

Pioneer Hi-Bred International, Inc. Petition (19-101-01p) for Determination of Nonregulated Status for Enhanced Grain Yield and Glufosinate-Ammonium Resistant DP202216 Corn

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Final Environmental Assessment

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ACRONYMS AND ABBREVIATIONS

| | |
|-----------------------|---|
| a.i. | active ingredient |
| AOSCA | Association of Official Seed Certifying Agencies |
| CAA | Clean Air Act |
| CEQ | Council on Environmental Quality |
| CFR | Code of Federal Regulations (United States) |
| CO | carbon monoxide |
| CO₂ | carbon dioxide |
| CWA | Clean Water Act |
| DNA | deoxyribonucleic acid |
| DP | in reference to DP202216 corn |
| EA | Environmental Assessment |
| EFSA | European Food Safety Agency |
| EO | Executive Order |
| EPA | U.S. Environmental Protection Agency |
| ESA | Endangered Species Act of 1973 |
| FDA | U.S. Food and Drug Administration |
| FFDCA | Federal Food, Drug, and Cosmetic Act |
| FIFRA | Federal Insecticide, Fungicide, and Rodenticide Act |
| FONSI | Finding of No Significant Impact |
| FQPA | Food Quality Protection Act |
| FWS | U.S. Fish and Wildlife Service |
| GE | genetically engineered |
| HR | herbicide resistant |
| IPCC | Intergovernmental Panel on Climate Change |
| IR | insect resistant |
| IWM | integrated weed management |
| lb | pound |
| N₂O | nitrous oxide |
| NAAQS | National Ambient Air Quality Standards |
| NEPA | National Environmental Policy Act of 1969 and subsequent amendments |
| NHPA | National Historic Preservation Act |
| NMFS | National Marine Fisheries Service |
| NOP | National Organic Program |

ACRONYMS AND ABBREVIATIONS

| | |
|----------------------------|---|
| NPS | non-point source (pollution) |
| NRC | National Research Council |
| NWQI | National Water Quality Initiative |
| OECD | Organization for Economic Cooperation and Development |
| PAT | phosphinothricin N-acetyltransferase (enzyme) |
| <i>pat / mo-pat</i> | gene from <i>Streptomyces viridochromogenes</i> that encodes the PAT enzyme |
| PIP | plant-incorporated protectant |
| PPRA | Plant Pest Risk Assessment |
| PPA | Plant Protection Act |
| TES | threatened and endangered species |
| TSCA | Toxic Substances Control Act |
| U.S. | United States |
| USDA | U.S. Department of Agriculture |
| USDA-AMS | U.S. Department of Agriculture- Agricultural Marketing Service |
| USDA-APHIS or APHIS | U.S. Department of Agriculture-Animal and Plant Health Inspection Service |
| USDA-ARMS | U.S. Department of Agriculture-Agricultural Resource Management Survey |
| USDA-ERS | U.S. Department of Agriculture-Economic Research Service |
| USDA-NASS | U.S. Department of Agriculture-National Agricultural Statistics Service |
| USC | U.S. Code |
| USFWS | U.S. Fish & Wildlife Service |
| WPS | Worker Protection Standard (40 CFR part 170) |
| <i>zm-gos2</i> | a constitutive promotor in corn that drives expression of the <i>zmm28</i> gene and ZMM28 protein |
| <i>zmm28</i> | <i>zmm28</i> gene |
| ZMM28 | ZMM28 protein, a transcription factor |

1 PURPOSE AND NEED

1.1 Background

In June 2019, Pioneer Hi-Bred International, Inc. (Pioneer) submitted a petition (19-101-01p) to the U.S. Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS), requesting that genetically engineered (GE) DP202216 corn (hereafter referred to as DP202216 corn), and any progeny derived from it, no longer be considered regulated articles under Title 7 of the Code of Federal Regulations part 340 (7 CFR part 340). DP202216 corn is currently regulated by APHIS because it was developed using the plant pest *Agrobacterium tumefaciens*, a regulated article under 7 CFR part 340.2.¹

As part of evaluation of Pioneer's petition APHIS has developed this final Environmental Assessment (EA) to consider the potential impacts of a determination of nonregulated status for DP202216 corn on the human environment.² This EA has been prepared in compliance with the National Environmental Policy Act (NEPA, 42 U.S.C. § 4321 et seq.); the Council of Environmental Quality's (CEQ) NEPA-implementing regulations (40 CFR parts 1500-1508); and USDA and APHIS NEPA-implementing regulations (7 CFR part 1b, and 7 CFR part 372, respectively).

1.2 Purpose of DP202216 corn

Pioneer genetically engineered DP202216 corn for increased grain yield potential and resistance to the herbicide active ingredient (a.i.) glufosinate-ammonium.³ Increased yield is conferred by modifying the expression of a transcription factor, ZMM28 protein, which is naturally present in corn (*Zea mays*). ZMM28 is a MADS-box type of transcription factor that shares 69%-96% amino acid sequence identity with some other plant MAD-box transcription factors that are known to control plant development, such as the transition from vegetative to reproductive growth, as well as the determination of floral meristems and floral organs (Wu et al. 2019). DP202216 corn contains a gene cassette with a constitutive promotor from the corn translation initiation factor *zm-gos2* gene, and an intron region from the corn ubiquitin gene, *ubiZM1* intron, which together drive expression of an extra inserted *zmm28* gene coding region and ZMM28 protein at an earlier growth stage in the leaf, and for an extended period of time in grain (Anderson et al. 2019). Both the *zm-gos2* and *zmm28* genes naturally occur in corn. The earlier and extended and overall increased expression of the ZMM28 protein in DP202216 corn results in higher grain yield potential, as demonstrated through multi-year field trials (Pioneer 2019c). DP202216 corn

¹ Disarmed *Agrobacterium* is commonly used in the genetic modification of plants. Disarmed means the *Agrobacterium* is non-virulent.

² Human environment includes the natural and physical environment and the relationship of people with that environment. When economic or social and natural or physical environmental effects are interrelated, the NEPA analysis may address these potential impacts as well (40 CFR §1508.14).

³ Note that "Resistance" to herbicides is defined by the Herbicide Resistance Action Committee (HRAC) as the inherited ability of a plant population to survive and reproduce following repeated exposure to a dose of herbicide normally lethal to the wild type. "Tolerance" is distinguished from resistance and defined by HRAC as the inherent ability of a plant to survive and reproduce following exposure to an herbicide treatment. This implies that there was no selection or genetic manipulation to make the plant tolerant; it is naturally tolerant. In reference to GE crops, the terms "resistance" and "tolerance" are often used interchangeably. Throughout this EA, APHIS will use the term "resistance" and "resistant", and "herbicide-resistant" (HR), when referring to GE corn.

hybrids produced higher yields than wild type control hybrids 79% of the time over 58 locations tested, with significantly greater yields in 19 locations in which yield increases of 4.8 – 6.4 bushels/acre were not uncommon (Wu et al. 2019). The overall average yield difference ranged from 2.3 to 6.3 bushels/acre depending on the year, and 3.4 bushels/acre across three years combined (Pioneer 2019c).

Resistance to glufosinate-ammonium is conferred by introduction of a gene cassette to express a modified, corn optimized version of the *pat* gene, derived from the soil bacterium *Streptomyces viridochromogenes*. The *pat* gene encodes for expression of the enzyme phosphinothricin acetyl transferase (PAT), which acetylates and inactivates the herbicidal activity of glufosinate-ammonium, rendering DP202216 corn resistant to the herbicide. The introduction of improved yield potential and herbicide-resistance in DP202216 corn is intended to provide growers an additional corn variety to meet market demand for corn based feed, food, and fuel products.

1.3 Coordinated Framework for the Regulation of Biotechnology

On June 26, 1986, the White House Office of Science and Technology Policy issued the Coordinated Framework for the Regulation of Biotechnology (Coordinated Framework), which outlined federal regulatory policy for ensuring the safety of biotechnology products. The primary federal agencies responsible for oversight of biotechnology products are the U.S. Department of Agriculture (USDA), the U.S. Environmental Protection Agency (EPA), and the U.S. Food and Drug Administration (FDA). On January 4, 2017, the USDA, EPA, and FDA released a 2017 update to the Coordinated Framework (USDA-APHIS 2018b), and an accompanying National Strategy for Modernizing the Regulatory System for Biotechnology Products (ETIPCC 2017).

The USDA-APHIS is responsible for protecting animal and plant health. The USDA-APHIS regulates products of biotechnology that may pose a risk to agricultural plants and agriculturally important natural resources under the authorities provided by the plant pest provisions of the Plant Protection Act (PPA), as amended (7 U.S. Code (U.S.C.) 7701–7772), and implementing regulations at 7 CFR part 340.

The purpose of EPA oversight is to protect public and environmental health. The EPA regulates pesticides, including pesticides that are produced by GE organisms, termed plant incorporated protectants, under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) (7 U.S.C. 136 *et seq.*). The EPA also sets tolerances (maximum limits) for pesticide residues that may remain on or in food and animal feed, or establishes an exemption from the requirement for a tolerance, under the Federal Food, Drug, and Cosmetic Act (FFDCA; 21 U.S.C. 301 *et seq.*). The USDA and FDA enforce tolerances to ensure the safety of the nation's food supply (US-EPA 2018; USDA-AMS 2019a; Wu et al. 2019). In addition, the EPA regulates certain GE microorganisms (agricultural uses other than pesticides) under the Toxic Substances Control Act (15 U.S.C. 53 *et seq.*).

The purpose of FDA oversight is to ensure human and animal foods and drugs are safe and sanitary. The FDA regulates a wide variety of products, including human and animal foods, cosmetics, human and veterinary drugs, and human biological products under the authority of the FFDCA and Food Safety Modernization Act (FSMA). The FDA created the Plant Biotechnology Consultation Program in the 1992 to cooperatively work with GE plant developers to help them ensure foods made from their new GE plant varieties are safe and lawful (US-FDA 1992a, 2006). In this program, the FDA evaluates the safety of

food/feed from the new GE crop before it enters the market. Although the consultation program is voluntary, GE plant developers routinely participate in it before bringing a new GE plant to market. The FDA completed its first plant biotechnology consultation in 1994. Thus far, the FDA has evaluated more than 150 GE plant varieties through this program.

A more detailed description of the roles and responsibilities of the USDA, the EPA, and FDA under the Coordinated Framework can be found on USDA's website (USDA-APHIS 2018b).

1.4 Purpose and Need for USDA-APHIS Action

APHIS regulations at 7 CFR part 340 govern the introduction (importation, interstate movement, and environmental release) of GE organisms that may pose a plant pest risk. A GE organism is considered a regulated article if the donor organism, recipient organism, vector, or vector agent used in engineering the organism belongs to one of the taxa listed in 7 CFR § 340.2, such as *Agrobacterium tumefaciens*, which was used in development of DP202216 corn. The regulations provide that any person may submit a petition to APHIS requesting that a GE organism should not be regulated, because it is unlikely to present a plant pest risk.⁴ As required by 7 CFR § 340.6 APHIS must respond to petitioners with a regulatory status decision. A GE organism is no longer subject to the requirements of 7 CFR part 340 or the plant pest provisions of the PPA if APHIS determines through conduct of a Plant Pest Risk Assessment (PPRA) that it is unlikely to pose a plant pest risk.

1.5 Public Involvement

APHIS seeks public comment on draft EAs through notices published in the *Federal Register*. On March 6, 2012, APHIS announced in the *Federal Register* updated procedures for the way it solicits public comment on petitions for determinations of nonregulated status.⁵ Details on policy and procedures for public participation in the petition review and NEPA process are available in the *Federal Register* notice and on the APHIS website (USDA-APHIS 2018a)

1.5.1 Public Involvement for Petition 19-101-01p

On July 25, 2019 APHIS announced in the *Federal Register* that it was making Pioneer's petition available for public review and comment to help identify potential environmental and interrelated economic issues that APHIS should consider in evaluation of the petition.⁶ APHIS accepted written comments on the petition for a period of 60 days, until midnight September 23, 2019. At the end of the comment period APHIS had received 4 comments on the petition. Two were opposed to deregulating

⁴ Petitioners are required (7 CFR § 340.6) to describe known and potential differences from the unmodified organism that would substantiate that the regulated article is unlikely to pose a greater plant pest risk than the unmodified organism from which it was derived.

⁵ FR Vol. 77, No. 44, Tuesday, March 6, 2012, p.13258: Biotechnology Regulatory Services; Changes Regarding the Solicitation of Public Comment for Petitions for Determinations of Nonregulated Status for Genetically Engineered Organisms. Available at: <http://www.gpo.gov/fdsys/pkg/FR-2012-03-06/pdf/2012-5364.pdf>

⁶ Federal Register, / Vol. 84, No. 143 / Thursday, July 25, 2019, p. 35850 Pioneer Hi-Bred International, Inc.; Availability of Petition for Determination of Nonregulated Status for Enhanced Grain Yield Potential and Glufosinate-Ammonium Resistant DP202216 Maize [Docket No. APHIS-2019-0040]. Available at <https://www.govinfo.gov/content/pkg/FR-2019-07-25/pdf/2019-15836.pdf>

DP202216 corn, one commenter was in favor of deregulation, and one comment was unrelated to the petition. None of the comments provide any substantive information that contributed to development of the draft EA. A full record of each comment received is available online at www.regulations.gov [Docket No. APHIS-2019-0040].

On July 20, 2020, APHIS announced in the *Federal Register* it was making available the draft EA for a 30-day public review and comment period⁷. By the end of the comment period closed, APHIS received 2 public comments in favor of the approval of the petition. These were from the National Corn Growers Association, and one anonymous person. The anonymous comment was in favor of approval of the petition. The comment from the National Corn Growers Association addressed that DP202216 corn variety can confer tolerance to glufosinate-ammonium, and potential for increased yield and yield stability. There was no comment received opposing the deregulation of this petition. No new information was presented to APHIS in the comments that contributed to or altered the analyses presented in the final EA. A full record of each comment received is available online at www.regulations.gov, [Docket ID: APHIS-2019-0040].

1.6 Issues Considered in this Final EA

APHIS developed a list of topics for consideration in this final EA based on issues identified in public comments on the draft EA and prior EAs and EISs evaluating petitions for nonregulated status, the scientific literature on agricultural biotechnology, and issues identified by APHIS specific to wild and cultivated *Zea* and *Tripsacum* species. The following topics were identified as relevant to the scope of analysis (40 CFR § 1508.25):

Agricultural Production

- Acreage and Areas of Corn Production
- Agronomic Practices and Inputs

Physical Environment

- Soils
- Water Resources
- Air Quality

Biological Resources

- Soil Biota
- Animal Communities
- Plant Communities
- Herbicide-Resistant Weeds
- Gene Flow and Weediness

⁷ Federal Register / Vol. 85, No. 139 / Monday, July 20, 2020 / Notices / p. 43807: Pioneer Hi-Bred International; Availability of a Preliminary Pest Risk Assessment and Draft Environmental Assessment. Available at <https://www.regulations.gov/document?D=APHIS-2019-0040>

- Biodiversity

Human Health Considerations

- Consumer Health and Worker Safety

Animal Health and Welfare**Socioeconomic Considerations**

- Domestic Economic Environment
- International Trade

In addition, potential cumulative impacts relative to these issues are also considered, potential impacts on threatened and endangered species (TES), as well as adherence of the action to Executive Orders, and environmental laws and regulations to which the action may be subject.

2 ALTERNATIVES

NEPA implementing regulations (40 C.F.R. § 1502.14) require agencies to evaluate all alternatives that appear reasonable and appropriate to the purpose and need for the Agency's action (in this case, a regulatory decision). Two alternatives are evaluated in this EA: (1) No Action, denial of the petition, which would result in the continued regulation of DP202216 corn, and (2) a determination of nonregulated status for DP202216 corn, approval of the petition, the Preferred Alternative.

2.1 No Action Alternative: Continuation as a Regulated Article

One of the alternatives that must be considered by APHIS is a "No Action Alternative," pursuant to CEQ regulations at 40 CFR part 1502.14. APHIS does not have the option to not respond to a petition, which could be considered "no action", because the regulations at 7 CFR § 340.6 require APHIS to respond to all petitioners with a regulatory status decision. Thus, for APHIS, No Action in this context means no change in regulatory status. Under the No Action Alternative APHIS would deny the petition request for nonregulated status and DP202216 corn and progeny derived from DP202216 corn would remain regulated articles under 7 CFR part 340. Permits issued or notifications acknowledged by APHIS would be required for the introduction of DP202216 corn. Because APHIS concluded in its draft PPRA that DP202216 corn is unlikely to pose a plant pest risk (USDA-APHIS 2019a), this alternative would not be an appropriate response to the petition for nonregulated status as it would not satisfactorily meet the purpose and need in providing a science based regulatory status decision to the petitioner, pursuant to 7 CFR § 340.6.

2.2 Preferred Alternative: Determination that DP202216 corn is No Longer a Regulated Article

Under this alternative APHIS would approve the petition request. DP202216 corn and progeny derived from it would no longer be subject to APHIS regulation under 7 CFR part 340 because it was determined that, based on the scientific evidence before the Agency, DP202216 corn is unlikely to pose a plant pest risk (USDA-APHIS 2019a). Permits issued or notifications acknowledged by APHIS would no longer be required for introductions of DP202216 corn. This alternative would best meet the purpose and need to respond appropriately to the petition for nonregulated status pursuant to the requirements of 7 CFR § 340.6, and the Agency's statutory authority under the PPA.

2.3 Alternatives Considered But Rejected from Further Consideration

APHIS has evaluated several other alternatives for consideration in this and other EAs for petitions for nonregulated status. APHIS has considered alternatives that would entail approving a petition request in part (7 CFR § 340.6(d)(3)(i)), mandatory isolation or geographic restriction of GE and non-GE cropping systems, and requirements for testing for the presence of GE crop plant material in non-GE crops and commodities.

Based on the PPRA for DP202216 corn (USDA-APHIS 2019a), and experience with GE and non-GE corn varieties, APHIS concluded that DP202216 corn is unlikely to pose a plant pest risk. Thus, the imposition of testing, release, and/or isolation requirements on DP202216 corn would be inconsistent with the Agency's statutory authority under the plant pest provisions of the PPA, implementing

regulations at 7 CFR part 340, and federal regulatory policies embodied in the Coordinated Framework. Because it would be unreasonable to evaluate alternatives absent any jurisdiction to implement them, these alternatives were dismissed from detailed analysis in this EA.

2.4 Summary of the No Action and Preferred Alternative Analyses

Table 2-1 presents a summary of the environmental impacts associated with the No Action Alternative and Preferred Alternative that are evaluated in this final EA. Detailed analysis of the affected environment and environmental impacts is discussed in Chapter 3 and Chapter 4, respectively.

| Table 2-1. Summary of Potential Impacts for the Alternatives Considered | | |
|--|--|--|
| Analysis | No Action Alternative: Continue to Regulate DP202216 corn as a Plant Pest | Preferred Alternative: Approve the Petition for Nonregulated Status for DP202216 corn |
| Meets Purpose and Need and Objectives | No | Yes |
| Unlikely to Pose a Plant Pest Risk | Determined via a Plant Pest Risk Assessment (USDA-APHIS 2019a). | Determined via a Plant Pest Risk Assessment (USDA-APHIS 2019a). |
| Management Practices | | |
| Acreage and Areas of Corn Production | Denial of the petition would have no effect on the areas or acreage utilized for corn production. Fluctuations in production areas and acreage would be relative to weed, insect pest, and disease pressures, and market demand for corn commodities. Regulated field trials would be conducted on lands allocated for this purpose. | Approval of the petition is unlikely to have a notable effect on an increase or decrease in total U.S. corn acreage. DP202216 corn, if adopted by growers, would be expected to replace other corn varieties currently cultivated, as opposed to augmenting current corn crops. As a higher yielding cultivar DP202216 corn could potentially require less acreage per bushel. |
| Agronomic Practices and Inputs | Agronomic practices and inputs used in corn crop production, to include regulated field trials, would remain unchanged. Denial of the petition would have no effect on weed management. | Agronomic practices and inputs would be the same as for other varieties of corn (Pioneer 2019c). DP202216 corn exhibits enhanced nitrogen uptake and assimilation capacity. How this may or may not affect fertilizer use with the corn variety is indeterminate. |
| Use of GE Corn | Approximately 80% of the U.S. corn crops are GE herbicide resistant (HR) varieties. Denial of the petition would have no effect on grower choice in the planting of GE and non-GE corn. | Approval of the petition would provide for cultivation of a GE corn resistant to glufosinate, and potentially increased yield. |
| Physical Environment | | |
| Soil Quality | Agronomic practices and inputs associated with corn production potentially impacting soils, to include regulated field trials, would continue along current trends. | The agronomic practices and inputs are the same for both DP202216 corn and existing corn varieties – potential impacts on soils would be unchanged. |
| Water Resources | Denial of the petition would have no effect on water resources in the | Because the agronomic practices and inputs utilized for DP202216 corn production |

Table 2-1. Summary of Potential Impacts for the Alternatives Considered

| Analysis | No Action Alternative: Continue to Regulate DP202216 corn as a Plant Pest | Preferred Alternative: Approve the Petition for Nonregulated Status for DP202216 corn |
|-----------------------------|---|---|
| | United States. Regulated field trials are limited on a spatiotemporal scale, and present negligible risks to water resources. | would be no different than currently used, sources of potential impacts on water resources, namely non-point source (NPS) pollutants in agricultural run-off, would not be expected to substantially differ. There are no novel impacts to water resources identified with cultivation of DP202216 corn. The EPA provides label use restrictions and guidance for pesticides, to include glufosinate based herbicides, that are intended to be protective of surface and groundwater. |
| Air Quality | Emission sources, namely tillage and machinery combusting fossil fuels, and the level of emissions associated with corn production, to include regulated field trials, would be unaffected by denial of the petition. | Because the agronomic practices and inputs used for corn, as well as acreage, would remain unchanged, no changes to emission sources nor any significant changes in the volume of emissions from U.S. corn production, would be expected. |
| Biological Resources | | |
| Soil Biota | Potential impacts of corn production/regulated field trials on soil biota would continue along current trends. | Commercial production of DP202216 corn or progeny is not expected to present any risks to soil biota. While DP202216 corn differs from non-GE corn varieties in the herbicide resistance and increased yield traits, these traits are not expected to have significant effects on soil biota or community structures. The introduced <i>zmm28</i> gene and ZMM28 protein product, which confers potentially increased yield, occurs naturally in corn and other plants. Glufosinate-ammonium resistance is conferred through introduction of a modified gene (<i>pat</i>) from <i>Streptomyces viridochromogenes</i> , a naturally occurring soil bacterium. Glufosinate-ammonium is readily degraded by soil microbiota, with no adverse effects on soil communities identified. |
| Animal Communities | Regulated field trials of DP202216 corn would present negligible risk to animal communities. | Approval of the petition, and subsequent commercial production of DP202216 corn, would not be expected to affect animal communities adjacent to or within DP202216 corn cropping systems any differently from that of current corn cropping systems. Neither the PAT nor ZMM28 proteins present any risk to wildlife. |
| Plant Communities | Regulated field trials of DP202216 corn would present negligible risks to | Because the agronomic practices and inputs that will be used for DP202216 corn |

Table 2-1. Summary of Potential Impacts for the Alternatives Considered

| Analysis | No Action Alternative: Continue to Regulate DP202216 corn as a Plant Pest | Preferred Alternative: Approve the Petition for Nonregulated Status for DP202216 corn |
|--------------------------------|---|---|
| | plant communities in proximity to DP202216 corn fields. | production are the same as/similar to that of other corn varieties, potential impacts on plant communities would be the same as that for other corn varieties currently cultivated. The EPA regulates and determines the use of glufosinate. Pesticide use requirements are intended to be protective of non-target plant communities and other plants, such as those in adjacent fields. |
| Gene Flow and Weediness | <i>Tripsacum</i> species are the only sexually compatible plants found in the United States. The potential for corn (<i>Zea mays</i>) to hybridize with wild relatives of <i>Tripsacum</i> is low; hybridization and successful introgression of <i>Z. mays</i> genes into <i>Tripsacum</i> is rare (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995). While transient hybrids have been observed (e.g., 2 or 3 generations), successful introgression of <i>Zea mays</i> genes into <i>Tripsacum</i> populations, successful gene flow in this direction, has not been observed (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995). | DP202216 corn, if grown for commercial purposes, would be cultivated as are current corn varieties and present the same potential risk for gene flow, specifically the propensity for and frequency of gene flow, as current corn varieties. In the unlikely event pollen flow from DP202216 corn to <i>Tripsacum</i> were to occur, it is unlikely the PAT trait extant in DP202216 corn would present any risk to communities of <i>Tripsacum</i> species in terms of plant fitness, or their ecological role in the communities of other plants. Conceptually, the ZMM28 increased yield trait could confer a fitness advantage to <i>Tripsacum</i> species in the event gene flow occurred. This considered, it is unlikely that any hybrid <i>Tripsacum</i> populations with the ZMM28 trait gene would develop. Occurrence of <i>Tripsacum</i> around DP202216 corn fields is expected to be rare. In the event outcrossing occurred, successful introgression of <i>Zea mays</i> genes into <i>Tripsacum</i> populations, successful gene flow in this direction, has not been observed in the wild (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995)). |
| Biodiversity | Denial of the petition, and further regulated field trials of DP202216 corn, conducted on lands allocated for this purpose, would have little effect on biodiversity in an around DP202216 corn crops. | Commercial production of DP202216 corn would affect biodiversity in and around DP202216 corn crops no differently than other corn cropping systems. The ZMM28 and PAT trait proteins are unlikely to present any risks to plant, animal, fungal, or bacterial communities. The same or functionally similar ZMM28 proteins are ubiquitous among plants, and PAT among soil dwelling <i>Streptomyces</i> species. All pesticide use would be subject to EPA registration and use requirements. |

| Table 2-1. Summary of Potential Impacts for the Alternatives Considered | | |
|---|--|--|
| Analysis | No Action Alternative: Continue to Regulate DP202216 corn as a Plant Pest | Preferred Alternative: Approve the Petition for Nonregulated Status for DP202216 corn |
| Human and Animal Health | | |
| Human Health and Worker Safety | Denial of the petition would have no effect on human health. DP202216 corn would remain a regulated article and would not be available for food or uses. | Approval of the petition would provide for the use of DP202216 corn products in the food and feed industries. As part of the FDA's voluntary biotechnology consultation program, Pioneer submitted a safety and nutritional assessment for DP202216 corn to the FDA's Center for Food Safety and Applied Nutrition in 2018 (Pioneer 2019c). The EPA regulation of pesticides, and worker protection standards, would be no different than that of the No Action Alternative. |
| Animal Health and Welfare | Denial of the petition would have no effect on animal health and welfare. DP202216 corn would remain a regulated article and would not be available for feed uses. | DP202216 corn would provide for animal feed products. Pioneer is consulting with the FDA as to the safety of feed derived from DP202216 corn. |
| Socioeconomic | | |
| Domestic Economic Environment | Denial of the petition would preclude DP202216 corn being available for food, feed, industrial, and fuel uses. This, however, would have no effect on domestic or international markets. | DP202216 corn may be cultivated to produce corn based food, feed, industrial, and fuel products. DP202216 corn may provide increased yields compared to other varieties, and as a glufosinate resistant variety is intended to facilitate the management of weeds, and herbicide resistant (HR) weeds and their development. Consequently, this variety may be competitive in grower selection of corn varieties. These factors considered, the impacts of DP202216 corn on domestic markets would be considered potentially beneficial. There are no adverse impacts associated with the introduction of DP202216 corn to commercial markets. |
| Trade Economic Environment | Denial of the petition would have no impacts on the trade of corn commodities. | Approval of the petition is unlikely to have a substantial effect on the trade of U.S. corn commodities. As discussed above, DP202216 corn, a field corn variety, is expected to be used for provision standard corn based food, feed, and fuel products. |
| Cumulative Impacts | | |
| Agriculture, Physical and Biological Resources, Public Health, Socioeconomic | There are no cumulative impacts on any aspect of the human environment evaluated identified with denial of the petition. | DP202216 corn production would entail the use of pesticides and fertilizers, and to some extent tillage, which will contribute to potential cumulative impacts on water, soil, and air quality, as does current corn production. If DP202216 corn is adopted by |

| Table 2-1. Summary of Potential Impacts for the Alternatives Considered | | |
|---|--|--|
| Analysis | No Action Alternative: Continue to Regulate DP202216 corn as a Plant Pest | Preferred Alternative: Approve the Petition for Nonregulated Status for DP202216 corn |
| | | growers, no increase in acreage is expected, thus, no increase in level of total U.S. agricultural inputs or NAAQS emissions. |
| Coordinated Framework | | |
| U.S. Regulatory Agencies | Denial of the petition would have no effect on the roles of the FDA and EPA in the oversight of DP202216 corn. | Pioneer is consulting with the FDA on the food and feed safety of DP202216 corn. Glufosinate and other pesticide use will be subject to EPA registration and label use requirements. |
| Regulatory and Policy Compliance | | |
| ESA, CWA, CAA, SDWA, NHPA, EOs | Fully compliant | Fully compliant |

3 AFFECTED ENVIRONMENT

This chapter provides an overview of those aspects of the human environment potentially affected by APHIS' decision to either approve or deny the petition. Broadly, those aspects considered are U.S. corn production, the physical environment, biological resources, public health, animal health and welfare, and socioeconomics. Because the introduced genes are involved in weed management and crop yield, the primary focus of this EA is on: (1) weed and herbicide resistant (HR) weed management, (2) the potential effects of exposure to the introduced trait genes and gene products on human health and wildlife, and (3) gene flow and the potential weediness of DP202216 corn.

3.1 Agricultural Production of Corn

3.1.1 Acreage and Area of U.S. Corn Production

There are three primary varieties of corn cultivated in the United States: Field (or grain) corn, sweet corn, and popcorn. Grain corn is classified into four main types: dent corn, flint corn, flour or soft corn, and waxy corn.

Sweet corn (*Zea saccharata* or *Zea rugosa*) is primarily used for food purposes, eaten as corn on the cob, or it can be canned or frozen for future consumption. Sweet corn is seldom used for feed or flour. Flint corn (*Zea mays indurata*) has not been commonly grown in the United States. It is more often cultivated in South America, Latin America, and southern Europe, regions that use flint corn as food and feed. Flour corn (*Zea mays amylacea*) is used for making corn flour, baked goods. Waxy corn (*Zea mays* var. *ceratina*) serves as a raw material for the production of amylopectin starch, which is processed for industrial and food uses (e.g., amylopectin or waxy starch is used in food products, and the textile, adhesive, and corrugating and paper industries). Popcorn (*Zea mays everta*), a type of flint corn, is only used for food purposes.

DP202216 corn is a dent corn (*Zea mays indenata*) variety. Dent corn, at maturity, has an obvious depression (or dent) at the crown of the kernels—thus its name. Dent corn, also referred to as "field" corn, is primarily used for animal feed, fuel ethanol stock, and industrial uses and comprises the bulk of U.S. production, in excess of 90% of corn acres annually. Among dent corn commodities, animal feed accounts for around 40%, and stock for the production of fuel ethanol for around 35% (NCGA 2019; PRX 2019; USDA-ERS 2019a). The remainder is processed into a variety of food and industrial products such as starch, sweeteners, corn oil and corn syrup, and beverage and industrial alcohol (Figure 3-1).

Around 10% of harvested corn is used for direct human consumption. Production of popcorn and sweet corn, those varieties used for direct human consumption, comprise about 0.2 and 0.5 million acres, respectively (< 1% of corn acreage on an annual basis) (USDA-NASS 2019a).

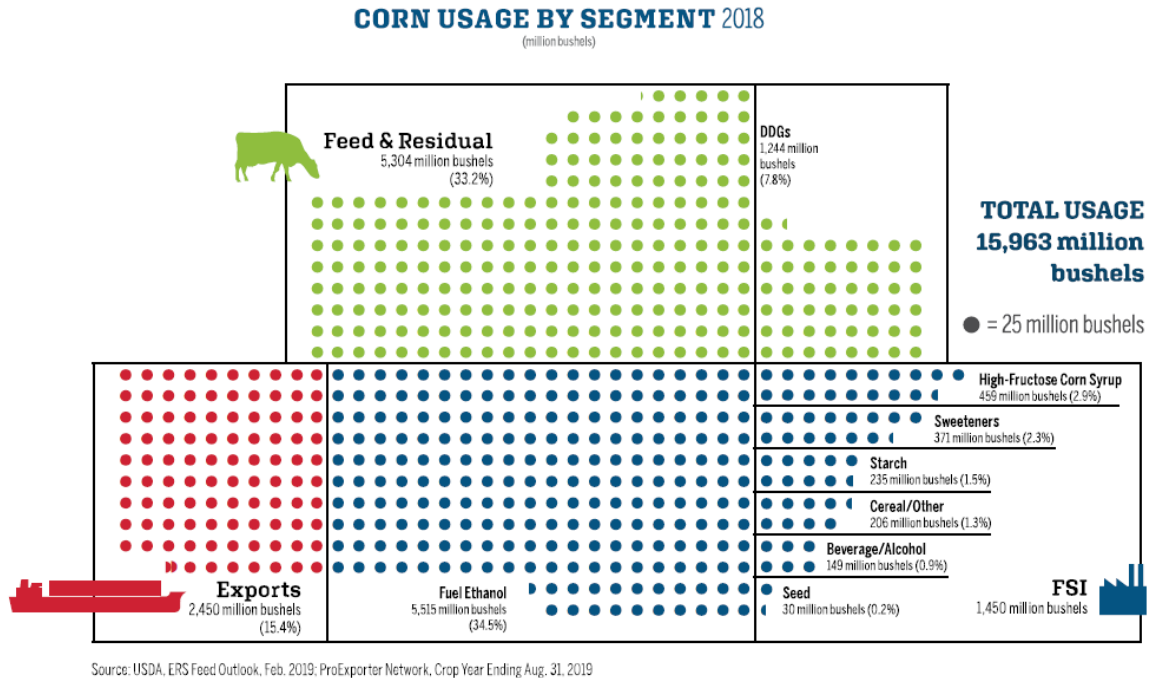


Figure 3-1. Corn Uses in the United States

Note that these statistics/percent allocations of corn vary annually, they are general approximations. Feed is comprised of grain, and distillers' dried grains with solubles (DDGS). Feed for both dairy and beef has been the primary use of DDGS, but increasingly larger quantities of DDGS are making their way into the feed rations of hogs and poultry. FSI refers to food, seed, and industrial uses. Source: (USDA-ERS 2018; NCGA 2019; PRX 2019)

Over the last ten years a total of around 85 to 95 million acres of corn have been planted in the United States on an annual basis (USDA-NASS 2019b). This comprises approximately 25% of total U.S. cropland (~394 million acres). While commercial corn crops are grown in all states to some extent (except Alaska), the majority of production occurs in the Corn Belt, generally defined as Illinois, Iowa, Indiana, southern and western Minnesota, eastern South Dakota and Nebraska, western Kentucky and Ohio, and the northern two-thirds of Missouri. The leading corn-producing states of Illinois, Iowa, and Nebraska account for approximately 40 % of the annual U.S. harvest (USDA-NASS 2019a). Substantial production also occurs in the Pacific Northwest, California's Central Valley, along the Mississippi River, and up the Eastern Seaboard from Georgia to Upstate New York (Figure 3-2).

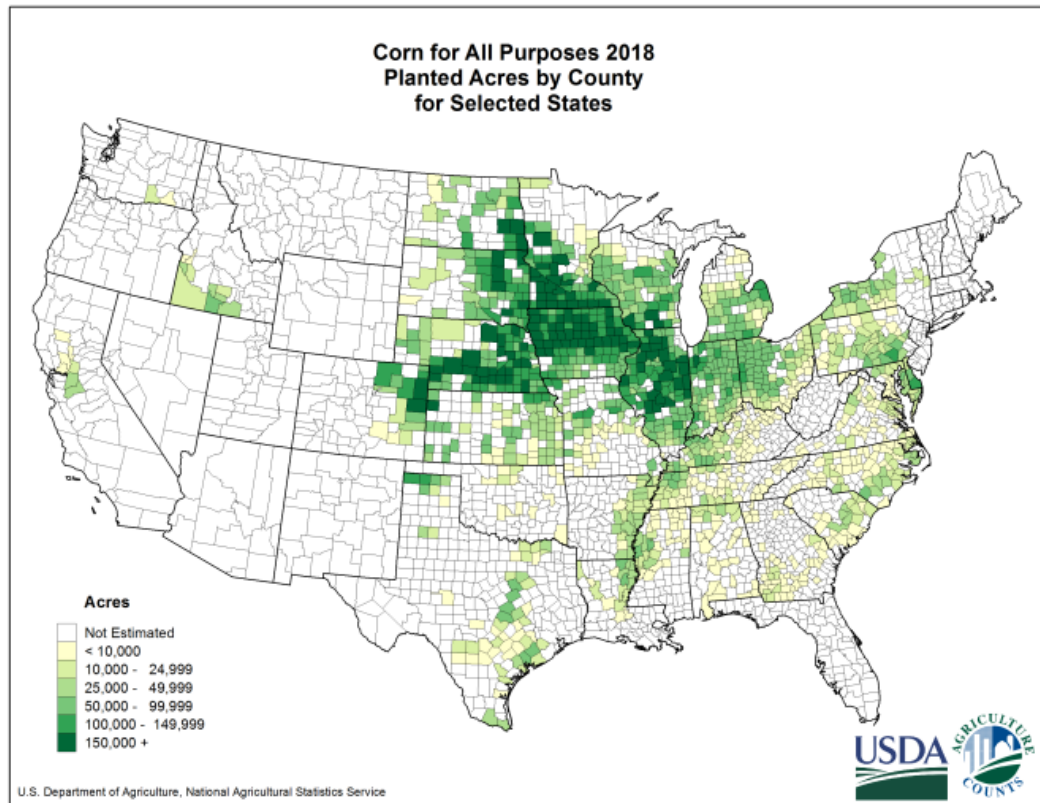


Figure 3-2. Planted and Harvested Acreage of Corn in the U.S., 1998-2018
(Source (USDA-NASS 2019c))

Around 90% of the corn produced in the United States is comprised of GE varieties (Figure 3-3), the majority of this dent/field corn, with limited quantities of GE sweet corn being grown. Most GE-corn varieties are stacked-trait herbicide-resistant (HR) and insect-resistant (IR). Stacked-trait varieties with both HR and IR traits accounted for 80% of the 2018 crop (USDA-ERS 2019b). Only 10% contained a single HR trait, and 2% a single IR in 2018 (USDA-ERS 2019b). Of the ~90 million corn acres planted in 2018, around 7,130 were non-GE.

Biotech Share of U.S. Corn Acres Planted 2018

(1,000 acres)

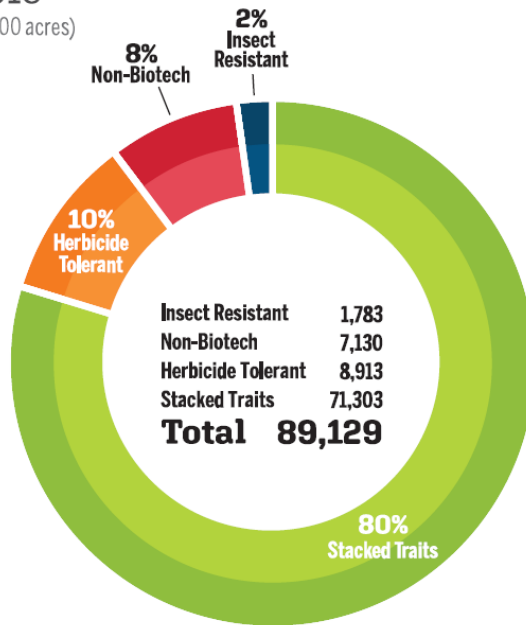


Figure 3-3. GE Corn Traits Planted in the United States, 2017

Source: (USDA-ERS 2019b)

3.1.2 Agronomic Practices and Inputs

Corn production (excluding organic) utilizes a variety of agronomic practices and inputs that aim to achieve optimal yield/acre, product quality, and grower net returns. These include the occasional or regular application of manure and synthetic fertilizers; pesticides; tillage; crop rotation; and cover crops. Organic farming systems are required to exclude certain inputs, such as use of synthetic pesticides and GE crop varieties. Some of these practices (e.g., tillage) and inputs (e.g., fertilizers, pesticides) can, when applied in excess or improperly, or as a result of cumulative effects, present environmental challenges in maintaining air, soil, and water quality. Pesticide and fertilizer use can also present risks to wildlife and human health. The relationship between these practices and inputs, and potential impacts on the physical environment, biological resources, and human health are discussed in the subsequent sections of this chapter.

Apart from the herbicide-, insect-, or disease-resistant trait(s), there are little differences in the agronomic practices and inputs used for GE and non-GE crops. GE HR crops will influence the types of herbicides used. GE IR generally reduce the overall application rate of insecticides, and disease resistant crops the use rates of fungicides and similar chemicals targeting plant pathogens. The agronomic practices, and current uses of the herbicides that will be used with DP202216 corn, are reviewed below.

3.1.2.1 Agronomic Practices

Growers employ several practices for the management of pests and weeds, summarized below. Scouting for weeds was the most widely reported monitoring practice in 2018, used on 94% of corn planted acres (Table 3-1). Among pest management practices, crop rotation was practiced on 84% of planted acres. The most widely used prevention practice was no-till or minimum till (65%). Maintaining ground cover, mulching, or using other physical barriers was the most reported suppression practice (45%). Tillage is the primary practice that can have environmental impacts, and this topic, in relation to DP202216 corn, discussed in more detail below.

| Table 3-1. Top Practices in Pest Management, 2018 Crop Year | |
|---|-------------------------|
| | % of corn planted acres |
| Monitoring: Scouted for weeds | 94 |
| Avoidance: Rotated crops during last three years | 84 |
| Prevention: Used no-till or minimum till | 65 |
| Suppression: Maintained ground cover, mulched, or used other physical barriers | 45 |

The USDA-NASS survey asked growers to report on the practices they used to manage pests, defined as weeds, insects, or diseases. Corn growers reported practices in four categories: prevention, avoidance, monitoring, and suppression. Only the top practice in each category is shown. Source: (USDA-NASS 2019d)

3.1.2.1.1 Tillage

Tillage is used to control weeds and soil-borne pests and disease, and prepare the seedbed. Tillage types are classified as conventional, reduced, and conservation tillage (to include no-till), which are characterized in part by the amount of plant material left on the field after harvest and the degree of soil disturbance they cause. Conventional tillage involves intensive plowing leaving less than 15% crop residue in the field; reduced tillage leaves 15 to 30% crop residue; conservation tillage involves leaving at least 30% crop residue; and no-till systems leave all crop residue on the field (Claassen et al. 2018; OSU 2019).

