current"	244	Advanced	Display Results More	
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<input type="checkbox"/>	13 7 or 8 or 9 or 10 or 11 or 12	1	Advanced	Display Results More	
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<input type="checkbox"/>	20 (Cry1F* or Cry 1F* or Cry 1 F* or Cry1 F* or Cry1F* or Cry 1F* or Cry 1 F* or Cry1 F*).mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	115	Advanced	Display Results More	
<input type="checkbox"/>	21 ("eCry3.1AB" or "e-Cry3.1AB" or eCry3 1AB or e-Cry3 1AB or "eCry3.1 AB" or "e-Cry3.1 AB" or eCry3 1 AB or e-Cry3 1 AB or "eCry 3.1AB" or "e-Cry 3.1AB" or eCry 3 1AB or e-Cry 3 1AB or "eCry 3.1 AB" or "e-Cry 3.1 AB" or eCry 3 1 AB or e-Cry 3 1 AB).mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	7	Advanced	Display Results More	
<input type="checkbox"/>	22 (14 and 21) or (14 and 17) or (15 and 21) or (15 and 17)	2	Advanced	Display Results More	
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<input type="checkbox"/>	24 18 and 21	6	Advanced	Display Results More	
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<input type="checkbox"/>	26 (21 and 19) or (17 and 19)	1	Advanced	Display Results More	
<input type="checkbox"/>	27 16 and 18 and 20	0	Advanced	Save More	

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<input type="checkbox"/>	30	Herculex*.mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	9	Advanced	Display Results More	
<input type="checkbox"/>	31	Duracade*.mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	1	Advanced	Display Results More	
<input type="checkbox"/>	32	29 or 31 or (29 and 30)	8	Advanced	Display Results More	
<input type="checkbox"/>	33	((Insect or Insects or coleoptera* or lepidoptera* or pest or pests or stalkborer* or stalk borer* or borer* or cornborer* or corn borer* or noctuidae or Crambidae or Chrysomelidae or earworm* or ear worm* or armyworm* or army worm* or cutworm* or cut worm* or rootworm* or root worm* or Ostrinia or O nubilalis or Diatraea or D grandiosella or D crambidoides or Helicoverpa or H zea or Spodoptera or S frugiperda or S exigua or Papaipema or P nebris or Elasmopalpus or E. Ignosellus or D saccharalis or Striacosta or S albicosta or Agrotis or A ipsilon or Feltia or F jaculifera or Pseudaletia or P unipuncta or Diabrotica or D virgifera or D barberi or ECB or SWCB or SCSB or CEW or FAW or SCB or WBC or WCRW or WCR or NCRW or MCR or MCRW) adj2 (toleran* or resistan* or protect* or controf*)) or B thuringiensis or Bacillus thuringiensis or ((glufosinate* or gluphosinate* or Basta* or Liberty* or Ignite* or Rely* or Finale* or Challenge* or gl?phosate or gl?fosate or roundup* or round up* or herbicide* or pesticide*) adj2 (toleran* or resistan* or protect*))) .mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	103071	Advanced	Display Results More	
<input type="checkbox"/>	34	(GMO* or LMO* or GM or GE or transgen* or ((genetic* or living or biotech*) adj3 (modif* or transform* or manipul* or improv* or engineer* or deriv*)) or stack*).mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	59345	Advanced	Display Results More	
<input type="checkbox"/>	35	33 and 34	6072	Advanced	Display Results More	
<input type="checkbox"/>	36	(GMHT or GEHT or GMHR or GEHR or GMHTs or GEHTs or GMHRs or GEHRs).mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	74	Advanced	Display Results More	
<input type="checkbox"/>	37	35 or 36	6113	Advanced	Display Results More	
<input type="checkbox"/>	38	(Maize* or corn* or Zea mays or Z mays).mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	156714	Advanced	Display Results More	
<input type="checkbox"/>	39	37 and 38	1064	Advanced	Display Results More	
<input type="checkbox"/>	40	((Bt or Bacillus thuringiensis or B thuringiensis) adj5 (maize* or corn* or mays) or Btmaize* or Btcorn*).mp. [mp=abstract, title, original title, broad terms, heading words, identifiers, cabicodes]	677	Advanced	Display Results More	
<input type="checkbox"/>	41	13 or 32 or 28 or 39 or 40	1302	Advanced	Display Results More	
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1. **Abundance of non-target predators in genetically modified corn.**

Hernandez-Juarez, A. Aguirre, L. A. Cerna, E. Flores, M. Frias, G. A. Landeros, J. Ochoa, Y. M.

Florida Entomologist; 2019. 102(1):96-100. 37 ref.

