NATIONAL ENVIRONMENTAL POLICY ACT DECISION:

FINDING OF NO SIGNIFICANT IMPACT

Permit Application 16-076-101r
Submitted by Dr. Anthony Shelton of Cornell University

Field Release of Genetically Engineered
Diamondback Moth Strain OX4319L-Pxy

United States Department of Agriculture
Animal and Plant Health Inspection Service
Biotechnology Regulatory Services

The United States Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS), has prepared this decision document that is consistent with the requirements of the National Environmental Policy Act of 1969 (NEPA), as amended, the Council of Environmental Quality (CEQ) regulations implementing NEPA, and USDA departmental and APHIS NEPA-implementing regulations and procedures. This NEPA decision document describes the Agency's NEPA decision and explains the rationale for making it.

On March 16, 2016, APHIS received a permit application from Dr. Anthony Shelton of Cornell University (APHIS Permit Number 16–076–101r) seeking authorization for the field release of a single strain of a genetically engineered (GE) diamondback moth (DBM), designated as OX4319L-Pxy, in both open and caged releases on experimental sites within the boundaries of the New York State Agricultural Experiment Station (NYSAES) in Geneva, New York. The purpose of the proposed releases is to allow the applicant to assess the efficacy of GE DBM strain OX4319L-Pxy for reducing pest populations of non-GE DBMs. Information provided in the application indicated that controlled releases of these GE DBMs may offer a species-specific management alternative for controlling economically significant DBM outbreaks in crucifers (*Brassica* spp.) that does not rely on insecticide applications.

In accordance with APHIS procedures implementing the Agency's NEPA Regulations described in Title 7 of the Code of Federal Regulations part 372 (7 CFR part 372), APHIS conducted an Environmental Assessment (EA) to evaluate and determine if any significant impacts to the human environment are likely to occur if environmental releases of a plant pest are made as described in the permit application. The EA analyzed alternatives for issuing a permit with conditions that would allow experimental field releases of GE DBM strain OX4319L-Pxy to reduce pest populations of non-GE DBMs. The field considered in the EA, which is proposed for experimental releases, is limited to a maximum of ten acres. A single point within this field will be selected as the location for unrestricted releases of GE DBMs. The permit would also allow the permit holder to conduct caged field studies in the experimental field designated as the release site, but outside of the plot containing the single release point. Because crop rotation practices may require moving the 10-acre experimental field to another location within the

NYSAES in subsequent growing seasons, if the permit is renewed, the EA considered the entire NYSAES as the action area.

The current application (16-076-101r) is a revision of an application that was submitted on October 24, 2013 from Dr. Anthony Shelton of Cornell University (APHIS Permit Number 13–297–102r**)** seeking the permitted field release of three imported strains of GE DBMs. An EA was prepared for the previous permit application and a Finding of No Significant Impact (FONSI) was signed by APHIS on November 7, 2014. The permit was issued and caged releases were made in 2015. However, the permit was subsequently withdrawn before unrestricted field releases (not sequestered in cages) occurred. APHIS also previously issued permits authorizing the applicant to import GE DBMs into an Agency-regulated containment facility to assess efficacy and possible environmental impacts under controlled conditions within quarantined confinement.

The current proposed action by USDA APHIS, Biotechnology Regulatory Services (BRS) is to issue the APHIS field release permit for the specified GE DBM strain with supplemental permit conditions in accordance with Agency regulations at 7 CFR part 340.4.[1] APHIS will publish the availability of the Finding of No Significant Impact (FONSI) in the Federal Register and post it on the APHIS web site and subsequently issue the permit.

In 1986, the Office of Science and Technology Policy (OSTP) issued the Coordinated Framework for the Regulation of Biotechnology (cited as the Coordinated Framework in the remainder of this document), which describes the comprehensive Federal regulatory policy for ensuring the safety of biotechnology products (51 FR 23302). Since 1986, the Environmental Protection Agency (EPA), Food and Drug Administration (FDA), and USDA have regulated GE organisms consistent with this framework. The Coordinated Framework is based on several important guiding principles: (1) agencies will define those GE organisms subject to review to the extent permitted by their respective statutory authorities; (2) agencies will focus on the characteristics and risks of the biotechnology product, not the process by which it is created; (3) agencies will exercise oversight of GE organisms when there is evidence of "unreasonable" risk.