Which tillage practices are used and to what extent can have substantial impacts on soil quality, soil erosion, and water and air quality (discussed in Section 3.2–Physical Environment). Over the long-term conventional tillage reduces soil quality, and results in soil erosion and runoff that can adversely affect surface waters (Wallander 2015). Conservation tillage systems are the least intensive and, as the name implies, aim to conserve topsoil and soil quality. Conservation tillage (including no-till) provides a variety of agronomic and economic benefits, such as reductions in fuel use and crop production costs, preservation of soil organic matter and moisture, and reductions in soil erosion and water pollution (Fernandez-Cornejo et al. 2012; Claassen et al. 2018). However, conservation tillage, especially no till, can also cause production problems such as increased soil compaction, perennial weeds or weed shifts, buildup of plant pathogens or pests in crop residue, and slow early crop growth due to cooler soil temperatures (Roth 2015). A systematic use of crop rotations can improve the success of conservation tillage by eliminating some of these stresses observed in continuous no-till corn (Roth 2015).

Decisions concerning the amount, timing, and type of tillage to employ are some of the most important crop producers make. These decisions involve consideration of a wide range of interrelated factors such as crop rotation practices, the variety and extent of weeds and crop pests present, soil erosional capacity,

fuel and other input costs, anticipated weather patterns, and potential air and water quality issues (Roth 2015).

The use of conservation tillage systems increased steadily throughout the 1980s and 1990s, and continues to do so. While approximately 33% of corn acres were produced using conservation tillage systems in 1990, 40% of corn acres were produced using conservation tillage systems in 2006 (Fernandez-Cornejo et al. 2012), and 65% in 2016 (Claassen et al. 2018). No-till accounted for around 42% of conservation tillage on U.S. corn acres in 2016 (27% overall) (Figure 3-4). In part, an increase in conservation tillage was facilitated by the availability (since the 1980s) of post-emergent herbicides (Fernandez-Cornejo et al. 2012), which can be applied over crops throughout the growing season (not just before planting, as had previously been the case). Another factor has been the implementation of soil conservation programs, which began in the mid-1980s, and have encouraged/incentivized conservation tillage practices to help conserve soils (USDA-NRCS 2006). Continued increases in conservation tillage since the late 1990s have also been attributed to, in part, the use of GE herbicide resistant (HR) crops, which can facilitate effective weed management and reduce the need for mechanical weed control (Towery and Werblow 2010; USDA-ERS 2012).

Trends in conservation tillage adoption

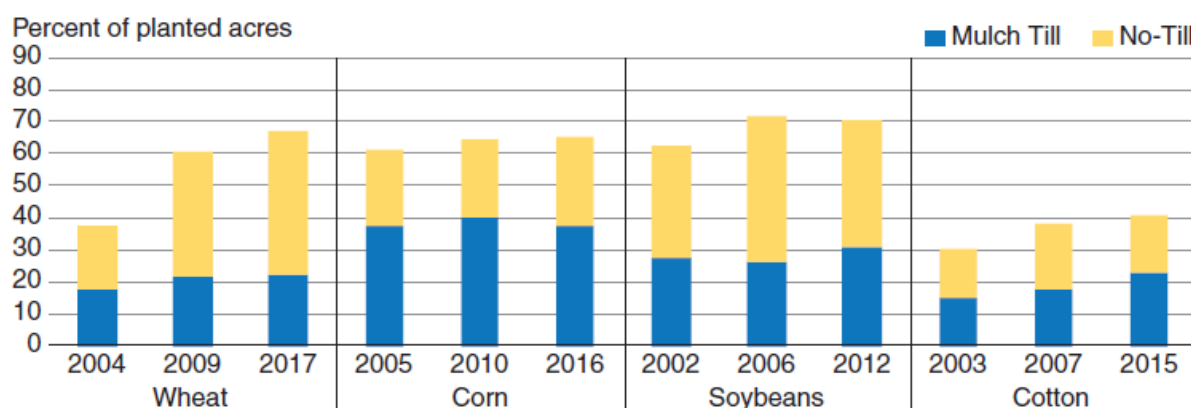


Figure 3-4. Conservation Tillage Practices in Corn, 2005 – 2016

Mulch till is a type of conservation tillage that leaves more than 30% crop residue cover after planting, and the soil surface disturbed by noninversion tillage. This practice benefits soil by increasing organic matter, improving soil tilth and increases productivity as the constant supply of organic material left on the soil surface is decomposed by a healthy population of earth worms and other organisms. Source: (Claassen et al. 2018)

3.1.2.2 Agronomic Inputs

Apart from the agronomic practices described, chemical inputs for control of insect pests, nematodes, pathogens, weeds, and the addition of plant nutrients to soils are an integral part of corn production, GE and non-GE cropping systems alike. Agronomic inputs relative to DP202216 corn production are discussed following.

3.1.2.2.1 Fertilizers

The majority of corn acreage is treated with fertilizer. Soils in many areas of the United States where corn is produced are naturally deficient in nitrogen, phosphorus, and other nutrients, requiring fertilizer inputs, to include manure, to produce crops efficiently, and the yields necessary, to meet market demand. Given the importance of nutrient availability to corn growth, fertilization with nitrogen, phosphorus, and potassium is practiced widely in the United States.

Since 1975, around 95% to 97% of acreage has been treated with nitrogen, with the average rate of application increasing from around 110 to 145 lbs/acre from 1975 to 2016 (USDA-ERS 2019c). Phosphate use has remained steady over this period, at ~ 80% to 85% of acreage, at an average rate of 60 lbs/acre (USDA-ERS 2019c). The acreage treated with potash slightly declined since 1975 to currently around 60% of acres, where the application rate has remained steady at ~ 80 to 85 lbs/acre (USDA-ERS 2019c). Inputs for the 2018 crop year (latest data) are provided in (Table 3-2). While nitrogen and phosphorus are important agricultural inputs in crop production, the introduction of amounts exceeding recommended thresholds can have a number of undesirable impacts on water and air quality (discussed in Section 3.2 – Physical Environment).

| Table 3-2. Fertilizer Applied to Corn Acres, 2018 Crop Year | | | |
|---|--------------------|----------------------------------|--------------------------------|
| Fertilizer | % of Planted Acres | Avg. Rate for Year (lbs/acre) | Total Applied (billion lbs) |
| Nitrogen (N) | 98 | 149 | 12.0 |
| Phosphate (P2O5) | 79 | 69 | 4.5 |
| Potash (K2O) | 63 | 87 | 4.5 |
| Sulfur (S) | 32 | 18 | 0.5 |

Source: (USDA-NASS 2019d)

3.1.2.2.2 Pesticides

Pesticides contribute to higher yields, optimal product quality, and grower net returns by controlling weeds, insects, nematodes, and plant pathogens. Herbicides, in particular, reduce the amount of labor, machinery, and fuel required for the manual control of weeds. However, some pesticides may be potentially harmful to humans and wildlife when not properly used.

The primary pesticides used on corn are herbicides, insecticides, and fungicides (USDA-NASS 2019d). In 2018, herbicides were applied to 97% of planted corn acres, fungicides to 17%, and insecticides to 13% of planted corn acres (USDA-NASS 2019d) (Figure 3-5). Common corn pests include *Coleoptera* species (beetles), *Lepidoptera* species (moth and butterfly larvae), pathogenic fungi (e.g., Corn Leaf Blight), bacteria (e.g., stalk rot), and viruses (e.g., Dwarf Mosaic Virus) (UMinn 2019). Numerous populations of weed species across the United States require annual management in corn cropping systems. Because DP202216 corn is an herbicide-resistant variety, emphasis here is given to herbicide use and weed management.

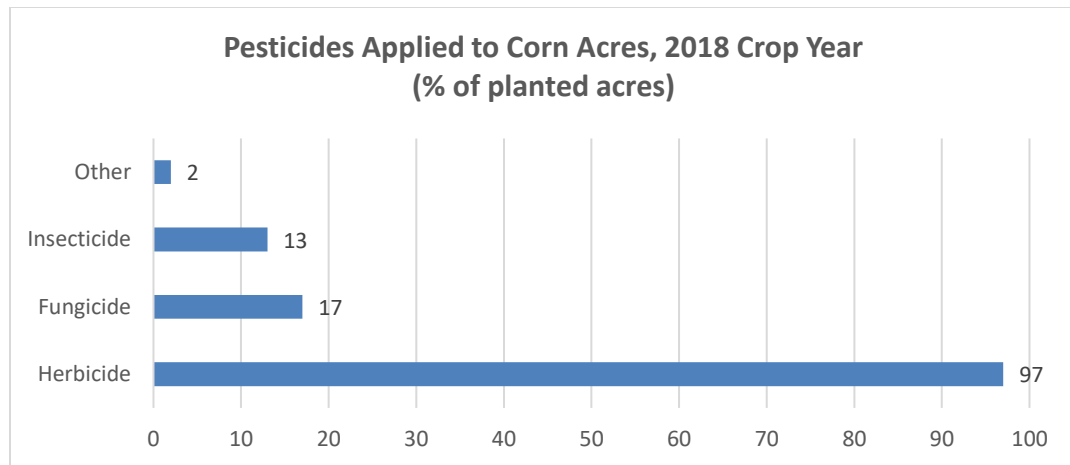


Figure 3-5. Pesticides Applied to Corn, 2018

Source: (USDA-NASS 2019d)

3.1.2.2.3 Weed and Herbicide Resistant Weed Management

There are around 50 major weeds among U.S. cornfields (Jhala et al. 2014), and around 17 principle weeds that can have substantial impacts on corn production in the U.S. due to herbicide resistance (Heap 2020). Weeds have been and will remain a problem in corn crop production; they are difficult to control, competitive, and use up resources — soil moisture, nutrients, access to sunlight — that would otherwise be available to the corn plant. Their presence can reduce yields and product quality, increase harvest costs, and reduce net economic returns by:

- increasing insect and disease damage in crops by serving as hosts for pests and pathogens;
- reducing seedbed soil moisture and structure as a result of increased tillage needed to kill weeds prior to seeding;
- increasing dockage with higher cleaning and transportation costs; and
- contamination, resulting in reduced grades and quality from similar inseparable size and shape weed seeds (cleavers, for example).

In terms of the potential impact of weeds on yield and net returns: One recent study found that, averaged across seven years, weed interference in corn in the United States and Canada caused a 50% yield loss, which equates to a loss of 148 million tons of corn valued at over \$26.7 billion annually (Soltani et al. 2017).

Weed Management with GE Herbicide Resistant Corn and Glufosinate

Prior to the development of synthetic pesticides in the 1940s farmers controlled weeds by tillage, mowing, site selection, crop rotation, use of crop seed free of weed seeds, and hoeing or pulling by hand. U.S. farmers began using synthetic herbicides after their commercial introduction in the 1940's because they were inexpensive, effective, easy to apply, reduced labor costs, reduced the need for tillage, and increase crop yields (Fernandez-Cornejo et al. 2014b). Because herbicides are effective, they remain the

most commonly used among weed management tools, and are expected to remain so for the foreseeable future. In the United States, herbicides are currently used on more than 90% of U.S. cropland.

Currently, around \$15 billion is spent annually on pesticides, representing a five-fold increase since 1960 when adjusting for inflation (GROi 2018). Sixty years ago, herbicides accounted for around 18% of pesticide use by volume on U.S. crops, and insecticides 58% percent. These figures are much different now, with herbicide and insecticide use accounting for approximately 76% and 6% percent of total pesticide applications, respectively (GROi 2018). Adoption of herbicides expanded due to low prices and availability of different chemicals, while insecticide use decreased as formulations became more effective and less product was needed to achieve the intended result. Presently, corn, soybeans, wheat, and cotton receive about 80% of total pesticides applied in the United States (GROi 2018).

In discussing herbicide use it is important to clarify that simply looking at the total pounds of active ingredient (a.i.) used per year, pounds a.i. per acre used, and trends in increase or decrease in lbs per year—evaluation of these metrics in isolation of other factors—is not particularly useful for assessment of environmental or human health risks. Environmental and human health risk requires evaluation of the specific herbicide a.i., its mode of action, the potential toxicity of the a.i. to various taxa, its environmental mobility and persistence, as well as the toxicity of herbicide formulations as there may be synergistic effects that derive from such formulations. Bearing these factors in mind, as weight and use rates are some of the most commonly reported metrics, and in response to comments received on EISs and EAs requesting evaluation of pesticide use, provided below are usage data on glufosinate, which will be used with DP202216 corn. Glufosinate toxicity in relation to use rates is also addressed, in Section 4.1 – Scope of Analysis.

Glyphosate has been one of the most widely used herbicides with GE HR corn, although dicamba, glufosinate, and 2,4-D resistant varieties, particularly stacked-trait varieties, are increasing in commercial use. The only broad spectrum herbicides (with effective control of both broadleaf and grass species) that can be used with GE HR corn today are glufosinate-ammonium and glyphosate. Glufosinate is a non-selective, foliar-applied herbicide that is registered for pre-plant and post-emergence control of over 120 grass and broadleaf weeds in crop and non-crop sites. It is marketed in herbicide formulations such as Basta, Rely, Finale, Ignite, Challenge, and Liberty.

Glufosinate is registered for use on a variety of crops, including apples, berries, canola, citrus, corn, cotton, currants, grapes, grass grown for seed, potatoes, rice, soybeans, sugar beets, and tree nuts (US-EPA 2016c). Among these, glufosinate is registered for use on GE canola, corn, cotton, and soybeans. The current voluntary request for termination on the use of glufosinate on rice is being processed by the US-EPA. Non-crop use sites include golf course turf, residential lawns, industrial and residential landscape plantings, utility and roadside rights-of-way, and timber site preparation for tree plantings.

The crops which account for the most glufosinate use are soybean, cotton, and corn (together, over 75% of usage) (US-EPA 2016c); these crops have GE varieties that are resistant to glufosinate. In 2017, 6.42 million lbs and 1.15 million lbs of glufosinate were applied to soybean and cotton, respectively (USDA-NASS 2019e). For corn, 234,000 lbs were applied in 2016, and 488,000 lbs were applied in 2018 (USDA-NASS 2019e). Glufosinate use is significant on several other crops, including canola (also GE for

glufosinate resistance) and pistachios, with more than 40% of these crops treated with glufosinate. Over 20% of almonds, wine grapes, and hazelnut crops are treated with glufosinate (US-EPA 2016c).

Currently, over 8 million pounds of glufosinate are used annually on more than 10 million agricultural acres (US-EPA 2016c; USDA-NASS 2019e); most of which are in the central United States, Southeast, California's central valley, Oregon, and Washington state (Figure 3-6).

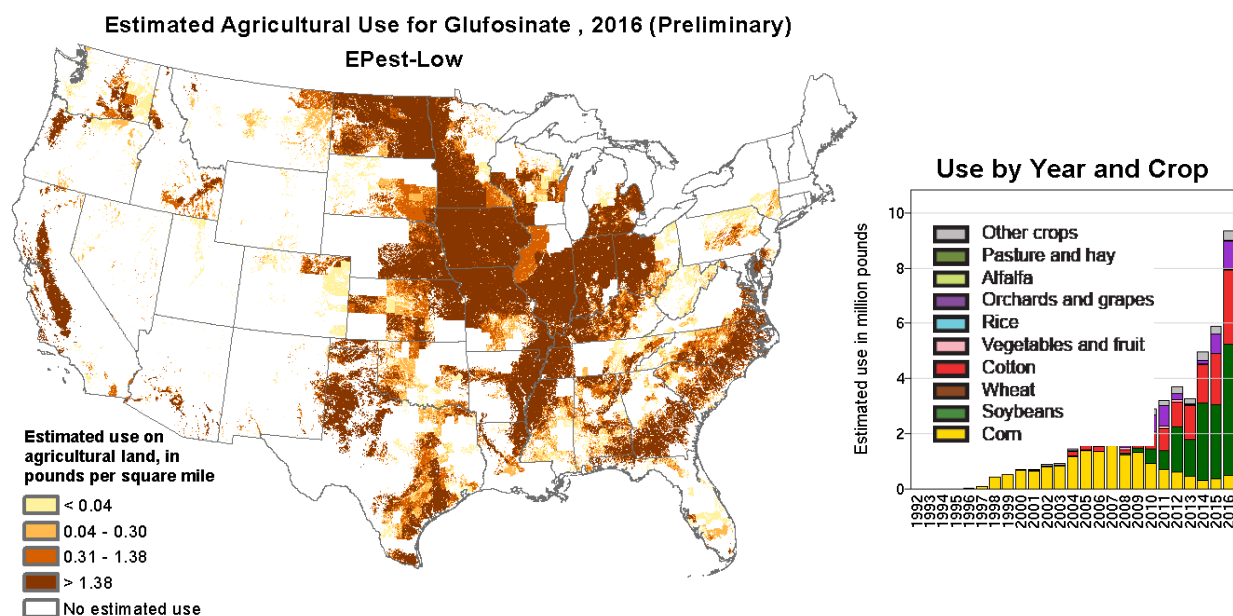


Figure 3-6. Glufosinate Use in the Conterminous United States - 2016

Source: (USGS 2016)

Currently, 90% of the corn and cotton acres are treated with glufosinate at an average rate of 0.4 lbs.a.i./acre (USDA-NASS 2019f), while 90% of the almond acres are treated at a rate of 1.4 lbs. a.i./acre (US-EPA 2016c; USDA-NASS 2019e). Several other crops, including apples, cherries, grapes (raisin, table, and wine), oranges, peaches, pears, pistachios, and walnuts are treated with glufosinate at rates greater than 1.0 lb a.i./acre (US-EPA 2016c; USDA-NASS 2019e). Overall, use of glufosinate on corn, even with GE glufosinate resistant corn varieties available, has been relatively limited. As of 2018, glufosinate use comprised 0.23% of total herbicides applied to corn, and was used on only around 1% of corn acres (Table 3-3).

| Table 3-3. Herbicide Use in U.S. Corn Production – 2018 | | | | |
|--|--------------------|--|---|---------------------------------------|
| Herbicide a.i. | lbs a.i./Yr | Application: lbs a.i./acre/Yr (Average) | Treated Acres, % of Area Planted | Portion of Total Herbicide Use |
| ATRAZINE | 55,899,000 | 1.037 | 65 | 26.03% |
| ACETOCHLOR | 38,757,000 | 1.433 | 33 | 18.05% |
| S-METOLACHLOR | 28,259,000 | 1.198 | 29 | 13.16% |
| GLYPHOSATE ISO. SALT | 27,691,000 | 0.993 | 34 | 12.90% |
| GLYPHOSATE POT. SALT | 25,306,000 | 1.187 | 26 | 11.79% |

| Table 3-3. Herbicide Use in U.S. Corn Production – 2018 | | | | |
|--|--------------------|-------|-----|-------|
| GLYPHOSATE | 10,081,000 | 1.018 | 12 | 4.69% |
| MESOTRIONE | 4,177,000 | 0.121 | 42 | 1.95% |
| GLYPHOSATE DIM. SALT | 3,508,000 | 1.158 | 4 | 1.63% |
| DIMETHENAMID-P | 3,410,000 | 0.676 | 6 | 1.59% |
| 2,4-D, 2-EHE | 3,024,000 | 0.526 | 7 | 1.41% |
| 2,4-D, DIMETH. SALT | 2,284,000 | 0.546 | 5 | 1.06% |
| METOLACHLOR | 1,817,000 | 1.392 | 2 | 0.85% |
| DICAMBA, DIGLY. SALT | 1,348,000 | 0.312 | 5 | 0.63% |
| CLOPYRALID | 859,000 | 0.082 | 13 | 0.40% |
| DICAMBA, DIMET. SALT | 742,000 | 0.287 | 3 | 0.35% |
| TRIFLURALIN | 678,000 | 1.655 | (Z) | 0.32% |
| DICAMBA, SODIUM SALT | 660,000 | 0.103 | 8 | 0.31% |
| PARAQUAT | 642,000 | 0.682 | 1 | 0.30% |
| SIMAZINE | 572,000 | 1.107 | 1 | 0.27% |
| GLUFOSINATE-AMMONIUM | 488,000 | 0.411 | 1 | 0.23% |
| TEMBOTRIONE | 472,000 | 0.084 | 7 | 0.22% |
| ISOXAFLUTOLE | 426,000 | 0.064 | 8 | 0.20% |
| PENDIMETHALIN | 342,000 | 0.896 | (Z) | 0.16% |
| CLOPYRALID MONO SALT | 301,000 | 0.082 | 4 | 0.14% |
| FLUMETSULAM | 267,000 | 0.031 | 10 | 0.12% |
| DIFLUFENZOPYR-SODIUM | 230,000 | 0.04 | 7 | 0.11% |
| BICYCLOPYRONE | 226,000 | 0.031 | 9 | 0.11% |
| SAFLUFENACIL | 220,000 | 0.055 | 5 | 0.10% |
| METRIBUZIN | 159,000 | 0.212 | 1 | 0.07% |
| THIENCARBAZONE-METHY | 123,000 | 0.022 | 7 | 0.06% |
| PYROXASULFONE | 113,000 | 0.12 | 1 | 0.05% |
| DICAMBA, POT. SALT | 106,000 | 0.202 | 1 | 0.05% |
| TOPRAMEZONE | 91,000 | 0.018 | 6 | 0.04% |
| FOMESAFEN SODIUM | 88,000 | 0.218 | (Z) | 0.04% |
| DICAMBA | 73,000 | 0.181 | (Z) | 0.03% |
| RIMSULFURON | 52,000 | 0.016 | 4 | 0.02% |
| FLUROXYPYR 1-MHE | 41,000 | 0.083 | 1 | 0.02% |
| SULFENTRAZONE | 35,000 | 0.153 | (Z) | 0.02% |
| FLUMIOXAZIN | 33,000 | 0.165 | (Z) | 0.02% |
| BROMOXYNIL OCTANOATE | 32,000 | 0.22 | (Z) | 0.01% |
| CLETHODIM | 19,000 | 0.078 | (Z) | 0.01% |
| NICOSULFURON | 19,000 | 0.051 | (Z) | 0.01% |
| THIFENSULFURON | 19,000 | 0.014 | 2 | 0.01% |
| IMAZETHAPYR | 9,000 | 0.02 | 1 | 0.00% |
| HALOSULFURON | 7,000 | 0.032 | (Z) | 0.00% |
| PRIMISULFURON | 4,000 | 0.021 | (Z) | 0.00% |
| FLUTHIACET-METHYL | 3,000 | 0.008 | (Z) | 0.00% |
| DIQUAT DIBROMIDE | 2,000 | 0.022 | (Z) | 0.00% |
| Total | 214,721,000 | | | |

*Z = no data. Herbicides in bold type are used with GE HR corn varieties.

Source: (USDA-NASS 2019f)

Glufosinate Mode of Action and Phosphinothricin Acetyltransferase

Bialaphos is an herbicidal compound produced by certain species of soil bacterium among the genus *Streptomyces* (Dayan et al. 2009; Dayan and Duke 2014). Bialaphos is a prototoxin, an inactive (non-toxic) precursor of the phytotoxin L-phosphinothricin (referred to as phosphinothricin from here out); bialaphos itself is nontoxic. Bialaphos is converted in plant cells to phosphinothricin, which inhibits

glutamine synthetase, an enzyme necessary for the production of glutamine (an essential amino acid) and ammonia detoxification. Accumulation of ammonia and reduced levels of glutamine in plant tissues interferes with photosynthesis and other metabolic activities, resulting in plant death. Glufosinate is a synthetic form of phosphinothricin that has the same herbicidal activity. Glufosinate is the only commercial herbicide that targets glutamine synthetase.

Resistance to glufosinate is conferred by the enzyme phosphinothricin acetyltransferase (PAT), which inhibits the phytotoxic activity of glufosinate by acetylation. Genes encoding PAT have been isolated from *Streptomyces hygroscopicus* (e.g., the *bar* gene, which is short for bialaphos resistance) (Thompson et al. 1987) and from *S. viridochromogenes* (*pat* gene) (Wohlleben et al. 1988). The *pat* gene, that was introduced into DP202216 corn, is very similar to the *bar* gene with 87 % identity at the nucleotide sequence level; both *bar* and *pat* encode PAT proteins with similar substrate affinity and biochemical activity (Wehrmann et al. 1996).

Herbicide Resistant Weeds

Herbicide resistant (HR) weeds are more of an agronomic than ecological concern, however, HR weed populations can potentially contribute to environmental harms when herbicide use or/and tillage is increased for their control, namely through potential increased soil erosion and run-off of non-point source pollutants (e.g., sediments, pesticides). Thus, HR weeds relative to glufosinate use are discussed below.

While herbicides are an important tool in the control/killing of weeds, they impart selection pressure on plants inherently/naturally resistant to the herbicide, resulting in survival of those particular plants (Owen 2011; Owen 2012; Vencill et al. 2012). HR plants occur naturally within weed populations. They differ slightly in genetic makeup from the rest of the population but remain reproductively compatible. HR plants initially are present in a weed population in extremely small numbers; about 1 in 100,000 to less than 1 in 1,000,000 (Campbell et al. 2015). The repeated use of one herbicide mode of action (MOA)⁸ allows these few resistant plants to survive and reproduce; selects for the naturally resistant weeds (Sherwani et al. 2015). The number of resistant plants then increases in the population.

Weed populations can also develop “evolved” resistance to an herbicide a.i.; where the weed adapts to an external chemical stressor. Key to understanding the evolution of herbicide-resistance traits is identifying the mechanism(s) of herbicide resistance, which are broadly classified into target-site resistance (TSR) and/or non-target-site resistance (NTSR). TSR mechanisms largely involve mutations in the target site of action of an herbicide, resulting in an insensitive or less sensitive target protein of the herbicide (Jugulam and Shyam 2019). For example, evolved TSR can emerge from mutations in genes/proteins involved in the herbicide’s MOA. A mutation can cause a minor change in a protein’s structure, resulting in an herbicide no longer having an adverse effect on the protein, rendering the plant “resistant” to the herbicide (e.g., (Yang et al. 2016; Rey-Caballero et al. 2017)). Additionally, TSR can also evolve as a result of the over-expression or amplification of the target gene (Jugulam and Shyam 2019).

⁸ The MOA is the unique biological mechanism at the cellular/molecular level by which an herbicide is lethal to a plant.

NTSR to herbicides in weeds can be conferred as a result of the alteration of one or more physiological processes, such as herbicide absorption, translocation, sequestration, and/or metabolism. The mechanisms of NTSR are generally more complex to decipher than TSR, can impart cross-resistance to herbicides with different modes of action, and complicates resistance management strategies (Jugulam and Shyam 2019).

Over-reliance on herbicides for weed control in lieu of other methods, and continued issues with the development of HR weed populations, has sparked debate on how to best incorporate herbicides into sustainable cropping systems (Vencill et al. 2012; Duke 2015; Korres et al. 2019). Currently, 48 states report the presence of HR weed populations. This is not a recent concern, nor is it unique to GE crops. Herbicide resistant weed populations have been occurring since the advent and wide-spread use of chemical herbicides in the 1950s. As illustrated in Figure 3-7, significant increases in HR weed populations began to occur in the mid-1980s. Currently, there are 166 unique cases of HR weeds in the United States (weed species by herbicide mode of action (MOA)) (Heap 2020).

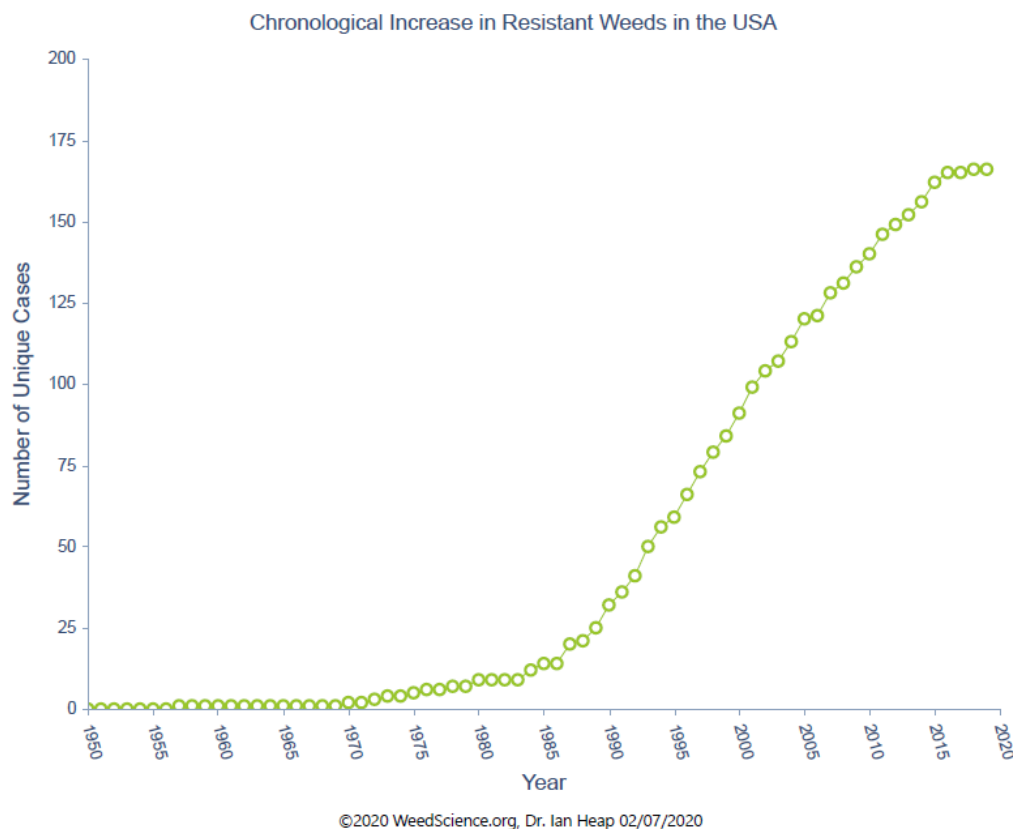


Figure 3-7. HR Weeds in the United States, 1950 – 2015

Source: (Heap 2020)

Globally, weeds have developed resistance to 23 of the 26 herbicide MOAs that are currently available, which reduces the number of weed control options available to growers (Figure 3-8). Most corn growing states have from around 7 to 28 different species of weeds that are herbicide resistant (Heap 2020). There have been no herbicides with completely novel MOAs developed and commercialized over the last

several decades (Duke 2012; Green 2014). Consequently, there are no herbicides with novel MOAs with which to control HR weeds.

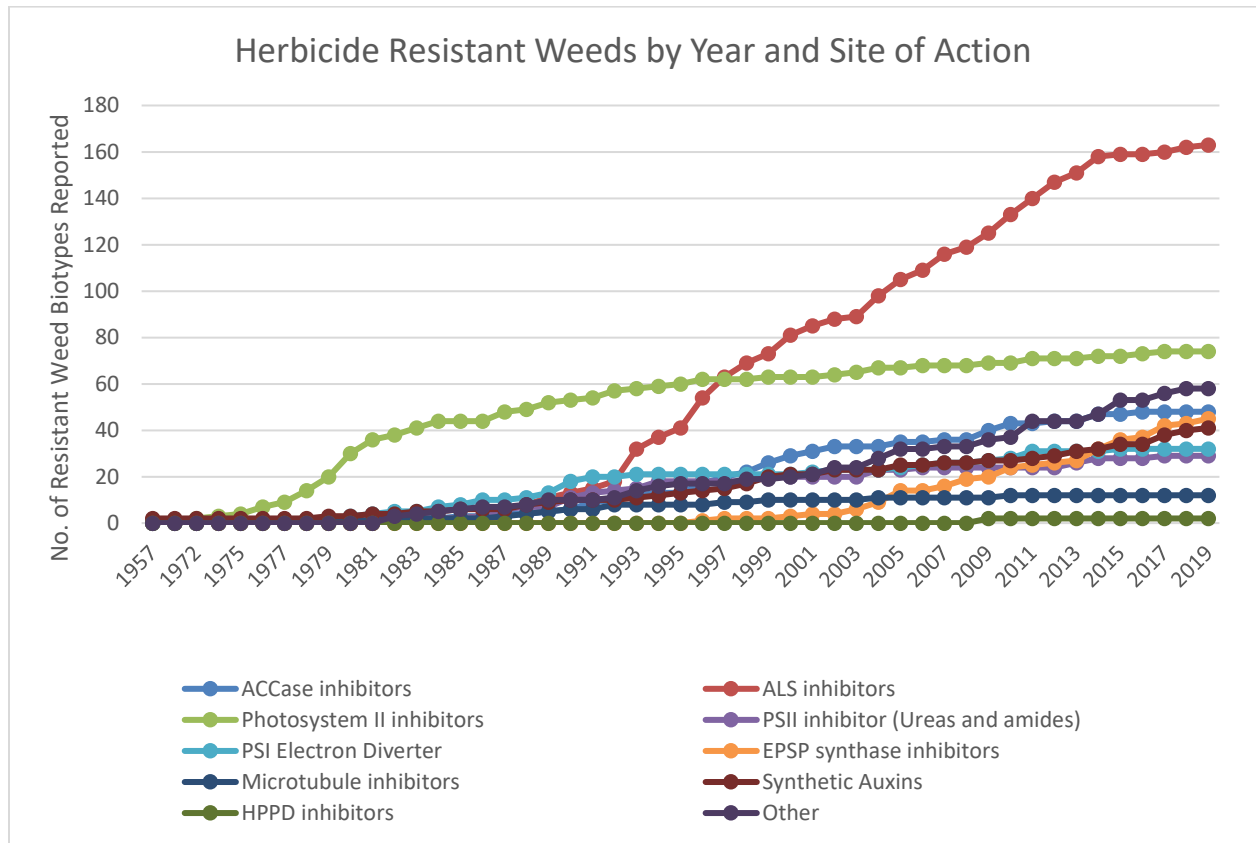


Figure 3-8. Increase in the Development of Herbicide Resistance: Herbicide Modes of Action

The herbicide groups with the most HR weeds are acetolactate synthase (ALS) inhibitors (i.e., imidazolinone and imazethapyr); ACCase inhibitors (i.e., phenylpyrazoline); triazine based photosynthesis II inhibitors (i.e., atrazine); synthetic auxins (i.e, dicamba, 2,4-D); bipyridilium based photosynthesis I inhibitors such as paraquat; glycines, which include the EPSP synthase inhibitor glyphosate; various ureas and amides that inhibit the photosynthesis II process; and dinitroaniline based microtubule inhibitors such as trifluralin.

Source: (Heap 2020)

Problematic is that fact that many HR weed populations in the United States have developed resistance, and continue to, to more than one herbicide MOA. For example, in U.S. corn crops: 16 weed species have populations with confirmed resistance to 2 MOAs, 5 species with confirmed resistance to 3 MOAs, 3 species with confirmed resistance to 4 MOAs, and 1 species with confirmed resistance to 5 MOAs (Heap 2020).

Relative to DP202216 corn: To date there is only one weed species in the United States reported to be resistant to herbicides that contain glufosinate; Italian ryegrass (*Lolium perenne* ssp. *multiflorum*), with populations in orchards in California and Oregon (Heap 2020).

Herbicide Resistant Weed Management

Strategies for managing weeds and avoiding the development of HR weed populations in U.S. agriculture are steadily being refined (e.g., (Norsworthy et al. 2012; Vencill et al. 2012; Garrison et al. 2014; Owen 2016), and others). A combination of preventive, cultural, mechanical, biological, and chemical methods are required for effective weed, and weed resistance management. The coordinated use of these is termed integrated weed management (IWM). Crop producers are advised to, and are implementing, IWM strategies to address the development of HR weeds, strategies developed and recommended by the crop protection and seed industries, the USDA, university extension services, the EPA, state departments of agriculture, the Weed Science Society of America (WSSA), and others. In 2017, the EPA issued PR Notice 2017-2, *Guidance for Herbicide-Resistance Management, Labeling, Education, Training and Stewardship* (US-EPA 2017a), which provides registrants and growers detailed information on slowing the development and spread of HR weeds. The EPA guidance is part of a more holistic, proactive approach involving crop consultants, agricultural commodity organizations, professional /scientific societies, researchers, and the pesticide registrants themselves.

3.2 Physical Environment

3.2.1 Soil Quality

Overview

Relative to crop production, concerns regarding soils are the potential for agronomic practices and inputs to affect soil fertility; erosional capacity; off-site transport of topsoil (sediments), pesticides, and fertilizers; and disturbance of soil biodiversity. Tillage, cover crops, crop rotation, and pesticide and fertilizer inputs can influence the biological, physical, and chemical properties of soil and have a substantial impact on soil fertility, crop yield potential, and erosion (Baumhardt et al. 2015). Soil quality loss occurs through declines in soil organic matter (SOM), minerals (e.g., magnesium, calcium), essential nutrients (e.g., nitrogen, phosphorus, potassium), soil biota, and physical alteration of soil structure (compaction).

Soil Erosion on U.S. Croplands

Due to the rate of soil formation, on the order of millimeters per year, soil is considered a nonrenewable resource that requires conservation and stewardship for sustainable crop production. Soil erosion not only increases fertilizer requirements and production costs, it leads to impaired air and water quality. Soil erosion occurs in all areas of the United States but is more concentrated in those regions where the percentage of total area in cropland is highest and a larger proportion of the land is highly erodible (Magleby et al. 1995; USDA-NRCS 2010; Baumhardt et al. 2015). Excessively eroding cropland soils are concentrated in the Midwest, Southern High Plains of Texas, and Northern Plain States, to include the Corn Belt (Figure 3-9). Where soil erosion occurs through natural processes, the rates of which are determined by soil type, local ecology, and weather, certain tillage and cover crop practices have substantial impacts on the erosion potential of soils.

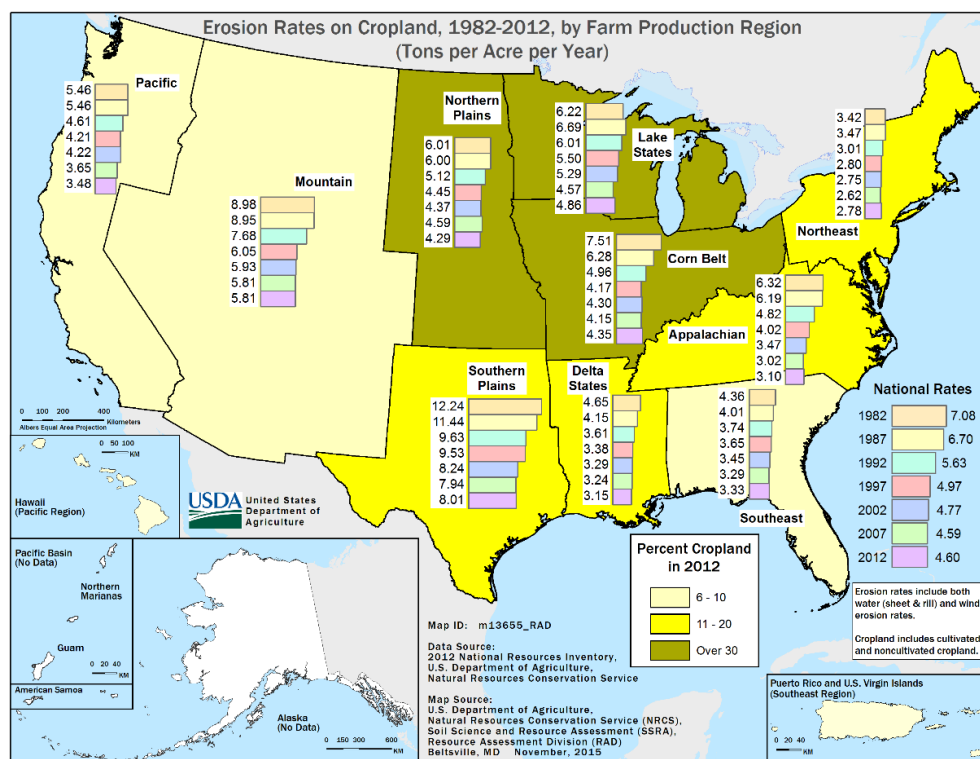


Figure 3-9. Locations and Status of U.S. Croplands Subject to Water and Wind Erosion
Source: (USDA-NRCS 2018b)

As conservation tillage and cover cropping practices have increased since the early 1980s, soil erosion has declined (USDA-NRCS 2010, 2018a). In 1982, total annual water erosion (sheet and rill) on cultivated cropland was 4.18 ± 0.04 tons per acre per year, versus 3.03 ± 0.04 in 2002. Since 2002, water sheet and rill erosion has remained fairly steady at around 2.90 ± 0.05 to 3.03 ± 0.05 . For wind erosion, erosion rates reduced from 3.53 ± 0.06 to 2.15 ± 0.08 over the same time period (USDA-NRCS 2018a). Any decrease in erosion of cropland soils carries with it a corresponding decrease in run-off and introduction of non-point source pollution (NPS) pollutants such as sediments, fertilizer, and pesticides into surface waters.

Because susceptibility to erosion is a key concern on more than half of U.S. cropland (USDA-NRCS 2010), soil management and conservation is a basic component of crop production. Since 1985, conservation programs have specifically targeted highly erodible lands in the United States. A 2017 survey found that 41% of surveyed farmers applied continuous no-till practices, 14% rotational no-till, 27% reduced tillage, and 4% vertical tillage (a type of conservation tillage), with only 14% using conventional tillage (SARE/CTIC 2017). Overall, surveyed farmers applied reduced tillage on 97.7 million acres, conventional tillage on 80 million acres, and no-till on 104.4 million acres (Table 3-4). In addition, they are adopting the use of cover crops to conserve soils and soil quality (SARE/CTIC 2017).