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<input type="checkbox"/>	3 (MIR604 or MIR 604 or SYN-IR674-5).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	37	Advanced	Display Results More	
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<input type="checkbox"/>	5 ("5307" or SYN-75377-1).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	226	Advanced	Display Results More	
<input type="checkbox"/>	6 (GA21 or GA 21 or GA21 or GA 21 or "MON ØØØ21" or MON OOO21* or MON 00021* or MON empty setempty setempty set21* or "MONØØØ21" or MONOOO21* or MON00021* or MONempty setempty setempty set21* or "MON ØØØ21" or MON OOO21* or MON 00021* or MON empty setempty setempty set21* or "MONØØØ21" or MONOOO21* or MON00021* or MONempty setempty setempty set21*).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	90	Advanced	Display Results More	
<input type="checkbox"/>	7 1 and 5	3	Advanced	Display Results More	
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<input type="checkbox"/>	11 5 and 6	2	Advanced	Display Results More	
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<input type="checkbox"/>	13 7 or 8 or 9 or 10 or 11 or 12	12	Advanced	Display Results More	
<input type="checkbox"/>	14 (Cry1Ab* or Cry 1Ab* or Cry1 Ab* or Cry 1 Ab* or Cry1Ab* or Cry 1Ab* or Cry1 Ab* or Cry 1 Ab*)mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	947	Advanced	Display Results More	
<input type="checkbox"/>	15 (Phosphinothricin N acetyltransferase or Phosphinothricin N acetyl transferase or Phosphinothricin acetyltransferase or Phosphinothricin acetyl transferase or PPT acetyltransferase or PPT acetyl transferase or PT N acetyltransferase or PT N acetyl transferase or Glufosinate acetyltransferase or Glufosinate acetyl transferase or Glufosinate acetyltransferase or Glufosinate acetyl transferase or pat or 111069-93-3 or "EC 2.3.1.183" or "E.C. 2.3.1.183").mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	5704	Advanced	Display Results More	
<input type="checkbox"/>	16 (Vip3AA20* or Vip3 AA20* or Vip3 AA 20* or Vip3A A 20*).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	21	Advanced	Display Results More	
<input type="checkbox"/>	17 (Phosphomannoisomerase or Mannose 6-phosphate isomerase or Phosphomannoisomerase or Phosphomannose isomerase or 9023-88-5 or AAA24109 or "EC 5.3.1.8" or "E.C. 5.3.1.8").mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	500	Advanced	Display Results More	
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supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]					
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<input type="checkbox"/>	25 20 and 21	1	Advanced	Display Results More	
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<input type="checkbox"/>	28 22 or 23 or 24 or 25 or 26 or 27	25	Advanced	Display Results More	
<input type="checkbox"/>	29 Agrisure*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	9	Advanced	Display Results More	
<input type="checkbox"/>	30 Herculex*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	17	Advanced	Display Results More	
<input type="checkbox"/>	31 Duracade*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	1	Advanced	Display Results More	
<input type="checkbox"/>	32 29 or 31 or (29 and 30)	9	Advanced	Display Results More	
<input type="checkbox"/>	33 (((Insect or Insects or coleoptera* or lepidoptera* or pest or pests or stalkborer* or stalk borer* or borer* or cornborer* or corn borer* or noctuidae or Crambidae or Chrysomelidae or earworm* or ear worm* or armyworm* or army worm* or cutworm* or cut worm* or rootworm* or root worm* or Ostrinia or O nubilalis or Diatraea or D grandiosella or D crambidoides or Helicoverpa or H zea or Spodoptera or S frugiperda or S exigua or Papaipema or P nebris or Elasmopalpus or E lgnosellus or D saccharalis or Striacosta or S albicosta or Agrotis or A ipsilon or Feltia or F jaculifera or Pseudaletia or P unipuncta or Diabrotica or D virgifera or D barberi or ECB or SWCB or SCSB or CEW or FAW or SCB or WBC or WCRW or WCR or NCRW or MCR or MCRW) adj2 (toleran* or resistan* or protect* or control*)) or B thuringiensis or Bacillus thuringiensis or ((glufosinate* or glufosinate* or Basta* or Liberty* or Ignite* or Rely* or Finale* or Challenge* or gl?fosate or gl?fosate or roundup* or roundup* or herbicide* or pesticide*) adj2 (toleran* or resistan* or protect*))) .mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	46389	Advanced	Display Results More	
<input type="checkbox"/>	34 (GMO* or LMO* or GM or GE or transgen* or ((genetic* or living or biotech*) adj3 (modif* or transform* or manipul* or improv* or engineer* or deriv*)) or stack*).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	469644	Advanced	Display Results More	
<input type="checkbox"/>	35 33 and 34	6123	Advanced	Display Results More	
<input type="checkbox"/>	36 (GMHT or GEHT or GMHR or GEHR or GMHTs or GEHTs or GMHRs or GEHRs).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms]	709	Advanced	Display Results More	
<input type="checkbox"/>	37 35 or 36	6798	Advanced	Display Results More	
<input type="checkbox"/>	38 (Maize* or corn* or Zea mays or Z mays).mp. [mp=title, abstract, original title, name of	260378	Advanced		

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1. **The impact of using genetically modified (GM) corn/maize in Vietnam: Results of the first farm-level survey.**

Brookes G; Dinh TX.
21645698. *12(1):71-83, 2021 Jan 02.*
[Journal Article]
UI: 32997586

Authors Full Name
Brookes, Graham; Dinh, Tran Xuan.

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AB This study assessed the farm-level economic and environmental impacts from the use of genetically modified (GM) corn in Vietnam (resistant to Lepidopteran pests of corn and tolerant to the herbicide glyphosate). It was largely based on a farmer survey conducted in 2018-19. The GM varieties out-performed conventional varieties in terms of yield by +30.4% (+15.2% if the yield comparison is with only the nearest performing equivalent conventional varieties) and reduced the cost of production by between US \$26.47 per ha and US \$31.30 per ha. For every extra US \$1 spent on GM seed relative to conventional seed, farmers gained between an additional US \$6.84 and US \$12.55 in extra income. The GM maize technology also reduced insecticide and herbicide use. The average amount of herbicide active ingredient applied to the GM crop area was 26% lower (1.66 kg per ha) than the average value for the conventional corn area (2.26 kg/ai per ha) and in terms of the associated environmental impact of the herbicide use, as measured by the Environmental Impact Quotient (EIQ) indicator, it was lower by 36% than the average value applicable to the conventional