In 2015, the EPA, FDA, and USDA began an effort to modernize the regulatory system for biotechnology products to accomplish three tasks: (1) clarify the current roles and responsibilities of the EPA, FDA, and USDA in the regulatory process; (2) develop a long-term strategy to ensure that the Federal regulatory system is equipped to efficiently assess the risks, if any, of the future products of biotechnology; and (3) commission an expert analysis of the future landscape of biotechnology products. The *Update to the Coordinated Framework for the Regulation of Biotechnology* was released on January 4, 2017[2], representing the first time in more than 20 years that the Federal government has produced a comprehensive summary of the roles and responsibilities of the three principal regulatory agencies with respect to regulating biotechnology products. This update provides the public with a comprehensive description of a robust and flexible regulatory structure that provides appropriate oversight for all products of modern biotechnology. Within that regulatory structure the federal agencies maintain high standards that, based on the best available science, protect health and the environment, while also

[1]Regulations are available for review at:
http://www.gpo.gov/fdsys/granule/CFR-2012-title7-vol5/CFR-2012-title7-vol5-sec340-4/content-detail.html
[2]See https://www.epa.gov/regulation-biotechnology-under-tsca-and-fifra/update-coordinated-framework-regulationbiotechnology

establishing transparent, coordinated, predictable and efficient regulatory practices. The authorities and regulatory roles for APHIS, the EPA, and FDA are briefly summarized below.

USDA-APHIS- Biotechnology Regulatory Services (BRS)

APHIS is authorized to regulate GE organisms under the plant pest provisions in the Plant Protection Act of 2000, as amended (7 USC § 7701 *et seq.*), to prevent or minimize plant pest risks. APHIS regulations at 7 CFR part 340, regulate the introduction (importation, interstate movement, and release into the environment) of certain GE organisms and their products. 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 the regulations (7 CFR 340.2) and is also considered a plant pest. A GE organism is also regulated under 7 CFR part 340 if APHIS has reason to believe that the GE organism may be a plant pest or APHIS does not have sufficient information to determine if the GE organism is unlikely to pose a plant pest risk. Diamondback moth, the recipient organism in 16-076-101r, is a plant pest.

When APHIS receives an application for a permit for environmental release and movement, the application is evaluated to determine if the environmental release and movement, with appropriate conditions imposed, can be implemented while preventing the dissemination and establishment of plant pests. The receipt of a permit application to introduce a GE organism requires a response from the Administrator as set forth in the regulations:

> *Administrative action on applications*. After receipt and review by APHIS of the application and the data submitted pursuant to paragraph (a) of this section, including any additional information requested by APHIS, a permit shall be granted or denied (7 CFR 340.4(e)).

The applicant provided the required information associated with the request in the permit application (16-076-101r). This information was reviewed by APHIS-BRS and analyzed in the EA.

FDA

The FDA regulates GE organisms pursuant to the authority of the Federal Food, Drug, and Cosmetic Act (21 USC § 301 *et seq.*). The FDA is responsible for ensuring the safety and proper labeling of all plant-derived foods and feeds, including those that are genetically engineered. To help developers of food and feed derived from GE crops comply with their obligations under Federal food safety laws, FDA encourages them to participate in a voluntary consultation process. The FDA policy statement concerning regulation of products derived from new plant varieties, including those genetically engineered, was published in the Federal Register on May 29, 1992 (57 FR 22984-23005). Under this policy, FDA uses a consultation process to ensure that human food and animal feed safety issues or other regulatory issues are resolved prior to commercial distribution of bioengineered foods.

EPA

The EPA regulates pesticides, including plant-incorporated protectants pursuant to the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) (7 USC § 136 *et seq.*). Specifically, the EPA sets tolerance limits for residues of pesticides on and in food and animal feed, or establishes an exemption from the requirement for a tolerance, pursuant to the Federal Food, Drug, and Cosmetic Act (FFDCA), and regulates certain biological control organisms under the Toxic

Substances Control Act (TSCA). The EPA is responsible for regulating the sale, distribution, and use of pesticides, including pesticides that are produced by an organism through techniques of modern biotechnology.

Because GE DBM is not used for food or feed purposes and does not contain any GE plant-incorporated protectants with insecticidal properties or GE traits that convey herbicide resistance, neither FDA nor EPA regulatory action is required prior to issuing the APHIS permit (16-076-101r) that is the subject of this FONSI. Under 7 CFR part 340, APHIS only has authority to regulate a GE organism if the Agency believes it may pose a plant pest risk (7 CFR § 340.1). APHIS has no regulatory jurisdiction over any other risks associated with GE organisms including those resulting from the use of pesticides on GE organisms, or those that occur as a result of their use for other purposes.