The use of conservation tillage, to include no-till, is attributed, in part, to cultivation of GE HR crops, which provide for effective chemical means of weed control, and can reduce reliance on tillage for control of weeds (Fernandez-Cornejo et al. 2014a). However, the availability of GE HR crops is not the only

driving factor in adoption of conservation tillage practices, as many growers adopted conservation tillage well before GE HR varieties were introduced to the market (Givens et al. 2009).

| Table 3-4. Tillage Practice on U.S. Cropland, 2012 – 2017 | | | | | |
|--|---|---|--|--|---|
| | Total Harvested Cropland | Cropland with reduced tillage, excluding no-till | Cropland with no-till practices | Cropland with intensive tillage practices | Cropland planted to a cover crop (excluding CRP) |
| 2017 | 320,041,858 | 97,753,854 30.54% | 104,452,339 32.64% | 80,005,292 25.00% | 15,390,674 4.81% |
| 2012 | 314,964,600 | 76,639,804 24.33% | 96,476,496 30.63% | 105,707,971 33.56% | 10,280,793 3.26% |

Source: (USDA-NASS 2014, 2019a)

In summary, land management practices for corn cultivation can affect soil quality and erosion relative to the tillage, pesticide application, crop rotation, soil amendment, and cover cropping practices applied. GE HR crops are correlated with use of conservation tillage practices, which sustain soil health and water retention, and reduce runoff (Fernandez-Cornejo et al. 2014a; Claassen et al. 2018); there are no adverse effects on soil health unique to of GE HR crops that have been identified since their adoption in the late 1990s. All growers producing crops on highly erodible land are required to maintain and implement a soil conservation plan that substantially reduces soil loss, and is approved by the USDA National Resources Conservation Service (NRCS). These plans are prepared by the grower pursuant to the Food Security Act of 1985 (P.L. 99-198, Farm Bill), which included a number of provisions designed to conserve soil and water resources, and minimize erosion. The 2014 and 2018 Farm Bills have continued the requirement that producers adhere to conservation compliance guidelines to be eligible for conservation programs administered by USDA-FSA and USDA-NRCS. State agencies likewise provide assistance in development and implementation of soil conservation plans.

3.2.2 Water Resources

Agronomic inputs, and in many areas tillage and irrigation, are necessary for efficient crop production. These practices and inputs can, however, lead to the impairment of surface waters through runoff of pesticides, fertilizers (nutrients), and soil sediment (Bricker et al. 2008; CENR 2010). Groundwater can also be impacted by agronomic inputs via leaching, as well as through irrigation withdraw. In many area of Midwest corn yields can either be increased by irrigation, or is necessary for production. Irrigated corn accounts for 58% of total annual corn production in the Western U.S. Corn Belt (Grassini et al. 2011).

While pollutants come from various sources, the National Water Quality Assessment indicate that agricultural nonpoint source (NPS) pollution is a leading cause of impairment of surveyed rivers and streams, the third largest source for lakes/ponds, the second largest source of impairments to wetlands, and a major contributor to contamination of surveyed estuaries, coastal areas, and ground water (US-EPA 2019f). The most common NPS contaminants in agricultural run-off are sediment, nutrients such as nitrogen and phosphorus, and pesticides (Table 3-5), all of which can adversely affect aquatic ecosystems.

| Table 3-5. Causes of Impairment in Assessed Waters, 2019 | | | |
|---|-------------------------------------|------------------------|-----------------|
| Rivers, Streams | Lakes, Reservoirs, Ponds | Bays, Estuaries | Wetlands |

| | Miles | Rank | Acres | Rank | Miles | Rank | Acres | Rank |
|-------------------|--------|------|---------|------|-------|------|-------|------|
| Nutrients | 118831 | 3rd | 3943395 | 2nd | 18279 | 2nd | 67849 | 6th |
| Sediment | 138874 | 2nd | 502200 | 12th | 400 | 18th | 1237 | 15th |
| Pesticides | 18069 | 16th | 412672 | 13th | 7543 | 8th | 202 | 21st |

Shown are national water quality data reported by the States to EPA under Section 305(b) and 303(d) of the Clean Water Act. The data shown is the most current available, which varies widely among states, spanning the years from 2004 to 2016. The EPA lists around 34 different factors that are the cause impairment of U.S. waters. For rivers and streams, the EPA lists sediments as the second most frequent cause of impairment, nutrients third, and pesticides sixteenth. For lakes, reservoirs, and ponds, nutrients are second, sediments twelfth, and pesticides thirteenth. For bays and estuaries, nutrients are second, sediments eighteenth, and pesticides 8th. For wetlands, nutrients are sixth, sediments fifteenth, and pesticides twenty-first. Source: (US-EPA 2019f)

Excess sediment in runoff can adversely affect aquatic ecosystems by covering fish breeding substrates, increased turbidity, and impairing growth of aquatic plants. Nutrient runoff (e.g., nitrogen and phosphorus) runoff from agricultural fields can contribute to eutrophication of surface waters. Nearly two-thirds of the U.S. estuaries have moderate to high levels of eutrophication. Eutrophic symptoms cause impairments to human uses and to living resources, including harmful algal blooms and hypoxic/anoxic conditions⁹ that lead to fish kills, fish consumption warnings (to prevent human health problems), declines in tourism, and impacts on fisheries (Bricker et al. 2008; CENR 2010). These conditions occur in estuaries along all coasts, but are most prevalent in estuaries along the Gulf of Mexico and Mid-Atlantic coasts (Figure 3-10). Watersheds with a high potential to discharge nitrogen from agriculture to estuaries are located primarily in the Heartland, Mississippi Portal, and Southern Seaboard regions (Wiebe and Gollehon 2006a; CENR 2010).

⁹ Hypoxia means low dissolved oxygen concentrations. Anoxia means a total depletion of dissolved oxygen. Both conditions are harmful to aquatic biota.

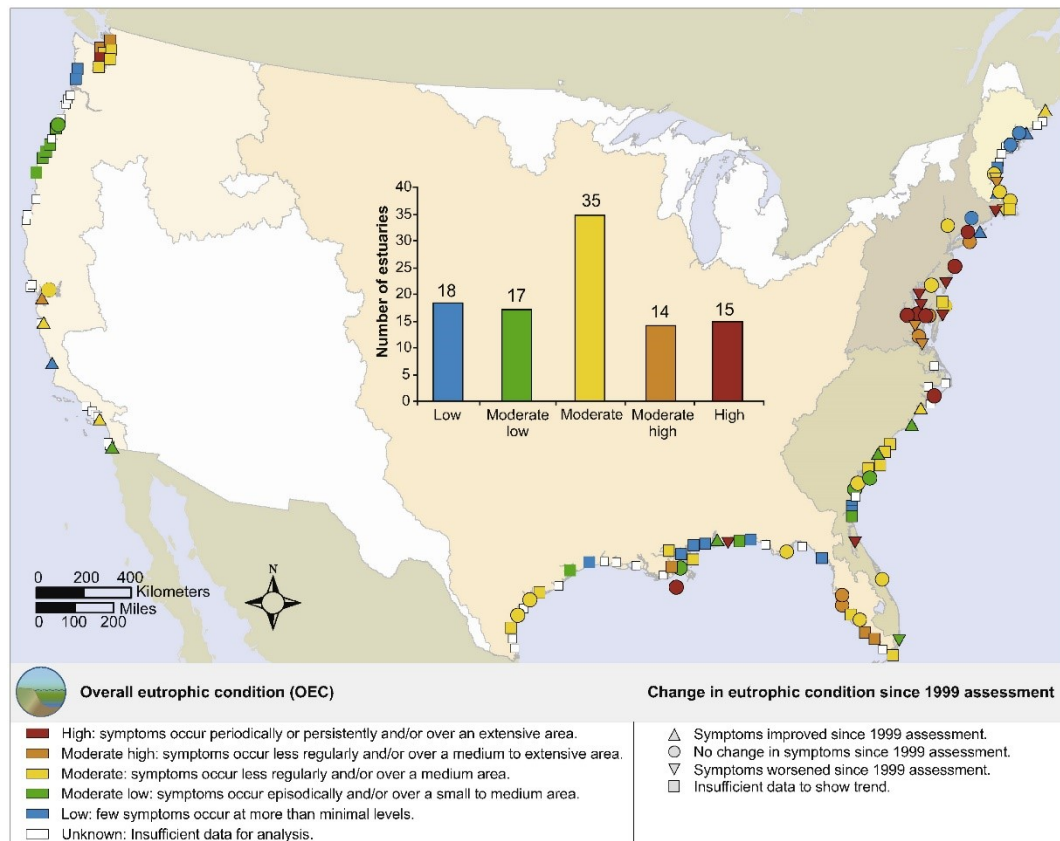


Figure 3-10. Overall Eutrophic Conditions on a National Scale

Source: (Bricker et al. 2008)

Human uses impacted by impairment of surface waters for all regions include commercial and recreational fishing, shellfish harvesting, fish consumption, swimming, aesthetics, and tourism (CENR 2010). The overall top four causes of these use impairments were listed as agriculture (crops and animal operations), wastewater treatment plants, urban runoff, and atmospheric deposition (Bricker et al. 2008; Boesch 2019). Animal operations and crop agriculture were noted mostly for systems in the mid- and South Atlantic regions while exurban development (outside boundaries of urban areas) was reported in the South Atlantic region. In all regions except for the North Atlantic, non-point sources remain a primary focus (CENR 2010).

The re-engineering of the Midwest over the past 50 years with tile drainage systems that allow farmers to control subsurface water levels has benefitted U.S. agriculture through increased yields, but has negatively affected water quality by facilitating water and its solutes—such as nitrogen, phosphorus, pesticides, and sediment—into streams and rivers without allowing natural elimination/attenuation processes to occur (CENR 2010; Ribaud et al. 2011).

Use of tile drainage for corn can greatly contribute to nitrogen loss. Tiles, however, provide a rapid conduit for soluble nitrate, effectively bypassing any attenuation that may occur in the soil. USDA-ARMS data indicate that nearly 26% of treated cropland is tiled, most of this in corn production (Ribaud et al. 2011).

et al. 2011). USDA- ARMS data indicate that about 71% of tiled acres do not meet nitrogen management criteria.

Nitrogen and phosphorus run-off is particularly problematic for Gulf of Mexico ecosystems and fisheries (Wiebe and Gollehon 2006b; US-EPA 2019g). Much of the tile-drained cropland is located in the Mississippi River Basin, which has implications for hypoxia in the Gulf of Mexico (Ribaud et al. 2011). The most heavily tile-drained areas of North America are also the largest contributing source of nitrate to the Gulf of Mexico, leading to seasonal hypoxia (David et al. 2010). Agricultural sources contribute around 70% of the nitrogen and phosphorus delivered to the Gulf of Mexico, versus 9% to 12% contribution from urban sources (Alexander et al. 2008). Corn, specifically, accounts for about 45% of U.S. crop acreage receiving manure, and 65% of the 8.7 million tons of nitrogen fertilizer applied by farmers each year (Ribaud et al. 2011). Nitrogen run-off from cornfields in the Mississippi River Basin is the single largest source of nutrient pollution to the Gulf of Mexico's "dead zone" (Figure 3-11).

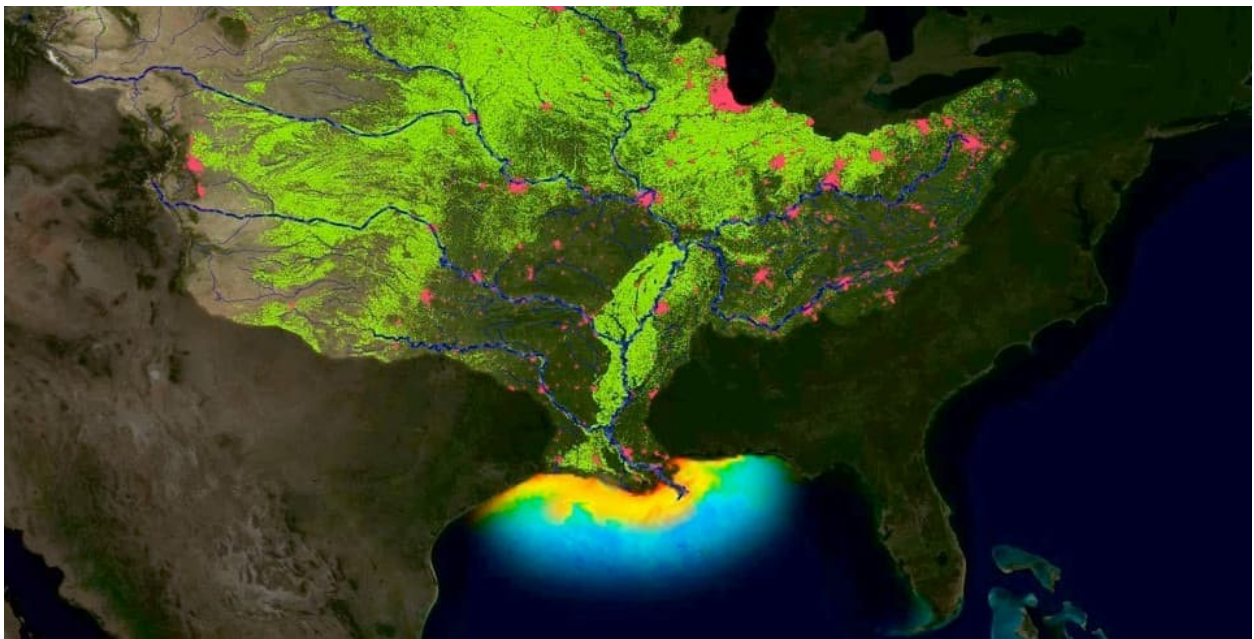


Figure 3-11. Agricultural Run-Off: Mississippi River Watershed

This image from a NOAA Environmental Visualization Lab animation illustrates how run-off from farms (green areas) and cities (red areas) drains into the Gulf of Mexico (GOM). This run-off contains nutrients from fertilizers, wastewater treatment plants, and other sources, which leads to hypoxic "dead zones" on an annual basis; areas in the GOM where the oxygen concentration is so low that aquatic biota can suffocate and die. The largest hypoxic zone in the United States, and the second largest hypoxic zone worldwide, forms in the northern Gulf of Mexico near New Orleans. Source: (NOAA 2019)

3.2.2.1 Water Quality Regulation and Improvement

Point and Non-Point Source Discharges

Under Section 404 of the Clean Water Act (CWA), it is unlawful to discharge any pollutant from a point source into navigable waters, unless a permit authorized under the CWA was obtained. The EPA's National Pollutant Discharge Elimination System (NPDES) permit program controls these point source

discharges (US-EPA 2019i). NPS pollution, which is the primary type of discharge from cropping systems, is not regulated under the CWA; rather, it is left largely to voluntary controls implemented by states and local authorities. Thus, many crop production activities do not require a Section 404 permit, even where they involve discharges of dredged or fill materials into waters of the United States. To be exempt, the farming activity must be part of an ongoing farming operation and cannot be associated with bringing a wetland into agricultural production or converting an agricultural wetland to a non-wetland area.

Diffuse runoff from nonpoint sources, such as agriculture fields, can be difficult to control, although improved production methods that reduce tillage, optimize fertilizer application, and buffer fields from waterways can mitigate water quality impairments. Due to the potential impacts of agriculture on water resources, various national and regional efforts are underway to reduce NPS contaminants in agricultural run-off, and run-off itself, such as the EPA's Mississippi River/Gulf of Mexico Hypoxia Task Force (US-EPA 2019g) and USDA-NRCS National Water Quality Initiative (NWQI) (USDA-NRCS 2019a). For example, through the NWQI, the NRCS and partners (e.g., local and state agencies, nongovernmental organizations) work with producers and landowners to implement voluntary conservation practices that improve water quality. The NWQI program is in its 8th year and extended through 2023. It provides funding for financial and technical assistance for conservation practices, and in 2018 NRCS invested \$30 million in targeted assistance to help farmers and ranchers improve water quality in high priority streams and rivers. State water quality agencies and other partners contribute additional resources for watershed planning, program implementation, and outreach, and for monitoring efforts to track water quality improvements over time.

Several other legislative drivers also influence how Federal agencies work on coastal water quality including the Clean Water Act; the Food, Conservation, and Energy Act ("Farm Bill"); the Energy Independence and Security Act of 2007; and the Coastal Zone Management Act, and The Harmful Algal Bloom and Hypoxia Research and Control Act. Responsibility for resolving hypoxia spans several Federal agencies (U.S. Department of Agriculture, U.S. Geological Survey, U.S. Environmental Protection Agency, and National Oceanic and Atmospheric Administration), which oversee research and management/control programs. States play a critical role in monitoring and managing eutrophication (CENR 2010).

Pesticides

The EPA determines use requirements for pesticides that are intended to be protective of water quality, including drinking water, and to protect aquatic life (US-EPA 2019l, c). The EPA provides label use restrictions and guidance for product handling intended to prevent impacts to surface and groundwater.

3.2.3 Air Quality

National Ambient Air Quality Standards

Because air pollution directly affects human health and can cause adverse environmental impacts maintaining air quality is a primary U.S. regulatory goal. The EPA establishes National Ambient Air Quality Standards (NAAQS) pursuant to the Clean Air Act (CAA) that are intended to protect public health and the environment. NAAQS are established for six criteria pollutants: ozone (O₃), nitrogen dioxide (NO₂), carbon monoxide (CO), sulfur dioxide (SO₂), lead (Pb), and particulate matter (PM). In

addition to criteria pollutants, the EPA regulates 187 hazardous air pollutants, such as ammonia and hydrogen sulfide.

All areas of the United States are classified as to their consistency with the NAAQS; for example, having attained NAAQS, or not. States enforce the NAAQS through creation of state implementation plans, which are designed to achieve EPA-established NAAQS. The EPA designates a region as being in attainment for a criteria pollutant if atmospheric concentrations of that pollutant are below the NAAQS, or being in nonattainment if criteria pollutant concentrations violate the NAAQS.

Crop production practices can generate air pollutants that can contribute to challenges in maintaining regional NAAQS. Agricultural emission sources include: smoke from agricultural burning (PM); fossil fuel consumption associated with equipment used in tillage, pesticide application, and harvest (CO₂, NO_x, SO_x); soil particulates from tillage (PM); soil nitrous oxide (N₂O) emissions from the use of fertilizers/manure; and atmospheric emissions through the volatilization of pesticides, and gases from manure (Aneja et al. 2009; US-EPA 2020a).

Prescribed burning is a land treatment used under controlled conditions to accomplish resource management objectives. Open combustion produces particles of widely ranging size, depending to some extent on the rate of energy release of the fire (US-EPA 2019b). The extent to which agricultural and other prescribed burning may occur is regulated by individual state implementation plans to achieve compliance with the NAAQS. Prescribed burning of fields would likely occur only as a pre-planting option based on individual farm characteristics.

While the EPA establishes NAAQS, the standards do not set emission control requirements for any particular industry, including agriculture. The USDA and EPA provide guidance for regional, state, and local regulatory agencies, and farmers, on how to best manage agricultural emissions sources (USDA-EPA 2012). These measures allow stakeholders the flexibility in choosing which measures are best suited for their specific situations/conditions and desired purposes. The EPA has developed USDA-approved measures to help manage air emissions from cropping systems to help satisfy SIP requirements. The EPA recommends that in areas where agricultural activities have been identified as a contributor to a violation of NAAQS, USDA-approved conservation systems and activities may be implemented to limit emissions. The USDA Environmental Quality Incentives Program Air Quality Initiative provides financial and technical assistance to help farmers and ranchers limit air pollution (USDA-NRCS 2019c).

Other conservation practices, as required by USDA to qualify for crop insurance, and federal loans and programs, effectively reduce crop production impacts to air quality through the use of windbreaks, shelterbelts, reduced tillage, and cover crops that promote soil protection on highly erodible lands.

Pesticides

Apart from NAAQS emissions, spray drift, and volatilization of pesticides from soil and plant surfaces, can result in the introduction of constituent chemicals into the air; which can present human health risks, and risks to nearby crops. Thus, drift and volatilization of pesticides can be a source of concern to both farmers and the general public in regard to potential environmental and human health effects.

Volatilization is dependent on pesticide chemistry, soil wetness, and temperature (US-EPA 2019k). Drift

is dependent on wind conditions and applicator practices, to include application equipment features such as nozzle size (US-EPA 2019j).

The EPA's Office of Pesticide Programs, which regulates the use of pesticides, introduced initiatives to help pesticide applicators minimize off-target pesticide drift. The EPA's voluntary Drift Reduction Technology Program was developed to encourage the manufacture, marketing, and use of spray technologies that reduce pesticide drift. The EPA is also working with pesticide manufacturers through the registration and registration review programs on improvements to pesticide label instructions to reduce drift and volatilization (US-EPA 2019k, j).

3.3 Biological Resources

3.3.1 Soil Biota

Soil biota consist of micro-organisms (bacteria, fungi, archaea and algae), soil animals (protozoa, nematodes, mites, springtails, spiders, insects, and earthworms), and plants (e.g., algae) living all or part of their lives in or on the soil, or pedosphere (Fortuna 2012). Soil biota play a key role in the formation and turnover of soil organic matter (including mineralization), biodegradation of anthropogenic substances (e.g., pesticides), nutrient cycling, suppression of plant diseases, promotion of plant growth, soil structure formation, and most biochemical soil processes (Gupta et al. 2007; Fortuna 2012; Parikh and James 2012). Plant roots, including those of corn, release a variety of compounds into the soil creating a unique environment for microorganisms in the rhizosphere (root zone). Millions of species of soil organisms exist but only a fraction of them have been cultured and identified (Fortuna 2012).

Some microorganisms can cause plant diseases, which can result in substantial economic losses in crop production, and soils treated to control plant pathogens. Soil borne corn crop diseases include fungal corn rusts, corn leaf blights, ear smuts, ear and kernel rot fungi, and maize mosaic viruses.

Potential changes to the soil microbial community as a result of cultivating GE crops has been of much research interest since their introduction in the late 1990s (e.g., (Motavalli et al. 2004; Locke et al. 2008; Kremer and Means 2009)). The majority of these studies have focused in Bt crops due to their insecticidal activity. Potential direct impacts could possibly include changes to the structural and functional community near the roots of GE plants due to altered root exudation or the transfer of novel proteins into soil, or a change in microbial populations due to the changes in agronomic practices used to produce GE crops (e.g., pesticides, fertilizers, and tillage practices). Most studies show no significant effect of Bt crop traits on soil community structures (Kowalchuk et al. 2003; Hannula et al. 2014; Zaman et al. 2015; Xie et al. 2016; Yasin et al. 2016).

Relative to crop production, the main factors affecting soil biota populations and diversity are soil type (texture, structure, organic matter, aggregate stability, pH, and nutrient content), plant type (providers of specific carbon and energy sources into the soil), and agricultural management practices (crop rotation, tillage, pesticide and fertilizer application, and irrigation (Kowalchuk et al. 2003; Garbeva et al. 2004; Gupta et al. 2007)). Pesticides used on corn crops can, relative to the application rates, toxicity, and frequency of exposure of soil biota to a pesticide, potentially impact soil communities (discussed further

below). Climate, particularly the water and heat content of soil, is a principal determinant of soil biological activity.

Pesticides

The continued use of pesticides are considered necessary to commercial crop production if the projected global demands for food are to be met (FAO 2017). The capacity of the soil to filter, buffer, degrade, immobilize, and detoxify pesticides is a function of soil biota, particularly soil microbiota.

Some pesticides used on corn crops can, relative to the application rates, mode of action (potential toxicity), and frequency of exposure of soil biota to a pesticide, potentially impact soil communities (Stevenson et al. 2002; Locke and Zablotowicz 2004). A recent global assessment of the impact of plant protection products on soil functions and soil ecosystems concluded that most agricultural inputs can cause changes in the amount, activity, diversity, and community structures of soil organisms (FAO 2017). Changes in community structure are in fact the most common type of effects observed with pesticides (FAO 2017).

In general, the effects of pesticides can lead to both significant decreases and increases in the attributes of soil organisms, such as biomass, enzymatic activity, soil respiration, and species composition. The challenge lies in interpreting these changes relative to whether such changes reflect adaptive responses in soil organisms/communities, or potentially harmful effects, such as decreased species diversity, impeded soil functions, and diminished soil productivity (FAO 2017). Changes in diversity and community structure might not always lead to changes in ecosystem function, soil processes, as the relationship between the diversity of soil organisms and soil ecosystem functioning is complex, and there exists redundancy among soil organisms and soil processes (Nielsen et al. 2011; Hannula et al. 2014). There is very limited evidence that the observed effects of pesticides on soil organisms have led to significant and long-lasting decreases in soil functions (FAO 2017). However, the inability to clearly link the observed effects of pesticides on organisms with soil functions is a major limitation of the current literature (FAO 2017).

There is more evidence for harmful effects of pesticides on earthworms. Specifically, the negative effects of copper-based fungicides are well-established, and recent evidence indicates that neonicotinoids are particularly toxic to earthworms (FAO 2017). Generally, earthworms are subject to pronounced, long-term effects when exposed to fungicides and insecticides (targeting taxa among Fungi and Animalia kingdoms), while herbicides (targeting taxa among the Planta kingdom) have limited or no effects (FAO 2017).

While the application of a pesticides may lead to the local suppression of a taxonomic unit of soil organisms, the resilience of soil organisms, or ability to adapt, and functional redundancy across taxa, serve to limit the effects of pesticides on soil ecosystem processes (FAO 2017). Fundamentally, the vast majority of soil organisms have yet to be identified, and hence, a comprehensive assessment of the effects of pesticides on soil biota is not possible at this time (FAO 2017).

Studies on the potential effects of glufosinate, specifically, on soil microbial communities has yielded differing results (see, e.g., (Bartsch and Tebbe 1989; Gyamfi et al. 2002; Lupwayi et al. 2004; Wibawa et al. 2010). For example, Sessitsch et al. (2005) concluded that rhizosphere bacteria associated with

glufosinate tolerant oilseed rape were affected by the genetic modification of the plant; however, the effects were considered minor as compared to the influence of the plant growth stage. Gyamfi et al. (2002) suggest that some of the observed microbial population shifts associated with glufosinate may be caused by an increase in herbicide-degrading soil microbes following application, due to use of glufosinate by microbes as a source of nitrogen (Bartsch and Tebbe 1989). Other research suggests that glufosinate may beneficially inhibit the activity of crop pathogens such as bacterial blight (Pline 1999) and grapevine downy mildew (Kortekamp 2011). Glufosinate has also been suggested to inhibit glutamine synthetase activity in pathogenic fungi or fungal like organisms, similar to inhibition of glutamine synthetase in plants (Kortekamp 2011). In general, glufosinate applied at recommended rates is not recognized as having significant or consistent adverse effects on soil microbial diversity (e.g., see (Gyamfi et al. 2002; Lupwayi et al. 2004; Wibawa et al. 2010)).

In most cases, crop and soil management practices that increase soil organic matter and plant residues, such as conservation tillage, impart attributes to the soil environment that enhance microbial degradation of herbicides (Locke and Zablotowicz 2004, Locke, Zablotowicz et al. 2008), and hinder herbicide movement into surface and groundwater.

3.3.2 Animal Communities

3.3.2.1 Birds and Mammals

Intensively cultivated lands, such as commercial cornfields, provide less suitable habitat for wildlife than natural areas. As such, the types and numbers of animal species found in and near cornfields will be less diverse as compared to unmanaged lands. Cornfields can, however, provide both food and cover for wildlife, including a variety of birds as well as large and small mammals. Blackbirds, grackles, and crows often feed on developing ears in corn fields following pollination early in the grain filling period. Large flocks of these birds can cause a significant damage to corn crops. Damage is often most prevalent along field edges and nearby wooded areas, but can extend throughout a large field. The types and numbers of birds that inhabit cornfields vary regionally and seasonally but for the most part the numbers are low. Bird species commonly observed in corn fields include (Best et al. 1990):

- Red-winged blackbird (*Agelaius phoeniceus*)
- Grackle (*Quiscalus quiscula*)
- Horned lark (*Eremophila alpestris*)
- Brown-headed cowbird (*Molothrus ater*)
- Vesper sparrow (*Pooecetes gramineus*)
- Ring-necked pheasant (*Phasianus colchicus*)
- Wild turkey (*Meleagris gallopavo*)
- American crow (*Corvus brachyrhynchos*)
- Blackbird (*Turdus merula*)
- Various quail species.

Following harvest, it is also common to find large flocks of migratory bird species foraging in cornfields, such as Canada geese (*Branta canadensis*), snow geese (*Chen caerulescens*), sandhill cranes (*Grus canadensis*), and various other species (Taft and Elphick 2007; Sherfy et al. 2011).

A variety of larger mammals forage on corn at various stages of plant growth. Large- to medium-sized mammals that are common foragers of cornfields include (Fleharty and Navo 1983; ODNR 2001):

- White-tailed deer (*Odocoileus virginianus*)
- Raccoon (*Procyon lotor*)
- Wild boar (*Sus scrofa*)
- Woodchuck (*Marmota monax*)

The most notable of these is the white-tailed deer which often inhabit woodlots adjacent to cornfields and frequent these fields for both food and cover, especially in mid-summer. Agricultural crops, particularly corn and soybean, comprise a major portion of deer diets in Midwestern agricultural regions; deer are considered responsible for more corn damage than any other wildlife species (MacGowan et al. 2006). Cornfields are vulnerable to deer damage from emergence through harvest, although damage to corn at the tasseling stage most directly impacts yield (Stewart et al. 2007). Losses to crop yield from feeding by raccoons have also been documented (Beasley and Rhodes Jr. 2008). Mature corn has been shown to constitute up to 65% of the diet of raccoons in some areas prior to harvest (MacGowan et al. 2006).

As with larger mammals, small mammal use cornfields for shelter and forage. Some of the more common small mammals common to corn fields are (USDA-NRCS 1999; U-Illinois-Ext 2000; Sterner et al. 2003).

- Deer mouse (*Peromyscus maniculatus*)
- House mouse (*Mus musculus*)
- Meadow vole (*Microtus pennsylvanicus*)
- Thirteen-lined ground squirrel (*Spermophilus tridecemlineatus*)

3.3.2.2 Invertebrates

Although certain invertebrates in corn fields are considered pests, such as the European corn borer (*Ostrinia nubilalis*) and corn rootworm (*Diabrotica* spp.), the majority are beneficial, performing valuable functions; they pollinate plants, contribute to the decay and processing of organic matter, reduce weed seed populations through predation, cycle soil nutrients, and prey on other insects and mites that are considered to be plant pests (Landis et al. 2005). Some of these beneficial species include the convergent lady beetle (*Hippodamia convergens*), carabid beetles, the caterpillar parasitoids (e.g., *Macrocentrus cingulum*), and the predatory mite (*Phytoseiulus persimilis*) (Landis et al. 2005; Shelton 2011). Earthworms, termites, ants, beetles, and millipedes contribute to the decay of organic matter and the cycling of soil nutrients (Ruiz et al. 2008).

3.3.3 Plant Communities

Plant diversity in surrounding field areas is an important component of a sustainable agricultural system (Scherr and McNeely 2008; CBD 2019b). Hedgerows, woodlands, fields, and other surrounding habitat serve as important reservoirs for beneficial insects, as well as plant pests. Corn fields and field edges are also habitat for weeds that adversely impact corn production directly through interference and resource competition (discussed below), and can also harbor both beneficial or damaging insects and plant microbes. Most weeds, however, provide valuable ecosystem services. By providing habitat, pollen and nectar resources, and serving as hosts, plants adjacent to corn fields can support a suite of beneficial arthropod species that serve as pollinators of insect-pollinated crops, and biological control agents, insects

that prey on corn plant pests, such as lady beetles, spiders, and parasitic wasps (Scherr and McNeely 2008; Nichols and Altieri 2012). However, for corn production, pollinators would not be as valued from an agronomic perspective, as corn is primarily wind and hand pollinated. Surrounding plant communities can also help regulate run-off, reduce soil erosion, and improve water quality. Hence, effective management of surrounding plant communities can provide benefits to corn crop production via control of some insect pests and agricultural run-off (Altieri and Letourneau 1982), and support pollinator services to other plants that benefit from insect pollination (Nichols and Altieri 2012).

Members of the plant communities in and around cornfields that adversely affect corn cultivation are generally characterized as weeds, and weed control programs are fundamental components of crop production in maximizing crop yield and quality (see 3.1.2.2.3–Weed and Herbicide Resistant Weed Management). The types of weeds in and around a cornfield vary according to the region in which the corn is planted, although some of the most common weeds in U.S. cornfields include (U-Missouri 2009):

- Giant foxtail (*Setaria faberi*);
- Giant ragweed (*Ambrosia trifida*);
- Velvetleaf (*Abutilon theophrasti*);
- Common lambsquarters (*Chenopodium album*);
- Common ragweed (*Ambrosia artemisiifolia*);
- Common cocklebur (*Xanthium pensylvanicum*);
- Canada thistle (*Cirsium arvense*);
- Johnsongrass (*Sorghum halepense*);
- Fall panicum (*Panicum dichotomiflorum*); and
- Marestalk (*Conyza canadensis*).

In addition to the more common weed species listed above, the following species are also noted as problematic in corn growing regions of the U.S. (ISU-Ext 2003; USDA-NRCS 2015):

- Cressleaf groundsel (*Senecio glabellus*);
- Purple deadnettle (*Lamium purpureum*);
- Biennial wormwood (*Artemisia biennis*);
- Asiatic dayflower (*Commelina communis*);
- Hophornbeam copperleaf (*Acalypha ostryfolia*);
- Burcucumber (*Sicyos angulatus*);
- Wild buckwheat (*Polygonum convolvulus*);
- Kochia (*Kochia scoparia*);
- Waterhemp (*Amaranthus rudis*);
- Palmer amaranth (*Amaranthus palmeri*);
- Star-of-Bethlehem (*Ornithogalum umbellatum*);
- White campion (*Silene latifolia*);
- Wild four o' clock (*Mirabilis nyctaginea*); and
- Pokeweed (*Phytolacca americana*).

Weed populations among corn fields change in response to agricultural management decisions. New weeds emerge as cropping practices change and when growers may fail to recognize or properly identify a

plant as a weed. Collectively, management decisions will impart selection pressures¹⁰ on the weed communities, that can result in shifts of weed species on a local level (i.e., field level) (Owen 2011; Owen 2012; Vencill et al. 2012). Weed shifts are generally most pronounced when a single or small group of weeds increases in abundance relative to other weed populations. As discussed in more detail in 3.1.2.2.3–Weed and Herbicide Resistant Weed Management, herbicide resistance in weeds naturally evolves when a plant survives and reproduces after exposure to a dose of herbicide, usually lethal to the wild type, passing this ability on to future generations of the plant.

Most relevant to environmental review of GE crop plants are those sexually compatible plant communities with which the GE crop plant can interbreed, discussed following.

3.3.4 Gene Flow and Weediness of Corn

Gene flow as a mechanism for the unintended movement of GE plant transgenes to non-GE crops, other GE crops, and wild or feral plants has been a topic of interest and research since the advent of GE crops in the 1990s. Factors such as the particular type of GE plant being grown, adjacent cropping systems, occurrence of wild relative(s) with which the GE plant may crossbreed, and GE trait all require consideration when evaluating the potential environmental impacts that could result from gene flow (Warwick et al. 2009; Ellstrand 2014). Gene flow among GE crops and conventional and organic cropping systems is of particular interest to farmers, food processors, and international, federal, and state regulators, as such gene flow can adversely affect crop management, net returns on crops and their products, and domestic and international trade. Gene flow from GE plants to wild relative species is a topic of interest among ecologists and environmentalists, as well as federal and state regulators, due to concerns that a transgene may confer herbicide resistance or weediness traits to, or alter the fitness of, wild relative species.

Of particular interest to APHIS is the possible occurrence of gene flow from a GE plant to sexually compatible wild relative species that could lead to introgression of the trait gene into a wild population, and development of a phenotype that could adversely affect agricultural interests and/or the environment.

3.3.4.1 Definition and Types of Gene Flow

Gene flow between sexually compatible GE crop plants and non-GE crop plants, as well as wild plants, is possible, and an important consideration in the design of field trials, seed production, and cultivation of GE and non-GE crops. The term “gene flow” can be synonymous with “outcrossing”¹¹ and the terms are used here interchangeably. Neither term implies the long-term persistence or introgression¹² of gene(s)

¹⁰ Selection pressure may be defined as any event or activity that reduces the reproductive likelihood of an individual in proportion to the rest of the population of that one individual. In agriculture, selection pressure may be imparted by any facet of management in the production of a crop, including the type of crop cultivated, strategy of pest management, or when and how a crop is planted or harvested.

¹¹ Outcrossing refers to the natural occurrence of gene flow among sexually compatible plants, or intentional introduction of genetic material into a breeding line with pollen from a different plant of the same species, often one that is a different variety.

¹² Introgression is the permanent incorporation of a gene(s) from one species into the genome of another. Introgression typically follows hybridization and the repeated backcrossing of interspecific hybrids. A prerequisite for introgression from a crop plant gene to a wild relative population is the occurrence of hybrids sufficiently fit to produce progeny, and for such

into a recipient population. A hybrid is the offspring of two genetically dissimilar but sexually compatible species, generally within the same genus, although hybrids between different genera are possible.

In commercial corn crops gene flow occurs via pollination. Corn plants contain both male and female reproductive structures and reproduce by both cross-pollination and self-pollination. The physical and biological processes and mechanisms by which pollen mediated gene flow occurs is not unique or different for GE or non-GE corn plants. Hence, the likelihood of occurrence of pollen mediated gene flow among all crop types (conventional, GE, and organic) and among corn plants and wild relative species is the same.¹³ As a group, GE plants are no more or less likely to hybridize with wild relatives than their non-GE counterparts (Ellstrand 2014). The rate and success of pollen mediated flow is dependent on numerous factors such as the: presence, abundance, and distance (proximity) of sexually-compatible plant species; overlap of flowering times among populations; method of pollination; biology and amount of pollen produced; and, weather conditions, including temperature, wind, and humidity.

Gene flow is also mediated by seed inadvertently entering the environment during transport or being incorporated into a crop field's soil as a result of crop plant residue (e.g., volunteer GE plants, discussed below). Seed-mediated gene flow depends on many factors, including the absence, presence, and extent of seed dormancy; various dispersal pathways (animals, humans, water, wind); and environmental conditions—all of which facilitate or deter seed germination. Seed mediated gene flow from the occurrence of unintended volunteer GE plant populations is an important consideration, as GE volunteer plants can occur where seed is spilled or otherwise dispersed in or outside the crop field, and GE volunteer plants can occur in subsequent crops planted in the same field as the GE crop. Such volunteer GE plant populations can serve as reservoirs from which a transgene could be passed into the genome of a wild relative or subsequent crop if the volunteer population is not detected and eliminated.

3.3.4.2 Factors Governing Gene Flow among Crop Plants and Wild Relative Species

GE crops, as well as non-GE, will vary in their propensity to outcross depending on several factors, including whether they are self-pollinated or cross-pollinated, the presence or absence of sexually compatible wild relatives, and the physical and spatial barriers separating GE crops from sexually compatible wild relatives.

The salient environmental concern is whether the flow of a GE trait gene (i.e., in the case of DP202216 corn, increased yield and glufosinate herbicide resistance) to a wild relative will have adverse ecological consequences. For a significant environmental impact to occur, gene flow would have to lead to the production of a fertile hybrid plant that produces viable offspring, and the resulting GE-wild plant hybrid having some type of competitive advantage that can lead, ultimately, to introgression of the GE trait gene into a wild plant population (Ellstrand et al. 2007; Ellstrand 2014; Goldstein 2014). The ecological consequences of a GE trait gene in a wild species depends on the type of trait, the stability of the gene in

hybrids to repeatedly reproduce in the wild. In order for introgression to occur first generation hybrids have to backcross with wild parental plants and produce fertile progeny.

¹³ An exception would be with plants bred for sterility. For example, some varieties of crop plants have been bred for male sterility and used in seed production.

the genome, whether fitness is conferred to the hybrid through expression of the trait gene, and ecological factors in the area of the hybrid (Felber et al. 2007; Ellstrand 2014).

It is generally assumed traits that impart increased fitness will persist in populations and those that impart negative effects on plant fitness will not. If a resulting GE-wild type hybrid had a competitive advantage over wild populations, it could persist in the environment and potentially disrupt the local ecology. In this case, there will be significant concerns regarding the potential invasiveness or weediness of GE-wild type hybrids, and the potential for hybrids to modify ecosystem dynamics. Where the transgenic trait does not provide fitness, and is not deleterious to survival of the hybrid, the transgene may still persist in wild populations with no effects on the local ecology. This could be the case for a number of introduced traits.

Whether the otherwise benign hybridization with sexually compatible wild relatives would itself constitute an environmental harm (e.g., the transgene did not enhance fitness), it would require evaluation on a case-by-case basis relative to the species' involved, and the ecosystem where the hybrid(s) occurred (e.g., the presence of sensitive species or habitats).

In respect to the occurrence of a GE-wild type hybrid, gene flow from a GE crop plant to wild or weedy relative species does not necessarily constitute an environmental harm in and of itself, nor does it inherently imply environmental damage (Ellstrand 2014). The salient issue is what the resultant ecological consequences of such gene flow to a wild population may be (Ellstrand 2014). Current understanding suggests that the presence of a GE trait outside the area of cultivation will likely have little or no adverse consequences unless:

- (1) the GE trait confers novel or enhanced fitness or weediness to the GE-wild relative hybrid, resulting in the evolution of increased weediness or invasiveness in wild type hybrids, or
- (2) the GE trait confers to GE-wild relative hybrid progeny reduced fitness, resulting in a selective disadvantage in wild relative populations (Kwit et al. 2011; Ellstrand 2014).

Hence, in evaluating potential environmental impacts it is not the risk of gene flow itself that is the chief concern, but rather the environmental consequences that could occur as the result of such an event; whether the transgene will persist in a wild population, and whether hybrid or introgressed populations will have adverse effects on ecosystem dynamics.

3.3.4.3 Gene Flow among Corn (*Zea mays* L.) and Wild Relative Species

The genus *Zea* is composed of annual and perineal grasses native to Mexico and Central America (Sánchez González et al. 2018a). The genus *Zea* includes both wild taxa known by the common name “teosinte” and cultivated varieties. Corn (*Zea mays* L. ssp. *mays*) is one of the oldest domesticated plants in the world, the origins of which date back to around 5,000 – 3,600 years ago in southern Mexico (de Wet et al. 1978; Eubanks 1995). There are two classifications for teosinte (see review by (Sánchez González et al. 2018a)).

Wilkes (1967) identified geographic populations of *Zea* associated with different environments and described four races of teosinte for Mexico (Nobogame, Central Plateau, Chalco and Balsas) and two for Guatemala (Guatemala and Huehuetenango). More recently, Iltis and Doebley (1980), Doebley and Iltis

(1980) and Doebley (1990) proposed a hierarchical system of classification for *Zea* based on the morphological, ecological and molecular features of the taxa. They divided *Zea* into two sections.

Section *Luxuriantes* includes *Zea perennis* (Hitch.) Reeves & Mangelsdorf, *Zea diploperennis* Iltis, Doebley & Guzmán, and *Zea luxurians* (Durieu & Ascherson) Bird. The newly described *Zea vespertilio* (Gómez-Laurito 2013) and *Zea nicaraguensis* (Iltis and Benz 2000) are also considered members of this section.

Section *Zea* includes *Zea mays* L., which was divided into *Zea mays* ssp. *mexicana* (Schrader) Iltis for races Chalco, Central Plateau and Nobogame, *Zea mays* ssp. *parviglumis* Iltis & Doebley that includes race Balsas, *Zea mays* ssp. *huehuetenangensis* (Iltis & Doebley) Doebley for race Huehuetenango, and *Zea mays* L. ssp. *mays* for cultivated maize.

Recently, Sánchez G et al. (2011), using evidence from multiple independent sources, reported three new taxa from Mexico within section *Luxuriantes* from the Mexican states Nayarit, Michoacán and Oaxaca.

How corn (*Zea mays* L. subsp. *mays*) evolved is still a matter of investigation, although most investigators agree that corn most likely descended from an annual species of teosinte (*Zea mays* ssp. *parviglumis*), a closely related wild grass endemic to Mexico (Piperno and Flannery 2001). Cultivated corn (*Zea mays* L. subsp. *mays*), is sexually compatible with other members of the genus *Zea* (Teosinte), with a few exceptions, and to a much lesser degree with members of the genus *Tripsacum*.

Teosinte

Teosinte is the common name applied to several distinct wild *Zea* species closely related to corn (*Zea mays* L. ssp. *mays*). Wild teosinte relatives of corn comprise a group of annual and perennial species that occur within the tropical and subtropical areas of Mexico, Guatemala, Costa Rica, Honduras, El Salvador, and Nicaragua (Sánchez González et al. 2018b). The natural geographic distribution of teosinte extends from the Western Sierra Madre of the State of Chihuahua, Mexico to the Pacific coast of Nicaragua and Costa Rica, including the western part of Mesoamerica. The Mexican annuals *Zea mays* ssp. *parviglumis* and *Zea mays* ssp. *mexicana* show a wide distribution in Mexico, while *Zea diploperennis*, *Zea luxurians*, *Zea perennis*, *Zea mays* ssp. *huehuetenangensis*, *Zea vespertilio* and *Zea nicaraguensis* have more restricted and distinct ranges, representing less than 20% of the total occurrences from published sources for the period 1842-2016 (Sánchez González et al. 2018b).

Except for *Z. perennis*, *Zea mays* and teosinte cross readily, and their hybrids are fully fertile (de Wet and Harlan 1972). Hybridization and introgression between *Z. mays* and the subspecies *Z. mays* subsp. *mexicana* occurs in Mexico, and has probably been taking place since the advent of corn domestication wherever these two taxa are sympatric (de Wet et al. 1978; Ellstrand et al. 2007). Hybrids appear to maintain their unity of type in the wild (de Wet and Harlan 1972). In general, humans select in the direction of corn (*Zea mays*), and nature strongly favors teosinte over their hybrid, which is less well adapted for natural seed dispersal (de Wet and Harlan 1972). The rate at which domesticated corn crop genes may enter teosinte populations will be limited by genetic barriers, phenological differences, and the relative fitness of the hybrids (Ellstrand et al. 2007).

Teosinte do not appear to be present in the United States other than in botanical gardens or at research stations. The USDA Plants Database lists *Zea mexicana* (Syn. *Z. mays* ssp. *mexicana*) as present in Florida, Alabama, and Maryland, having been introduced from Mexico (USDA-NRCS 2019d). It has, apparently, occasionally been cultivated in the Southern United States for forage (Hitchcock 1951). The documentation cited for occurrence in Florida only shows distribution of native or naturalized populations in Miami-Dade, Orange, and Levy Counties (Wunderlin et al. 2019). While citations were provided in the Plants database for distribution in Maryland and Alabama, current Maryland plants databases have no listed *Zea* species, other than *Z. mays* (UMD 2005; MPAWG 2016), nor are any *Zea* species or subspecies other than *Z. mays* (corn) listed in Alabama (Kral et al. 2019).

Zea perennis (Syn. *Euchlaena perennis* Hitchc.) is listed as occurring in Texas and South Carolina. It is described as having been cultivated at academic research stations in Angeton, Texas and Sacaton, Arizona, and established on James Island, South Carolina (Hitchcock 1951). It is not known if the James Island population has persisted. There are no *Zea* species found in the comprehensive online South Carolina Plant Atlas (USC 2019); which catalogues over 3000 species.

Teosinte identified as *Zea mays* ssp. *parviglumis* is listed as having occurred in Miami-Dade County, Florida (Wunderlin et al. 2019), an area that is now largely urban. *Zea diploperennis* and *Zea luxurians* are also listed in the USDA Plants database, but there is no information about the presence of any wild populations in the United States.

Experts familiar with the teosinte collections in the United States, some of whom were involved with revision of the Manual of Grasses for North America (Roché et al. 2007), are not aware of any naturalized or native populations of teosinte currently growing in the United States (USDA-APHIS 2013).

Tripsacum

The closest relative of *Zea* in the United States is the genus *Tripsacum* (OECD 2003). Three species have been identified in the United States: *T. dactyloides*, Eastern gamagrass, is known to occur in the eastern half of the United States, *T. lanceolatum*, Mexican gamagrass, occurs in the southwest of the United States, and *T. floridanum*, Florida gamagrass, is native to South Florida and Cuba (OECD 2003; USDA-NRCS 2019d). *T. dactyloides* is the only *Tripsacum* species of widespread occurrence and agricultural importance in the United States, and has been commonly grown as a forage grass (USDA-NRCS 1996). *T. fasciculatum* and *T. latifolium* occur in Puerto Rico (USDA-NRCS 2019d). *Tripsacum* species (2n=18) can be represented by diploid, triploid, tetraploid, and higher ploidy levels.

Although not closely related cytologically (e.g., differing numbers of chromosomes), gene exchange can take place between *Z. mays* and *Tripsacum* (de Wet et al. 1978). Certain species of *Tripsacum* can and have been crossed with *Zea mays* or at least some accessions of each species can cross under experimental lab conditions, but only with difficulty. The resulting hybrids are male sterile and usually female sterile (de Wet et al. 1978; Leblanc et al. 1996; Lee et al. 2017; Iqbal et al. 2019). Hybrids between *T. dactyloides* and *Z. mays*, specifically, have been found to be male sterile, but female fertile (de Wet and Harlan 1972). Attempts at artificially induced introgression from *Tripsacum* species into *Z. mays* failed to produce either teosinte-like offspring or the combination of characteristics assumed to indicate introgression during the evolution of several South American races of corn (Mangelsdorf and Reeves

1959; de Wet and Harlan 1972). The probability of natural introgression from *Tripsacum* in the direction of *Z. mays* seems to be low (de Wet et al. 1978).

Hybrid combinations with *Z. mays* (as pollen donor) and *T. dactyloides* are known to give rise to recovered *Z. mays* within three or more further backcrosses with *Z. mays*. It is, however, not too likely that this process commonly occurs in nature (de Wet et al. 1978). With each successive backcross, the offspring become more *Z. mays* like, and less capable of surviving in competition without the help of humans. Hybrids have been observed to not only produce low yields, but are also partially female sterile (de Wet et al. 1978).

In summary, gene exchange is possible between *Zea* and *Tripsacum*, and several South American races of corn, where teosinte is absent, exhibit past evidence of hybridization (de Wet et al. 1978). Natural introgression between *Zea* and *Tripsacum*, however, appears unlikely (de Wet et al. 1978). Hybrids between *Z. mays* and *Tripsacum*, as well as their derivatives when backcrossed with *Z. mays*, are poorly adapted for survival in competition with both their wild and cultivated parents (de Wet et al. 1978). Although hybridization of *Tripsacum* and *Z. mays* has been accomplished in the laboratory using special techniques under highly controlled conditions (Wozniak 2002; Lee et al. 2017), pollen-directed gene flow from corn (*Zea mays*) to wild *Tripsacum* species is considered an unlikely event (Wozniak 2002; Lee et al. 2017). APHIS is unaware of any reported cases of hybridization among naturally occurring *Tripsacum* and *Z. mays* in the United States.

3.3.4.4 Corn as a Weed or Volunteer

In the United States, there are no *Zea* species listed as on the Federal Noxious Weed List (7 CFR part 360) (USDA-NRCS 2019e). Corn (*Zea mays*), as a highly domesticated crop plant with limited seed dispersal and dormancy, does not readily form persistent feral populations; does not present as a weed outside of areas of cultivation (USDA-APHIS 2019a).

Corn can and periodically does occur as a volunteer plant in subsequent crops planted in the same field. Corn seed can remain in fields as a result of harvester inefficiency, dispersal by birds and other foraging wildlife, or from fallen ears. When seeds survive to the next growing season, volunteer plants may develop within subsequent crops rotated with corn, such as soybean, dry beans, sugar beets, as well as subsequent corn crops. Volunteer corn is more of agronomic/economic than environmental concern; the presence of volunteers can result in minor to significant yield impacts on subsequent crops planted in the same field, interfere with harvest, and cause unacceptable levels of contamination in harvested soybean (Nicolai et al. 2018), depending on the density of the volunteer corn (Nicolai et al. 2018; Jhala et al. 2019).

In controlled agronomic studies, volunteer corn densities ranging from 800 to 13,000 plants per acre resulted in yield losses of 0 to 54% in soybean and 0 to 13% in corn (Nicolai et al. 2018). In addition, clumps of volunteer corn plants emerging from dropped ears are more competitive than individual plants. Similarly, soybean yield reductions have been found to range from 10% to 41% where early-emerging volunteer corn densities ranged from 0.5 to 16 plants m², although no soybean yield loss occurred with a late-emerging cohort of volunteer corn (Marquardt et al. 2012). Thus, the potential impact of volunteer corn on the yield of subsequent crops can be substantial. Volunteer corn can also encourage dispersal and survival of western corn rootworm and gray leaf spot disease limiting the benefits of a corn-soybean

rotation (Jhala and Rees 2018). Successful control of volunteer corn is accomplished with the use of various combinations of cultivation practices and use of herbicides with differing modes of action (Jeschke and Doerge 2010; Nicolai et al. 2018).

3.3.5 Biodiversity

Biological diversity relative to agricultural ecosystems (agro-ecosystems) encompasses the variety and variability of animals, plants, and microorganisms, at the genetic, species, and community levels that are necessary to sustain key functions of the agro-ecosystem (CBD 2019b). Various species contribute to essential ecological functions upon which agriculture depends, such as sustaining soil fertility and plant pest control. One invaluable function of biodiversity is the support of diverse populations of insects on farms. In one study of corn farms across the Northern Great Plains, Lundgren and Fergen (2014) found that farms with lower insect biodiversity had more plant pests, and that more bio-diverse cornfields had fewer plant pests. The results from their study also suggest that designing cropping systems with high diversity requires fewer insecticide inputs and can save farmers money. Thus, farming practices that promote insect biodiversity can facilitate control of plant pests.

Relative to GE crops, specifically, by facilitating conservation tillage (HR crops), decreasing insecticide use (IR crops), use of more environmentally benign herbicides, and increasing yield—which alleviates pressure to convert additional land into agricultural use—some GE crops can contribute to reducing the impacts of agriculture on biodiversity (Carpenter 2011). A U.S. National Research Council assessment of the relationship between GE crop adoption and farm sustainability in the United States concluded that, generally, GE crops have had fewer adverse effects on the environment than non-GE crops produced conventionally (NRC 2010).

While biodiversity will be inherently limited in commercial corn crops due to frequent disturbance, tillage, mechanized planting, planting of a monoculture crop, and application of fertilizers and pesticides, growers, as well as federal and state programs, well recognize the need for maintenance of some degree of cropland biodiversity. A variety of federally supported programs, such as the USDA funded Sustainable Agriculture Research and Education Program (SARE), and partnership programs among the EPA and the agricultural community support sustainable agricultural practices that are intended to protect the environment, conserve natural resources, and promote cropland biodiversity (i.e., (USDA-NIFA 2017; US-EPA 2019d)). The USDA Natural Resources Conservation Service, through its Conservation Stewardship Program, Landscape Initiatives, Environmental Quality Incentives Program, Landscape Planning, and other services provides technical and financial support to growers to assist in managing the complex interaction of cropping systems and habitat (USDA-NRCS 2019b).

3.4 Human Health

Human health considerations associated with GE crops are those related to (1) the safety and nutritional value of GE crops and their products for consumers, and (2) the potential health effects of pesticides that may be used in association with GE crops. As for food safety, consumer health concerns are in regard to the potential toxicity or allergenicity of the introduced genes/proteins, the potential for altered levels of existing allergens in modified plants, or the expression of new antigenic proteins. Consumers may also be

concerned about the potential consumption of pesticides on/in foods derived from GE crops. Occupational exposure to pesticides is also considered.

The safety assessment of GE crop plants, summarized following, includes characterization of the physicochemical and functional properties of the introduced gene(s) and gene products, determination of the safety of the gene products (e.g., proteins, enzymes), and evaluation of the potential health effects of food derived from the GE crop plant. The introduced *pat* gene and gene product, and *zmm28* gene from corn (*Zea mays*), which encodes for a transcription factor that can potentially increase yield, are reviewed below.

3.4.1 Food Safety

In addition to direct consumption (e.g. grits, corn on the cob), humans consume corn products such as corn starch, corn meal, corn flour, corn oil, and corn syrup. Various food items are comprised of corn products, which include cereals, bakery mixes, muffins, tortillas, salad dressings, snack foods, and processed meats. Dent corn (DP202216 corn variety) is only used for manufacture of processed corn products, such corn oil and corn syrup.

As summarized in Section 1.3—Coordinated Framework for the Regulation of Biotechnology, the FDA regulates the safety of plant-derived foods pursuant to the FFDCA and FSMA. The FDA created a voluntary plant biotechnology consultation process in the 1990's, which developers can use to ensure the safety of food derived from GE plants before they enter the market (US-FDA 1992b, 2006). In such a consultation, a developer who intends to commercialize food or feed derived from a GE plant meets with the FDA to identify and discuss relevant safety, nutritional, or other regulatory issues regarding the food product(s). The FDA evaluates the food safety data presented by developers and responds to the developer by letter with any concerns it may have or additional information it may require. Pioneer is consulting with the FDA in regard to the safety of food and feed products derived from DP202216 corn (Pioneer 2019c).

In addition to the FDA consultation, foods derived from GE plants undergo a safety evaluation among international agencies before entering foreign markets, such as reviews by the European Food Safety Agency and the Australia and New Zealand Food Standards Agency. The Codex Alimentarius, established by the World Health Organization and Food and Agriculture Organization of the United Nations, is a set of international standards, principles, and guidelines for the safety assessment of foods derived from modern biotechnology. These standards help countries coordinate and harmonize review and regulation of foods derived from GE plants to ensure public safety and facilitate international trade (WHO-FAO 2009). Currently, the Codex Alimentarius Commission is comprised of 188 member countries, to include the United States. Most governments incorporate Codex principles and guidelines in their review of foods derived from GE crop plants.

There are multiple ways in which crop plants can be and have been genetically modified; through traditional cross breeding, somatic hybridization, chemical or radiation-induced mutagenesis, microprojectile bombardment, and agrobacterium mediated genetic engineering (NRC 2004). Modification to produce desired traits in plants used for food began about 10,000 years ago. Development of new plant varieties, along with natural evolutionary changes, have resulted in common species of food

plants that are now genetically different from their ancestors (NRC 2004). Advantages derived from these genetic modifications include increased food production, reliability, and yields; enhanced taste and nutritional value; and decreased losses due to various biotic and abiotic stresses, such as fungal and bacterial pathogens (NRC 2004). These objectives continue to motivate plant breeders and food scientists to develop/improve methods for the development of new plant varieties.

Food safety reviews for GE crop plants commonly compare the compositional characteristics of the GE crop plant with non-transgenic, conventional varieties of that crop. Compositional analyses include characteristics such as moisture, protein, fat, carbohydrates, ash, minerals, dietary fiber, essential and non-essential amino acids, fatty acids, vitamins, and antinutrients (NAS 2016). The food and feed safety reviews of GE crops introduced into the market to date have generally concluded that there are no significant nutritional differences in conventional versus GE plants for food or animal feed, beyond those intended (NAS 2016; Delaney et al. 2017). The safety of the HR and enhanced yield traits are reviewed below.

3.4.1.1 Safety of PAT and ZMM28 Yield Trait

PAT Safety Evaluations

Phosphinothricin N-acetyltransferase (PAT) has a history of safe use in several commercially available plant products. The FDA has previously consulted on PAT, and the *pat* and *bar* genes encoding for expression of PAT, for over 30 glufosinate resistant crop varieties (US-FDA 2019); these include canola, corn, cotton, soybean, and sugar beet. None of these modified crop varieties was identified as presenting any risk to human or animal health.

By 2011, regulatory authorities in 11 different countries had issued approvals for the environmental release of GE plants expressing the PAT protein, either by itself or in combination with other GE traits. This represents approximately 38 products, and 8 crop varieties (ILSI-CERA 2011). Previous evaluations of PAT have shown that it does not share amino acid sequence similarity to known toxins, nor does it possess characteristics associated with food allergens (Herouet et al. 2005; ILSI-CERA 2011). APHIS has evaluated and deregulated 11 varieties of GE corn comprised of either the *pat* or *bar* genes, as well glufosinate resistant varieties of soybean, cotton, canola, beet (USDA-APHIS 2019b).

ZMM28 Transcription Factor

The *zmm28* gene, which encodes the ZMM28 protein, is endogenous to corn. Based on bioinformatic studies the introduced ZMM28 protein in DP202216 corn is identical to that of the native ZMM28 protein in non-modified corn (Anderson et al. 2019; Pioneer 2019c). The amino acid sequence of the ZMM28 protein in DP202216 corn also shares varying degrees of homology with the amino acid sequence of closely related proteins in various other commonly consumed grain crops, fruits and vegetables (Anderson et al. 2019; Pioneer 2019c).

The total amount of ZMM28 protein expressed in DP202216 corn tissues remains low (parts per billion (ppb) range). The ZMM28 protein is present in sweet corn kernels and in vegetative tissues of conventional corn lines at levels comparable to those found in DP202216 corn kernels and vegetative tissues. Thus, the estimated acute and chronic dietary exposures to the ZMM28 protein from consumption

of DP202216 corn products are comparable to the acute and chronic dietary exposures to the ZMM28 protein from consumption of non-GE sweet corn varieties (Pioneer 2019c).

3.4.2 Pesticides, Tolerance Limits for Foods, and Exemption from the Requirement for a Tolerance

The EPA regulates the sale, distribution, and use of pesticides under FIFRA (Section 1.3—Coordinated Framework). The EPA also regulates certain GE microorganisms used as biofertilizers, bioremediation agents, and for the production of various industrial compounds including biofuels under the TSCA. Before a pesticide may legally be used in the United States, the EPA must evaluate the pesticide to ensure that it will not result in an unreasonable risk to human health or the environment. Pesticides that complete this evaluation are issued a "registration" that permits their sale and use according to requirements set by the EPA.

To ensure the continued safety of pesticides and public health the EPA conducts pesticide registration reviews so that, as the ability to assess risk evolves and as policies and practices change, all registered pesticides continue to meet the statutory standard of no unreasonable adverse effects. As part of this program, the EPA recently conducted a human health risk assessment for glufosinate registration review; specifically, for dietary, residential, occupational, and aggregate risks associated with the use of glufosinate. The EPA concluded that with uniform implementation of label statements clarifying restrictions on residential lawn applications, there are no dietary, residential, or aggregate risks of concern for glufosinate from exposure to residues in food and drinking water, or from residential handler and post-application exposure (US-EPA 2016c).

Before a pesticide can be used on a food crop, the EPA, pursuant to the FFDCA and Food Quality Protection Act of 1996 (FQPA), establishes a tolerance limit, which is the amount of pesticide residue allowed to remain in or on each treated food commodity (21 U.S. Code § 346a - Tolerances and exemptions for pesticide chemical residues). Pesticide tolerance limits established by the EPA are to ensure the safety of foods and feed for human and animal consumption (US-EPA 2019h). If pesticide residues are found above the tolerance limit, the commodity will be subject to seizure by the government.

Section 408(c)(2)(A)(i) of the FFDCA allows the EPA to establish an exemption from the requirement for a tolerance (the legal limit for a pesticide chemical residue in or on a food) if EPA determines that the exemption is "safe." Safe is defined as meaning that there is a "reasonable certainty that no harm will result from aggregate exposure to the pesticide residue." To make a safety finding, the EPA considers, among other things: the toxicity of the pesticide and its break-down products, aggregate exposure to the pesticide in foods and from other sources of exposure, and any special risks posed to infants and children. Some pesticides are exempted from the requirement to have a tolerance. The EPA may grant exemptions in cases where the pesticide residues do not pose a dietary risk under reasonably foreseeable circumstances.

Both the FDA and USDA monitor foods for pesticide residues to enforce these tolerance limits, and ensure protection of human health. By example, the USDA Pesticide Data Program (PDP) collects data on pesticides residues on agricultural commodities in the U.S. food supply, with an emphasis on those commodities highly consumed by infants and children (USDA-AMS 2019a). The program is

implemented through cooperation with state agriculture departments and other federal agencies. The EPA uses PDP data to prepare pesticide dietary exposure assessments pursuant to the FQPA. PDP data enable the EPA to assess dietary exposure; facilitate the global marketing of U.S. agricultural products; and provide guidance for the FDA and other governmental agencies to make informed decisions.

The EPA has established tolerance limits for glufosinate at 40 CFR §180.473. Pesticide tolerance levels for glufosinate have been established for a wide variety of commodities, including field corn for grain and forage, as described in 40 CFR §180.473 (US-EPA 2019a). PAT has been considered a plant-incorporated protectant (PIP) inert ingredient when included with genetic constructs that encoded pesticidal substances intended as PIP active ingredients. In such cases the EPA has issued permanent exemptions from food and feed tolerance limits for the PAT protein in all food commodities in the United States recognizing the negligible risk it poses to human health (US-EPA 2007).

The EPA also sets limits for potential drinking water contaminants that need to be regulated in order to protect public health (40 CFR part 141). These contaminant limits are required by the Safe Drinking Water Act (SDWA). The EPA works with States, Tribes, and many other partners to implement SDWA standards.

3.4.3 Worker Safety

Agriculture is considered one of the most hazardous industries in the United States. Worker hazards common to all types of agricultural production include those associated with the operation of farm machinery, vehicles, and pesticide application. Agricultural operations are covered by several Occupational Safety and Health standards including Agriculture (29 CFR 1928), General Industry (29 CFR 1910), and the General Duty Clause. Further protections are provided through the National Institute of Occupational Safety and Health (NIOSH), which in 1990 began development of an extensive agricultural safety and health program to address the high risks of injuries and illnesses experienced by workers and families in agriculture.

In consideration of the risk of pesticide exposure to field workers, the EPA's Worker Protection Standard (WPS) (40 CFR Part 170) was issued in 1992 to require actions to reduce the risk of pesticide poisonings and injuries among agricultural workers and pesticide handlers. The WPS contains requirements for pesticide safety training, notification of pesticide applications, use of personal protective equipment, restricted entry intervals following pesticide application, decontamination supplies, and emergency medical assistance. The Occupational Safety and Health Administration (OSHA) also requires employers to protect their employees from hazards associated with pesticides and herbicides.

On November 2, 2015, EPA revised the WPS to implement more protections for agricultural workers, handlers, and their families (80 FR 211, November 2, 2015, p. 67495). The WPS revisions are intended to decrease pesticide exposure incidents among farmworkers and their family members. Fewer incidents means a healthier workforce and avoiding lost wages, medical bills, and absences from work and school. Most of the revised WPS requirements became effective on January 2, 2017 (US-EPA 2016b). Farmworkers are required to use pesticides consistent with the instructions provided on the EPA-approved pesticide labels, which may include instruction on personal protective equipment, specific handling requirements, pesticide equipment application specifications, and field reentry procedures.

3.5 Animal Health and Welfare

Dent corn accounts for around 95% of feed grain production in the United States, a primary feed source for beef cattle, poultry, hogs, and dairy cattle. Animal feed derived from corn comes not only from the unprocessed grain, but also from silage (the above-ground portions of the corn plant), stalk residues in fields that might be grazed, and residuals derived from corn refining and milling, such as corn gluten feed, corn gluten meal, corn germ meal, corn steep liquor, and amino acids.

The FDA's Center for Veterinary Medicine (CVM) is responsible for the regulation of animal food (feed) products under the FFDCA and FSMA—as discussed for human health. Pioneer submitted a safety and nutritional assessment for DP202216 corn to the FDA's Center for Food Safety and Applied Nutrition (FDA CFSAN) in 2018 (Pioneer 2019c).

3.6 Socioeconomics

3.6.1 Domestic and International Markets

U.S. Corn Commodities

The production of corn and the various commodities utilized by the food, feed, fuel and industrial sectors is major component of the U.S. economy. The United States is the largest producer and exporter of corn in the world, with around 85 to 95 million acres of corn planted on an annual basis over the last ten years (USDA-NASS 2019b). Corn is primarily used for feed grain and fuel ethanol, which account for approximately 40% and 35% of corn use, respectively (Figure 3-12). The remainder of harvested corn is processed for food and industrial products such as sweeteners, corn oil, and beverage and industrial alcohol. In 2017, the market value of U.S. corn crops was \$51.2 billion (USDA-NASS 2019a).

During processing, corn is either wet or dry milled depending on the desired end products:

- Wet millers process corn into high fructose corn syrup (HFCS), glucose and dextrose, starch, corn oil, beverage alcohol, industrial alcohol, and fuel ethanol.
- Dry millers process corn into flakes for cereal, corn flour, corn grits, corn meal, and brewers grits for beer production.

Both the dry-milling and wet-milling methods of corn processing generate economically valuable co-products, the most prominent of which are distillers' dried grains with solubles (DDGS), which can be used as a feed ingredient for livestock (USDA-ERS 2019a). In the United States, feed for both dairy and beef has been the primary use of DDGS, but increasingly larger quantities of DDGS are making their way into the feed rations of hogs and poultry (USDA-ERS 2019a).

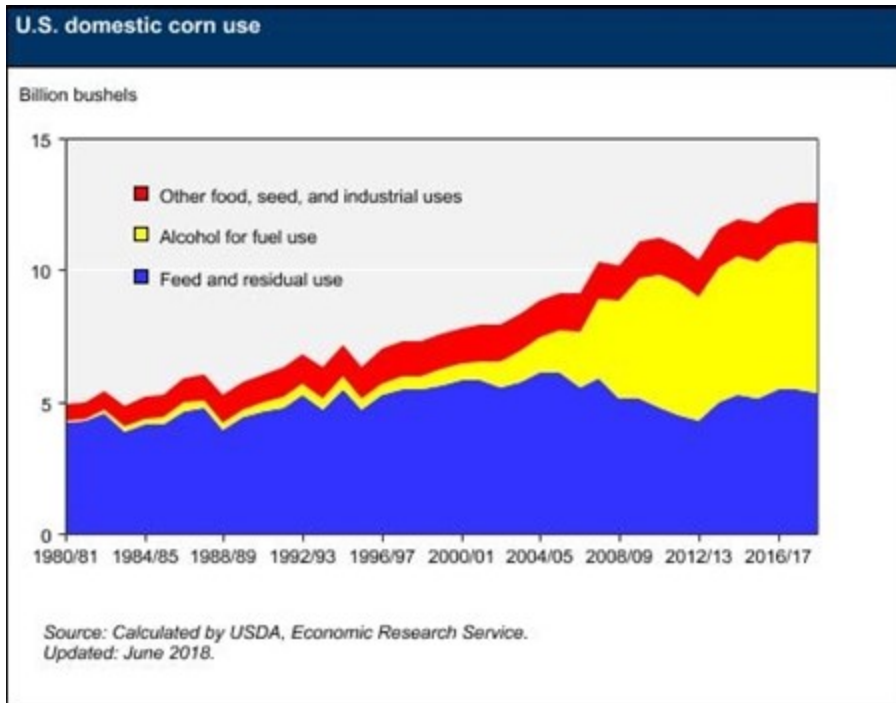


Figure 3-12. Uses of Corn in the United States, 1980 – 2018

Source: (USDA-ERS 2019a)

Corn is the most widely produced feed grain in the United States, accounting for more than 95% of total production and use. The other three major feed grains are sorghum, barley, and oats (Figure 3-13). Feed use, a derived demand, is closely related to the number of animals (cattle, hogs, and poultry) that are fed corn. The amount of corn used for feed also depends on the crop's supply and price, the amount of supplemental ingredients used in feed rations, and the supplies and prices of competing ingredients (USDA-ERS 2019a). Swine and poultry are the major consumers of both feed grains and protein meals, while cattle and dairy cows are the major consumers of roughage; this is largely due to the fact that monogastric species (e.g., hogs) or avian species (e.g., poultry) are not able to easily digest cellulosic material (Schnepf 2011).

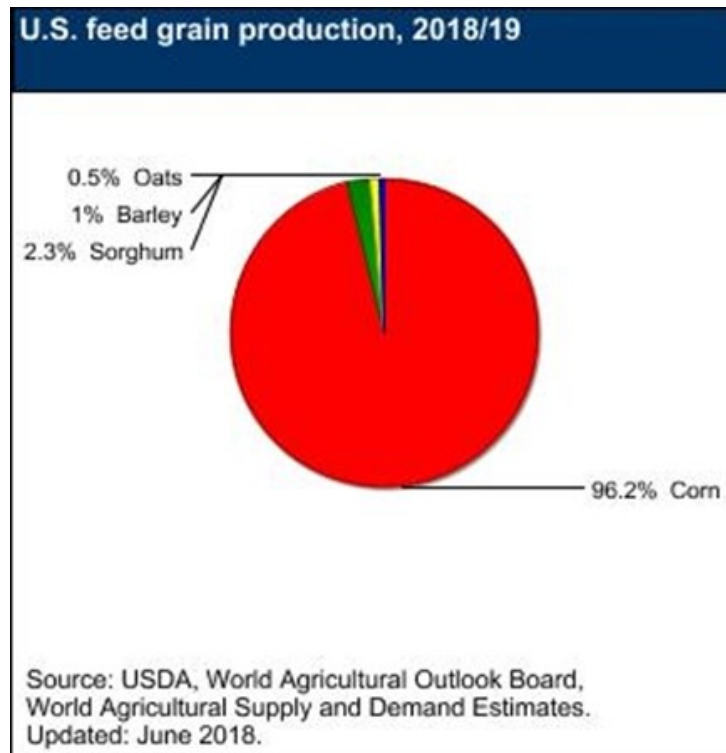


Figure 3-13. Feed Grain Use in the United States, 2018/2019

Source: (USDA-ERS 2019a)

International Trade

The United States is the world's largest corn producer, and provides over a third of the total supply of corn in the world market. Field corn is the largest component of global coarse grain (corn, sorghum, barley, oats, rye, millet, and mixed grains) trade, generally accounting for about two-thirds of the volume over the past decade (USDA-ERS 2019a). Field corn grain exports represent a principal source of demand for U.S. producers and make the largest net contribution to the U.S. agricultural trade balance of all the agricultural commodities, reflective of the importance of corn exports to the U.S. economy. The United States currently exports between 10% and 20% of its annual production (USDA-ERS 2019a).

In the 2018/2019 crop marketing year, (Sept. 1- Aug. 31) the United States grew more than 14.42 billion bushels (366 million metric tons) of corn. Roughly 14.3 percent of production was exported to more than 73 different countries (USGC 2020). In 2018, corn grain and feed exports totaled 63.5 million metric tons, with a value of \$11.2 billion (USDA-ERS 2019d). Only around 240,765 metric tons of sweet corn was exported in 2018 (USDA-ERS 2019f). Argentina, Brazil, and Ukraine are other major exporters, with 33.5, 38.5, and 30 million metric tons projected for 2019/2020 (USDA-FAS 2019).

As the global demand for meat increases, so does the demand for livestock feed, and in turn, corn. Projected increase in U.S. corn exports over the next decade is largely due to a strong global demand for feed grains in support of meat production, particularly in those countries where climate and geography restrict local production of these feed materials (Westcott and Hansen 2015; USDA-ERS 2019e).

Ethanol

The Renewable Fuel Standard (RFS) is a federal program that requires transportation fuel sold in the United States to contain a minimum volume of renewable fuels. It originated with the Energy Policy Act of 2005 and was expanded and extended by the Energy Independence and Security Act of 2007. Congress created the RFS program to reduce greenhouse gas emissions and expand the nation's renewable fuels sector while reducing reliance on imported oil. The EPA implements the program in consultation with U.S. Department of Agriculture and the Department of Energy (US-EPA 2020b).

The RFS requires renewable fuel to be blended into transportation fuel in increasing amounts each year, escalating to 36 billion gallons by 2022. The four renewable fuel categories under the RFS are: biomass-based diesel, cellulosic biofuel, advanced biofuel, and total renewable (conventional) fuel, which typically refers to ethanol derived from corn starch. The RFS began mandating the use of corn ethanol in U.S. fuels in 2006.

U.S. corn processing capabilities allow for production of fuel ethanol and DDGS on a level currently unmatched by any other country. The ethanol industry was comprised of approximately 210 plants in 27 states with a capacity of 16.1 billion gallons as of 2018 (RFA 2019). As a result, the United States dominates trade in these two corn based commodities.

In marketing year 2017/18, over 14 million tons of corn-equivalent ethanol were exported, nearly the same level as bulk corn exports to Japan and Mexico. U.S. corn processed into fuel ethanol and DDGS generates around \$4-5 billion in trade annually, even as the European Union and China – formerly the largest markets, respectively, for these products – imposed prohibitive duties (USDA-FAS 2019).

Identity Preservation

Identity preservation (IP) refers to a system of production, handling, and marketing practices that maintains the integrity and purity of agricultural commodities (Sundstrom et al. 2002). In its simplest form, IP has been employed since the beginning of agriculture when the seeds and grain of different crops were first traded separately. As the seed and food industries developed, the purity and quality expectations of buyers and processors increased and standards were established. Identity preservation typically involves independent, third-party verification of the identification, segregation, and traceability of their product's unique, value-added characteristic (USDA-AMS 2019b). Verification is provided at every stage, including seed, production, processing, and distribution. Buyers are assured that the identity of the product is preserved from the requested stage of production. For example, seed certification programs such as that used by the Association of Official Seed Certifying Agencies (AOSCA) play a major role in maintaining seed purity standards at levels established by the industry for national and international trade (Sundstrom et al. 2002). Similarly, commodity traders, marketing organizations, and food processors have established purity and quality standards for specific end-product uses. Farmers who grow specialty corn in the same general area need to communicate and plan with their neighbors growing different specialty corn to ensure these crops (commodity) identities are preserved and premiums can be realized.

Identity protection is important in international trade. The low level presence (LLP) or adventitious presence (AP) of GE trait material in internationally traded conventional, organic, or other GE crop

commodities are important considerations in the trade of corn. LLP refers to the unintended presence, at low levels, of GE crop material that is authorized for commercial use or sale in one or more countries, but not yet authorized in an importing country. AP refers to instances when trace amounts of GE crop material that has not been approved for commercial use by any country is found in the commercial crop or food supply.

Asynchronous approvals and zero tolerance policies can result in the diversion of trade by some exporters (Van Eenennaam and Young 2014), and rejection or market withdrawals by importers of corn (e.g., (FOEU 2014; Frisvold 2015)). Consequently, incidents of LLP or AP can lead to income loss for exporters and consequently for producers, and consumers in importing countries can potentially face higher domestic prices when an import is deterred or directed to another trading partner (Atici 2014).

The challenges associated with maintaining variety identity in international trade can increase costs, as well as the premiums paid, for some GE crops. GE corn is excluded by some countries sensitive to the importation of food or feed derived from GE plants, and other countries may lag approval of new GE corn varieties. In general, LLP or compromise of corn commodity identity can cause disruptions in international trade when GE trait material is inadvertently incorporated into food or feed shipments. As such, GE crop producing countries are required to take those measures necessary in the production, harvesting, transportation, storage, and post-harvest processing of GE crops to avoid the potential for LLP in conventional or organic commodities.

Pioneer is a member of Excellence Through Stewardship (ETS)®, a global non-profit organization that promotes the universal adoption of product stewardship programs and quality management systems for the full life cycle of agricultural technology products. Pioneer products are commercialized in accordance with ETS Product Launch Policy Stewardship Guidance and in compliance with the Pioneer policies regarding stewardship of those products. Part of these stewardship practices involve systems and processes for maintaining plant product integrity, including product identity and traceability (Pioneer 2019c).

4 ENVIRONMENTAL CONSEQUENCES

This chapter provides an evaluation of the potential environmental impacts that could derive from the alternatives considered in this EA; denying the petition, or issuing a determination of nonregulated status for DP202216 corn. Pursuant to CEQ regulations APHIS considers the direct, indirect, and cumulative impacts of both alternatives. Potential direct and indirect impacts are discussed here, and potential cumulative impacts in Chapter 5.

4.1 Scope of Analysis

An impact would be any change, beneficial or adverse, from existing (baseline) conditions described for the affected environment in Chapter 3. A direct impact is one that derives immediately from an Agency decision on the petition, without intermediate steps or processes. A direct impact would be, on approval of the petition, the availability of DP202216 corn to commercial markets, subject to any EPA requirements, FDA consultation, and/or state requirements. Indirect impacts are those related to, but removed from the Agency's decision in space and time. Examples would include emissions of air pollutants from farm equipment used in DP202216 corn production, and potential impacts on water quality resulting from agricultural run-off comprised of soil sediment, pesticides, and fertilizers.

Where possible, APHIS used data that supported a quantitative analysis of the potential impacts that may derive from selecting either the No Action Alternative or the Preferred Alternative. When data were not available or were insufficient to support a quantitative assessment, APHIS provides qualitative assessments of the impacts of an Agency regulatory decision for DP202216 corn. APHIS focused its environmental analyses to the geographic areas that currently support U.S. corn production.

DP202216 corn: Assumptions Used in Analysis

It is assumed that, in the event Pioneer's petition is approved, that DP202216 corn would be produced commercially for food, feed, and fuel purposes. It is also assumed that the only potential impacts that could derive from production of DP202216 corn that could be considered unique, as compared to other GE and non-GE corn varieties, are relative to the trait genes and gene products and their corresponding impact on the phenotype of DP202216 corn. These aspects of DP202216 corn are summarized below.

DP202216 corn Phenotypes

DP202216 corn hybrids have been demonstrated to have increased yield potential compared to other commercially relevant control hybrids from multiyear yield field trials in numerous locations managed to optimize yield (Pioneer 2019c; Wu et al. 2019). It was reported that the extended and increased expression of the ZMM28 transcription factor as a result of the introduced *ZMGos2-zmm28* gene results in corn plants with increased plant growth, photosynthesis capacity, and nitrogen utilization that are associated with this significant increase in grain yield. It is proposed that the ZMM28 protein transcription factor forms proteins complexes that interact with target genes to regulate their expression resulting in downstream impacts on biochemical and physiological processes including nitrogen assimilation, photosynthesis and growth-regulating plant hormone reception leading to enhanced plant growth and increased grain yield. Increased yield potential and potential increase in nitrogen uptake and utilization could potentially influence corn acreage and agricultural inputs such as nitrogen fertilizer.

DP202216 corn is also resistant to glufosinate-ammonium herbicide through introduction a modified maize optimized version of the *pat* gene and expression of the PAT protein. Glufosinate resistance could encourage the use of glufosinate in lieu of or in addition to other herbicides with subsequent indirect impacts on biotic and abiotic resources.

ZMM28 protein

As discussed in 3.4–Human Health, ZMM28 is a MADS-box protein (transcription factor) that naturally occurs in corn (Anderson et al. 2019; Wu et al. 2019). MADS-box genes are key regulators of both reproductive and vegetative plant development (De Bodt et al. 2003), and common in various plant based foods such as rice, wheat, and barley (Callens et al. 2018). ZMM28 shares 69%-96% amino acid sequence identity with some other plant MAD-box transcription factors that are known to control plant development, such as the transition from vegetative to reproductive growth, as well as the determination of floral meristems and floral organs (Wu et al. 2019). There are many genes homologous to *zmm28* in various taxa of plants (Becker and Theissen 2003; De Bodt et al. 2003). The amino acid sequence of the ZMM28 protein in DP202216 corn is identical to that in selected varieties of sweet corn, and shares homology with proteins whose genes are expressed in consumed plant parts of many other food crops, fruits, and vegetables, with sorghum and rice having homologs with 95 and 84 percent similarity, respectively (Anderson et al. 2019). Thus, ZMM28 and structurally/functionally similar proteins are commonly consumed by humans and wildlife; it is unlikely that the native ZMM28 corn protein presents any risk to human health, or other animals that consume DP202216 corn. As part of the FDAs voluntary consultation program, Pioneer submitted a safety and nutritional assessment for DP202216 corn to the FDA’s Center for Food Safety and Applied Nutrition (FDA CFSAN) in 2018 (Pioneer 2019c).