PUBLIC INVOLVEMENT

Prior to taking a permitting action and conducting an EA, APHIS seeks public involvement and input by making the EA available for public review and comment. In a notice published April 19, 2017 in the *Federal Register* (FR Vol. 82, No. 74, pp. 18416-18417) APHIS announced the availability of the EA (Docket No. APHIS-2014-0056-0293) for public review and comment on the proposed field release of GE DBMs. Comments were accepted during a 30-day comment period that closed on May 19, 2017. APHIS received just over 670 comments. Both the EA and the comments remain available[3] for public viewing. This FONSI will be posted in the *Federal Register* and on the APHIS BRS web site[4].

All comments were analyzed to identify new issues, alternatives, or information. Responses to substantive comments are included as an attachment to this FONSI.

MAJOR ISSUES ADDRESSED IN THE EA

Relevant issues were identified and described in the EA, and two alternatives were considered and evaluated in relation to the identified issues. The list of resource areas considered in the EA was developed by APHIS from its experience in considering public concerns and issues identified in public comments submitted for other similar NEPA documents (USDA-APHIS 2017) prepared by the Agency including those for the release of GE insects (USDA-APHIS 2008a, 2009, 2011a). The resource areas also addressed concerns identified in previous legal actions related to GE organisms regulated by APHIS, and issues previously identified by various stakeholders. A summary of resource areas considered in this EA follows.

Environmental Considerations:

- Soil resources
- Water resources
- Air quality
- Climate change

[3]https://www.regulations.gov/document?D=APHIS-2014-0056-0291
[4]https://www.aphis.usda.gov/brs/biotech_ea_permits.html

- Plant communities
- Wildlife
- Biological diversity

Human Population Considerations:

- Farmworker health
- Public health

AFFECTED ENVIRONMENT

The experimental area for releases of GE DBM as described in the permit (16-076-101r) application and reviewed in Chapter 3 of the EA consists of a single 10-acre field site. However, the action area for the permit includes the entire NYSAES in Geneva, NY, since the 10-acre field site may need to be moved within the NYSAES in subsequent growing seasons for crop rotation practices if the permit is renewed. The NYSAES encompasses 870 acres located on the northwestern boundary of Geneva, NY, approximately two miles from the population center of the city.

The proposed release site is surrounded by other agricultural fields. The NYSAES has been used for agricultural activities for much of its 134-year history (NYSAES 2014). More than 700 acres are currently planted in row/vegetables crops, orchards, and vineyards (NYSAES 2014), including the proposed field release site.

Studies on dispersal of DBMs have examined both local, short-range movement and long-distance migration. Long-distance migration has been attributed to be the source of spring populations of DBMs where they cannot overwinter (Talekar and Shelton 1993; Furlong et al. 2013). Evidence for long-distance migration is both indirect (*e.g.,* Wei et al. 2013; Yang et al. 2015) and direct (*e.g.,* Chapman 2002; Fu et al. 2014), but long-distance dispersal is dependent on weather patterns (Hopkinson and Soroka 2010; Leskinen et al. 2011). If long-distance dispersal of GE DBMs were to occur from the proposed release site, then the predominant winds in the region of the proposed release site would likely move them to regions of similar latitude or further north (*i.e.*, to regions that experience winter months as cold or colder than Geneva, NY). Prevailing wind patterns near Geneva, NY, when releases of GE DBMs are most likely to occur (NOAA-NCEI 2016), will prevent DBM movement into regions where they may successfully overwinter.

Local movement, in contrast, is the primary dispersal pattern of DBMs, the directionality of which is not influenced by the weather (*i.e.,* a random pattern of dispersal) (Schellhorn et al. 2008; Shirai & Nakamura, 1994; Mo et al. 2003). DBMs rarely leave an area with suitable host plants to disperse beyond 100 m (Shirai and Nakamura 1994; Mo et al. 2003). However, because local dispersal of DBMs has some level of uncertainty, APHIS adopted a conservative approach and set a 150 m boundary zone surrounding the release site. This boundary zone will not be planted with host plants for DBMs to prevent attracting moths to leave the release site.

ALTERNATIVES

The EA analyzed the potential environmental consequences of APHIS issuing an environmental release permit with supplemental permit conditions to allow the field release of GE DBMs to control pest populations of non-GE DBMs. Based on the information in the permit application (16-076-101r), two alternatives were considered and analyzed in the EA: (1) no action: deny the permit; (2) preferred alternative: issue the APHIS permit.

Alternative A: No Action – Deny the Permit

Under the No Action Alternative, APHIS would deny the permit application (16-076-101r). The applicant would not be authorized to release the GE DBM strain OX4319L-Pxy. This alternative is the appropriate one for APHIS to choose if sufficient evidence demonstrates that this GE DBM strain either presents an unacceptable plant pest risk, would not remain confined to the release area described in the application, or APHIS lacks sufficient information to make a determination about possible risks associated with releasing GE DBM.