Pat Gene and the Enzyme Phosphinothricin N-Acetyltransferase

Phosphinothricin N-acetyltransferase (PAT) is an enzyme that confers glufosinate resistance in GE plants. PAT acetylates glufosinate, which inhibits its herbicidal activity. PAT, as an introduced trait in crop plants, can be encoded by the *bar* gene derived from the naturally occurring soil bacterium *Streptomyces hygroscopicus*, and by the *pat* gene derived from *S. viridochromogenes* (ILSI-CERA 2011). *Streptomyces* are gram-positive bacteria of the actinomycetal order with more than 600 described species in soils, sediments, and seawater (Labeda et al. 2012; Bontemps et al. 2013). The gene encoding the PAT protein in DP202216 corn was isolated from *Streptomyces viridochromogenes*, which occurs predominantly in (Labeda et al. 2012). Because the PAT enzyme occurs naturally in soils (Liu et al. 2013), it stands to reason that wildlife and humans, globally, have been and are potentially exposed, incidentally, to the *pat* gene and PAT enzyme through environmental sources on a daily basis (e.g., ingestion, inhalation). As reviewed in Chapter 3 (Human Health, Biological Resources), there are no human health or ecological risks associated with exposure to PAT; the enzyme has a long history of safe use in commercially produced GE corn, soybean, and cotton varieties, which have been evaluated by the FDA (US-FDA 2019). Due to the negligible human health risks associated with PAT, the EPA granted an exemption from the requirement of a tolerance for PAT when used as plant-pesticide inert ingredients in all food commodities (US-EPA 2007). The EPA also conducted an ecological risk assessment for PAT and concluded that no unreasonable adverse effects on non-target organisms are expected from exposure to the PAT protein (US-EPA 2005).

Regulatory reviews of PAT by the United States and international agencies encompass 11 countries and 8 varieties of crop plants. Risk assessments associated with these regulatory reviews concluded that the expression of PAT in crop plants does not alter the potential for persistence or spread of GE plants in the environment, does not alter the reproductive biology or potential for gene flow, and does not present risks for adverse effects in other organisms (ILSI-CERA 2011).

Glufosinate

The EPA is currently conducting its required periodic registration review for glufosinate and issued a preliminary ecological risk assessment in 2014 (US-EPA 2014, 2016c). The EPA's initial screening studies indicated that chronic risk quotients (RQs) exceeded levels of concern for mammals and birds at the maximum labeled single application rate, for most crops. The EPA conducted more detailed assessments using average application rates and estimated environmental concentrations (EECs) that may occur from glufosinate use. This refined assessment identified potential chronic risk to mammals from most registered uses of glufosinate; however, chronic risk estimates for birds, reptiles, and terrestrial amphibians fell below the Agency's level of concern (LOC) (US-EPA 2014, 2016c). The toxicity of glufosinate formulations to various taxa are provided in Table 4-1.

| Table 4-1. Taxonomic Groups and Example Test Species for Potential Effects of Glufosinate | | | | |
|--|---|--|--|--|
| Taxonomic Group | Example(s) of Surrogate Species | Glufosinate: Technical Grade a.i. | Tested Glufosinate Herbicide Formulations | Degradates |
| Birds | Mallard duck (<i>Anas platyrhynchos</i>), Bobwhite quail (<i>Colinus virginianus</i>) | Practically nontoxic | No data | No data |
| Mammals | Laboratory rat (<i>Rattus norvegicus</i>) | Practically nontoxic | Moderately toxic | Practically nontoxic |
| Insects | Honey bee (<i>Apis mellifera</i> L.) | Practically nontoxic | Practically nontoxic | No data |
| Freshwater fish ³ | Bluegill sunfish (<i>Lepomis macrochirus</i>), Rainbow trout (<i>Oncorhynchus mykiss</i>) | Practically nontoxic | Slightly to moderately toxic | Practically nontoxic up to limit of solubility |
| Freshwater invertebrates | Waterflea (<i>Daphnia magna</i>) | Practically nontoxic | Slightly to moderately toxic | Practically nontoxic to slightly toxic (acidification) up to limit of solubility |
| Estuarine/marine fish | Sheepshead minnow (<i>Cyprinodon variegatus</i>) | Practically nontoxic | Moderately toxic | No data |
| Estuarine/marine invertebrates | Mysid (<i>Americamysis bahia</i>), Eastern oyster (<i>Crassostrea virginica</i>) | Practically nontoxic to moderately toxic | Moderately to highly toxic | No data |
| Terrestrial plants ⁴ | Monocots – onion (<i>Allium cepa</i>), Dicots – carrot (<i>Daucus carota</i>) | Not classified | Not classified | No data |
| Aquatic plants and algae | Duckweed (<i>Lemna gibba</i>), Freshwater blue-green alga (<i>Anabaena flos-aquae</i>) | Not classified | Not classified | No data |

Source: (US-EPA 2014)

Mammals: Glufosinate is classified as moderately toxic to mammals. The EPA believes that chronic risk to mammals from glufosinate use may be less than what has been estimated at the screening level. On a national basis, the amount of time during the growing season that actual mammalian exposure to glufosinate would lead to risks above the LOC is less than what was modeled. Additionally, the availability of glufosinate treated foliage to mammals as a food item may be limited due to the fast-acting nature of glufosinate on target weeds, and signs of phytotoxicity in plants, such as wilting and necrosis. Consequently, while mammals could eat plants that have been treated with glufosinate, they may not get all of their diet from treated plants, as simulated in the risk assessment (US-EPA 2016c).

Invertebrates (pollinators): Glufosinate is practically nontoxic to honey bees on both an acute contact and oral exposure basis (adults only), but there is uncertainty regarding the toxicity of glufosinate to bee larvae since no data on this age group of honey bees were available at the time of EPA review (US-EPA 2016c).

Aquatic Plants, Fish, and Aquatic Invertebrates: The EPA's preliminary risk assessment identified no risks of concern for aquatic plants, fish, or aquatic invertebrates, except for the use of glufosinate on rice. Since the time of that assessment, the Agency has determined that the use of glufosinate on rice is limited to nonexistent in the United States; the registrant for use on rice has submitted a voluntary request to terminate the use of glufosinate on rice (US-EPA 2016c). Thus, the Agency has concluded at the present time that the use of glufosinate poses no ecological risks of concern for aquatic organisms. Glufosinate degradates are generally less toxic than parent glufosinate, although results of chronic aquatic toxicity tests with glufosinate degradates are variable (US-EPA 2016c).

Terrestrial Plants: Consistent with its herbicidal mode of action, glufosinate adversely affects most monocotyledonous (monocot) and dicotyledonous (dicot) species at all treatment levels (US-EPA 2016c).

Use and Use Rates

Application rates for glufosinate vary considerably among crops. The highest registered application rate for broadcast spray (ground) is 1.5 lbs a.i./acre for control of understory weeds for orchard nuts and fruits (e.g., almonds, apples, and hickory), grapes, grasses grown for seed, and golf course turf (US-EPA 2019e). The maximum rate for cotton applications is 0.79 lbs a.i./acre (also labeled at 0.52 lbs a.i./acre), for rice 0.73 lbs a.i./acre, and for sugar beets 0.55 lbs a.i./acre (US-EPA 2019e). Corn and soybean broadcast applications are a maximum of 0.44 lbs a.i./A. The maximum seasonal application for corn production is 0.80 lbs a.i./acre, and 0.66 lbs a.i./acre for burndown (US-EPA 2019e). There are numerous other applications, including turf and patio applications, labeled for 0.03 lbs a.i./acre. Multiple applications are allowed by most labels, with an annual maximum rate for agricultural crops of up to 4.5 lbs a.i./acre for orchards and vineyards. Any use of glufosinate must comply with EPA label use requirements, modifications to which were implemented in 2016 (US-EPA 2016c).

4.2 No Action Alternative – Deny the Petition

Because APHIS concluded in its PPRA that DP202216 corn is unlikely to pose a plant pest risk (USDA-APHIS 2019a), and that APHIS has not identified any significant impacts on the human environment that would derive from approval of the petition (discussed following in Section 4.3), denial of the petition for nonregulated status would be inconsistent with the Agency's statutory authority under the plant pest

provisions of the PPA, implementing regulations at 7 CFR part 340, and federal policies embodied in the Coordinated Framework. Because it would be unreasonable to implement an alternative absent any jurisdiction to do so, this alternative is not a practicable option.

While implementing the No Action alternative is not feasible, APHIS provides a summary evaluation for denial of the petition – where DP202216 corn would remain a regulated article and require APHIS authorization for importation, interstate movement, or release into the environment.

APHIS' continued regulation of DP202216 corn, which would effectively preclude commercial production of this variety, would have no effect on the acreage and areas used for U.S. corn production, nor the current practices and inputs used for the commercial production of corn. Likewise, denial of the petition would have no effect on the physical environment, biological resources, human or animal health, or domestic or international corn commodities markets.

Any field testing or interstate movement of DP202216 corn would require APHIS authorization, which would be provided via permit, or an acknowledgment of notification pursuant to 7 CFR part 340 and APHIS guidance. For both permits and notifications, USDA-BRS prescribes criteria and conditions that must be met in order to ensure that the regulated article is introduced in such a way that it is not inadvertently released beyond the proposed introduction site, and it or its progeny do not persist in the environment. Applicants submit documents for releases, such as design protocols, that address how the required conditions will be met.

The regulations in § 340.4(a) provide that any person may submit an APHIS permit application for the introduction of a regulated article, for example, field testing of a GE crop plant. When APHIS receives a permit application, the Agency is required to make a decision to either grant or deny the permit after review of the application and any data submitted with the application.

Notification is an administratively-streamlined alternative to the permit. GE plants must meet specified eligibility criteria, and the introduction must meet certain pre-defined performance standards. For notifications, the notifier must meet eligibility requirements specified in 7 CFR part 340.3 (b)(4)(ii) and 7 CFR part 340.3 (6). APHIS reviews notifications to verify that the GE plants meet the eligibility criteria, and evaluates whether the proposed importation, interstate movement, or environmental release can be done in a manner that meets the required performance standards described in the regulation. When APHIS receives a notification application, it is reviewed by APHIS for completeness to verify that the GE organisms proposed for introduction meet the eligibility criteria for a notification and that performance standards can be met. If APHIS completes the review process and finds that all regulatory requirements have been met, the notification is authorized in a process termed “acknowledgement,” and the applicant may proceed with the proposed introduction under the terms of the notification as prescribed in 7 CFR §340.3.

If a regulated article does not meet the eligibility criteria for notification, a more stringent APHIS permit is required (7 CFR § 340.4). For organisms that present a risk of establishment or persistence in the environment (e.g., are related to wild or weedy plants, insects, or microorganisms), a permit is required so that APHIS can specify appropriate conditions for confinement and monitoring. In addition to the information required for notification, permit applicants must describe how developers of GE organisms

will perform field testing, including specific measures to keep the GE organism confined to the authorized field site and measures to ensure that it does not persist after completion of the field test. The permitting provisions found in § 340.4 describe the information required for permit applications, the standard permit conditions, and administrative information. Standard permit conditions are listed in the regulation, and APHIS can supplement these with additional conditions or requirements, as necessary.

Actions taken by APHIS on permit applications and notifications are subject to NEPA. APHIS performs a variety of functions to ensure compliance with NEPA. Issuance of permits and acknowledgement of notifications are typically authorized under a categorical exclusion from the requirement to conduct an EA or EIS,¹⁴ consistent with APHIS' NEPA implementation regulations (7 CFR part 372). This process complies with CEQ and USDA regulations for implementing NEPA.¹⁵

There are no anticipated impacts on the human environment that would derive from denial of the petition. To the extent individuals comply with current APHIS notification and permit requirements, EPA requirements for pesticide use, and ESA requirements, there would be little risk of harm to wildlife or natural resources as a result of APHIS authorized field testing of DP202216 corn. Interstate movement of DP202216 corn would present negligible environmental risks.

4.3 Preferred Alternative – Approve the Petition

4.3.1 Agricultural Production of Corn

4.3.1.1 Acreage and Area of Corn Production

Approval of the petition is expected to have little to no effect on increases or decreases in total U.S. corn acreage; acreage is determined primarily by market demand for corn based food, feed, fuel, and industrial commodities, versus other agricultural commodities, both domestically and internationally, independent of APHIS' regulatory status decision.

DP202216 corn is a field corn variety, similar to 90% of the other field corn varieties currently produced. It will be used to produce common food, feed, and fuel products. DP202216 corn, if adopted by growers, would be expected to replace some percentage of other corn varieties currently cultivated, namely other GE HR field corn varieties, as opposed to augmenting current corn crops. Other GE corn hybrids with glufosinate resistance are already available to growers.

DP202216 corn hybrids exhibited greater yield than wild type commercially relevant control hybrids (i.e. lacking the transgene for modified expression of ZMM28) 79% of the time in multiyear trials over 58 locations tested (Wu et al. 2019). The overall average yield difference ranged from 2.3 to 6.3 bushels/acre depending on the year, and 3.4 bushels/acre across three years combined (Pioneer 2019c). To put this into perspective, from 1971 to 2016, U.S. corn yields have increased at an annual rate of 1.8 bushels/acre/yr, from about 85 bu/ac to 175 bushels/acre/yr, but the yield increases are not tightly correlated with corn acreage harvested in the United States during this period, and are influenced by other factors such as market demand versus other crops (Specht et al. 2017). Local biotic and abiotic stress

¹⁴ See 7 CFR 372.5(c) for more information about the APHIS categorical exclusion process.

¹⁵ CEQ regulations for implementing NEPA at 40 CFR 1500); USDA regulations implementing NEPA at 7 CFR part 1b; and APHIS regulations at 7 CFR part 372.

conditions could also influence corn acreage planted. As noted in Section 3.1.1, U.S. corn acreage planted annually during the past 10 years has been at 85-95 million acres, and is expected to remain steady at around 90 million acres annually through 2025 (Westcott and Hansen 2015).

4.3.1.2 Agronomic Practices and Inputs

Corn growers will select a particular GE HR crop based on weed/HR weed populations present; the potential for high yield; efficacy of the herbicide(s) used with the GE HR crop; costs of pesticide inputs; and ease and flexibility in management of weeds. Information contained within the Pioneer (2019a) petition (Pioneer 2019c) demonstrates that the cultivation practices needed for growing DP202216 corn are similar to practices used to grow conventional corn and no changes in either insect or disease control measures are foreseen.

It was demonstrated that, compared to wild type control plants, nitrogen uptake was significantly greater by 16% in DP202216 corn when analyzed at the V8 growth stage in hydroponically grown plants in growth chambers, and nitrogen assimilation was significantly greater by 10% in leaf and 23% in the root in field pot-grown plants at the R1 growth stage (Wu et al. 2019). In addition, the specific activity of nitrate reductase, a key enzyme catalyzing a rate limiting step in nitrate assimilation—by reducing nitrate to organic forms—was significantly increased in the leaves, but not the roots, of DP202216 corn at the V4 and V11 growth stages (Wu et al. 2019). It was postulated that the enhanced nitrogen utilization in plants such as DP202216 corn with the ZmGos2-zmm28 trait could serve as a starting point to create more environmentally sustainable corn hybrids with increased yield (Wu et al. 2019). However, since the nitrogen uptake and utilization studies were done under artificial conditions, and the multiyear yield field trials were conducted under conditions to optimize yields with no information provided on nitrogen fertilizer inputs at specific sites, and the yield at those sites, there is insufficient information to accurately determine whether nitrogen fertilizer rates per acre would increase or decrease to achieve optimal performance of DP202216 corn hybrids.

As with previously deregulated GE glufosinate resistant corn varieties that have been commercialized, adoption of DP202216 corn by growers would facilitate use of glufosinate on corn acres. As summarized in Section 3.1.2.2.3, glufosinate is but one of around 40 herbicides used in corn production, comprising 0.23% of total herbicide use on corn in 2018 and was used on only around 1% of corn acres (Table 3-3). Of 19 GE HR corn varieties APHIS has previously deregulated, 11 are glufosinate resistant (USDA-APHIS 2019b). There are several glufosinate resistant corn products currently available to growers (e.g, LibertyLink® and Genuity™SmartStax™ corn). DP202216 corn would present growers and breeders another option among currently available GE HR varieties.

At the national level, adoption rates for GE corn have plateaued in recent years at around 90%. However, adoption rates will vary at the State level on an annual basis due to weed and pest pressures, among other factors. As of 2018, approximately 80% of the U.S. corn acres were planted GE stacked-trait seeds comprised of both HR and IR traits (Figure 4-1).

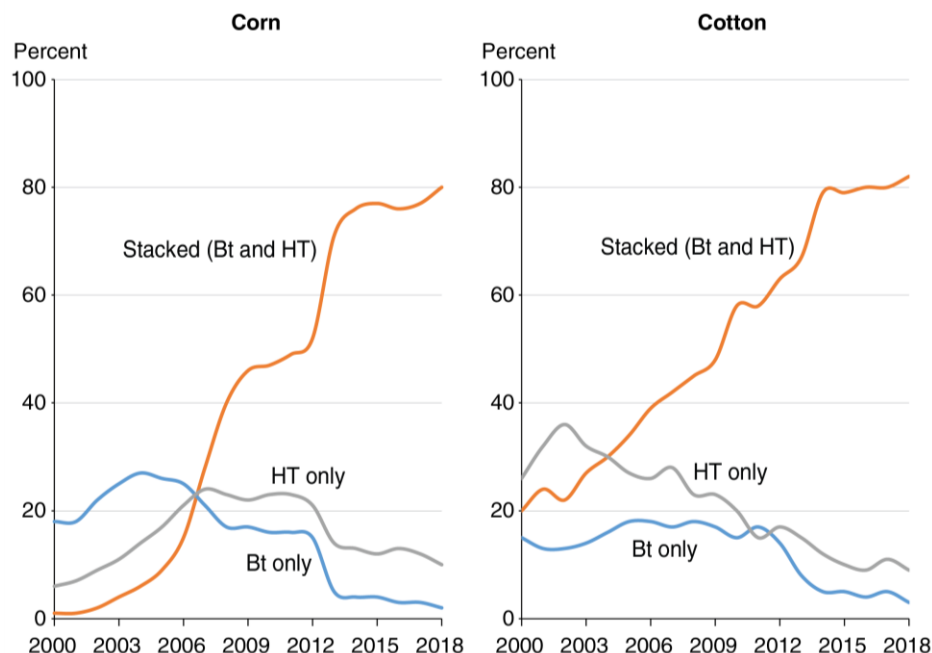


Figure 4-1. Share of Corn and Cotton Acreage Planted with Stacked-Trait Seed – 2018

HT = HR, Bt = IR

Source: (USDA-ERS 2019b)

It is expected that, in the event DP202216 corn is available for use and adopted by growers, that it would result in either (1) growers opting to produce DP202216 corn in lieu of other GE glufosinate resistant varieties, or (2) substitution, use of DP202216 corn in lieu of other GE HR varieties that are resistant to herbicides other than or in addition to glufosinate. In the second case, an increase in glufosinate use and decrease/substitution in use of other herbicides could occur in corn production. No significant increase in total herbicide use on corn would be expected in either case (e.g., Table 3-3). Minor annual fluctuations in total herbicide use are expected due to variances in the use rates in lbs a.i./acre/year among herbicides, herbicide rotation, and annual acreage planted to corn.

HR Weeds

As reviewed in Section 3.1.2, the development of HR weeds continues in many areas of the United States and problematic for growers (Heap 2020). Most corn growing states have from around 7 to 26 different species of weeds that are herbicide resistant (Heap 2020). Given the persistent risk of herbicide-resistance developing in weeds, glufosinate is considered an important tool for grower's in the management of weeds, and weed resistance (US-EPA 2016c). This is generally attributed to its mode of action (MOA; a glutamine synthetase inhibitor), to which there is only one weed species in the United States that has developed resistance; Italian ryegrass (*Lolium perenne* ssp. *multiflorum*), with populations in orchards in California and Oregon (Heap 2020). Glufosinate is also as a broad spectrum post-emergence herbicide.

Certain herbicide MOAs are more susceptible to development of resistance. For example, development of resistance to ALS-inhibitors is most common (161 cases reported worldwide), a trend that is attributed to the relative ease with which plants can evolve resistance. Several single amino acid substitutions that are

sufficient to confer resistance to ALS-inhibitors have been identified in genes that are nuclear inherited. In contrast, development of resistance to the PSII-inhibitor herbicide group is more difficult because in most cases reported it requires a specific point mutation in the *psbA* gene, which is maternally inherited (4 cases reported worldwide) (Holt et al. 2013).

There are only 4 reported cases worldwide of weeds developing resistance to glutamine synthetase inhibition (Heap 2020), and the physiological and genetic mechanisms that underlie resistance are currently poorly understood (Avila-Garcia et al. 2012; Karn et al. 2018). Both glufosinate and glyphosate are two important herbicides used for grass suppression in orchard and vineyard settings (Karn et al. 2018). Populations of an Italian ryegrass collected in a hazelnut orchard in Oregon resistant to glyphosate were also found to exhibit resistance to glufosinate, despite the fact that there was no record of glufosinate use in the orchard where the populations were collected (Avila-Garcia et al. 2012). In this case it was hypothesized that a single non-target site based mechanism may affect translocation of both glyphosate and glufosinate and confer resistance to both herbicides (Avila-Garcia et al. 2012). However, this does not appear to be the case in California orchard populations of Italian ryegrass, which contained individual plants that were resistant to glufosinate, glyphosate, or both, while some individuals were susceptible to both herbicides. It was concluded that multiple resistance mechanisms, rather than cross resistance from a single mechanism, are likely responsible (Karn et al. 2018).

From a practicable standpoint, glufosinate use does present a risk for development of herbicide resistant weed populations, as do all herbicides (Avila-Garcia et al. 2012). The frequency of resistant populations, both with individual or multiple herbicide resistance, is likely to increase with continued herbicide applications if no resistance management strategies are implemented (Karn et al. 2018). Glufosinate-resistant weeds may become more abundant in crops resistant to this herbicide as these crops are more widely adopted; and variability in response of plants to glufosinate under different seasonal or environmental conditions may be problematic, as the increase in survival of plants may not appear uniformly over time, thus making resistance monitoring more difficult (Karn et al. 2018). Total herbicide area-treatments¹⁶ in U.S. corn have increased in corn from 1990 to 2014, and herbicide mode of action diversity also steadily increased until 2005 and then slightly decreased (Kniss 2018). But from about 2010 -2014 the herbicide area-treatments with glyphosate have declined slightly while herbicide area-treatments with non-glyphosate herbicides (as a group) increased during that same period (Kniss 2018). Herbicide use data for corn in 2018 demonstrates that many herbicides with different MOA are used (Table 3-3), with glufosinate use very low; only on 1% of U.S. corn acreage planted, so there has been relatively low selection pressure for glufosinate resistant weeds in corn thus far.

Successful management of development of glufosinate resistant weeds in DP202216 corn would be relative to implementation of EPA resistance management guidance (US-EPA 2017a), Pioneer product stewardship requirements (Pioneer 2019c, b), and recommended non-chemical IWM strategies relevant to corn cropping systems (Mortensen et al. 2012; Norsworthy et al. 2012; Owen 2016; Beckie et al. 2019). In 2017, the EPA issued PR Notice 2017-2, *Guidance for Herbicide-Resistance Management, Labeling,*

¹⁶ One area-treatment is roughly defined as the number of times one herbicide is applied to one field. Area-treatments are used as a way to standardize herbicide application rates in terms of selection pressure for resistant weeds, since simply presenting the weight of each active ingredient would be misleading. To calculate area-treatments, the total amount of each herbicide active ingredient applied per crop per year is divided by the average application rate within each crop for each year, then further divided by the number of planted hectares of that crop in that year.

Education, Training and Stewardship (US-EPA 2017a), which provides registrants and growers information on slowing the development and spread of HR weeds. In addition, the EPA has issued specific recommendations for glufosinate resistance management (US-EPA 2016a). Pioneer implements a product stewardship program that aims to manage the life cycle of products for maximum value and longevity (e.g., DP202216 corn). Pioneer is a member of Excellence Through Stewardship®, a global non-profit organization that promotes the universal adoption of product stewardship programs and quality management systems for the full life cycle of agricultural biotechnology products (Pioneer 2019c). Corteva Agriscience, the parent company of Pioneer, is a member of the global Herbicide Resistance Action Committee (HRAC 2020), an industry-based group administrated by CropLife International (CLI 2020). HRAC supports and equips regional offices to help educate farmers, agronomists, and industry members in the management of on herbicide-resistant weeds. HRAC also identifies and organizes working groups that tackle key herbicide-resistance challenges.

It is expected that the referenced EPA recommendations and label use requirements, and recommended IWM strategies, to include herbicide resistance weed management, would be implemented in production of DP202216 corn. The EPA has recently revised its label language for spray drift management requirements, herbicide resistance management recommendations, and application requirements.

4.3.2 Physical Environment

4.3.2.1 Soil Quality

The agronomic practices and inputs used for DP202216 corn production (see 4.3.1.2) that can impact soil quality would be no different from those currently used, thus, any potential impacts on soil quality resulting from DP202216 corn cultivation would be the same or similar as for other corn varieties. DP202216 corn differs only in the trait genes, gene products, and phenotype, which, as reviewed in the scope of analysis for this Chapter (4.1), are unlikely to affect soil quality.

For all cropping systems, GE and non-GE alike, growers producing crops on highly erodible land would be required to maintain and implement a soil conservation plan that reduces soil loss—a plan approved by the USDA National Resources Conservation Service (NRCS).

4.3.2.2 Water Quality

Cultivation of DP202216 corn is not expected to have any effects on the agronomic practices and inputs used for corn production, other than, where DP202216 corn is grown it could facilitate the use of glufosinate in lieu of other herbicides. The availability of DP202216 corn to the commercial market would have no effect on increases or decreases in total U.S. corn acreage. Consequently, no significant increase in total herbicide use (lbs a.i./year) on corn would be expected. Because the agronomic practices and inputs utilized for DP202216 corn production would be similar to or no different than those currently used (~90% of corn crops are HR), the sources of potential impacts on water resources, namely NPS pollutants in agricultural run-off, would not substantially differ (e.g., sediments, fertilizers, insecticides, herbicides, fungicides). Glufosinate use would be subject to EPA label and other use requirements, which are in part established to be protective of water quality (US-EPA 2019c).

Various National and regional efforts are underway to reduce NPS contaminants in agricultural run-off, and run-off itself (US-EPA 2019g; USDA-NRCS 2019a). For example, in 2012, the USDA Natural Resources Conservation Service (NRCS) launched the National Water Quality Initiative (NWQI), in

collaboration with the EPA and state water quality agencies, to reduce nonpoint sources of nutrients, sediment, and pathogens related to agriculture in high-priority watersheds in each state (USDA-NRCS 2019a).

4.3.2.3 Air Quality

Because the agronomic practices and inputs for DP202216 corn production are the same as/similar to other corn varieties, and there would be no increase in acreage resulting from DP202216 corn production, no changes to emission sources (i.e., tillage, fossil fuel burning equipment, the application of fertilizers and pesticides), or volume of emissions from U.S. corn production, are expected.

4.3.3 Biological Resources

4.3.3.1 Soil Biota

While DP202216 corn differs from non-GE corn varieties in the herbicide resistance and potential increased yield traits, these traits are not expected to have any effects on soil biota or community structures in corn fields. The introduced genes/gene products and regulatory sequences in DP202216 corn are derived from common soil-borne biota or plants. The *zm-gos2* promoter and *zmm28* transcription factor are native to corn (*Zea mays*). The *pinII*, terminator region is from potato (*Solanum tuberosum*). The ubiquitin (*ubiZM1*) sequences used for plasmid construction are derived from corn (*Zea mays*), and homologs are found in most tissues of eukaryotic organisms, (e.g., it occurs ubiquitously). The T-DNA also contains two flippase (Flp) recombinase target sites, FRT1 and FRT87 as well as two loxP and four attB recombination sites. Flippase recombinase (FRT1, FRT87), which catalyzes DNA exchange reactions, was derived from the yeast *Saccharomyces cerevisiae*, which occurs in soils and commonly used in winemaking, baking, and brewing. The flippase, loxP, and attB recombination sites were derived from *Escherichia coli* bacteriophage lambda (Cheo et al. 2004). Glufosinate-ammonium resistance is conferred through introduction of a modified gene (*pat*) from *Streptomyces viridochromogenes*, a naturally occurring soil bacterium. Thus, the genetic elements, and organisms from which they were derived, are prevalent in soils; there are no adverse effects on soil biota that would likely derive from these genetic elements, or gene products.

Use of herbicides on DP202216 corn is not expected to substantially differ from than that currently used in corn production, save for the preferential use of glufosinate with DP202216 corn, in lieu of other herbicides. Herbicides differ from each other with regard to their effects on soil biota relative to their toxicological profile, mode of action, and environmental behavior (Wolmarans and Swart 2014; Dennis et al. 2018). Most herbicides used at normal field rates are generally considered to have no major or long-term effect on soil microbial activities (Busse et al. 2001; Zabaloy et al. 2008; Rose et al. 2016; Nguyen et al. 2018). Some herbicides serve as sources of carbon and energy for microorganisms, and are degraded and assimilated by soil biota, while others can effect soil organisms, and soil biochemical processes (Marin-Morales et al. 2013). For glufosinate, soil microbial degradation is the primary process by which it is degraded in the environment. The aerobic half-life for glufosinate in soil is typically 3-11 days with an anaerobic half-life of 5-10 days. Field dissipation half-lives of 6-20 days (avg. 13 days) are typical (TOXNET 2015). Studies on the effects of glufosinate based herbicides on soil communities are limited. Among those that have been conducted, no significant adverse effects on the composition of soil bacterial and archaeal communities, or the composition of nematode communities, have been identified (Tothova et al. 2010; Dennis et al. 2018).

Many species of soil bacteria are resistant to glufosinate, and able to degrade the herbicide. Tothova et al. (2010) found several species naturally resistant to glufosinate occur in soils, in spite of the fact that the sampled soil was never before exposed to the herbicide or seeded with any GE crops. It has also been observed that repeated field application of glufosinate can increase the occurrence of glufosinate-resistant bacteria in soils. Bartsch and Tebbe (1989) found that more than 90% of 300 microbial isolates from a barley field, which had been exposed to two annual applications of Basta (glufosinate-ammonium), tolerated high concentrations of soil glufosinate, whereas less than 5% were inhibited by glufosinate. Similarly, studies by Hsiao et al. (2017) also suggests that long-term herbicide exposure is a promotive factor in generating bacterial strains having high degradation efficiency of glufosinate.

4.3.3.2 Wildlife Communities

DP202216 corn cropping systems would not be expected to affect animal communities any differently from that of current corn cropping systems; namely corn and other crops using glufosinate. DP202216 corn is agronomically and phenotypically similar to other corn varieties apart from the PAT and ZMM28 traits. Thus, conceptually, the only potential risk to wildlife, as a matter of hazard assessment, would be from exposure to the trait genes and gene products via consumption of the kernel or other plant parts, this type of feeding largely limited to granivorous insects, foraging birds, rodents, and larger mammals. As discussed in 4.1–Scope of Analysis, neither the PAT nor ZMM28 present any known hazard to wildlife.

The environmental risks of pesticide use on wildlife and wildlife habitat are assessed by the EPA in the pesticide registration process and are regularly reevaluated by EPA for each pesticide to maintain its registered status under FIFRA. The EPA’s most recent ecological risk assessment for glufosinate indicated a moderate level of concern for mammals when exposed to the maximum labeled single application rate for most crops (4.1–Scope of Analysis). Chronic risk estimates for other taxa fell below the Agency’s level of concern (LOC) (US-EPA 2016c). Glufosinate use with DP202216 corn would be subject to EPA label and other use requirements, which are in part based on risks posed to wildlife (US-EPA 2016c).

4.3.3.3 Plant Communities

DP202216 corn production would be expected to have the same impacts on vegetation proximate to corn fields as currently cultivated corn varieties, relative to the particular herbicides used. Consistent with its herbicidal mode of action, glufosinate adversely affects monocotyledonous (monocot) and dicotyledonous (dicot) species (US-EPA 2016c). Glufosinate spray drift may inadvertently impact non-target plants proximate to DP202216 corn fields. Consequently, glufosinate based herbicides must be used in accordance with the spray drift management precautions on the label to minimize off-site exposures (US-EPA 2016c). Herbicides that contain glufosinate are often applied with a boom-mounted sprayer that dispenses a medium droplet size. However, they can be occasionally applied aerially on corn, cotton, potato, and soybean. Generally, less than 1% of the total amount of glufosinate-containing herbicides that were applied were applied from the air, with the largest part on potatoes (22% of potato crops are treated with glufosinate by air) (US-EPA 2016c). The EPA provides extensive guidance and label use requirements for reducing the probability of herbicide spray drift and volatilization (US-EPA 2015).

4.3.3.4 Gene Flow and Weediness

DP202216 corn, if grown for commercial purposes, would be cultivated as are current corn varieties and present the same potential risk for gene flow, specifically the propensity for and frequency of gene flow,

as current field corn varieties. Accordingly, a determination of nonregulated status for DP202216 corn and subsequent commercial production would not be expected to present more or less risk for gene flow to wild relative species, or other corn crops, as do current corn varieties.

The T-DNA inserted into DP202216 corn is comprised of two gene cassettes, one conferring increased yield potential, and the other glufosinate resistance. It is assumed for the purposes of this analysis that the entire T-DNA segment, both trait gene cassettes, would be transferred in the event gene flow occurred. The observed inheritance pattern in corn predicts the segregation of these genes and/or traits as a single unit, and at a single genetic locus (Pioneer 2019c).

Relative to the ZMM28 trait that confers increased yield potential: DP202216 corn exhibited increased grain yield by an average 3.4 bushels/acre across a 3 year study, as compared to control corn (Pioneer 2019c). DP202216 corn is otherwise agronomically similar to non-GE corn comparators with respect to reproductive parameters, and susceptibility to pest, disease, and abiotic stress (Wu et al. 2019).

Tripsacum species are the only sexually compatible plants known to occur in the United States. Conceptually, the increased yield trait could confer a potential fitness advantage to *Tripsacum* species that acquired the trait via pollination by DP202216 corn. The extended and increased *zmm28* gene expression results in DP202216 corn plants with greater vegetative stage plant height, leaf biomass, and total leaf area (Wu et al. 2019). Overexpression of *zmm28* also increases photosynthesis across various light levels, as well as nitrogen uptake. Studies by (Wu et al. 2019) demonstrated that N uptake was significantly greater by 16% in DP202216 corn to control corn varieties. Overall, phenotypic differences described by (Wu et al. 2019) include an increase in early plant vigor—measured as an increase in plant height and leaf biomass, as well as an increase of total leaf area—, nitrogen utilization, and photosynthesis (Wu et al. 2019). There were no undesirable secondary phenotypes observed.

The *pat* transgene would confer resistance to glufosinate-ammonium. In the event gene flow from DP202216 corn to *Tripsacum* species occurred, the HR trait would not be expected to confer any fitness advantage, or disadvantage, in areas where glufosinate was not regularly used.

As reviewed in 3.3.4.3–Gene Flow among Corn (*Zea mays* L.) and Wild Relative Species, teosinte do not appear to be present in the United States other than in botanical gardens or at research stations. The closest relative of *Zea mays* in the United States is the genus *Tripsacum*. Three species have been identified: *T. dactyloides*, Eastern gamagrass, in the eastern half of the United States, *T. lanceolatum*, Mexican gamagrass, occurs in the southwest of the United States, and *T. floridanum*, Florida gamagrass, is native to South Florida and Cuba. *T. dactyloides* is the only *Tripsacum* species of widespread occurrence and agricultural importance in the United States, and commonly grown as a forage grass.

While conceptually possible, the likelihood of *Tripsacum* populations comprising the *zmm28* and *mo-pat* trait genes developing is considered remote, for two reasons. First, in contrast with corn and teosinte (*Zea* spp.), which may hybridize relatively easily under certain conditions, as discussed in Subsection 3.3.4 - Gene Flow and Weediness of Corn, the potential for hybridization and successful introgression of *Z. mays* genes into *Tripsacum* populations is rare (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995). Special techniques are required to hybridize *Z. mays* and *Tripsacum*; hybrids of *Tripsacum* species with *Zea* species do not commonly occur outside of a laboratory. Offspring are often sterile or have reduced

fertility, and are unable to withstand even mild winter conditions (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995).

Second, while corn pollen can travel as far as 1/2 mile (800 m) in 2 minutes in a wind of 15 miles per hour (27 km/h) (Nielsen 2016), most pollen is deposited within a short distance of the corn plant. Numerous studies show the majority (84-92%) of pollen grains travel less than 16 feet (5 meters) (Pleasant et al. 2001). At a distance of 200 feet (60 m) from the corn plant, the pollen concentration averages only about 1%, compared with pollen samples collected about 3 feet (0.9 m) from the pollen source (Brittan 2006). The number of outcrosses is reduced to one-half at a distance of 12 feet (3.6 m) from the pollen source, and at a distance of 40 to 50 feet (12 to 15 m), the number of outcrosses is reduced by 99% (Brittan 2006). Thomison (2004) showed cross-pollination between cornfields could be limited to 1% or less by a separation distance of 660 feet (200 m), and to 0.5% or less by a separation distance of 984 feet (300 m). However, cross-pollination frequencies could not be reduced to 0.1% consistently, even with isolation distances of 1,640 feet (500 m). More stringent measures such as 2 weeks of temporal isolation in flowering and isolation distances of 750 m may be needed to achieve 0% outcrossing between nearby corn plantings (Halsey et al. 2005; Goggi et al. 2006).

In the United States, neither corn (*Zea mays*) nor *Tripsacum* is listed as a weed on the Federal Noxious Weed List (7 CFR part 360) (USDA-NRCS 2019e). Corn, domesticated *Zea mays*, has been cultivated throughout the United States without any evidence it forms persistent feral populations.

Based on these factors, it is unlikely the PAT and ZMM28 traits extant in DP202216 corn present any risk to communities of *Tripsacum* species or their ecological role in the communities of other plants.

4.3.3.5 Biodiversity

Commercial production of DP202216 corn would affect biodiversity in and around DP202216 corn crops no differently than other corn cropping systems. As discussed through this Section (4.3.3), the ZMM28 and PAT trait proteins are unlikely to present any risks to plant, animal, fungal, or bacterial communities. The same or functionally similar ZMM28 proteins are ubiquitous among plants, and PAT trait among soil dwelling *Streptomyces* species.

4.3.4 Human Health

There are no risks to public health that would derive from approval of the petition for DP202216 corn. DP202216 corn is a field (dent) corn variety (e.g., *Zea mays indentata*), so it is less likely to be directly consumed by humans, as most such corn is used for feed, fuel production, and other non-food uses as described in Section 3.1. Direct consumption of corn is generally limited to sweet corn (*Zea mays* convar. *saccharata* var. *rugosa*), popcorn (*Zea mays everta*), and flint corn (*Zea mays indurata*) (e.g., polenta). DP202216 corn could, however, be used for production of cornmeal flour (used in the baking of cornbread), and manufacture of corn chips, tortillas, and taco shells, and other processed corn products. In this case, as reviewed in Chapter 3 and Scope of Analysis (4.1), the PAT and ZMM28 traits present negligible risks to human health. Due to the negligible risk PAT poses to human health, residues of PAT are exempt from the requirement of a tolerance when used as plant-incorporated protectant inert ingredients in all food commodities (US-EPA 2007). The EPA has established tolerance limits for glufosinate at 40 CFR §180.473. Pesticide tolerance levels for glufosinate have been established for a

wide variety of commodities, including field corn for grain and forage, as described in 40 CFR §180.473 (US-EPA 2019a). The ZMM28 protein naturally occurs in corn. As part of the FDA voluntary consultation program, Pioneer submitted a safety and nutritional assessment for DP202216 corn to the FDA's Center for Food Safety and Applied Nutrition (FDA CFSAN) in 2018 (Pioneer 2019c).

As described in Subsection 3.4.3—Worker Safety, EPA WPS regulations provide protections to agricultural workers, pesticide handlers, and other persons via training, pesticide safety and hazard communication requirements, personal protective equipment requirements, and provision of supplies for routine washing and emergency decontamination. Agricultural workers and handlers, owners/managers of agricultural establishments, commercial (for-hire) pesticide handling establishments, and crop production consultants are provided guidance for compliance with WPS regulations (US-EPA 2016b).

4.3.5 Animal Health and Welfare

There are no risks to animal health and welfare that are associated with PAT and ZMM28 traits present in DP202216 corn. Food safety reviews frequently will compare the compositional characteristics of the GE crop with non-transgenic, conventional varieties of that crop. This comparison also evaluates the composition of the modified crop under actual agronomic conditions, including various agronomic inputs. The nutrient composition of grain and forage samples derived from DP202216 corn and a near-isogenic non-GE control was evaluated. The compositional analyses of grain included crude protein, crude fat, crude fiber, acid detergent fiber and neutral detergent fiber, ash, carbohydrates, fatty acids, total amino acids, vitamins, minerals, key anti-nutrients, and key secondary metabolites. Compositional analyses of forage included crude protein, crude fat, crude fiber, ash, carbohydrates, calcium, and phosphorus (Pioneer 2019c). The majority of compositional metrics measured between DP202216 corn and its non-GE, near isogenic control did not result in any significant differences in forage or grain. While significant differences were observed in grain for three amino acids (glycine, methionine, and serine) and two vitamins (vitamin B1 (thiamine) and vitamin B3 (niacin)), the observed values of these components were within the range of variation observed in other commercially-available corn varieties (ILSI-CERA 2020). Pioneer is consulting with the FDA as to the safety of feed derived from DP202216 corn (Pioneer 2019c; US-FDA 2019).

4.3.6 Socioeconomic Impacts

4.3.6.1 Domestic Economic Environment

DP202216 corn may be cultivated to produce corn based food, feed, and fuel products (Pioneer 2019c). DP202216 corn, were it adopted, would be expected to replace some GE HR varieties of corn currently cultivated, commensurate with benefits DP202216 corn provided growers. The selection and cultivation of corn varieties is based on efficiencies in crop production. As described in Section 4.3.1.1, DP202216 corn may provide increased yields as compared to other varieties, and as a glufosinate resistant variety, may prove useful in the management of weeds, and HR weeds and their development. Agronomic inputs in corn production as described in section 3.1.2.2 are expected to be unchanged, with the exception of possible substitution of glufosinate for some other herbicides. Information contained within the Pioneer (2019a) petition (Pioneer 2019b) demonstrates that the cultivation practices needed for growing DP202216 corn are similar to practices used to grow conventional corn, and no changes in either insect or disease control measures, e.g. pesticide inputs are foreseen. Consequently, this variety may be

competitive in grower selection of corn varieties. These factors considered, the potential impacts of DP202216 corn on domestic markets would be considered largely beneficial. There are no adverse economic impacts associated with the introduction of DP202216 corn to commercial markets, and grower adoption of DP202216 corn.

4.3.6.2 International Trade

Approval of the petition is unlikely to have substantial effect on the trade of corn based food, feed, and fuel products. It is expected that DP202216 corn, a field corn variety (e.g., *Zea mays indentata*), would be used for production of common feed and industrial commodities, and stock for fuel ethanol. It could also be used for processed food commodities.

As with all GE crop commodities, there exist the potential for low level presence (LLP) occurring in countries importing U.S. agricultural commodities. The issue of asynchronous approval (AA), and resulting LLP situations, can lead to trade delays, shipment rejection, and costs to traders (FAO 2014). GE crop producing countries are required to take the measures necessary in the production, harvesting, transportation, storage, and marketing of GE crop commodities to avoid LLP/AP. International trade is facilitated by the World Trade Organization (WTO) and the Organization for Economic Cooperation and Development (OECD) (OECD 2015; WTO 2019a). Standards and guidelines for the safety evaluation and trade of GE crop commodities are established under international policy and agreements such as the Codex Alimentarius (FAO 2009), the WTO International Plant Protection Convention (WTO 2019b), WTO Sanitary and Phytosanitary Measures (WTO 2019a), WTO Technical Barriers to Trade Agreement (WTO 2019c) and the Cartagena Protocol on Biosafety (CBD 2019a).

DP202216 corn would be subject to the same international regulatory requirements, discussed above, as currently traded corn varieties. In general, developers have various legal, quality control, and marketing motivations to implement rigorous stewardship measures to ensure IP, prevent commingling, and avoid AA and LLP. By necessity, all international regulatory and industry standards and requirements must be met for marketing of DP202216 corn commodities.

Pioneer also implements a product stewardship program that aims to manage the life cycle of products for maximum product value, benefits, and longevity (e.g., DP202216 corn). This stewardship program in part helps growers understand and meet their grain and grain byproduct marketing responsibilities and export approvals (Pioneer 2019b).

5 CUMULATIVE IMPACTS

CEQ NEPA implementing regulations (40 CFR 1508.7) define a cumulative impact as an “impact on the environment which results from the incremental impact of the action when added to other past, present, and reasonably foreseeable future actions regardless of what agency [federal or non-federal] or person undertakes such other actions”. Emissions of air pollutants from a multitude of individual sources is an example of a cumulative environmental impact.

5.1 Assumptions Used for Cumulative Impacts Analysis

If there are no direct or indirect impacts associated with those aspects of the human environment discussed in Chapter 4 APHIS assumes there can be no cumulative impacts. Further assumptions and uncertainties that are part of evaluation of potential cumulative impacts are summarized as follows.

Upon a decision to deregulate DP202216 corn, it may be used to generate, through traditional breeding methods, stacked-trait corn varieties that are both HR and IR, or resistant to two or more herbicides. Stacked-trait varieties possessing both HR and IR are the most commonly produced GE varieties (Euginius 2015; CLI 2019). Whether DP202216 corn or progeny will be stacked with traits from any particular nonregulated GE corn variety, or non-GE cultivar, is uncertain. It is however assumed that DP202216 corn would be used for this purpose; to expand grower choice and production efficiencies in the management of plant pests, pathogens, and agricultural weeds. Adoption rates of stacked-trait corn varieties have increased in recent years, with stacked-trait corn expanding from 1% of planted acres in 2000, to 80% in 2019. In 2019, only around 9% of corn acres are planted with a single HR trait, and 3% a single IR trait (USDA-ERS 2019b). GE varieties incorporating three or four traits are now common, in both corn and cotton.

The increase in adoption of stacked-trait GE varieties is due in part to the fact that stacked-trait varieties have been found to potentially generate higher yields relative to conventional seeds, or seeds with only one GE trait (Fernandez-Cornejo et al. 2014a). This is particularly true when pest or weed pressures are high. For example, USDA 2010 ARMS data indicate that conventional corn seeds had an average yield of 134 bushels per acre, while seeds with two types of herbicide tolerance (glyphosate and glufosinate) and three types of insect resistance (corn borer, corn rootworm, and corn earworm) had an average yield of 171 bushels per acre (Fernandez-Cornejo et al. 2014a).

The adoption level of crossbred progeny of DP202216 corn would depend on the extent to which producers valued the traits offered by such stacked-trait DP202216 corn varieties over other stacked-trait corn varieties, and the pricing and production efficiencies of such stacked-trait DP202216 corn varieties relative to other corn varieties (of which there are a substantial number).

5.2 Cumulative Impacts: Agricultural Production of Corn

Areas and Acreage of Corn Production

The commercial availability of DP202216 corn would have no effect on the acreage or area devoted to corn production in the United States, thus there are no reasonably foreseeable cumulative impacts on corn acreage, or the areas of corn production, that would derive from approval of the petition.

5.2.1 Herbicide Use

DP202216 corn may contribute to a shift in the types of herbicides used, with DP202216 corn facilitating more glufosinate use (in lieu of other herbicide active ingredients). However, this wouldn't be expected to contribute to any significant cumulative effect on the total pounds of herbicide a.i./acre used in corn production. Herbicide use, in terms of lbs a.i./acre/year, will fluctuate relative to the particular GE HR variety, or non-GE variety, growers elect to produce, and the herbicides used with the variety selected. Herbicide selection by growers naturally fluctuates to some degree relative to the severity of weed and herbicide resistant weed species present. Thus, while DP202216 corn would drive the use of glufosinate, and perhaps other herbicides relative to stacked-trait progeny, this would not be expected to contribute to any significant cumulative increase in total herbicide use on corn.

The above considered, relative to glufosinate use nationally, as discussed in Section 3.1.2.2.2 (see Fig 3-7), glufosinate is applied on canola, corn, cotton, potato, and soybean. Several other crops, including almonds, apples, cherries, grapes, oranges, peaches, pears, pistachios, and walnuts are treated with glufosinate at rates greater than 1.0 lb a.i./acre (US-EPA 2016c). Thus, DP202216 corn production could possibly contribute to an increase in the nationwide use of glufosinate. This would be relative to which GE HR corn varieties growers choose to produce, and the particular herbicides used with those GE HR corn varieties. Thus, as DP202216 corn is adopted, there could potentially be an increase in the nationwide use of glufosinate, and decrease in use of other herbicides.

5.2.2 Weed and Herbicide Resistant Weed Management

As discussed in 4.3.1.2, glufosinate, a broad-spectrum herbicide with only 2 resistant weed populations (same species) reported in the United States, is considered an important tool for growers in the management of weeds and weed resistance management. In this respect, DP202216 corn could potentially contribute in a cumulative manner to helping allay the development of resistant weed biotypes in corn, and management of current herbicide resistant weed populations. Management for development of glufosinate resistant weeds would be subject to EPA recommendations (US-EPA 2016a), and Pioneer product stewardship requirements (Pioneer 2019c, b).

While the need for management of development of weeds and HR weed populations is well recognized, and glufosinate considered an important tool, how to best do so continues to be debated. Although HR-trait stacking can offer growers increased flexibility in the management of HR weeds, many weed scientists and academics are of the opinion that this solution is not sustainable in the long-term using current practices, which will inevitably lead to increased incidences of multiple-HR weed populations (Heap and Duke 2018; Beckie et al. 2019). With the rapid transition towards corn and other crop cultivars with stacked (two- and three-trait) HR varieties, some weed scientists argue that over reliance on these stacked-trait HR crops will result in weed populations resistant to multiple herbicide MOAs (Heap and Duke 2018; Beckie et al. 2019). Current consensus among many weed scientists is that weeds must be managed employing IWM practices that limit herbicide use, while employing multiple other non-

chemical strategies such as harvest weed seed control to reduce weed seed banks (Heap and Duke 2018; Beckie et al. 2019).

While stacked-trait crop varieties using mixtures of herbicides with multiple MOAs are emerging as the main strategy for weed and HR weed management for many growers, it is unlikely these crop varieties alone, absent non-chemical strategies, will be sufficient to keep up with the capacity of weeds to evolve resistance to herbicides (Green 2018; Heap and Duke 2018; Beckie et al. 2019). Owen (2016) is of the opinion that unless diverse chemical and non-chemical IWM approaches to HR weed management are widely adopted, it is inevitable that evolved herbicide resistances in key weeds will continue to increase and costs to agriculture and consumers will continue to escalate.

Within the context of using stacked-trait cultivars for weed and weed resistance management, DP202216 corn, with the traits of glufosinate resistance and increased yield, could be used for development of GE HR stacked-trait/pyramided corn varieties. The efficacy of such stacked-trait hybrids in the management of weeds will depend on various factors; namely the extent to which such stacked-trait varieties are utilized as part of a diversified IWM program employing various non-chemical strategies. It is unknown as to how future stacked-trait hybrids, derived from DP202216 corn, may be successfully or unsuccessfully incorporated into sustainable IWM programs.

5.3 Cumulative Impacts: Physical Environment

Production of corn entails the use of pesticides, fertilizers, and tillage, which can contribute to potential cumulative impacts on water, soil, and air quality. DP202216 corn would be no exception. The agronomic practices and inputs that would be used in the cultivation of DP202216 corn, and the contribution to the cumulative impacts of these practices and inputs, would be no different than that of currently cultivated corn varieties.

5.3.1 Soils

Any contribution to cumulative impacts on soil quality resulting from practices and inputs used in DP202216 corn cultivation would be the same as or similar to that with other corn varieties. Cumulative impacts would be relative to the tillage practices and chemical inputs employed, as well as crop rotation and cover cropping practices. As discussed in 4.3.2.1–Soil Quality, DP202216 corn cultivation would be the same or similar as for other corn varieties, and trait genes, gene products, and phenotype, are unlikely to affect soil quality.

5.3.2 Water Quality

Cumulative impacts on water resources derive from point source and non-point source (NPS) pollutants. NPS contaminants in runoff originate from sources such as construction sites (e.g., residential and commercial development, construction of roads/highways), impervious surfaces (parking lots, roads/highways, rooftops), and crop fields and livestock rearing facilities. NPS pollutants include fertilizers and pesticides applied to residential, commercial, and agricultural sites, and sediments from the built environment as well as unmanaged landscapes. As discussed in 3.2.2–Water Resources, the most common NPS contaminants in agricultural run-off are sediment, nutrients such as nitrogen and phosphorus, and pesticides, all of which can adversely affect aquatic ecosystems.

Point source pollutants are discharged from any identifiable, singular source, such as a pipe, drain, tunnel, conduit, or vessel. Factories and sewage treatment plants are examples of point sources. Factories, such as oil refineries, pulp/paper mills, and chemical, electronics and automobile manufacturers typically discharge one or more pollutants in EPA regulated effluents. Some factories discharge effluents directly into a waterbody, others treat it themselves before it is released, and some send their wastes to sewage treatment plants. Livestock rearing facilities (e.g., dairy and beef cows, pigs, chickens) are other sources of point source pollution. These types of operations are identified as concentrated animal feeding operations (CAFOs). Waste from agricultural livestock operations has been a long-standing concern with respect to contamination of water resources, particularly in terms of nutrient pollution, microbial pathogens, and pharmaceuticals present in the waste (Burkholder et al. 2007).

To control point source discharges, the Clean Water Act established the National Pollutant Discharge Elimination System (NPDES). Under the NPDES program, factories, CAFOs, sewage treatment plants, and other point sources must obtain a permit from the state and EPA before they can discharge their waste or effluents into any body of water. Prior to discharge, the point source must use the latest technologies available to treat its effluents and reduce the level of pollutants. If necessary, a second, more stringent set of controls can be placed on a point source to protect a specific waterbody.

As discussed in 3.2.2–Water Resource, tillage and agronomic inputs, on a regional scale, can and do contribute to the impairment of surface waters through soil erosion and runoff of pesticides and fertilizers (nutrients). Agricultural inputs can also impact groundwater through leaching. Agricultural runoff, to include from corn fields, is a primary contributor to NPS pollutants that impact streams, rivers, lakes, and estuaries. Cultivation of DP202216 corn and progeny would potentially contribute to cumulative impacts on water quality as do other cropping systems. DP202216 corn is expected to replace other corn varieties in the event of adoption (no increase in acreage); thus, the sources of potential cumulative impacts on water resources, namely NPS pollutants in agricultural run-off, would not be expected to substantially differ between the No Action and Preferred Alternative. Any contribution of DP202216 corn production to cumulative impacts on water quality would be similar to that which currently occurs.

The development of HR weeds continues in many areas of the United States (Heap 2020). Where HR weeds are particularly problematic and other strategies are not effective, growers may have to forego conservation tillage and use more aggressive tillage practices to control HR weeds, which can increase soil erosional capacity, and, potentially, agricultural run-off. This is in fact the case; in some areas of the South growers have returned to more aggressive conventional tillage to control resistant weed populations (Morrison 2014; Sosnoskie and Culpepper 2014). As discussed above, DP202216 corn could be used for development of GE HR stacked-trait/pyramided corn varieties to potentially help manage the development of HR weed populations. There is some uncertainty, however, as to how future stacked-trait hybrids derived from DP202216 corn, or any other HR variety, in the long-term, will help allay or prevent the development of HR weed populations. This will be relative to the diversity of chemical and non-chemical strategies employed in IWM programs.

5.3.3 Air Quality

Air pollution is inherently a problem resulting from the cumulative emissions of various sources. The EPA has categorized primary emissions sources into: point, mobile, biogenic, and area. Point sources

include major industrial facilities such as chemical plants, oil refineries, and power plants. Mobile sources include cars, trucks and buses and off-road equipment such as ships, airplanes, and agricultural and construction equipment. Area sources are defined as smaller operations such as dry cleaners and gas stations. Biogenic sources are comprised of vegetation, soils, and animals.

The Clean Air Act (CAA) establishes a number of permitting programs designed to carry out the goals of the Act. Some of these programs are directly implemented by EPA through its Regional Offices but most are carried out by states, local agencies, and approved tribes. As discussed in 3.2.3–Air Quality, the EPA establishes NAAQS pursuant to the CAA that are intended to protect public health and the environment. The EPA has also identified over 187 HAPs, including substances that cause cancer, neurological, respiratory, and reproductive effects. While the EPA establishes NAAQS, the standards do not set emission control requirements for any particular industry, including agriculture. The USDA and the EPA provide guidance for regional, state, and local regulatory agencies, and farmers, on how to best manage agricultural emissions sources. Agricultural emission sources include: PM from tillage and agricultural burning; CO₂, NO₂, SO₂ from fossil fuel consumption associated with equipment used in tillage, pesticide application, and harvest; and soil nitrous oxide (N₂O) emissions from the use of fertilizers/manure. Volatilization of pesticides can also be a concern.

As discussed in 4.3.2.3–Air Quality, the agronomic practices and inputs for DP202216 corn production are the same as/similar to other corn varieties, there would be no increase in acreage resulting from DP202216 corn production, no changes to emission sources (i.e., tillage, fossil fuel burning equipment, the application of fertilizers and pesticides), nor to volume of emissions from U.S. corn production. Because there would be no increase in acreage resulting from DP202216 corn production, nor changes to emission sources (i.e., tillage, fossil fuel burning equipment, the application of fertilizers and pesticides), and DP202216 corn would be expected to replace other corn varieties, there are no cumulative effects on emissions of NAAQS pollutants unique to the production of DP202216 corn. If DP202216 corn were produced, air quality would continue to be affected along current trends by emission sources such as tillage (PM), pesticide application (aerosols, spray drift), and use of farm equipment that combusts fossil fuels (NAAQS pollutants). The EPA and USDA efforts to reduce emissions, along with state and local efforts would likewise continue (US-EPA 2017b).

5.4 Cumulative Impacts: Biological Resources

There were no direct or indirect impacts on soil biota, wildlife communities, or plant communities identified in Chapter 4 that differ from those associated with corn varieties currently produced. Consequently, there are no unique contributions to cumulative impacts on biological resources, beyond those already known to occur with corn production; both conventional and GE corn. Neither DP202216 corn nor its progeny would require any change in agronomic practices or inputs used to cultivate corn. As discussed in Chapter 4, the PAT and ZMM28 traits pose no known risk to biological resources. Glufosinate use would be no different for DP202216 corn, subject to EPA label and weed resistant management recommendations.

5.4.1 Gene Flow and Weediness

Gene movement from DP202216 corn or progeny derived from this variety to *Tripsacum* species is considered a highly unlikely event, as concluded in the PPRA (USDA-APHIS 2019a) and Section 4.3.3.4.

While DP202216 corn, if commercially produced, would introduce an additional novel trait (ZMM28) to the agricultural community/corn market, this is not expected to contribute in any cumulative manner to an increased (compound) risk for gene flow from GE corn to wild or cultivated *Tripsacum* species. DP202216 corn and progeny would present no more or less risk for gene flow to *Tripsacum* species, or other corn crops, than do current corn varieties.

5.5 Cumulative Impacts: Human Health, Worker Safety, and Animal Health and Welfare

Risks to public health primarily derive from the potential effects of crop production and other agricultural activities on air and water quality (pollutants), and pesticide application. There were no DP202216 corn associated risks to public health or worker safety identified in Chapter 4 that differ from production of other corn crops. The introduced trait genes and gene products present negligible risk to human and food animal health. Consequently, there are no potential cumulative impacts on human or animal health that would derive from approval of the petition, and subsequent commercial use of DP202216 corn.

5.6 Cumulative Impacts: Socioeconomics

DP202216 corn would entail entry of another GE corn variety into the agricultural commodities markets. DP202216 corn, as a new GE corn variety (HR and Hi-yield potential), would require segregation from the organic food/feed supply chain, and DP202216 corn seed segregation to capture the added value of hi-yield to growers. DP202216 corn seed and grain would also require segregation to prevent entry into export markets that haven't yet approved it—incidences of LLP. These, however, would not be considered production and marketing requirements that presented unusual or unique risks in a cumulative sense. New varieties of corn and specialty commodities are expected to be continually developed and marketed to meet market demands for food, feed, and fuel. Thus, entry of DP202216 corn into domestic and foreign markets would contribute to cumulative impacts on the need for segregation of seed corn, segregation from organically produced commodities, and from export supply chains moving to markets in which DP202216 corn is not yet approved in the same way as other corn varieties that have and will enter the market. This type of cumulative impact is not considered unique to DP202216 corn, rather, it is inherent to the GE, conventional, and organic corn commodities markets. Identity preservation certification programs are well developed and inherent part of crop production in the United States, to include GE crops (Sundstrom et al. 2002). Pioneer implements a product stewardship program that in part helps growers meet their grain and grain byproduct marketing responsibilities, to include export approvals (Pioneer 2019b).

5.6.1 Weed and Weed Resistance Management Costs

Managing weed resistance is more cost effective than ignoring resistance (Fernandez-Cornejo and Osteen 2015; Livingston et al. 2015). Studies by Livingston et al. (2015) suggest that corn growers who had reported a glyphosate resistant weed infestation in 2010 realized significantly lower total returns (-\$67.29/acre) than similar corn growers who had not reported such an infestation. The results suggest that lower yields and higher chemical and fuel costs might have contributed to the shortfall in returns, although the differences in yields and chemical and fuel costs themselves were not statistically significant at the 10% level. These findings suggest weed resistance contributed to a substantial reduction in returns on affected corn growers. The studies further suggest that corn growers who used glyphosate and at least

one different herbicide MOA had higher operating costs (due in part to higher chemical costs), but also higher yields, total returns (\$15.72/acre), and operating returns (\$20.74/acre) than did corn growers who were very similar, but used glyphosate by itself. In general, corn producers who used at least one other herbicide in combination with glyphosate, even while the crop producers had higher production costs, had higher yields and net returns on their crops (Fernandez-Cornejo and Osteen 2015).

For growers, nationally, herbicide-resistant weeds incur significant costs. Some growers are required to add additional herbicide applications to protect their investments, which by some estimates can increase production costs by 30%-40% (Weaver 2019). Growers are also hiring crews to hand weed their fields (Sfiligoj 2014). In both cases, estimates from the University of Wisconsin research suggest this is adding approximately \$2 billion to U.S. growers' annual crop production bills (Sfiligoj 2014).

USDA research suggests that use of stacked-trait corn seeds, utilizing herbicides with differing MOAs, can provide higher yields than conventional seeds or seeds with only one GE trait. An analysis of the 2010 USDA Agricultural Resource Management Survey found that conventional corn seeds yielded 134 bushels per acre in 2010. In contrast, seeds with two types of herbicide resistance MOAs (glyphosate and glufosinate) and three types of insect resistance (corn borer, corn rootworm, and corn earworm) yielded 171 bushels per acre (Fernandez-Cornejo et al. 2014a). In general, current data indicate that, while stacked-trait HR seed varieties may incur more costs in some instances, the use of IWM practices in tandem with stacked-trait varieties can improve yields and gains in net-returns (Fernandez-Cornejo and Osteen 2015; Livingston et al. 2015; Westcott and Hansen 2015). DP202216 corn, with the traits of glufosinate resistance and increased yield, could contribute to the options available for development of GE stacked-trait corn varieties, potentially providing for new varieties that could increase grower yields and net returns.

This potential benefit considered, as discussed in Section 5.2, although HR-trait stacking/pyramiding offers growers increased flexibility to manage HR weeds, and can contribute to increased yields, many weed scientists and academics are of the opinion that reliance on this approach alone to weed management may not be sustainable in the long-term, and will inevitably lead to increased incidences of HR weed populations resistant to multiple herbicide MOAs (Heap and Duke 2018; Beckie et al. 2019). Owen (2016) is of the opinion that unless diverse IWM approaches to HR weed management are widely adopted, it is inevitable that evolved herbicide resistances in key weeds will continue to increase, and in turn costs to agriculture and consumers. The efficacy of such stacked-trait hybrids in management of weeds and costs to growers and consumers will depend on the extent to which such stacked-trait varieties are utilized as part of an IWM program employing various other non-chemical strategies.

6 THREATENED AND ENDANGERED SPECIES

The Endangered Species Act (ESA) of 1973, as amended, is a far-reaching wildlife conservation law. Congress passed the ESA to prevent extinctions facing many species of fish, wildlife and plants. The purpose of the ESA is to conserve endangered and threatened species and the ecosystems on which they depend as key components of America's heritage. The U.S. Fish & Wildlife Service (USFWS) and the National Marine Fisheries Service (NMFS) together comprise "the Services" and implement the ESA by working with other federal, state, and local agencies, Tribes, non-governmental organizations, and private citizens.

Before a plant or animal species can receive the protection provided by the ESA, it must be added to the federal list of threatened and endangered wildlife and plants. Threatened and endangered species (TES) are those plants and animals at risk of becoming extinct throughout all or part of their geographic ranges (endangered species) or species likely to become endangered in the foreseeable future throughout all or a significant portion of their ranges (threatened species).

The Services add a species to the list when they determine the species to be endangered or threatened because of any of the following factors:

- The present or threatened destruction, modification, or curtailment of its habitat or range;
- Overutilization for commercial, recreational, scientific, or educational purposes;
- Disease or predation;
- The inadequacy of existing regulatory mechanisms; and
- The natural or manmade factors affecting its survival.

Once a species is added to the list, in accordance with the ESA, protective measures apply to the species and its habitat. These measures include protection from adverse effects of federal activities.

6.1 Requirements for Federal Agencies

Section 7(a)(2) of the ESA requires that federal agencies, in consultation with USFWS and/or the NMFS, ensure that any action they authorize, fund, or carry out is "not likely to jeopardize the continued existence of a listed species or result in the destruction or adverse modification of designated critical habitat." It is the responsibility of the federal agency taking the action to assess the effects of their action and to consult with the USFWS and NMFS if it is determined that the action "may affect" listed species or designated critical habitat (a process is known as a Section 7 Consultation).

To facilitate the development of its ESA consultation requirements, APHIS met with the USFWS from 1999 to 2003 to discuss factors relevant to APHIS' regulatory authority and effects analysis for petitions for nonregulated status of GE crop lines. By working with USFWS, APHIS developed a process for conducting an effects determination consistent with the Plant Protection Act (PPA) of 2000 (Title IV of Public Law 106-224). APHIS uses this process to help fulfill its obligations and responsibilities under Section 7 of the ESA for biotechnology regulatory actions.

As discussed in Chapters 1 and 2, APHIS regulatory authority over GE organisms under the PPA is limited to those where the Agency has reason to believe the GE organisms could pose a plant pest risk, or where APHIS does not have sufficient information to determine that the GE organism is unlikely to pose a plant pest risk. In this case, Pioneer has requested that the USDA-APHIS consider that DP202216 corn is not a plant pest as defined by the PPA. After completing a PPRA, if APHIS determines that DP202216 corn seeds, plants, or parts thereof do not pose a plant pest risk, then this article would no longer be subject to the plant pest provisions of the PPA or to the regulatory requirements of 7 CFR part 340, and therefore, APHIS must reach a determination that this article is no longer regulated. As part of this EA, APHIS analyzed the potential effects of DP202216 corn on TES and/or critical habitat. As part of this process, APHIS thoroughly reviewed data related to DP202216 corn to inform the ESA effects analysis and, if necessary, the biological assessment. For each transgene/transgenic plant the following information, data, and questions are considered by APHIS:

- A review of the biology, taxonomy, and weediness potential of the crop plant and its sexually compatible relatives;
- Characterization of each transgene with respect to its structure and function and the nature of the organism from which it was obtained;
- A determination of where the new transgene and its products (if any) are produced in the plant and their quantity;
- A review of the agronomic performance of the plant including disease and pest susceptibilities, weediness potential, and agronomic and environmental impact;
- Determination of the concentrations of known plant toxicants (if any are known in the plant); and
- Analysis to determine if the transgenic plant is sexually compatible with any threatened or endangered plant species (TES) or a host of any TES.
- Any other information that may inform the potential for an organism to pose a plant pest risk.

APHIS met with USFWS officials on June 15, 2011, to discuss and clarify whether APHIS has any obligations under the ESA regarding analyzing the effects on TES that may occur from use of pesticides associated with GE crops. As a result of these joint discussions, USFWS and APHIS have agreed that it is not necessary for APHIS to perform an ESA effects analysis on pesticide use associated with GE crops because the EPA has both regulatory authority over the labeling of pesticides under FIFRA, and the necessary technical expertise to assess pesticide effects on the environment. APHIS has no statutory authority to authorize or regulate the use of pesticides by corn growers. Under APHIS' current Part 340 regulations, APHIS only has the authority to regulate DP202216 corn or any other GE organism as long as APHIS believes they may pose a plant pest risk (7 CFR § 340.1). APHIS has no regulatory jurisdiction over any other risks associated with GE organisms including risks resulting from the use of pesticides on those organisms.

6.2 Potential Effects of DP202216 corn on TES

APHIS evaluated the potential effects that a determination of nonregulated status for DP202216 corn may have, if any, on federally listed TES and species proposed for listing, as well as designated critical habitat and habitat proposed for designation. As described in further detail elsewhere in this EA, in the petition

(Pioneer 2019c), and in the PPRA (USDA-APHIS 2019a), Pioneer engineered DP202216 corn to provide increased yield, and for resistance to the herbicide active ingredient glufosinate-ammonium.

Based on the information submitted by the applicant and reviewed by APHIS, DP202216 corn, with the exception of increased yield and resistance to glufosinate-ammonium, is agronomically and compositionally comparable to conventional corn (Pioneer 2019c). Pioneer has presented results of agronomic field trials for DP202216 corn. The results of these field trials demonstrate that there are no differences in agronomic practices between DP202216 corn and conventional corn (Pioneer 2019c). The common agricultural practices that would be carried out in the cultivation of DP202216 corn are not expected to deviate from current practices, including the use of EPA-registered pesticides. DP202216 corn is not expected to directly cause a change in agricultural acreage or area devoted to corn production in the United States (see Subsection 4.3.1). It is expected that DP202216 corn will replace other HR varieties without expanding the acreage or area of corn production.

Corn can be grown in all 50 states and U.S. territories. The issues discussed herein focus on the potential environmental consequences of approving the request for nonregulated status of DP202216 corn on TES and critical habitat in the areas where corn is currently cultivated. APHIS obtained and reviewed the USFWS list of TES species (listed and proposed) for all 50 states and U.S. territories where corn is produced from the USFWS Environmental Conservation Online System .

For its analysis on TES plants and critical habitat, APHIS focused on the agronomic differences between the regulated article and corn varieties currently grown; the potential for increased weediness; and the potential for gene movement to native plants, listed species, and species proposed for listing.

For its analysis of potential effects on TES animals, APHIS focused on the implications of exposure to the ZMM28 and PAT proteins expressed in DP202216 corn as a result of the transformation, and the ability of the GE plants to serve as a host for a TES.

6.2.1 Threatened and Endangered Plant Species and Critical Habitat

The agronomic data provided by Pioneer were used in the APHIS analysis of the weediness potential for DP202216 corn, and evaluated for the potential to impact TES and critical habitat. Agronomic studies conducted by Pioneer evaluated the weediness and invasiveness potential of DP202216 corn with respect to conventional corn (Pioneer 2019c). No substantive differences were detected between DP202216 corn and conventional corn in hardiness, persistence, or susceptibility to pests and diseases, other than the intended effect of resistance to glufosinate-ammonium and increased yield potential. As discussed in this EA and APHIS' PPRA (USDA-APHIS 2019a), there are no weed risks associated with corn. Corn has been cultivated around the globe without any report that it occurs as a serious weed or that it forms persistent feral populations. While the ZMM28 trait confers increased yield potential, this is not a trait that would introduce a weediness risk with DP202216 corn (USDA-APHIS 2019a). Volunteer corn plants can be easily controlled if needed, either with herbicides or manual removal.

APHIS evaluated the potential of DP202216 corn to cross with a listed species. As discussed in Gene Flow and Weediness (Subsections 3.3.4 and 4.3.3.4), the potential for gene movement between DP202216 corn and related *Zea* and *Tripsacum* species is limited. While *Zea mays* may hybridize with other *Zea* (teosinte) species, teosinte species do not appear to be present in the United States other than in

botanical gardens or at research stations. The closest relative of *Zea* in the United States is the genus *Tripsacum*. Three species have been identified: *T. dactyloides*, Eastern gamagrass, is known to occur in the eastern half of the United States, *T. lanceolatum*, Mexican gamagrass, occurs in the southwest of the United States, and *T. floridanum*, Florida gamagrass, is native to South Florida and Cuba. *T. dactyloides* is the only *Tripsacum* species of widespread occurrence and agricultural importance in the United States, and commonly is grown as a forage grass. As discussed in Subsection 3.3.4, gene flow from *Zea mays* to *Tripsacum* species in the United States is improbable. There are no federally listed *Zea* or *Tripsacum* species in the United States (USFWS 2020).

Based on all of these factors, APHIS determined that DP202216 corn will have no effect on threatened or endangered plant species or on critical habitat in the United States.

6.2.2 Threatened and Endangered Animal Species

Threatened and endangered animal species that may be exposed to the gene products from DP202216 corn would be those TES that inhabit corn fields and feed on DP202216 corn. As discussed in Subsection 4.3.3, cornfields are generally considered poor habitat for birds and mammals in comparison with uncultivated lands, but the use of cornfields by birds and mammals is not uncommon. Some birds and mammals use cornfields at various times throughout the corn production cycle for feeding and reproduction. Most birds and mammals that utilize cornfields are ground-foraging omnivores that feed on corn seed, sprouting corn, and the corn remaining in the fields following harvest.

Of the TES birds, whooping crane (*Grus americana*), Mississippi sandhill crane (*Grus canadensis pulla*), piping plover (*Charadrius melodus*), and interior least tern (*Sterna antillarum*) may transit and feed in corn fields during migration (Krapu et al. 2004; Sherfy et al. 2011; USFWS 2011). The whooping crane, in particular, spends the majority of its foraging time during migration in agricultural fields (CWS-USFWS 2007; Jorgensen and Dinan 2016). During migration, about 90% of the sandhill crane diet consists of corn when corn is available (NGP 2019).

As discussed in Section 4.3.3.2—Wildlife Communities, many mammals may feed on corn, particularly white tailed deer, raccoons, mice, and voles. There are no listed raccoon species in the United States. There are two listed deer species in the United States. Key deer (*Odocoileus virginianus clavium*) are highly localized in the Florida Keys (USFWS 1999). Listed populations of Columbian white-tailed deer (*Odocoileus virginianus leucurus*) are found in certain areas associated with the Columbia River in Washington (USFWS 2020). These locations are well south and west, respectively, of the regions where corn crops are typically planted (see Subsection 3.1.1—Acreage and Area of U.S. Corn Production). Of the mice, voles, and their relatives in the Cricetidae family, listed species include: the Amargosa vole (*Microtus californicus scirpensis*), which is listed as endangered and occurs in California (USFWS 2020), as well as the Florida salt marsh vole (*Microtus pennsylvanicus dukecampbelli*), which occurs in salt marsh habitat on the Gulf Coast of Florida (USFWS 2020), the endangered Key Largo woodrat (*Neotoma floridana smalli*) of Florida Key's climax hardwood hammocks (USFWS 2020), and the northern and southern subspecies of the endangered, tidal marsh dwelling, salt marsh harvest mouse (*Reithrodontomys raviventris*) (USFWS 2013, 2020) .

APHIS considered the risks to threatened and endangered animals from consuming DP202216 corn. Pioneer presented information on the food and feed safety of DP202216 corn, comparing the DP202216 corn variety with conventional varieties currently grown. There are no toxins or allergens associated with this plant (Pioneer 2019c). Compositionally, DP202216 corn was determined to be the same as conventional varieties. Nutrient composition analysis of DP202216 corn included proximates, fiber, minerals, fatty acids, amino acids, vitamins, secondary metabolites, and anti-nutrients. The analytes included for the compositional assessment were based on the OECD consensus document on compositional considerations for new varieties of maize (OECD 2003). Results presented by Pioneer show that the introduced genetic material in DP202216 corn does not result in any significant compositional differences between DP202216 corn and non-transgenic corn.

As discussed in those sections addressing human health, animal health and welfare, and wildlife, there are no health hazards associated with the ZMM28 and PAT proteins. The FDA has previously consulted on PAT in various food crops; these include canola, corn, cotton, soybean, and sugar beet. None of these modified crop varieties were identified as presenting a human health risk (US-FDA 2019). PAT is exempt from the requirement for food or feed tolerances in all crops. As part of the FDA voluntary biotechnology consultation program, Pioneer submitted a safety and nutritional assessment for DP202216 corn to the FDA's Center for Food Safety and Applied Nutrition (FDA CFSAN) in 2018 (Pioneer 2019c). There is no expectation that exposure to the PAT and ZMM28 proteins will have any effect on TES that may be exposed to DP202216 corn.

APHIS considered the possibility that DP202216 corn could serve as a host plant for a threatened or endangered species (i.e., a listed insect or other organism that may use the corn plant to complete its lifecycle). APHIS is not aware of any TES for which corn serves as a host plant (USFWS 2011, 2020).

Considering there are no risks to humans or other animals associated with DP202216 corn (discussed in Chapter 4), and the nutritional similarity between DP202216 corn to other varieties currently grown—apart from modified ZMM28 and PAT traits—APHIS has concluded that exposure to and consumption of DP202216 corn would have no effect on threatened or endangered animal species.

6.3 Summary

After reviewing the possible effects of a determination of nonregulated status, and subsequent commercial production of DP202216 corn, APHIS has not identified any stressor that could affect listed TES or species proposed for listing. APHIS also considered the potential effect of DP202216 corn on designated critical habitat and habitat proposed for designation and could identify no differences from effects that would occur from the production of other corn varieties. Corn is not sexually compatible with, nor does it serve as a host species for, any listed species or species proposed for listing. Considering that discussed in Chapter 4, consumption of DP202216 corn by any listed species or species proposed for listing would pose no health risks.

Based on these factors, APHIS has concluded that a determination of nonregulated status of DP202216 corn, and subsequent commercial production of this corn variety, will have no effect on listed species or species proposed for listing, and would not affect designated habitat or habitat proposed for designation.

Because of this no-effect determination, consultation under Section 7(a)(2) of the Act or the concurrences of the USFWS or NMFS is not required.

7 CONSIDERATION OF FEDERAL AND STATE LAWS AND REGULATIONS, EXECUTIVE ORDERS, STANDARDS, AND TREATIES

7.1 Federal Laws and Regulations

The laws most relevant to APHIS determinations of regulatory status are the National Environmental Policy Act of 1969 (NEPA), the Clean Water Act of 1972 (CWA), the Safe Drinking Water Act of 1974 (SDWA), the Clean Air Act of 1970 (CAA), the Endangered Species Act of 1973 (ESA), and the National Historic Preservation Act of 1966 (NHPA). Compliance with the requirements of the ESA has been addressed in Chapter 6. Compliance with the requirements of NEPA, CWA, SDWA, CAA, and NHPA, are specifically addressed in the following subsections.

7.1.1 National Environmental Policy Act (NEPA)

NEPA (42 United States Code (U.S.C) 4321, *et seq.*) is designed to ensure transparency and communication of the possible environmental effects of federal actions prior to implementation. The Act and implementing regulations require federal agencies to document, in advance and in detail, the potential effects of their actions on the human environment, so as to ensure that there is a full understanding of the possible environmental outcomes of federal actions by both the decision-makers and the public. This EA documents the potential environmental outcomes of the alternatives considered, approval or denial of Pioneer's petition, consistent with the requirements of NEPA and Council on Environmental Quality implementing regulations at 40 CFR parts 1500-1508.

7.1.2 Clean Air Act, Clean Water Act, and Safe Drinking Water Act

The CAA, CWA, and SDWA authorize the EPA to regulate air and water quality in the United States. Because DP202216 corn is agronomically equivalent to currently utilized corn varieties, the potential sources of impacts on water resources and air quality are the same under both the No Action and Preferred Alternatives. DP202216 corn production would entail the use of pesticides and fertilizers, and to some extent tillage, which will contribute to potential cumulative impacts on air quality, and potentially water quality. The sources and degree of potential impacts would be no different than that which occurs with current corn production. APHIS assumes use of all pesticides on DP202216 corn will be compliant with EPA registration and label use requirements. As discussed in Chapters 3 & 4, the transgenes and gene products extant in DP202216 corn present no known risks to water or air quality. Considering these factors, approval of the petition would not lead to circumstances that resulted in non-compliance with the requirements of the CWA, CAA, and SDWA.

7.1.3 National Historic Preservation Act

The NHPA of 1966 (NHPA; Public Law 89-665; 16 U.S.C. 470 *et seq.*) and its implementing regulations (36 CFR part 800) requires federal agencies to: 1) determine whether activities they propose constitute "undertakings" that have the potential to cause effects on historic properties and 2) if so, to evaluate the effects of such undertakings on such historic resources and consult with the Advisory Council on Historic

Preservation (i.e., State Historic Preservation Office, Tribal Historic Preservation Officers), as appropriate.

Approval of the petition is not a decision that would directly or indirectly result in alteration of the character or use of historic properties protected under the NHPA, nor would it result in any loss or destruction of cultural or historical resources. As with other corn, for DP202216 corn, standard agricultural practices for land preparation, planting, irrigation, and harvesting of plants would be used on these agricultural lands, including the use of EPA-registered pesticides. Adherence to the EPA label use restrictions for pesticides will mitigate impacts to the human environment, including historic and cultural resources. There may be the potential for increased noise during the operation of machinery and other equipment, as with all corn crop production, however, these activities would have only temporary effects on historic sites in the way of noise, with no consistent long-term effects on the enjoyment of a historical site.

7.2 Executive Orders Related to Domestic Issues

The following executive orders (EO) require consideration of the potential impacts of federal actions on human health, cultural resources, wildlife, and the environment.

- **EO 12898 – Federal Actions to Address Environmental Justice in Minority Populations and Low-Income Populations**

This EO requires federal agencies to conduct their programs, policies, and activities that substantially affect human health or the environment in a manner so as not to exclude persons and populations from participation in or benefiting from such programs. It also enforces existing statutes to prevent minority and low-income communities from being subjected to disproportionately high and adverse human health or environmental effects.

- **EO 13045 – Protection of Children from Environmental Health Risks and Safety Risks**

Children may suffer disproportionately from environmental health and safety risks due to their developmental stage, higher metabolic rates, and behavior patterns, as compared to adults. This EO to the extent permitted by law and consistent with the agency's mission requires each federal agency to identify, assess, and address the potential environmental health and safety risks that may disproportionately affect children.

- **EO 13175 – Consultation and Coordination with Indian Tribal Governments**

Executive departments and agencies are charged with engaging in consultation and collaboration with tribal governments; strengthening the government-to-government relationship between the United States and Indian tribes; and reducing the imposition of unfunded mandates upon Indian tribes. The EO emphasizes and pledges that federal agencies will communicate and collaborate with tribal officials when proposed federal actions have potential tribal implications.

Neither alternative evaluated in this EA is expected to have disproportionate adverse impacts on minorities, low-income populations, or children, or adversely affect tribal entities. As reviewed in Chapter 4, there are no risks to human health, nor to food animal health and welfare, associated with the

introduced genes and gene products in DP202216 corn. DP202216 corn would be cultivated as are all other corn varieties, using the same agronomic practices and inputs.

Tribal entities are recognized as independent governments and agricultural activities on tribal lands would only be conducted if approved by the tribe. Tribes would have control over any potential conflict with cultural resources on tribal properties. Neither approval nor denial of the petition would have any effect on Indian tribal self-governance or sovereignty, tribal treaties, or other rights.

- **EO 13751 – Safeguarding the Nation from the Impacts of Invasive Species**

Invasive species are a significant issue in the United States, causing both adverse economic and environmental impacts. This EO directs actions to continue coordinated federal prevention and control efforts related to invasive species. This order maintains the National Invasive Species Council (Council) and the Invasive Species Advisory Committee; expands the membership of the Council; clarifies the operations of the Council; incorporates considerations of human and environmental health, climate change, technological innovation, and other emerging priorities into federal efforts to address invasive species; and strengthens coordinated, cost-efficient federal action.