Alternative B: Preferred Alternative – Issue the APHIS Permit

Under the Preferred Alternative, APHIS would issue an environmental release permit in accordance with 7 CFR part 340 to allow the release of GE DBM strain OX4319L-Pxy within the experimental field area described in the permit application. This alternative is the appropriate one for APHIS to choose if sufficient evidence demonstrates that this GE DBM strain would neither present an unacceptable plant pest risk nor allow for its establishment and persistence in the environment if released in accordance with APHIS-prescribed conditions described in 7 CFR part 340.4.

Under the Preferred Alternative, the permit, if issued, would be valid until the end of 2017. Under the Preferred Alternative, the applicant would be allowed to gather data on the performance of GE DBMs for reducing populations of non-GE DBMs until the expiration of the permit.

ENVIRONMENTAL CONSEQUENCES OF THE SELECTED ACTION

Table 1 includes a summary of the findings made by APHIS from the Agency's analysis of each of the issues considered in the Environmental Consequences chapter of the final EA in response to the permit application (16-076-101r). Reviewers of this FONSI are referred to the final EA for full descriptions of the analysis made for each issue considered.

Table 1. Comparison of Alternatives

Attribute / Measure	Alternative A: No Action Alternative Deny the permit request	Alternative B: Preferred Alternative Grant the permit request
Meets Purpose and Need and Objectives	**No**	**Yes**
Unlikely to pose a plant pest risk	No plant pest risk.	Satisfied through use of regulated field trials, including APHIS-imposed permit conditions and monitoring for compliance. Impacts would be similar to the No Action Alternative.
Physical Environment		
Soil Quality	Common agricultural activities related to field preparation/maintenance that impact soil (e.g., tillage, pesticide application, etc.) will continue unchanged under the No Action Alternative.	The permitted field release of GE DBMs is not anticipated to change common agricultural activities related to preparing and maintaining an agricultural field that are already occurring under the No Action Alternative. Transfer of non-native DNA from decomposing GE DBMs to other soil microflora is not likely under the Preferred Alternative. Therefore, impacts on soil resources would be similar to the No Action Alternative.
Water Resources	Agronomic practices that could impact water resources (e.g., irrigation, tillage practices, and the application of agronomic inputs) would be expected to continue unchanged under the No Action Alternative. The use of pesticides in accordance with EPA-approved label directions assures no unreasonable risks to water quality from their use.	The permitted field release of GE DBMs is not anticipated to change common agricultural activities related to preparing and maintaining an agricultural field that are already occurring under the No Action Alternative. Therefore, impacts on water resources would be similar to the No Action Alternative.
Air Quality	Common agricultural activities (e.g., tillage; use of mechanized equipment that emits exhaust pollutants, and applications of pesticides and fertilizers) would continue unchanged under the No Action Alternative. The use of pesticides in accordance with EPA-approved labels minimizes drift and reduces environmental impacts.	The permitted field release of GE DBMs is not anticipated to change common agricultural activities currently used for fields as described under the No Action Alternative. Therefore, impacts on air quality would be similar to the No Action Alternative.

Attribute / Measure	Alternative A: No Action Alternative Deny the permit request	Alternative B: Preferred Alternative Grant the permit request
Climate Change	Common agricultural activities possess the potential to impact climate change, through the release of CO_2 to the atmosphere from tillage; machinery powered by fossil fuel; and NO_2 emissions associated with nitrogen fertilizers use. These activities are already occurring, and are likely to continue occurring, under the No Action Alternative.	The permitted field release of GE DBMs is not anticipated to change common agricultural activities related to preparing and maintaining an agricultural field as are already occurring under the No Action Alternative. Therefore, the impact on GHG emissions and climate change would be similar to the No Action Alternative.
Biological Environment		
Wildlife	Common agricultural activities such as such as tillage, cultivation, pesticide and fertilizer applications, and the use of agricultural equipment may impact wildlife communities. The use of EPA-registered pesticides and herbicides in accordance with EPA-approved labels minimizes potential impacts to animal communities.	The permitted field release of GE DBMs are not anticipated to change common agricultural activities related to preparing and maintaining agricultural fields that are currently occurring under the No Action Alternative. The introduced traits in GE DBMs do not encode for any known allergens or toxins, and GE DBMs are not anticipated to persist because they cannot overwinter in the action area. Horizontal gene transfer of DNA from GE DBMs to wildlife that may consume them is also unlikely. Therefore, impacts on wildlife would be similar to the No Action Alternative.
Plant Communities	Under the No Action Alternative, the plant community within the action area will continue to generally consist of planted crops (cruciferous and non-cruciferous) and weeds of those planted crops. As a result of this simplified agricultural ecosystem, planted crops will continue to be potentially harmed by pests and weeds, and growers will continue to manage populations of pests and weeds.	The permitted field release of GE DBMs is not anticipated to change common agricultural practices currently used on fields as described for the No Action Alternative. Adult DBMs do not damage plant tissues and DBM larvae only feed on cruciferous plants. Damage from GE DBM larvae on planted cruciferous plants is not anticipated to be substantial because of the ubiquity of non-GE DBMs in the action area and their ability to persist within the action area. Damage from GE DBM larvae on cruciferous weeds is also not anticipated to be substantial because they are likely to be managed through cultural or chemical methods, so any damage from GE DBMs to