One concern with the cultivation of certain transgenic crops is their potential dispersal, persistence, and spread into non-agricultural areas. Field corn (*Zea mays*) is a crop plant that was domesticated, bred for thousands of years, for large-scale food production. Domestication of *Zea mays* has rendered this cultivar less capable of survival in outside areas of cultivation, it is largely dependent on humans for persistence in the environment (OECD 2003). Due the size and weight of the seed, it is not easily dispersed by wind, water, or wildlife (Mallory-Smith and Zapiola 2008; Mallory-Smith and Sanchez Olguin 2011). Corn seed also lacks dormancy, which limits its ability to persist in soil seed bank. Because of these factors, field corn does not easily establish, naturalize, and spread.

APHIS evaluated the potential weediness and invasiveness of DP202216 corn and concluded that it is unlikely that DP202216 corn will become weedy or invasive in areas where it is grown (USDA-APHIS 2019a). As discussed in Subsections 3.3.4 and 4.3.3.4, Gene Flow and Weediness, there are a few populations of closely related species of *Tripsacum* within the U.S.; however, the potential for a weedy or invasive species of corn to develop as a result of outcrossing of DP202216 corn with other sexually compatible species of corn, or wild *Tripsacum* species, is considered remote. Hybridization and successful introgression of *Z. mays* genes into *Tripsacum* populations is rare (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995). Special techniques are required to hybridize *Z. mays* and *Tripsacum*; hybrids of *Tripsacum* species with *Zea* species do not commonly occur outside of a laboratory. Offspring are often sterile or have reduced fertility, and are unable to withstand even mild winter conditions (de Wet and Harlan 1972; de Wet et al. 1978; Eubanks 1995). There are no known wild hybrids comprised of *Zea mays* and *Tripsacum* species in the United States.

- **EO 13186 – Responsibilities of Federal Agencies to Protect Migratory Birds**

The United States has recognized the critical importance of migratory birds as a shared resource by ratifying international, bilateral conventions for the conservation of migratory birds. These conventions impose substantive obligations on the United States for the conservation of migratory birds and their habitats. Through the Migratory Bird Treaty Act (Act) the United States has

implemented these conventions with respect to the United States. This Executive Order directs executive departments and agencies to take certain actions to further implement the Act.

Migratory birds may transit corn fields and forage on corn, namely residual corn cobs/kernels left in the field post-harvest (Sherfy et al. 2011). For example, during migration, about 90% of the sandhill crane diet consists of corn, when corn is available (NGP 2019). As reviewed in this EA, it is highly unlikely the PAT and ZMM28 proteins, which naturally occur in soil bacteria and corn, respectively, present any risks to the health of migratory birds. Pioneer submitted a safety and nutritional assessment for DP202216 corn to the FDA's Center for Food Safety and Applied Nutrition in 2018 (Pioneer 2019c). The FDA response is pending.

The potential environmental impacts associated with glufosinate are summarized in the EPA's ecological risk assessment for the herbicide. The available data indicate that glufosinate (technical grade glufosinate-ammonium, the active ingredient) is practically non-toxic to birds on an acute oral and subacute dietary exposure basis and is practically non-toxic to mammals on an acute oral exposure basis (US-EPA 2014, 2016c). There is no data on formulations of glufosinate based herbicides. All glufosinate use would have to comply with EPA label use requirements, which are in part intended to be protective of wildlife.

Based on the Agency's assessment of DP202216 corn, it is unlikely that that approval of the petition, and subsequent production of DP202216 corn, would present any hazard to migratory bird populations. Rather, DP202216 corn would likely provide a food source for some species of migratory birds.

7.3 Executive Orders related to International Issues

- **EO 12114 - Environmental Effects Abroad of Major Federal Actions**

This EO requires federal officials to take into consideration any potential environmental effects that may occur outside the United States, its territories, and possessions, that may result from actions being taken.

As discussed in this EA, all crop production can potentially have adverse impacts on soils, and air and water quality. Any cultivation of DP202216 corn outside of the United States, its territories, or possessions would utilize the same (or similar) agronomic practices and inputs as those utilized in the United States. Consequently, the sources and degree of environmental impacts that derive from crop production abroad would be no different than those described for United States. In the event APHIS approves the petition for DP202216 corn, significant adverse environmental impacts outside the United States as a result of cultivation of this corn variety are unlikely.

The United States is a member of the World Trade Organization (WTO), which facilitates harmonizing the global rules of trade between nations. The Agreement on the Application of Sanitary and Phytosanitary Measures (the "SPS Agreement"), entered into force with the establishment of the WTO on January 1, 1995, sets out the basic rules for food safety and animal and plant health standards. The SPS agreement recognizes three international organizations/frameworks that have established standards and guidelines related to SPS measures (WTO 2019a), these are; the Codex Alimentarius Commission (Codex), the World Organization for Animal Health (OIE), and the International Plant Protection Convention (IPPC). Any international trade of DP202216 corn or products derived from it following a

determination of nonregulated status would be subject to national phytosanitary requirements and be in accordance with international SPS standards; Codex (food safety) international food standards, guidelines, and codes of practice that contribute to safety, quality, and fairness in the international trade of food; and the IPPC, the purpose of which is to protect the world's plant resources from the spread and introduction of pests, and promote safe trade.

7.4 State and Local Requirements

The PPA contains a preemption clause (7 U.S.C. § 7756) that prohibits state regulation of any, “plant, biological control organism, plant pest, noxious weed, or plant product” to protect against plant pests or noxious weeds if the Secretary (USDA) has issued regulations to prevent the dissemination of biological control organisms, plant pests, or noxious weeds within the United States. The PPA preemption clause does however allow states to impose additional prohibitions or restrictions based on special needs supported by sound scientific data or risk assessment. Consequently, while the PPA limits states' issuance of laws and regulations governing GE organisms and bars conflicting state regulation, it does allow state oversight when there is a special need for additional prohibitions or restrictions.

States use a variety of mechanisms to regulate the movement or release of GE organisms within their jurisdiction. For example, South Dakota simply authorizes holders of a federal permit issued under 7 CFR part 340 to use it within the state (SD Stat § 38-12A-31 (2015)). Minnesota issues state permits for release of GE organisms only after federal applications or permits are on file (MN Stat § 18F.07 (2015)). Nebraska may rely on APHIS or other experts before they issue their permit (NE Code § 2-10,113 (2015)). These illustrative examples show the range of state approaches to regulating the movement and release of GE organisms within state boundaries.

Neither of the alternatives considered would affect APHIS partnerships with states in the oversight of GE organisms, specifically in regulation of interstate movement and environmental releases. Under both alternatives, APHIS would continue working with states. The range of state legislation addressing agricultural biotechnology, namely in the way of permitting, crop protection, seed regulation, and economic development, would be unaffected by denial or approval of the petition.

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9 REFERENCES

- Alexander RB, Smith RA, Schwarz GE, Boyer EW, Nolan JV, and Brakebill JW. 2008. *Differences in Phosphorus and Nitrogen Delivery to The Gulf of Mexico from the Mississippi River Basin*. Environmental Science & Technology 42, pp. 822-830. Retrieved from <http://dx.doi.org/10.1021/es0716103>
- Altieri MA and Letourneau DK. 1982. *Vegetation management and biological control in agroecosystems*. Crop Protection 1, pp. 405-430. Retrieved from <http://www.sciencedirect.com/science/article/pii/0261219482900230>
- Anderson JA, Brustkern S, Cong B, Deege L, Delaney B, Hong B, Lawit S, Mathesius C, Schmidt J, Wu J, Zhang J, and Zimmermann C. 2019. *Evaluation of the history of safe use of the maize ZMM28 protein*. Journal of Agricultural and Food Chemistry 67, pp. 7466-7474. Retrieved from <https://doi.org/10.1021/acs.jafc.9b00391>
- Aneja VP, Schlesinger WH, and Erisman JW. 2009. *Effects of Agriculture upon the Air Quality and Climate: Research, Policy, and Regulations*. Environmental Science & Technology 43, pp. 4234-4240. Retrieved from <http://dx.doi.org/10.1021/es8024403>
- Atici C. 2014. *Low Levels of Genetically Modified Crops in International Food and Feed Trade: FAO International Survey and Economic Analysis*. FAO Commodity and Trade Policy, Research Working Paper No. 44. Food and Agriculture Organization of the United Nations. Retrieved from <http://www.fao.org/docrep/019/i3734e/i3734e.pdf>
- Avila-Garcia WV, Sanchez-Olguin E, Hulting AG, and Mallory-Smith C. 2012. *Target-site mutation associated with glufosinate resistance in Italian ryegrass (Lolium perenne L. ssp. multiflorum)*. Pest management science 68, pp. 1248-1254.
- Bartsch K and Tebbe CC. 1989. *Initial Steps in the Degradation of Phosphinothricin (Glufosinate) by Soil Bacteria*. Applied and Environmental Microbiology 55, pp. 711-716. Retrieved from <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC184185/>
- Baumhardt R, Stewart B, and Sainju U. 2015. *North American Soil Degradation: Processes, Practices, and Mitigating Strategies*. Sustainability 7, pp. 2936. Retrieved from <http://www.mdpi.com/2071-1050/7/3/2936>
- Beasley JC and Rhodes Jr. OE. 2008. *Relationship between raccoon abundance and crop damage*. Human-Wildlife Conflicts 2, pp. 248-259. Retrieved from <http://digitalcommons.unl.edu/cgi/viewcontent.cgi?article=1039&context=hwj>
- Becker A and Theissen G. 2003. *The major clades of MADS-box genes and their role in the development and evolution of flowering plants*. Molecular phylogenetics and evolution 29, pp. 464-489.
- Beckie HJ, Ashworth MB, and Flower KC. 2019. *Herbicide Resistance Management: Recent Developments and Trends*. Plants (Basel, Switzerland) 8, pp. 1-13.
- Best LB, Whitmore RC, and Booth GM. 1990. *Use of Cornfields by Birds during the Breeding Season: The Importance of Edge Habitat*. American Midland Naturalist 123, pp. 84-99. Retrieved from <http://www.jstor.org/stable/2425762>
- Boesch DF. 2019. *Barriers and Bridges in Abating Coastal Eutrophication*. Frontiers in Marine Science 6. Retrieved from <https://www.frontiersin.org/article/10.3389/fmars.2019.00123>
- Bontemps C, Toussaint M, Revol PV, Hotel L, Jeanbille M, Uroz S, Turpault MP, Blaudez D, and Leblond P. 2013. *Taxonomic and functional diversity of Streptomyces in a forest soil*. FEMS microbiology letters 342, pp. 157-167.

- Bricker SB, Longstaff B, Dennison W, Jones A, Boicourt K, Wicks C, and Woerner J. 2008. *Effects of nutrient enrichment in the nation's estuaries: A decade of change*. Harmful Algae 8, pp. 21-32. Retrieved from <http://www.sciencedirect.com/science/article/pii/S1568988308001182>
- Brittan K. 2006. *Methods to Enable the Coexistence of Diverse Corn Production Systems*. University of California, Agricultural Biotechnology in California Series, Publication 8192. Retrieved from <https://anrcatalog.ucanr.edu/pdf/8192.pdf>
- Burkholder J, Libra B, Weyer P, Heathcote S, Kolpin D, Thorne PS, and Wichman M. 2007. *Impacts of waste from concentrated animal feeding operations on water quality*. Environ Health Perspect 115, pp. 308-312. Retrieved from <https://www.ncbi.nlm.nih.gov/pubmed/17384784>
<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1817674/>
- Busse MD, Ratcliff AW, Shestak CJ, and Powers RF. 2001. *Glyphosate toxicity and the effects of long-term vegetation control on soil microbial communities*. Soil Biology and Biochemistry 33, pp. 1777-1789. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0038071701001031>
- Callens C, Tucker MR, Zhang D, and Wilson ZA. 2018. *Dissecting the role of MADS-box genes in monocot floral development and diversity*. Journal of experimental botany 69, pp. 2435-2459.
- Campbell J, Mallory-Smith C, Hulting AG, and Weber CE. 2015. *Herbicide-Resistant Weeds and Their Management: Herbicide-Resistance Basics*. Oregon State University Extension Service. Retrieved from <https://catalog.extension.oregonstate.edu/pnw437/html>
- Carpenter JE. 2011. *Impact of GM crops on biodiversity*. GM Crops 2, pp. 7-23. Retrieved from <http://www.tandfonline.com/doi/abs/10.4161/gmcr.2.1.15086> Last accessed 2015/06/12.
- CBD. 2019a. *The Cartagena Protocol on Biosafety*. Convention on Biological Diversity Retrieved from <https://bch.cbd.int/protocol>
- CBD. 2019b. *Agricultural Biodiversity*. Convention on Biological Diversity (CBD). Retrieved from <https://www.cbd.int/agro/>
- CENR. 2010. *Committee on Environment and Natural Resources: Scientific Assessment of Hypoxia in U.S. Coastal Waters*. Interagency Working Group on Harmful Algal Blooms, Hypoxia, and Human Health of the Joint Subcommittee on Ocean Science and Technology. Washington, DC. Retrieved from <https://obamawhitehouse.archives.gov/sites/default/files/microsites/ostp/hypoxia-report.pdf>
- Cheo DL, Titus SA, Byrd DRN, Hartley JL, Temple GF, and Brasch MA. 2004. *Concerted assembly and cloning of multiple DNA segments using in vitro site-specific recombination: functional analysis of multi-segment expression clones*. Genome Res 14, pp. 2111-2120. Retrieved from <https://www.ncbi.nlm.nih.gov/pubmed/15489333>
<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC528927/>
- Claassen R, Bowman M, McFadden J, Smith D, and Wallander S. 2018. *Tillage Intensity and Conservation Cropping in the United States, Economic Information Bulletin Number 197*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <https://www.ers.usda.gov/webdocs/publications/90201/eib-197.pdf?v=7027.1>
- CLI. 2019. *CropLife International BioTrade Status Database*. Retrieved from <http://www.biotradestatus.com/>
- CLI. 2020. *CropLife International*. Retrieved from <https://croplife.org/>

- CWS-USFWS. 2007. *International Recovery Plan for the Whooping Crane*. Retrieved from http://ecos.fws.gov/docs/recovery_plan/070604_v4.pdf Last accessed May 2, 2014.
- David MB, Drinkwater LE, and McIsaac GF. 2010. *Sources of nitrate yields in the Mississippi River Basin*. Journal of environmental quality 39, pp. 1657-1667.
- Dayan FE and Duke SO. 2014. *Natural compounds as next-generation herbicides*. Plant Physiol 166, pp. 1090-1105.
- Dayan FE, Cantrell CL, and Duke SO. 2009. *Natural products in crop protection*. Bioorganic & medicinal chemistry 17, pp. 4022-4034.
- De Bodt S, Raes J, Van de Peer Y, and Theissen G. 2003. *And then there were many: MADS goes genomic*. Trends Plant Sci 8, pp. 475-483. Retrieved from <https://www.sciencedirect.com/science/article/pii/S1360138503002206?via%3Dihub>
- de Wet MJM and Harlan JR. 1972. *Origin of Maize: The Tripartite Hypothesis*. Euphytica 21, pp. 271-279. Retrieved from <https://doi.org/10.1007/BF00036767>
- de Wet MJM, Harlan JR, Stalker HT, and Randrianasolo AV. 1978. *The Origin of Tripsacoid Maize (Zea mays L.)*. Evolution 32, pp. 233-244. Retrieved from <http://www.jstor.org/stable/2407592>
- Delaney B, Goodman RE, and Ladics GS. 2017. *Food and Feed Safety of Genetically Engineered Food Crops*. Toxicological Sciences 162, pp. 361-371. Retrieved from <https://doi.org/10.1093/toxsci/kfx249> Last accessed 2/12/2020.
- Dennis PG, Kukulies T, Forstner C, Orton TG, and Pattison AB. 2018. *The effects of glyphosate, glufosinate, paraquat and paraquat-diquat on soil microbial activity and bacterial, archaeal and nematode diversity*. Scientific reports 8, pp. 2119-2119. Retrieved from <https://www.ncbi.nlm.nih.gov/pubmed/29391493>
- Doebley JF. 1990. *Molecular systematics of Zea (Gramineae)*. Maydica 35 35, pp. 143-150. Retrieved from <https://www.cabdirect.org/cabdirect/abstract/19901614567>
- Doebley JF and Iltis HH. 1980. *Taxonomy of Zea (Gramineae). I. A subgeneric classification with key to taxa*. American Journal of Botany 67, pp. 982-993.
- Duke SO. 2012. *Why have no new herbicide modes of action appeared in recent years?* Pest management science 68, pp. 505-512.
- Duke SO. 2015. *Perspectives on transgenic, herbicide-resistant crops in the United States almost 20 years after introduction*. Pest management science 71, pp. 652-657. Retrieved from <https://onlinelibrary.wiley.com/doi/pdf/10.1002/ps.3863>
- Ellstrand NC. 2014. *Is gene flow the most important evolutionary force in plants?* American Journal of Botany 101, pp. 737-753. Retrieved from <https://onlinelibrary.wiley.com/doi/pdf/10.3732/ajb.1400024>
- Ellstrand NC, Garner LC, Hegde S, Guadagnuolo R, and Blancas L. 2007. *Spontaneous Hybridization between Maize and Teosinte*. Journal of Heredity 98, pp. 183-187. Retrieved from <http://jhered.oxfordjournals.org/content/98/2/183.abstract>
- ETIPCC. 2017. *National Strategy for Modernizing the Regulatory System for Biotechnology Products, Product of the Emerging Technologies Interagency Policy Coordination Committee's Biotechnology Working Group, September 2016*. White House Office of Science and Technology Policy (OSTP), National Science and Technology Council (NSTC), Emerging Technologies Interagency Policy Coordination Committee (ETIPCC). Retrieved from https://www.aphis.usda.gov/biotechnology/downloads/biotech_national_strategy_final.pdf

- Eubanks M. 1995. *A cross between two maize relatives: Tripsacum dactyloides and Zea diploperennis (Poaceae)*. Economic Botany 49, pp. 172-182. Retrieved from <https://doi.org/10.1007/BF02862921>
- EUginius. 2015. *European GMO Initiative for a Unified Database System (EUginius)*. Retrieved from <http://www.euginius.eu/euginius/pages/home.jsf?sessionExpired=true>
- FAO. 2009. *Codex Alimentarius, Foods Derived from Modern Biotechnology, 2nd Edition*. World Health Organization, Food and Agriculture Organization of the United Nations. Retrieved from <http://www.fao.org/3/a-a1554e.pdf>
- FAO. 2014. *Technical Consultation on Low Levels of Genetically Modified (GM) Crops in International Food and Feed Trade. Technical Background Paper 2 [TC-LLP/2014/3]. Low levels of GM crops in international food and feed trade: FAO international survey and economic analysis*. Food and Agriculture Organization of the United Nations. Retrieved from http://www.fao.org/fileadmin/user_upload/agns/topics/LLP/AGD803_3_Final_En.pdf
- FAO. 2017. *Global assessment of the impact of plant protection products on soil functions and soil ecosystems*. Food and Agriculture Organization of the United Nations (FAO), Intergovernmental Technical Panel on Soils of the Global Soil Partnership. Retrieved from <http://www.fao.org/3/I8168EN/i8168en.pdf>
- Felber F, Kozłowski G, Arrigo N, and Guadagnuolo R. 2007. Genetic and Ecological Consequences of Transgene Flow to the Wild Flora. In: *Green Gene Technology* (Springer Berlin Heidelberg), pp. 173-205. Retrieved from http://dx.doi.org/10.1007/10_2007_050
- Fernandez-Cornejo J and Osteen C. 2015. *Managing Glyphosate Resistance May Sustain Its Efficacy and Increase Long-Term Returns to Corn and Soybean Production*. Amber Waves: U.S. Department of Agriculture, Economic Research Service. Retrieved from <https://www.ers.usda.gov/amber-waves/2015/may/managing-glyphosate-resistance-may-sustain-its-efficacy-and-increase-long-term-returns-to-corn-and-soybean-production/>
- Fernandez-Cornejo J, Wechsler S, Livingston M, and Mitchell L. 2014a. *Genetically Engineered Crops in the United States [Economic Research Report Number 162]*. U.S. Department of Agriculture, Economic Research Service. Retrieved from https://www.ers.usda.gov/webdocs/publications/45179/43668_err162.pdf
- Fernandez-Cornejo J, Hallahan C, Nehring R, Wechsler S, and Grube A. 2012. *Conservation Tillage, Herbicide Use, and Genetically Engineered Crops in the United States: The Case of Soybeans*. AgBioForum 15, pp. 231-241. Retrieved from <http://www.agbioforum.org/v15n3/v15n3a01-fernandez-cornejo.htm>
- Fernandez-Cornejo J, Nehring R, Osteen C, Wechsler S, Martin A, and Vialou A. 2014b. *Pesticide Use in U.S. Agriculture: 21 Selected Crops, 1960-2008 [EIB-124]*. U.S. Department of Agriculture, Economic Research Service. Retrieved from https://www.ers.usda.gov/webdocs/publications/43854/46734_eib124.pdf
- Fleharty ED and Navo KW. 1983. *Irrigated Cornfields as Habitat for Small Mammals in the Sand Sage Prairie Region of Western Kansas*. Journal of Mammalogy 64, pp. 367-379. Retrieved from <http://www.jstor.org/stable/1380349>
- FOEU. 2014. *GM food and the EU-US trade deal, September 2014. Friends of the Earth Europe (FOEU)*. Retrieved from http://www.foeeurope.org/sites/default/files/gm_food_eu-us_trade_deal.pdf
- Fortuna A. 2012. *The Soil Biota*. Nature Education Knowledge 3, pp. 1. Retrieved from <https://www.nature.com/scitable/knowledge/library/the-soil-biota-84078125/>

- Frisvold G. 2015. *Genetically Modified Crops: International Trade and Trade Policy Effect*. International Journal of Food and Agricultural Economics 3, pp. 1-13. Retrieved from <http://www.foodandagriculturejournal.com/vol3.no2.pp1.pdf>
- Garbeva P, van Veen JA, and van Elsas JD. 2004. *Microbial diversity in soil: selection of microbial populations by plant and soil type and implications for disease suppressiveness*. Annual review of phytopathology 42, pp. 243-270.
- Garrison AJ, Miller AD, Ryan MR, Roxburgh SH, and Katriona S. 2014. *Stacked Crop Rotations Exploit Weed-Weed Competition for Sustainable Weed Management*. Weed Science 62, pp. 166-176. Retrieved from <http://dx.doi.org/10.1614/WS-D-13-00037.1> Last accessed 2015/05/19.
- Givens WA, Shaw DR, Kruger GR, Johnson WG, Weller SC, Young BG, Wilson RG, Owen MDK, and Jordan D. 2009. *Survey of Tillage Trends Following The Adoption of Glyphosate-Resistant Crops*. Weed Technology 23, pp. 150-155. Retrieved from http://www.gri.msstate.edu/publications/docs/2009/03/6134givens_2009_tillage_trends.pdf
- Goggi AS, Caragea P, Lopez-Sanchez H, Westgate M, Artritt R, and Clark C. 2006. *Statistical analysis of outcrossing between adjacent maize grain production fields*. Field Crops Research 99, pp. 147-157. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0378429006000918>
- Goldstein DA. 2014. *Tempest in a Tea Pot: How did the Public Conversation on Genetically Modified Crops Drift so far from the Facts?* Journal of Medical Toxicology 10, pp. 194-201. Retrieved from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4057531/pdf/13181_2014_Article_402.pdf
- Gómez-Laurito J. 2013. *A new species of Zea (Poaceae) from the Murciélago Islands, Santa Elena Peninsula, Guanacaste, Costa Rica*. Brenesia, pp. 36-39.
- Grassini P, Thorburn J, Burr C, and Cassman KG. 2011. *High-yield irrigated maize in the Western U.S. Corn Belt: I. On-farm yield, yield potential, and impact of agronomic practices*. Field Crops Research 120, pp. 142-150. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0378429010002522>
- Green JM. 2014. *Current state of herbicides in herbicide-resistant crops*. Pest management science 70, pp. 1351-1357.
- Green JM. 2018. *The rise and future of glyphosate and glyphosate-resistant crops*. Pest management science 74, pp. 1035-1039.
- GROi. 2018. *A Look at Fertilizer and Pesticide Use in the U.S., 11 June 2018*. Gro Intelligence. Retrieved from <https://gro-intelligence.com/insights/articles/a-look-at-fertilizer-and-pesticide-use-in-the-us>
- Gupta VVSR, Neate SM, and Leonard E. 2007. *Life in the Soil - The Relationship Between Agriculture and Soil Organisms*. Cooperative Research Centre for Soil & Land Management. Retrieved from https://www.researchgate.net/publication/268800863_Life_in_the_soil_the_relationship_between_agriculture_and_soil_organisms
- Gyamfi S, Pfeifer U, Stierschneider M, and Sessitsch A. 2002. *Effects of transgenic glufosinate-tolerant oilseed rape (Brassica napus) and the associated herbicide application on eubacterial and Pseudomonas communities in the rhizosphere*. FEMS Microbiology Ecology 41, pp. 181-190. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0168649602002908>
- Halsey ME, Remund KM, Davis CA, Qualls M, Eppard PJ, and Berberich SA. 2005. *Isolation of Maize from Pollen-Mediated Gene Flow by Time and Distance*. Crop Science 45, pp. 2172-2185. Retrieved from <http://dx.doi.org/10.2135/cropsci2003.0664>

- Hannula SE, de Boer W, and van Veen JA. 2014. *Do genetic modifications in crops affect soil fungi? a review*. Biol Fertil Soils 50, pp. 433-446. Retrieved from <https://www.scopus.com/inward/record.uri?eid=2-s2.0-84897573198&doi=10.1007%2fs00374-014-0895-x&partnerID=40&md5=950c304533b183d68eda53f2482b052f>
- Heap I. 2020. *The International Survey of Herbicide Resistant Weeds*. Retrieved from www.weedscience.org
- Heap I and Duke SO. 2018. *Overview of glyphosate-resistant weeds worldwide*. Pest Manag Sci. 74, pp. 1040-1049.
- Herouet C, Esdaile DJ, Mallyon BA, Debruyne E, Schulz A, Currier T, Hendrickx K, van der Klis RJ, and Rouan D. 2005. *Safety evaluation of the phosphinothricin acetyltransferase proteins encoded by the pat and bar sequences that confer tolerance to glufosinate-ammonium herbicide in transgenic plants*. Regulatory toxicology and pharmacology : RTP 41, pp. 134-149.
- Hitchcock AS. 1951. *Manual of the Grasses of the United States, 2nd ed.* USDA Miscellaneous Publication No. 200. Retrieved from https://ia801709.us.archive.org/13/items/manualofgrasses0200hitc_0/manualofgrasses0200hitc_0.pdf
- Holt JS, Welles SR, Silvera K, Heap IM, Heredia SM, Martinez-Berdeja A, Palenscar KT, Sweet LC, and Ellstrand NC. 2013. *Taxonomic and life history bias in herbicide resistant weeds: implications for deployment of resistant crops*. PLoS One 8, pp. e71916.
- HRAC. 2020. *Herbicide Resistance Action Committee*. Retrieved from <https://www.hracglobal.com/>
- Hsiao C-L, Young C-C, and Wang C-Y. 2017. *Screening and Identification of Glufosinate-Degrading Bacteria from Glufosinate-Treated Soils*. Weed Science 55, pp. 631-637. Retrieved from <https://www.cambridge.org/core/article/screening-and-identification-of-glufosinatedegrading-bacteria-from-glufosinatetreated-soils/102B14AE445C6C1A578B5B55558CB673>
- ILSI-CERA. 2011. *A review of the environmental safety of the PAT protein*. International Life Sciences Institute, Center for Environmental Risk Assessment, Washington, D.C. . Retrieved from <https://ilsirf.org/publication/a-review-of-the-environmental-safety-of-the-pat-protein/>
- ILSI-CERA. 2020. *ILSI Crop Composition Database*. International Life Sciences Institute, Center for Environmental Risk Assessment, Washington, D.C. . Retrieved from https://www.cropcomposition.org/query/workflow.wiz?_flowExecutionKey=c7A6FFE29-565D-09A9-B48B-987C39EEAB6C_ka116D734-8713-18AF-7C83-D59401E68E82
- Iltis HH and Doebley JF. 1980. *Taxonomy of Zea (Gramineae). II. Subspecific categories in the Zea mays complex and a generic synopsis*. American Journal of Botany 67, pp. 994-1004.
- Iltis HH and Benz BF. 2000. *Zea nicaraguensis (Poaceae), a new teosinte from Pacific coastal Nicaragua*. Novon, pp. 382-390.
- Iqbal MZ, Cheng M, Su Y, Li Y, Jiang W, Li H, Zhao Y, Wen X, Zhang L, Ali A, Rong T, and Tang Q. 2019. *Allopolyploidization facilitates gene flow and speciation among corn, Zea perennis and Tripsacum dactyloides*. Planta 249, pp. 1949-1962. Retrieved from <https://doi.org/10.1007/s00425-019-03136-z>
- ISU-Ext. 2003. *Weeds to Watch: New Weed Threats for Corn and Soybean Fields*. Ed. University of Illinois Extension, Michigan State University Extension, University of Minnesota Extension Service, Purdue University Cooperative Extension, University of Wisconsin Cooperative Extension. Retrieved from <https://store.extension.iastate.edu/Product/Weeds-to-Watch-New-Weed-Threats-for-Corn-and-Soybean-Fields>

- Jeschke MJ and Doerge T. 2010. *Managing Volunteer Corn in Corn Fields*. Crop Insights 18, pp. 4. Retrieved from http://s3.amazonaws.com/zanran_storage/www.mccormickcompany.net/ContentPages/44064101.pdf
- Jhala A and Rees J. 2018. *Control of Volunteer Corn in Soybean and Corn*. CropWatch, University of Nebraska, Lincoln. Retrieved from <https://cropwatch.unl.edu/2018/control-volunteer-corn-soybean-and-corn>
- Jhala A, Wright B, and Chahal P. 2019. *Weed Science: Volunteer Corn in Soybeans*. University of Nebraska-Lincoln Extension. Retrieved from <https://cropwatch.unl.edu/volunteer-corn-soybean-impact-and-management>
- Jhala A, Knezevic S, Ganie Z, and Singh M. 2014. Integrated Weed Management in Maize. In: *Recent Advances in Weed Management*, pp. 177-196. Retrieved from <https://agronomy.unl.edu/documents/Integrated%20Weed%20Mana.%20in%20Corn.pdf>
- Jorgensen J and Dinan L. 2016. *Whooping Crane (Grus americana) behavior, habitat use and wildlife watching visitation during migratory stopover at two Wildlife Management Areas in Nebraska 2015-2016*. Nebraska Game and Parks Commission. Retrieved from <https://digitalcommons.unl.edu/cgi/viewcontent.cgi?article=1086&context=nebgamestaff>
- Jugulam M and Shyam C. 2019. *Non-Target-Site Resistance to Herbicides: Recent Developments*. Plants (Basel, Switzerland) 8, pp. 417. Retrieved from <https://www.ncbi.nlm.nih.gov/pubmed/31618956>
<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6843234/>
- Karn E, Beffa R, and Jasieniuk M. 2018. *Variation in Response and Resistance to Glyphosate and Glufosinate in California Populations of Italian Ryegrass (<i>Lolium perenne</i> ssp. <i>multiflorum</i>)*. Weed Science 66, pp. 168-179, 112. Retrieved from <https://doi.org/10.1017/wsc.2017.71>
- Kniss AR. 2018. *Genetically Engineered Herbicide-Resistant Crops and Herbicide-Resistant Weed Evolution in the United States*. Weed Science 66, pp. 260-273. Retrieved from <https://www.cambridge.org/core/article/genetically-engineered-herbicides-resistant-crops-and-herbicides-resistant-weed-evolution-in-the-united-states/22B3B07F8EB980D2CFEEE3AA36B7B2C1>
- Korres N, Burgos N, and Duke S. 2019. *Weed Control: Sustainability, Hazards and Risks in Cropping Systems Worldwide*. CRC Press. Retrieved from <file:///C:/Users/aprchardman/Downloads/WEEDCONTROL.pdf>
- Kortekamp A. 2011. *Unexpected Side Effects of Herbicides: Modulation of Plant-Pathogen Interactions*. DLR Agricultural Research Station, Department of Phytomedicine, Neustadt/Weinstrasse, Germany. InTech. Retrieved from <http://www.intechopen.com/books/herbicides-and-environment/unexpected-side-effects-of-herbicides-modulation-of-plant-pathogen-interactions>
- Kowalchuk GA, Bruinsma M, and van Veen JA. 2003. *Assessing responses of soil microorganisms to GM plants*. Trends in Ecology & Evolution 18, pp. 403-410. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0169534703001873>
- Kral R, Diamond Jr AR, Ginzburg SL, Hansen CJ, Haynes RR, Keener BR, Lelong MG, Spaulding DD, and Woods M. 2019. *Alabama Plant Atlas*. Retrieved from <http://www.floraofalabama.org/Plant.aspx?id=5097>
- Krapu GL, Brandt DA, and Cox Jr. RR. 2004. *Less Waste Corn, More Land in Soybeans, and the Switch to Genetically Modified Crops: Trends with Important Implications for Wildlife Management*. Wildlife Society Bulletin, 2004 32, pp. 127 - 136.

- Kremer RJ and Means NE. 2009. *Glyphosate and glyphosate-resistant crop interactions with rhizosphere microorganisms*. European Journal of Agronomy 31, pp. 153-161. Retrieved from <http://www.sciencedirect.com/science/article/pii/S1161030109000641>
- Kwit C, Moon HS, Warwick SI, and Stewart Jr CN. 2011. *Transgene introgression in crop relatives: molecular evidence and mitigation strategies*. Trends in Biotechnology 29, pp. 284-293. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0167779911000333>
- Labeda DP, Goodfellow M, Brown R, Ward AC, Lanoot B, Vannanneyt M, Swings J, Kim SB, Liu Z, Chun J, Tamura T, Oguchi A, Kikuchi T, Kikuchi H, Nishii T, Tsuji K, Yamaguchi Y, Tase A, Takahashi M, Sakane T, Suzuki KI, and Hatano K. 2012. *Phylogenetic study of the species within the family Streptomycetaceae*. Antonie van Leeuwenhoek 101, pp. 73-104.
- Landis DA, Menalled FD, Costamagna AC, and Wilkinson TK. 2005. *Manipulating plant resources to enhance beneficial arthropods in agricultural landscapes*. Weed Science 53, pp. 902-908. Retrieved from <http://dx.doi.org/10.1614/WS-04-050R1.1> Last accessed 2015/06/26.
- Leblanc O, Grimanelli D, Faridi N, Berthaud J, and Savidan Y. 1996. *Reproductive Behavior in Maize-Tripsacum Polyhaploid Plants: Implications for the Transfer of Apomixis Into Maize*. Journal of Heredity 87. Retrieved from https://www.researchgate.net/publication/31070697_Reproductive_Behavior_in_Maize-Tripsacum_Polyhaploid_Plants_Implications_for_the_Transfer_of_Apomixis_Into_Maize
- Lee MS, Anderson EK, Stojisin D, McPherson MA, Baltazar B, Horak MJ, de la Fuente JM, Wu K, Crowley JH, Rayburn AL, and Lee DK. 2017. *Assessment of the potential for gene flow from transgenic maize (Zea mays L.) to eastern gamagrass (Tripsacum dactyloides L.)*. Transgenic research 26, pp. 501-514.
- Liu Z, Zhao X, and Bai F. 2013. *Production of xylanase by an alkaline-tolerant marine-derived Streptomyces viridochromogenes strain and improvement by ribosome engineering*. Applied Microbiology and Biotechnology 97, pp. 4361-4368. Retrieved from <https://doi.org/10.1007/s00253-012-4290-y>
- Livingston M, Fernandez-Cornejo J, Unger J, Osteen C, Schimmelpfennig D, Park T, and Lambert D. 2015. *The Economics of Glyphosate Resistance Management in Corn and Soybean Production [Economic Research Report Number 184]*. U.S. Department of Agriculture, Economic Research Service. Retrieved from https://www.ers.usda.gov/webdocs/publications/45354/52761_err184.pdf?v=42207
- Locke MA and Zablotowicz RM. 2004. Chapter 14: Pesticides in Soil - Benefits and Limitations to Soil Health. In: *Managing Soil Quality: Challenges in Modern Agriculture* (U.S. Department of Agriculture, Agricultural Research Service. Southern Weed Science Research Unit). Retrieved from <http://www.cabi.org/cabebooks/ebook/20033208669>
- Locke MA, Zablotowicz RM, and Reddy KN. 2008. *Integrating soil conservation practices and glyphosate-resistant crops: impacts on soil*. Pest management science 64, pp. 457-469.
- Lundgren JG and Fergen JK. 2014. *Predator community structure and trophic linkage strength to a focal prey*. Molecular Ecology 23, pp. 3790-3798. Retrieved from <https://onlinelibrary.wiley.com/doi/abs/10.1111/mec.12700>
- Lupwayi NZ, Harker KN, Clayton GW, Turkington TK, Rice WA, and O'Donovan JT. 2004. *Soil microbial biomass and diversity after herbicide application*. Canadian Journal of Plant Science 84, pp. 677-685. Retrieved from <http://dx.doi.org/10.4141/P03-121> Last accessed 2015/06/19.
- MacGowan B, Humberg LA, Beasley JC, DeVault TL, Retamosa MI, and Rhodes OEJ. 2006. *Corn and Soybean Crop Depredation by Wildlife*. Department of Forestry and Natural Resources, Purdue

- University (FNR-265). Retrieved from <https://www.extension.purdue.edu/extmedia/FNR/FNR-265-W.pdf>
- Magleby R, Sandretto C, Crosswhite W, and Osborn CT. 1995. *Soil Erosion and Conservation in the United States. Agriculture Information Bulletin Number 718*. Retrieved from <http://naldc.nal.usda.gov/download/CAT10712833/PDF>
- Mallory-Smith C and Zapiola M. 2008. *Gene flow from glyphosate-resistant crops*. Pest management science 64, pp. 428-440. Retrieved from <https://onlinelibrary.wiley.com/doi/full/10.1002/ps.1517>
- Mallory-Smith CA and Sanchez Olguin E. 2011. *Gene Flow from Herbicide-Resistant Crops: It's Not Just for Transgenes*. Journal of Agricultural and Food Chemistry 59, pp. 5813-5818. Retrieved from <http://dx.doi.org/10.1021/jf103389v>
- Mangelsdorf PC and Reeves RG. 1959. *The Origin of Corn: I. Pod Corn, the Ancestral Form*. Botanical Museum Leaflets, Harvard University 18, pp. 329-356. Retrieved from <http://www.jstor.org/stable/41762197>
- Marin-Morales M, Ventura-Camargo B, and Hoshina M. 2013. Toxicity of Herbicides: Impact on Aquatic and Soil Biota and Human Health. In: *Herbicides: Current Research and Case Studies in Use*, pp. 399-443. Retrieved from <https://www.intechopen.com/books/herbicides-current-research-and-case-studies-in-use/toxicity-of-herbicides-impact-on-aquatic-and-soil-biota-and-human-health>
- Marquardt P, Krupke C, and Johnson WG. 2012. *Competition of Transgenic Volunteer Corn with Soybean and the Effect on Western Corn Rootworm Emergence*. Weed Science 60, pp. 193-198. Retrieved from <http://dx.doi.org/10.1614/WS-D-11-00133.1> Last accessed 2015/06/29.
- Morrison L. 2014. *Should you use tillage to control resistant weeds? Herbicide-resistant weeds: The tillage dilemma*. FarmProgress. Retrieved from <https://www.farmprogress.com/tillage/should-you-use-tillage-control-resistant-weeds>
- Mortensen DA, Egan JF, Maxwell BD, Ryan MR, and Smith RG. 2012. *Navigating a Critical Juncture for Sustainable Weed Management*. BioScience 62, pp. 75-84. Retrieved from <http://bioscience.oxfordjournals.org/content/62/1/75.abstract>
- Motavalli PP, Kremer RJ, Fang M, and Means NE. 2004. *Impact of genetically modified crops and their management on soil microbially mediated plant nutrient transformations*. Journal of environmental quality 33, pp. 816-824.
- MPAWG. 2016. *Maryland Plant Atlas Work Group, Digital Atlas of the Maryland Flora*. Retrieved from <https://www.marylandplantatlas.org/viewChecklist.php?genus=Zea>
- NAS. 2016. *Genetically Engineered Crops: Experiences and Prospects*. Washington, DC: National Academies Press. Retrieved from <http://www.nap.edu/catalog/23395/genetically-engineered-crops-experiences-and-prospects>
- NCGA. 2019. *World of Corn*. The National Corn Growers Association. Retrieved from <http://www.worldofcorn.com/#corn-usage-by-segment>
- NGP. 2019. *Sandhill Cranes*. Nebraska Game and Parks Commission. Retrieved from <http://outdoornebraska.gov/sandhillcrane/>
- Nguyen DB, Rose MT, Rose TJ, and van Zwieten L. 2018. *Effect of glyphosate and a commercial formulation on soil functionality assessed by substrate induced respiration and enzyme activity*. European Journal of Soil Biology 85, pp. 64-72. Retrieved from <https://www.scopus.com/inward/record.uri?eid=2-s2.0-85041592081&doi=10.1016%2fj.ejsobi.2018.01.004&partnerID=40&md5=c860625345da6557c494dbfd6d6d2c7>