Attribute / Measure	Alternative A: No Action Alternative Deny the permit request	Alternative B: Preferred Alternative Grant the permit request
		cruciferous weeds is likely to be less than that from deliberate efforts to control them. Therefore, the impact to plant communities would be similar to the No Action Alternative. Cruciferous plants do not pose a risk of entering or contaminating the food supply because: no harvesting or movement of plants/plant materials that can function as hosts for DBMs can be moved from the proposed release site and isolation perimeter unless they are double bagged before transiting to a secure laboratory within a quarantine containment facility, where they will eventually be destroyed prior to disposal, and none of the plant parts or other derivatives of crucifers capable of supporting DBMs will be used for food or feed. In New York crucifer production, it is common practice to destroy crop debris following harvest by plowing it under to kill eggs and larvae of DBMs and other insect pests (Extension and Markets 2015). DBMs cannot develop at temperatures below 2.1°C. (35.8 °F) (Bahar et al. 2014). Since average annual low temperatures are below this threshold for Geneva, New York during the months of November-March (Data 2016), this indicates that DBMs are highly unlikely to overwinter in Geneva, New York.
Biological Diversity	Under the No Action Alternative, biological diversity within the action area is reduced and will continue to be reduced when compared to environments that are less intensively managed.	The permitted field release of GE DBMs is not anticipated to change common agricultural activities related to preparing and maintaining agricultural fields already used as described for the No Action Alternative. Therefore, impacts to biological diversity from common agricultural activities would be similar to the No Action Alternative.

Attribute / Measure	Alternative A: No Action Alternative Deny the permit request	Alternative B: Preferred Alternative Grant the permit request
		The release of GE DBMs is not anticipated to substantially affect biological diversity because non-GE DBMs are already targeted for management/control in the action area, so both non-GE and GE DBMs are unlikely to persist within the action area after the end of a growing season.
Human Health Environment		
Human Health	No changes are anticipated to currently-adopted agricultural activities under the No Action Alternative. As a result, human exposure (e.g., to farmworkers or the general human population) from risks and hazards as a result of these common agricultural activities is also anticipated to continue occurring under the No Action Alternative. A variety of EPA-approved pesticides would continue to be used for pest management within the action area. Use of registered pesticides in accordance with EPA-approved labels protects human health and worker safety. EPA also establishes tolerances for pesticide residue that give a reasonable certainty of no harm to the general population and any subgroup from the use of pesticides at the approved levels and methods of application.	The permitted field release of GE DBMs is not anticipated to change common agricultural activities related to preparing and maintaining an agricultural field that is currently occurring under the No Action Alternative. Therefore, impacts to human health (e.g., farmworkers and the general human population) from common agricultural activities would be similar to the No Action Alternative. Cruciferous plants do not pose a risk of entering or contaminating the food supply because no harvesting or movement of plants/plant materials that can serve as hosts for DBMs can be moved from the proposed release site and isolation perimeter unless double bagged for secure transiting to the laboratory within the APHIS-regulated quarantine containment facility for examination before eventual destruction and disposal in accordance with APHIS regulations; no plant/plant materials that can serve as hosts for DBMs can be used for food or feed. Previous NEPA documents,(USDA-APHIS 2008a, 2011a), have analyzed and concluded that there is no unreasonable risk to humans associated with the introduced traits in the GE DBMs described in the permit application. These GE DBMs also do not differ otherwise taxonomically from naturally occurring DBMs, which belong to the lepidopteran Family

Attribute / Measure	Alternative A: No Action Alternative Deny the permit request	Alternative B: Preferred Alternative Grant the permit request
		Plutellidae, which is a group (taxon) that is not known to cause any allergic reactions in humans. Therefore, these GE DBMs are not anticipated to substantially affect human health differently from what may occur under the No Action Alternative.
Compliance with Other Laws		
CWA, CAA, EOs	Fully compliant	Fully compliant

FINDING OF NO SIGNIFICANT IMPACT

The analysis in the EA indicates that there will not be any individual or cumulative significant impacts on the quality of the human environment as a result of this proposed action. APHIS agrees with this conclusion and therefore finds that an EIS need not be prepared. This NEPA determination is based on the following context and intensity factors (40 CFR 1508.27):

Context – The term "context" recognizes potentially affected resources, as well as the location and setting in which the environmental impact would occur. This action would be limited to the environmental release of a GE DBMs on the single release site described in the permit application 16-076-101r (see Section 2.4 of the EA). The action area is contained within the boundaries of the NYSAES in Geneva, NY. The NYSAES itself consists of 870 acres located in the northwestern corner of the boundary encompassing Geneva, NY, approximately two miles from suburban/urban population center of the city. The proposed field releases have limited potential to affect resources outside of the field test sites. Permit conditions in 7 CFR part 340.4 and the supplemental permit conditions applied to this permit will effectively prevent any potentially adverse environmental impacts associated with the permitted field release of GE DBMs.

Intensity – Intensity is a measure of the degree or severity of an impact based on these ten factors. These following factors were used as a basis for this decision:

1. *Impacts that may be both beneficial and adverse.*
   According to the applicant, GE DBMs may serve as an insecticide-free means of controlling field populations of DBMs in a species-specific manner. Issuance of the field release permit by APHIS would allow research to assess the reduction of pest populations of non-GE DBMs. The release of GE DBM strain OX4319L-Pxy on sites within the boundaries of the NYSAES will allow the applicant to obtain data on performance of the GE DBMs. The field release will not have any impact on existing agricultural practices because the sites are solely for research purposes. Therefore, current agricultural practices will essentially remain unchanged.

2. *The degree to which the proposed action affects public health or safety.*
   The proposed action to issue the APHIS field release permit should not pose a risk to human health and therefore would have no significant impacts on human health. GE DBMs are not used for food or feed purposes and do not contain any GE pesticides or traits for resistance to herbicides. However, at the conclusion of each experiment, the release sites will be devitalized of any remaining DBMs by applying an EPA-registered insecticide, Coragen (chlorantraniliprole). Post-experiment monitoring of DBMs with traps will continue until no GE DBMs are recaptured for two consecutive weeks. Adherence to EPA label instructions will ensure that only negligible impacts occur as a result of pesticide use. Potential adverse impacts to public health or safety as a result of approving field releases of GE DBMs are negligible.

3. *Unique characteristics of the geographic area such as proximity to historic or cultural resources, park lands, prime farmlands, wetlands, wild and scenic rivers, or ecologically critical areas.*

This action would be limited to the environmental release of GE DBMs on a single release site described in the permit application 16-076-101r (see Section 2.4 of the EA). The action area is contained within the 870-acre boundary of the NYSAES in Geneva, NY. The field release has limited potential to affect resources beyond this action area. Issuing the permit for GE DBMs is not expected to impact unique characteristics of geographic areas such as park lands, prime farmlands, wetlands, wild and scenic areas, or ecologically critical areas. As analyzed in the Environmental Consequences section of the EA, no different agronomic activities within the action area are anticipated as a result of the Preferred Alternative. If the permit is issued, field releases will occur on land already under agricultural management, and they are not expected to alter land use patterns within the action area.

There are no proposed major ground disturbances; no new physical destruction or damage to property; no alterations of property, wildlife habitat, or landscapes; nor any prescribed sales, leases, or transfers of ownership of any property. This action is limited to issuing a permit to release GE DBMs until the end of 2017. This action would not convert land use to non-agricultural use and, therefore, would have no adverse impact on prime farmland. Standard agricultural practices for land preparation, planting, irrigation, and harvesting of plants would be used on agricultural lands planted under the Preferred Alternative, including the use of EPA-registered pesticides that will not differ from those used currently under the No Action Alternative. The inability of DBMs to overwinter in the action area and devitalization procedures required by permit conditions will ensure that any GE DBMs remaining at the conclusion of the growing season will not persist into the following season (see Section 3.2 of the EA).

Based on these findings, including recognition that EPA-label use restrictions are in place to protect unique geographic areas and that those label use restrictions will be adhered to, issuing a permit for the field release of GE DBMs is not expected to impact unique characteristics of geographic areas such as park lands, prime farm lands, wetlands, wild and scenic areas, or ecologically critical areas.