- Nichols CI and Altieri MA. 2012. *Plant biodiversity enhances bees and other insect pollinators in agroecosystems. A review*. Agronomic Sustainable Development 33, pp. 257-274. Retrieved from <https://link.springer.com/article/10.1007/s13593-012-0092-y> Last accessed 09/14/2016.
- Nicolai D, Stahl L, and Gunsolus J. 2018. *Managing the potential for volunteer corn in 2019*. Univ. of Minnesota Extension Retrieved from <https://blog-crop-news.extension.umn.edu/2018/10/managing-potential-for-volunteer-corn.html>
- Nielsen RL. 2016. *Tassel Emergence & Pollen Shed*. Purdue University Corny News Network Website. Retrieved from <https://www.agry.purdue.edu/ext/corn/news/timeless/Tassels.html>
- Nielsen UN, Ayres E, Wall DH, and Bardgett RD. 2011. *Soil biodiversity and carbon cycling: A review and synthesis of studies examining diversity-function relationships*. European Journal of Soil Science 62, pp. 105-116. Retrieved from <https://www.scopus.com/inward/record.uri?eid=2-s2.0-78751503817&doi=10.1111%2fj.1365-2389.2010.01314.x&partnerID=40&md5=e6b544ad01352d9e1d0e3e483f0f04fe>
- NOAA. 2019. *Dealing with Dead Zones: Hypoxia in the Ocean*. U.S. Department of Commerce, National Oceanic and Atmospheric Administration. Retrieved from <https://oceanservice.noaa.gov/podcast/feb18/nop13-hypoxia.html>
- Norsworthy JK, Ward SM, Shaw DR, Llewellyn RS, Nichols RL, Webster TM, Bradley KW, Frisvold G, Powles SB, Burgos NR, Witt WW, and Barrett M. 2012. *Reducing the Risks of Herbicide Resistance: Best Management Practices and Recommendations*. Weed Science 60, pp. 31-62. Retrieved from <http://dx.doi.org/10.1614/WS-D-11-00155.1>
- NRC. 2004. *Safety of Genetically Engineered Foods: Approaches to Assessing Unintended Health Effects. Chapter 2, Methods and Mechanisms for Genetic Manipulation of Plants, Animals, and Microorganisms*. National Research Council (US) Committee on Identifying and Assessing Unintended Effects of Genetically Engineered Foods on Human Health: National Research Council (US) Committee on Identifying and Assessing Unintended Effects of Genetically Engineered Foods on Human Health. Retrieved from <https://www.ncbi.nlm.nih.gov/books/NBK215771/>
- NRC. 2010. *National Research Council (NRC). The Impact of Genetically Engineered Crops on Farm Sustainability in the United States*. National Academies Press, Washington, DC. Retrieved from <http://www.nap.edu/catalog/12804/impact-of-genetically-engineered-crops-on-farm-sustainability-in-the-united-states>
- ODNR. 2001. *Wildlife Crop Damage Manual*. Ohio Department of Natural Resource, Division of Wildlife. Retrieved from <http://wildlife.ohiodnr.gov/portals/wildlife/pdfs/publications/wildlife%20management/Crop%20Damage%20Manual.pdf>
- OECD. 2003. *Consensus Document on the Biology of Zea mays subsp. mays (Maize) [ENV/JM/MONO(2003)11]. Series on Harmonisation of Regulatory Oversight in Biotechnology No. 27*. Organization for Economic Cooperation and Development. Retrieved from <http://www.oecd.org/env/ehs/biotrack/46815758.pdf>
- OECD. 2015. *Biotechnology Policies*. Organization for Economic Co-operation and Development (OECD). Retrieved from <http://www.oecd.org/sti/emerging-tech/>
- OSU. 2019. *Tillage Intensity to Maintain Target Residue Cover (NRCS 329, 345 & 346)*. AgBMPs: Ohio State University Extension. Retrieved from <https://agbmps.osu.edu/bmp/tillage-intensity-maintain-target-residue-cover-nrcs-329-345-346>

- Owen MDK. 2011. *Weed resistance development and management in herbicide-tolerant crops: Experiences from the USA* Journal of Consumer Protection and Food Safety 6, pp. 85-89. Retrieved from <http://link.springer.com/article/10.1007/s00003-011-0679-2>
- Owen MDK. 2012. *2013 Herbicide Guide for Iowa Corn and Soybean Production: Weed management update for 2013 (WC-94)*. Iowa State University Extension and Outreach. Retrieved from <http://www.weeds.iastate.edu/reference/WC94%202013.pdf>
- Owen MDK. 2016. *Diverse Approaches to Herbicide-Resistant Weed Management*. Weed Science 64, pp. 570-584. Retrieved from <https://www.cambridge.org/core/journals/weed-science/article/diverse-approaches-to-herbicideresistant-weed-management/C4771C62E6DBE92A834C33693BBE3B85>
- Parikh SJ and James BR. 2012. *Soil: The Foundation of Agriculture*. Nature Education Knowledge 3(10):2. Retrieved from <http://www.nature.com/scitable/knowledge/library/soil-the-foundation-of-agriculture-84224268>
- Pioneer Hi-Bred International, Inc. 2019a. *Petition for Determination of Nonregulated Status for Enhanced Grain Yield Potential and Glufosinate-ammonium Resistant DP202216 Maize*. Submitted by S.A. Catron, Registration Manager. Pioneer Hi-Bred International, Inc. Johnston, IA.
- Pioneer. 2019b. *Stewardship*. Pioneer Retrieved from <https://www.pioneer.com/home/site/us/products/stewardship/>
- Pioneer. 2019c. *Pioneer Hi-Bred International, Inc. Petition (19-337-01p) for the Determination of Nonregulated Status for Insect Resistant and Herbicide-Tolerant DP23211 Maize [OECD Unique Identifier: DP-Ø23211-2]*. DuPont-Pioneer Retrieved from <https://www.aphis.usda.gov/aphis/ourfocus/biotechnology/permits-notifications-petitions/petitions/petition-status>
- Piperno DR and Flannery KV. 2001. *The earliest archaeological maize (Zea mays L.) from highland Mexico: New accelerator mass spectrometry dates and their implications*. Proceedings of the National Academy of Sciences 98, pp. 2101-2103. Retrieved from <https://www.pnas.org/content/pnas/98/4/2101.full.pdf>
- Pleasant JM, Hellmich RL, Dively GP, Sears MK, Stanley-Horn DE, Mattila HR, Foster JE, Clark P, and Jones GD. 2001. *Corn pollen deposition on milkweeds in and near cornfields*. Proceedings of the National Academy of Sciences 98, pp. 11919-11924. Retrieved from <https://www.pnas.org/content/pnas/98/21/11919.full.pdf>
- Pline WA. 1999. *Effect of Temperature and Chemical Additives on the Efficacy of the Herbicides Glufosinate and Glyphosate in Weed Management of Liberty-Link and Roundup-Ready Soybeans* Virginia Tech. Dissertations and Theses. Retrieved from <https://vtechworks.lib.vt.edu/handle/10919/31699>
- PRX. 2019. *PRX: Grain Market Overview, U.S. Major Grains Crop Years 2018/19 & 2019/20 with USDA Oct 10, 2019 WASDE*. Proexporter Network. Retrieved from https://www.proexporter.com/clientfiles/assets/files/PRX_Overview.pdf
- Rey-Caballero J, Menéndez J, Osuna MD, Salas M, and Torra J. 2017. *Target-site and non-target-site resistance mechanisms to ALS inhibiting herbicides in Papaver rhoeas*. Pesticide Biochemistry and Physiology 138, pp. 57-65. Retrieved from <http://www.sciencedirect.com/science/article/pii/S0048357517300822>
- RFA. 2019. *Why is Ethanol Important? U.S. Ethanol Industry Has Grown by Leaps and Bounds*. Renewable Fuels Association Retrieved from <https://ethanolrfa.org/consumers/why-is-ethanol-important/>

- Ribaudo M, Delgado J, Hansen L, Livingston M, Mosheim R, and Williamson J. 2011. *Nitrogen in Agricultural Systems: Implications for Conservation Policy* [Economic Research Report Number 127]. United States Department of Agriculture, Economic Research Service Retrieved from <http://www.ers.usda.gov/media/117596/err127.pdf>
- Roché C, Vorobik L, Miller AD, Gunn B, Roberts C, and Blankenship M. 2007. *Manual of Grasses for North America*. University Press of Colorado. Retrieved from <http://www.jstor.org/stable/j.ctt4cgkq1>
- Rose MT, Cavagnaro TR, Scanlan CA, Rose TJ, Vancov T, Kimber S, Kennedy IR, Kookana RS, and Van Zwieten L. 2016. *Impact of Herbicides on Soil Biology and Function*. Retrieved from <https://www.scopus.com/inward/record.uri?eid=2-s2.0-84954286750&doi=10.1016%2fbs.agron.2015.11.005&partnerID=40&md5=09e83d5ebf93121a2d1be8e7eb5cc300>
- Roth G. 2015. *Crop Rotations and Conservation Tillage* [Publication Code: UC124]. Penn State Extension. Retrieved from <http://extension.psu.edu/plants/crops/soil-management/conservation-tillage/crop-rotations-and-conservation-tillage>
- Ruiz N, Lavelle P, and Jimenez J. 2008. *Soil Macrofauna Field Manual: Technical Level*. Food and Agriculture Organization of the United Nations. Retrieved from <http://www.fao.org/docrep/011/i0211e/i0211e00.htm>
- Sánchez G J, De La Cruz L L, Vidal M V, Ron P J, Taba S, Santacruz-Ruvalcaba F, Sood S, Holland J, Ruiz C J, and Carvajal S. 2011. *Three new teosintes (Zea spp., Poaceae) from México*. American journal of botany 98, pp. 1537-1548.
- Sánchez González JdJ, Ruiz Corral JA, García GM, Ojeda GR, Larios LDIC, Holland JB, Medrano RM, and García Romero GE. 2018a. *Ecogeography of teosinte*. PLOS ONE 13, pp. e0192676. Retrieved from <https://doi.org/10.1371/journal.pone.0192676>
- Sánchez González JdJ, Ruiz Corral JA, García GM, Ojeda GR, Larios LDIC, Holland JB, Medrano RM, and García Romero GE. 2018b. *Ecogeography of teosinte*. PloS one 13, pp. e0192676-e0192676. Retrieved from <https://www.ncbi.nlm.nih.gov/pubmed/29451888>
<https://www.ncbi.nlm.nih.gov/pmc/PMC5815594/>
- SARE/CTIC. 2017. *Annual Report 2016-2017: Cover Crop Survey, September 2017*. Sustainable Agriculture Research and Education (SARE) program and the Conservation Technology Information Center (CTIC). Retrieved from https://www.ctic.org/files/2017CTIC_CoverCropReport-FINAL.pdf
- Scherr SJ and McNeely JA. 2008. *Biodiversity conservation and agricultural sustainability: towards a new paradigm of 'ecoagriculture' landscapes*. Phil. Trans. R. Soc. B 363, pp. 477-494. Retrieved from <http://rstb.royalsocietypublishing.org/royptb/363/1491/477.full.pdf>
- Schnepf R. 2011. *U.S. Livestock and Poultry Feed Use and Availability: Background and Emerging Issues* Congressional Research Service, Report for Congress. Retrieved from <https://nationalaglawcenter.org/wp-content/uploads/assets/crs/R41956.pdf>
- Sessitsch A, Gyamfi S, Tschierko D, Gerzabek M, and Kandeler E. 2005. *Activity of microorganisms in the rhizosphere of herbicide treated and untreated transgenic glufosinate-tolerant and wildtype oilseed rape grown in containment*. Plant Soil 266, pp. 105-116. Retrieved from <http://dx.doi.org/10.1007/s11104-005-7077-4>
- Sfiligoi E. 2014. *The Weed Resistance Problem: A Matter Of Billions*. CropLife. Retrieved from <https://www.croplife.com/crop-inputs/herbicides/the-weed-resistance-problem-a-matter-of-billions/>

- Shelton A. 2011. *Biological Control: A Guide to Natural Enemies in North America*. Retrieved from <http://www.nysaes.cornell.edu/ent/biocontrol/>
- Sherfy MH, Anteau MJ, and Bishop AA. 2011. *Agricultural practices and residual corn during spring crane and waterfowl migration in Nebraska*. The Journal of Wildlife Management 75, pp. 995-1003. Retrieved from <http://dx.doi.org/10.1002/jwmg.157>
- Sherwani SI, Arif IA, and Khan HA. 2015. Modes of Action of Different Classes of Herbicides In: *Herbicides, Physiology of Action, and Safety*. Retrieved from <https://www.intechopen.com/books/herbicides-physiology-of-action-and-safety/modes-of-action-of-different-classes-of-herbicides>
- Soltani N, Dille JA, Burke IC, Everman WJ, VanGessel MJ, Davis VM, and Sikkema PH. 2017. *Potential Corn Yield Losses from Weeds in North America*. Weed Technology 30, pp. 979-984. Retrieved from <https://www.cambridge.org/core/article/potential-corn-yield-losses-from-weeds-in-north-america/4AFABD1F1F976034665D000FBA543C2F>
- Sosnoskie LM and Culpepper AS. 2014. *Glyphosate-Resistant Palmer Amaranth (Amaranthus palmeri) Increases Herbicide Use, Tillage, and Hand-Weeding in Georgia Cotton*. Weed Science 62, pp. 393-402. Retrieved from <https://doi.org/10.1614/WS-D-13-00077.1> Last accessed 2017/08/16.
- Specht J, Grassini P, Hoegemeyer T, Elmore R, Rees J, Mueller N, and Glewen K. 2017. *Soybean and Corn Yield and Acreage Trends through 2016*. University of Nebraska, Lincoln. Retrieved from <https://cropwatch.unl.edu/2017/soybean-and-corn-yield-and-acreage-trends-through-2016>
- Sterner RT, Petersen BE, Gaddis SE, Tope KL, and Poss DJ. 2003. *Impacts of small mammals and birds on low-tillage, dryland crops*. Crop Protection 22, pp. 595-602. Retrieved from http://digitalcommons.unl.edu/cgi/viewcontent.cgi?article=1271&context=icwdm_usdanwrc
- Stevenson K, Anderson RV, and Vigue G. 2002. *The density and diversity of soil invertebrates in conventional and pesticide free corn*. Transactions of the Illinois State Academy of Science 95, pp. 1-9. Retrieved from <http://ilacadofsci.com/wp-content/uploads/2013/08/095-01MS2113-print.pdf>.
- Stewart CM, McShea WJ, and Piccolo BP. 2007. *The Impact of White-Tailed Deer on Agricultural Landscapes in 3 National Historical Parks in Maryland*. Journal of Wildlife Management 71, pp. 1525-1530. Retrieved from <https://repository.si.edu/bitstream/handle/10088/6043/3CA614E8-A6EC-40A4-BE70-9F007630C058.pdf?sequence=1>
- Sundstrom FJ, Williams J, Van Deynze A, and Bradfor K. 2002. *Identity Preservation of Agricultural Commodities, Publication 8077*. Retrieved from <http://sbc.ucdavis.edu/files/200651.pdf>
- Taft OW and Elphick CS. 2007. *Waterbirds: Chapter 4 -Corn. Waterbirds on Working Lands: Literature Review and Bibliography Development*. National Audubon Society. Retrieved from http://web4.audubon.org/bird/waterbirds/pdf/Chapter_4_%20Corn.pdf
- Thomison P. 2004. *Managing "pollen drift" to minimize contamination of nonGMO Corn [AGF-153]*. Ohio State University Extension Fact Sheet Retrieved from <http://ohioline.osu.edu/agf-fact/0153.html>
- Thompson CJ, Movva NR, Tizard R, Crameri R, Davies JE, Lauwereys M, and Botterman J. 1987. *Characterization of the herbicide-resistance gene bar from Streptomyces hygrosopicus*. The EMBO journal 6, pp. 2519-2523. Retrieved from <https://www.ncbi.nlm.nih.gov/pubmed/16453790>
- Tothova T, Sobekova A, Holovska K, Legath J, Pristas P, and Javorsky P. 2010. *Natural glufosinate resistance of soil microorganisms and GMO safety*. Central European Journal of Biology 5, pp. 656-663. Retrieved from <https://doi.org/10.2478/s11535-010-0042-0>

- Towery D and Werblow S. 2010. *Facilitating Conservation Farming Practices and Enhancing Environmental Sustainability with Agricultural Biotechnology*. Conservation Technology Information Center (CTIC). Retrieved from <http://www.ctic.org/media/pdf/BioTechFINAL%20COPY%20SEND%20TO%20PRINTER.pdf>
- TOXNET. 2015. *Glufosinate-Ammonium*, CASRN: 77182-82-2. TOXNET. Toxicology Data Network, National Library of Medicine. Retrieved from <http://toxnet.nlm.nih.gov/cgi-bin/sis/search/a?dbs+hsdb:@term+@DOCNO+6666>
- U-Illinois-Ext. 2000. *Controlling Rodent Damage in Conservation Tillage Systems*. "2000 Illinois Agricultural Pest Management Handbook. Simpson, IL: University of Illinois Extension, Dixon Springs Agricultural Center. p 113-18. http://web.aces.uiuc.edu/vista/pdf_pubs/iapm2k/chap06.pdf.
- U-Missouri. 2009. *Practical Weed Science for the Field Scout: Corn and Soybean*. Plant Protection Programs, College of Agriculture, Food, and Natural Resources, University of Missouri. Retrieved from <http://www.ncga.com/upload/files/documents/pdf/Take%20Action%20on%20Weeds/Integrated-Pest-Management-Plant-Protection-Programs.pdf>
- UMD. 2005. *Native Plants of Maryland: What, When and Where* [Home and Garden Mimeo HG#120 3/2005]. University of Maryland, Cooperative Extension. Retrieved from https://extension.umd.edu/sites/extension.umd.edu/files/_images/programs/hgic/Publications/HG_120_Native_Plants%20of_MD.pdf
- UMinn. 2019. *Corn pest management*. University of Minnesota Extension. Retrieved from <https://extension.umn.edu/corn/corn-pest-management>
- US-EPA. 2005. *Environmental Risk Assessment of Plant Incorporated Protectant (PIP) Inert Ingredients - Phosphinothricin acetyltransferase (PAT)* U.S. Environmental Protection Agency. Retrieved from <https://archive.epa.gov/scipoly/sap/meetings/web/pdf/pipinertenvironmentalriskassessment11-18-05.pdf>
- US-EPA. 2007. *40 CFR §174.522 - Phosphinothricin Acetyltransferase (PAT); exemption from the requirement of a tolerance*. U.S. Environmental Protection Agency. Retrieved from <http://www.gpo.gov/fdsys/granule/CFR-2012-title40-vol25/CFR-2012-title40-vol25-sec174-522>
- US-EPA. 2014. *Environmental Fate and Ecological Risk Assessment for the Registration Review of Glufosinate*. Retrieved from <https://www.regulations.gov/document?D=EPA-HQ-OPP-2008-0190-0049>
- US-EPA. 2015. *Reducing Pesticide Drift*. U.S. Environmental Protection Agency. Retrieved from <http://www2.epa.gov/reducing-pesticide-drift>
- US-EPA. 2016a. *Glufosinate Resistance Management Recommendations* Retrieved from <https://cals.arizona.edu/apmc/docs/EPA-HQ-OPP-2008-0190-0048.pdf>
- US-EPA. 2016b. *Pesticide Worker Protection Standard "How to Comply" Manual*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/pesticide-worker-safety/pesticide-worker-protection-standard-how-comply-manual>
- US-EPA. 2016c. *Glufosinate Ammonium: Proposed Interim Registration Review, Decision Case Number 7224 [Docket Number EPA-HQ-OPP-2008-0190]*. Retrieved from <https://www.regulations.gov/document?D=EPA-HQ-OPP-2008-0190-0055>
- US-EPA. 2017a. *PR Notice 2017-2, Guidance for Herbicide-Resistance Management, Labeling, Education, Training and Stewardship*. U.S. Environmental Protection Agency. Retrieved from

- <https://www.epa.gov/pesticide-registration/prn-2017-2-guidance-herbicide-resistance-management-labeling-education>
- US-EPA. 2017b. *Air Monitoring at Agricultural Operations*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/afos-air>
- US-EPA. 2018. *Overview of Risk Assessment in the Pesticide Program*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/pesticide-science-and-assessing-pesticide-risks/overview-risk-assessment-pesticide-program>
- US-EPA. 2019a. *Indexes to Part 180 Tolerance Information for Pesticide Chemicals in Food and Feed Commodities*. U.S. Environmental Protection Agency. Retrieved from <http://www.epa.gov/opp00001/regulating/part-180.html>
- US-EPA. 2019b. *Agriculture and Air Quality*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/agriculture/agriculture-and-air-quality#prescribedburning>
- US-EPA. 2019c. *Drinking Water and Pesticides*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/safepestcontrol/drinking-water-and-pesticides>
- US-EPA. 2019d. *Agriculture and Sustainability*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/agriculture/agriculture-and-sustainability>
- US-EPA. 2019e. *Label Amendment –Add additional non-crop uses sites, weeds and adjust rates on Sublabel B. Product Name: WILLOWOOD GLUFOSINATE 280SL; EPA Registration Number: 87290-41; Application Date: 08/07/2019; Decision Number: 554230* U.S. Environmental Protection Agency Retrieved from https://www3.epa.gov/pesticides/chem_search/ppls/087290-00041-20191010.pdf
- US-EPA. 2019f. *Watershed Assessment, Tracking & Environmental Results, National Summary of State Information* U.S. Environmental Protection Agency. Retrieved from https://ofmpub.epa.gov/waters10/attains_nation_cy.control
- US-EPA. 2019g. *Mississippi River/Gulf of Mexico Hypoxia Task Force*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/ms-htf#citation>
- US-EPA. 2019h. *Pesticide Tolerances*. U.S. Environmental Protection Agency Retrieved from <http://www.epa.gov/opp00001/regulating/tolerances.htm>
- US-EPA. 2019i. *National Pollutant Discharge Elimination System (NPDES)*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/npdes>
- US-EPA. 2019j. *Reducing Pesticide Drift*. U.S. Environmental Protection Agency. Retrieved from <http://www2.epa.gov/reducing-pesticide-drift>
- US-EPA. 2019k. *Pesticide Volatilization*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/reducing-pesticide-drift/pesticide-volatilization>
- US-EPA. 2019l. *Aquatic Life Benchmarks and Ecological Risk Assessments for Registered Pesticides*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/pesticide-science-and-assessing-pesticide-risks/aquatic-life-benchmarks-and-ecological-risk>
- US-EPA. 2020a. *Inventory of U.S. Greenhouse Gas Emissions and Sinks*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/ghgemissions/inventory-us-greenhouse-gas-emissions-and-sinks>
- US-EPA. 2020b. *Renewable Fuel Standard Program*. U.S. Environmental Protection Agency. Retrieved from <https://www.epa.gov/renewable-fuel-standard-program>

- US-FDA. 1992a. *Statement of Policy - Foods Derived from New Plant Varieties*. U.S. Food and Drug Administration. Retrieved from <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/statement-policy-foods-derived-new-plant-varieties>
- US-FDA. 1992b. *Guidance to Industry for Foods Derived from New Plant Varieties*. Federal Register 57, pp. 22984. Retrieved from <https://www.fda.gov/food/guidanceregulation/guidancedocumentsregulatoryinformation/biotechnology/ucm096095.htm>
- US-FDA. 2006. *Guidance for Industry: Recommendations for the Early Food Safety Evaluation of New Non-Pesticidal Proteins Produced by New Plant Varieties Intended for Food Use*. U.S. Food and Drug Administration. Retrieved from <http://www.fda.gov/Food/GuidanceComplianceRegulatoryInformation/GuidanceDocuments/Biototechnology/ucm096156.htm>
- US-FDA. 2019. *Consultations on Food from GE Plant Varieties*. U.S. Food and Drug Administration. Retrieved from <http://www.accessdata.fda.gov/scripts/fdcc/?set=Biocon>
- USC. 2019. *South Carolina Plant Atlas*. John Nelson, Curator, A. C. Moore Herbarium, Department of Biological Sciences, University of South Carolina. Retrieved from <http://herbarium.biol.sc.edu/scplantatlas.html>; <https://docs.google.com/viewer?a=v&pid=sites&srcid=ZGVmYXVsdGRvbWFpbXxzYzBoZXJp dGFnZTB0cnVzdHxneDo0YjA1MjQzOWQ0YzlkxNTY5>
- USDA-AMS. 2019a. *Pesticide Data Program*. U.S. Department of Agriculture, Agricultural Marketing Service, Science and Technology Programs Retrieved from <http://www.ams.usda.gov/AMSV1.0/pdp>
- USDA-AMS. 2019b. *Identity Preservation Program*. U.S. Department of Agriculture, Agricultural Marketing Service. Retrieved from <https://www.ams.usda.gov/services/auditing/identity-preservation>
- USDA-APHIS. 2013. *Plant Pest Risk Assessment for HCEM485 Corn [09-063-01p]*. U.S. Department of Agriculture, Animal and Plant Health Inspection Retrieved from https://www.aphis.usda.gov/brs/aphisdocs/09_06301p_fpra.pdf
- USDA-APHIS. 2018a. *Enhancements to Public Input*. Retrieved from https://www.aphis.usda.gov/aphis/ourfocus/biotechnology/SA_Permits_Notifications_And_Petitions/SA_Petitions/CT_Pet_proc_imp_info
- USDA-APHIS. 2018b. *Coordinated Framework*. Retrieved from https://www.aphis.usda.gov/aphis/ourfocus/biotechnology/sa_regulations/ct_agency_framework_roles
- USDA-APHIS. 2019a. *Draft Plant Pest Risk Assessment: Pioneer Hi-Bred International, Inc. Petition (19-337-01p) for the Determination of Nonregulated Status for Insect Resistant and Herbicide-Tolerant DP23211 Maize [OECD Unique Identifier: DP-023211-2]*. Retrieved from <https://www.aphis.usda.gov/aphis/ourfocus/biotechnology/permits-notifications-petitions/petitions/petition-status>
- USDA-APHIS. 2019b. *Biotechnology: Petitions for Determination of Nonregulated Status* U.S. Department of Agriculture, Animal and Plant Health Inspection Service. Retrieved from http://www.aphis.usda.gov/biotechnology/petitions_table_pending.shtml
- USDA-EPA. 2012. *Agricultural Air Quality Conservation Measures: Reference Guide for Cropping Systems And General Land Management (October 2012)*. Retrieved from http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/stelprdb1049502.pdf

- USDA-ERS. 2012. *Agricultural Resources and Environmental Indicators, 2012 Edition [Economic Information Bulletin Number 98]*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <http://www.ers.usda.gov/publications/eib-economic-information-bulletin/eib98.aspx>
- USDA-ERS. 2018. *Corn and Other Feedgrains*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <https://www.ers.usda.gov/topics/crops/corn-and-other-feedgrains/background/>
- USDA-ERS. 2019a. *Feedgrains Sector at a Glance*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <https://www.ers.usda.gov/topics/crops/corn-and-other-feedgrains/feedgrains-sector-at-a-glance/>
- USDA-ERS. 2019b. *Adoption of Genetically Engineered Crops 2018*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <https://www.ers.usda.gov/data-products/adoption-of-genetically-engineered-crops-in-the-us/recent-trends-in-ge-adoption/>
- USDA-ERS. 2019c. *Fertilizer Use and Price - Datasets*. U.S. Department of Agriculture, Economic Research Service Retrieved from <https://www.ers.usda.gov/data-products/fertilizer-use-and-price.aspx>
- USDA-ERS. 2019d. *Outlook for U.S. Agricultural Trade: Situation and Outlook Report [AES-106]*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <https://www.ers.usda.gov/webdocs/publications/90902/aes-106.pdf?v=3468.1>
- USDA-ERS. 2019e. *USDA Agricultural Projections to 2028: Interagency Agricultural Projections Committee [Long-term Projections Report OCE-2019-1]*. U.S. Department of Agriculture, Office of the Chief Economist, World Agricultural Outlook Board. Retrieved from https://www.usda.gov/oce/commodity/projections/USDA_Agricultural_Projections_to_2028.pdf
- USDA-ERS. 2019f. *Sweet corn: U.S. exports by volume*. U.S. Department of Agriculture, Economic Research Service. Retrieved from https://data.ers.usda.gov/reports.aspx?programArea=veg&stat_year=2008&top=5&HardCopy=True&RowsPerPage=25&groupName=Vegetables&commodityName=Sweet%20corn&ID=17858#P472e928bf33643fe972f49514382e65c_2_203
- USDA-FAS. 2019. *Grain: World Markets and Trade, June 2019*. U.S. Department of Agriculture, Foreign Agricultural Service. Retrieved from <https://apps.fas.usda.gov/psdonline/circulars/grain.pdf>
- USDA-NASS. 2014. *2012 Census of Agriculture, United States, Summary and State Data, Vol. 1, Geographic Area Series, Part 51 [AC-12-A-51]*. U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from http://www.agcensus.usda.gov/Publications/2012/Online_Resources/Ag_Atlas_Maps/Crops_and_Plants/Field_Crops_Harvested/12-M160-RGBDot1-largetext.pdf
- USDA-NASS. 2019a. *2017 Census of Agriculture, United States, Summary and State Data, Vol. 1, Geographic Area Series, Part 51 [AC-17-A-51]*. U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from http://www.agcensus.usda.gov/Publications/2012/Online_Resources/Ag_Atlas_Maps/Crops_and_Plants/Field_Crops_Harvested/12-M160-RGBDot1-largetext.pdf
- USDA-NASS. 2019b. *National Statistics for Corn* U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from https://www.nass.usda.gov/Statistics_by_Subject/result.php?7B832B80-F468-398F-8A28-F9C76A50551B§or=CROPS&group=FIELD%20CROPS&comm=CORN

- USDA-NASS. 2019c. *Charts and Maps, Field Crops, Corn: Acreage by Year*. U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from https://www.nass.usda.gov/Charts_and_Maps/Field_Crops/
- USDA-NASS. 2019d. *2018 Agricultural Chemical Use Survey: Corn [No. 2019-1]*. U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from https://www.nass.usda.gov/Surveys/Guide_to_NASS_Surveys/Chemical_Use/2018_Peanuts_Soy_beans_Corn/ChemUseHighlights_Corn_2018.pdf
- USDA-NASS. 2019e. *Quick Stats*. U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from <https://quickstats.nass.usda.gov/>
- USDA-NASS. 2019f. *Quick Stats*. U.S. Department of Agriculture, National Agricultural Statistics Service. Retrieved from <http://quickstats.nass.usda.gov/#80DA2DF4-B605-3184-A045-AE595D8FF3D3>
- USDA-NIFA. 2017. *Sustainable Agriculture Program*. United States Department of Agriculture, National Institute of Food and Agriculture. Retrieved from <https://nifa.usda.gov/program/sustainable-agriculture-program>
- USDA-NRCS. 1996. *Eastern Gamgrass*. U.S. Department of Agriculture, Natural Resources Conservation Service Retrieved from https://www.nrcs.usda.gov/Internet/FSE_PLANTMATERIALS/publications/mopmcfseggrs.pdf
- USDA-NRCS. 1999. *Conservation Tillage Systems and Wildlife. Fish and Wildlife Literature Review Summary, Number 1*. U.S. Department of Agriculture, Natural Resources Conservation Service. Retrieved from http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs143_022212.pdf
- USDA-NRCS. 2006. *Conservation Resource Brief: Soil Erosion, Number 0602*. U.S. Department of Agriculture, Natural Resources Conservation Service. Retrieved from http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs143_023234.pdf
- USDA-NRCS. 2010. *2007 National Resources Inventory: Soil Erosion on Cropland*. Retrieved from http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs143_012269.pdf
- USDA-NRCS. 2015. *Introduced, Invasive, and Noxious Plants*. U.S. Department of Agriculture, Natural Resources Conservation Service.
- USDA-NRCS. 2018a. *Summary Report: 2015 National Resources Inventory*. U.S. Department of Agriculture, Natural Resources Conservation Service, Washington, DC, and Center for Survey Statistics and Methodology, Iowa State University, Ames, Iowa. Retrieved from <https://www.nrcs.usda.gov/wps/portal/nrcs/main/national/technical/nra/nri/>
- USDA-NRCS. 2018b. *Index of internet NRCS RCA maps*. U.S. Department of Agriculture, Natural Resources Conservation Service Retrieved from https://www.nrcs.usda.gov/Internet/NRCS_RCA/maps/m13655.png
- USDA-NRCS. 2019a. *National Water Quality Initiative (NWQI)*. U.S. Department of Agriculture, Natural Resources Conservation Service, Caribbean Area. Retrieved from <https://www.nrcs.usda.gov/wps/portal/nrcs/detail/national/programs/initiatives/?cid=stelprdb1047761>
- USDA-NRCS. 2019b. *Natural Resources Conservation Service: Programs*. U.S. Department of Agriculture, Natural Resources Conservation Service. Retrieved from <https://www.nrcs.usda.gov/wps/portal/nrcs/main/national/programs/>
- USDA-NRCS. 2019c. *Environmental Quality Incentives Program Initiatives - Overview*. U.S. Department of Agriculture, Natural Resources Conservation Service Retrieved from

- <https://www.nrcs.usda.gov/wps/portal/nrcs/detail/national/programs/financial/equip/?&cid=stelprd1047458>
- USDA-NRCS. 2019d. *USDA Plants Database*. U.S. Department of Agriculture, Natural Resources Conservation Service. Retrieved from <https://plants.sc.egov.usda.gov/java/>
- USDA-NRCS. 2019e. *Introduced, Invasive, and Noxious Plants*. U.S. Department of Agriculture, Natural Resources Conservation Service. Retrieved from <https://plants.usda.gov/java/noxiousDriver>
- USFWS. 1999. *South Florida multi-species recovery plan*. U.S. Fish & Wildlife Service. Retrieved from <https://www.fws.gov/verobeach/MSRPPDFs/ExecSum.pdf> Last accessed July 27, 2015.
- USFWS. 2011. *Environmental Assessment - Use of Genetically Modified, Glyphosate-Tolerant Soybeans and Corn on National Wildlife Refuge Lands in the Mountain–Prairie Region (Region 6)*. Lakewood, Colorado. Retrieved from http://www.fws.gov/mountain-prairie/planning/resources/documents/resources_gmo_ea.pdf
- USFWS. 2013. *Recovery Plan for Tidal Marsh Ecosystems of Northern and Central California*. U.S. Fish & Wildlife Service. Retrieved from https://www.fws.gov/sfbaydelta/documents/tidal_marsh_recovery_plan_v1.pdf Last accessed July 27, 2015.
- USFWS. 2020. *USFWS Environmental Conservation Online System*. U.S. Fish & Wildlife Service. Retrieved from <https://ecos.fws.gov/ecp/> Last accessed July 27, 2015.
- USGC. 2020. *Corn: Production and Exports*. U.S. Grains Council. Retrieved from <https://grains.org/buying-selling/corn/>
- USGS. 2016. *National Water-Quality Assessment (NAWQA) Program: Pesticide National Synthesis Project, Estimated Annual Agricultural Pesticide Use, Pesticide Use Maps - Glufosinate* U.S. Department of the Interior, U.S. Geological Survey. Retrieved from http://water.usgs.gov/nawqa/pnsp/usage/maps/show_map.php?year=2014&map=GLUFOSINATE&hilo=L
- Van Eenennaam AL and Young AE. 2014. *Prevalence and impacts of genetically engineered feedstuffs on livestock populations*. J. Anim. Sci. 92, pp. 4255-4278. Retrieved from <https://academic.oup.com/jas/article/92/10/4255/4702576>
- Vencill WK, Nichols RL, Webster TM, Soteres JK, Mallory-Smith C, Burgos NR, Johnson WG, and McClelland MR. 2012. *Herbicide Resistance: Toward an Understanding of Resistance Development and the Impact of Herbicide-Resistant Crops*. Weed Science 2012 Special Issue, pp. 2-30. Retrieved from <http://www.wssajournals.org/doi/pdf/10.1614/WS-D-11-00206.1>
- Wallander S. 2015. *Soil Tillage and Crop Rotation*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <http://www.ers.usda.gov/topics/farm-practices-management/crop-livestock-practices/soil-tillage-and-crop-rotation.aspx>
- Warwick SI, Beckie HJ, and Hall LM. 2009. *Gene flow, invasiveness, and ecological impact of genetically modified crops*. Annals of the New York Academy of Sciences 1168, pp. 72-99. Retrieved from <http://onlinelibrary.wiley.com/doi/10.1111/j.1749-6632.2009.04576.x/abstract;jsessionid=BD07B0AA60A7AD1E4C1388B6601DA74C.f01t02>
- Weaver M. 2019. *Growers face battle against herbicide resistance*. Capitalpress. Retrieved from https://www.capitalpress.com/nation_world/agriculture/growers-face-battle-against-herbicide-resistance/article_9521ac9e-5667-11e9-b00f-8fbfd342c31.html

- Wehrmann A, Van Vliet A, Opsomer C, Botterman J, and Schulz A. 1996. *The similarities of bar and pat gene products make them equally applicable for plant engineers*. Nature biotechnology 14, pp. 1274-1278.
- Westcott P and Hansen J. 2015. *USDA Agricultural Projections to 2024, Long-term Projections Report OCE-2015-1*. United States Department of Agriculture, Office of the Chief Economist, World Agricultural Outlook Board. Retrieved from https://www.usda.gov/oce/commodity/projections/USDA_Agricultural_Projections_to_2024.pdf
- WHO-FAO. 2009. *Codex Alimentarius: Foods derived from modern biotechnology*. Rome, Italy: World Health Organization (WHO), Food and Agriculture Organization of the United Nations (FAO). Retrieved from ftp://ftp.fao.org/codex/Publications/Booklets/Biotech/Biotech_2009e.pdf
- Wibawa W, Mohamad R, Omar D, Zain N, Puteh A, and Awang Y. 2010. *Comparative impact of a single application of selected broad spectrum herbicides on ecological components of oil palm plantation*. African Journal of Agricultural Research 5, pp. 2097-2102. Retrieved from http://www.academicjournals.org/article/article1381143469_Wibawa%20et%20al.pdf
- Wiebe K and Gollehon N. 2006a. *Agricultural Resources and Environmental Indicators [Economic Information Bulletin No. EIB-16]*. U.S. Department of Agriculture, Economic Research Service. Retrieved from <http://www.ers.usda.gov/publications/eib-economic-information-bulletin/eib16.aspx>
- Wiebe K and Gollehon N. 2006b. *Agricultural Resources and Environmental Indicators, 2006 Edition*. Retrieved from <http://www.ers.usda.gov/publications/eib-economic-information-bulletin/eib16.aspx>
- Wilkes HG. 1967. *Teosinte: the closest relative of maize*. The Bussey Institution of Harvard University.
- Wohlleben W, Arnold W, Broer I, Hillemann D, Strauch E, and Puhler A. 1988. *Nucleotide sequence of the phosphinothricin N-acetyltransferase gene from Streptomyces viridochromogenes Tu494 and its expression in Nicotiana tabacum*. Gene 70, pp. 25-37.
- Wolmarans K and Swart WJ. 2014. *Influence of glyphosate, other herbicides and genetically modified herbicide-resistant crops on soil microbiota: a review*. South African Journal of Plant and Soil 31, pp. 177-186. Retrieved from <https://doi.org/10.1080/02571862.2014.960485>
- Wozniak CA. 2002. *Gene Flow Assessment for Plant-Incorporated Protectants. Scientific Methods Workshop: Ecological and Agronomic Consequences of Gene Flow from Transgenic Crops to Wild Relatives*. Retrieved from <http://www.biosci.ohio-state.edu/~asnowlab/Proceedings.pdf>
- WTO. 2019a. *Sanitary and Phytosanitary Measures*. World Trade Organization (WTO). Retrieved from https://www.wto.org/english/tratop_e/sps_e/sps_e.htm
- WTO. 2019b. *International Plant Protection Convention (IPPC)*. World Trade Organization (WTO). Retrieved from https://www.wto.org/english/thewto_e/coher_e/wto_ippc_e.htm
- WTO. 2019c. *WTO Technical Barriers to Trade (TBT) Agreement*. World Trade Organization (WTO). Retrieved from https://www.wto.org/English/docs_e/legal_e/17-tbt_e.htm
- Wu J, Lawit SJ, Weers B, Sun J, Mongar N, Van Hemert J, Melo R, Meng X, Rupe M, Clapp J, Haug Collet K, Trecker L, Roesler K, Peddicord L, Thomas J, Hunt J, Zhou W, Hou Z, Wimmer M, Jantes J, Mo H, Liu L, Wang Y, Walker C, Danilevskaya O, Lafitte RH, Schussler JR, Shen B, and Habben JE. 2019. *Overexpression of zmm28 increases maize grain yield in the field*. Proceedings of the National Academy of Sciences of the United States of America 116, pp. 23850-23858. Retrieved from <https://www.pnas.org/content/116/47/23850>

- Wunderlin RP, Hansen BF, Franck AR, and Essig FB. 2019. *Atlas of Florida Plants*. University of South Florida (USF), Institute for Systematic Botany, Tampa. S. M. Landry and K. N. Campbell (application development). Retrieved from <http://florida.plantatlas.usf.edu/>
- Xie M, Zhang Y-J, Peng D-L, Wu G, Xu P, Zhao J-J, and Zhang Z-R. 2016. *Field studies show no significant effect of a CryIAb/Ac producing transgenic cotton on the fungal community structure in rhizosphere soil*. European Journal of Soil Biology 73, pp. 69-76. Retrieved from <http://www.sciencedirect.com/science/article/pii/S1164556316300061>
- Yang Q, Deng W, Li X, Yu Q, Bai L, and Zheng M. 2016. *Target-site and non-target-site based resistance to the herbicide tribenuron-methyl in flaxweed (Descurainia sophia L.)*. BMC Genomics 17, pp. 551. Retrieved from <https://bmcbgenomics.biomedcentral.com/track/pdf/10.1186/s12864-016-2915-8>
- Yasin S, Asghar HN, Ahmad F, Zahir ZA, and Waraich EA. 2016. *Impact of Bt-cotton on soil microbiological and biochemical attributes*. Plant Production Science 19, pp. 458-467. Retrieved from <https://www.scopus.com/inward/record.uri?eid=2-s2.0-85006427823&doi=10.1080%2f1343943X.2016.1185637&partnerID=40&md5=9338004631e3e4b779e2b7d4e577866d>
- Zabaloy MC, Garland JL, and Gómez MA. 2008. *An integrated approach to evaluate the impacts of the herbicides glyphosate, 2,4-D and metsulfuron-methyl on soil microbial communities in the Pampas region, Argentina*. Applied Soil Ecology 40, pp. 1-12. Retrieved from <http://www.sciencedirect.com/science/article/pii/S092913930800036X>
- Zaman M, Mirza MS, Irem S, Zafar Y, and Mehboob ur R. 2015. *A temporal expression of CryIAc protein in cotton plant and its impact on soil health*. International Journal of Agriculture and Biology 17, pp. 280-288. Retrieved from <https://www.scopus.com/inward/record.uri?eid=2-s2.0-84938995601&partnerID=40&md5=b511f933835cac2f995ba6ed03dfe008>