4. *The degree to which the effects on the quality of the human environment are likely to be highly controversial.*

The effects on the quality of the human environment are not highly controversial. Although there is opposition to APHIS issuing this field release permit, this action is not highly controversial in terms of size, nature or effect. This action would be limited to releases within the action area described in permit application 16-076-101r (see Section 2.4 of the EA). The action area is entirely encompassed by the boundaries (870-acre perimeter) of the NYSAES in Geneva, NY. The public comments received by APHIS in response to its EA did not establish any specific, substantial, factual discrepancies or other concerns about the data provided and analyzed by APHIS in its EA to support approval of this permit application. The Agency's responses to public comments about issues analyzed in its EA are included as an attachment to this FONSI.

5. *The degree to which the possible effects on the human environment are highly uncertain or involve unique or unknown risks.*

The effects of the proposed action to issue the APHIS field release permit are not highly uncertain and do not involve unique or unknown risks. Based on the analysis documented in the EA, the effects on the human environment would not be significant. APHIS does not anticipate any unknown risks to be associated with GE DBM strain OX4319L-Pxy, when

released into the environment. The field release of GE DBMs is not likely to present any unforeseen risks. Based on the analysis and information provided in the EA and supporting permit application, the new genes that are engineered into the specified GE DBM strain should not pose significant risks associated with field release. Adherence to the permit conditions by the applicant will effectively prevent any potential adverse impacts to the human environment.

6. *The degree to which the action may establish a precedent for future actions with significant effects or represents a decision in principle about a future consideration.*
The proposed action would not establish a precedent for future actions with significant effects or represent a decision in principle about a future decision. Similar to past permit applications reviewed and approved by APHIS, this decision on whether or not to issue a permit for environmental release will be based upon information provided in the permit application and the best available science. APHIS regulations at 7 CFR part 340 regulate the introduction (importation, interstate movement, and release into the environment) of certain GE organisms and products. In accordance with these regulations, when APHIS receives an application for a permit for importation, movement or environmental release, the application is evaluated to determine whether the importation, movement or environmental release with appropriate conditions imposed, can be authorized, while preventing the dissemination and establishment of plant pests. The applicant has provided the information associated with this request in the permit application, and APHIS must determine to either approve or deny the permit. Each permit application that APHIS receives undergoes this independent review to determine if APHIS should approve or deny the individual permit.

7. *Whether the action is related to other actions with individually insignificant but cumulatively significant impacts.*
No significant cumulative effects were identified during the analysis performed for the EA. As discussed in the cumulative effects analysis presented in the EA, APHIS has determined that there are no past, present, or reasonably foreseeable actions that would have aggregate effects from the proposed action that would result in cumulative impacts or reduce the long-term productivity or sustainability of any of the resources (soil, water, ecosystem quality, biodiversity, etc.) associated with the release sites or the ecosystem in which they are situated. No significant cumulative impacts were identified that would result from the proposed action.

8. *The degree to which the action may adversely affect districts, sites, highways, structures, or objects listed in or eligible for listing in the National Register of Historic Places or may cause loss or destruction of significant scientific, cultural, or historical resources.*
This action would be limited to the environmental release of a GE DBM strain within the NYSAES as described in permit application 16-076-101r (see Section 2.4 of the EA). The action area is contained within the NYSAES in Geneva, NY. The field release has limited potential to affect resources outside of field test sites. APHIS' proposed action, issuing a permit for one year to release GE DBMs, is not expected to adversely impact cultural resources on tribal properties. APHIS' Preferred Alternative would have no impact on districts, sites, highways, structures, or objects listed in or eligible for listing in the National Register of Historic Places, nor would it likely cause any losses or destruction of significant scientific, cultural, or historical resources. This action is limited to issuing a permit limited to one year for the field release of GE DBMs.

APHIS' proposed action is not an undertaking that may directly or indirectly cause alteration in the character or use of historic properties protected under the National Historic Preservation Act (NHPA). In general, common agricultural activities associated with this action do not have the potential to introduce visual, atmospheric, or noise elements to areas in which they are used that could result in effects on the character or use of historic properties. For example, there is potential for increased noise on the use and enjoyment of a historic property during the operation of tractors and other mechanical equipment close to such sites. An inherent mitigating factor for this issue is that virtually all of the methods involved would only have temporary effects on the audible nature of a site and can be ended at any time to restore the audible qualities of such sites to their original condition with no further adverse effects. These cultivation practices are also currently being conducted throughout the action area. This permit, limited to one year for field release of GE DBMs, is not expected to change any of these agronomic practices that would result in an adverse impact under the NHPA.

9. *The degree to which the action may adversely affect an endangered or threatened species or its habitat that has been determined to be critical under the Endangered Species Act of 1973.*
   APHIS evaluated the potential for negative effects on federally listed threatened and endangered species as listed by the U.S. Fish and Wildlife Service from the issuance of the field release permit with associated permit conditions and concluded that there would be no effect on federally listed threatened or endangered species or species proposed for listing, or on designated critical habitat or habitat proposed for designation *(see* section on Threatened and Endangered Species in the EA).

10. *Whether the action threatens a violation of Federal, state, or local law or requirements imposed for the protection of the environment.*
    The proposed action would be in compliance with all Federal, state, and local laws. The proposed action to issue the APHIS field release permit would be implemented in accordance with 7 CFR part 340. GE DBMs are not used for food or feed purposes and do not contain any genetically engineered pesticides or traits for herbicide resistance. Therefore, consultations with FDA and EPA are not required. At the conclusion of each experiment, the release sites will be devitalized of any remaining DBMs by application of the EPA-registered insecticide, Coragen (chlorantraniliprole) in accordance with EPA label instructions. There are no other Federal, state, or local permits that are needed prior to the implementation of this action.

NEPA Decision and Rationale

I have carefully reviewed the EA prepared for this NEPA determination and the input from the public involvement process. I believe that the issues identified in the EA are best addressed by selecting the preferred alternative - Issue the APHIS Permit.

As stated in the CEQ regulations, "the agency's Preferred Alternative is the alternative which the agency believes would fulfill its statutory mission and responsibilities, giving consideration to economic, environmental, technical and other factors." The Preferred Alternative has been selected for implementation based on consideration of a number of environmental, regulatory, and social factors. Based upon our evaluation and analysis, Alternative B is selected because (1)

it allows APHIS to fulfill its statutory mission to protect America's agriculture and environment using a dynamic and science-based regulatory framework that allows for the safe development and use of genetically engineered organisms; and (2) it allows APHIS to fulfill its regulatory obligations. Therefore, it is my decision to implement the preferred alternative as described in the EA. Based on all of the analysis and reasons above, I have determined that there would be no significant impact to the quality of the human environment from the implementation of the chosen alternative (the Preferred Alternative to issue the APHIS permit) and therefore, no EIS needs to be prepared. As such, APHIS will issue this permit to allow the field releases of a GE diamondback moths for scientific research, as described in the permit application, that are engineered to reduce pest populations of non-GE DBMs within the boundaries of the New York State Agricultural Experiment Station in Geneva, New York.

M. J. Firko

7/6/2017

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

Date

Summary of Comments Received from the Public for the Draft Environmental Assessment

I. Introduction

On April 19, 2017, APHIS published a notice in the *Federal Register* (82 FR 18416-18417) announcing the availability of the draft EA for a 30-day public review and comment period. Comments were accepted from the public until May 19, 2017, 11:59 PM ET. APHIS received just over 670 individual comments during the 30-day comment period for the DBM draft EA. The Agency expresses thanks to all those who participated in the public involvement process by reviewing the draft EA for the GE DBM permit request and providing comments. APHIS welcomes public involvement and considers public perspectives and input in its decision-making process.

APHIS evaluated all issues identified in comments received for the draft GE DBM EA. The Agency's responses to substantive comments opposing the field testing follow. Most of the comments opposing focused on the genetic modification of GE DBM, while other comments supporting the field test addressed issues related to the plant pest risk DBM poses to U.S. agriculture, and the efficacy GE DBM may provide in management of DBM. For matters of efficiency, the comments received are summarized by topic area, as provided by the regulations at 40 CFR §1503.4. Comment summaries are designed to efficiently address the salient topics identified. A full record of each comment received for the draft EA is available for public review at: www.regualtions.gov, Docket ID: APHIS-2014-0056.[5] The major themes in the comments opposing the field trial are:

- **Topic 1:** EIS versus EA
- **Topic 2:** Length of the Comment Period
- **Topic 3:** Containment of GE DBM – Potential Escape of Transgenic Insects into the Environment
- **Topic 4:** Impacts on Human Health and Non-Target Organisms
- **Topic 5:** Antibiotic Resistance
- **Topic 6:** Horizontal Gene Transfer to Other Species
- **Topic 7:** Unintentional Survival of Female GE DBM
- **Topic 8:** Response of Non-GE DBM and Non-Target Plant Pests to Release of GE DBM
- **Topic 9:** Potential Socioeconomic Impacts

Many of the comments supporting the proposed field testing focused on aspects of GE DBM considered beneficial, such as: that GE DBM may help control DBM via biological control methods; reduce the use of pesticides; help reduce the development of pesticide resistance in

[5] http://www.regulations.gov/#!docketDetail;D=APHIS-2014-0056

DBM populations; and reduce adverse impacts of synthetic chemicals on pollinators and natural enemies of DBM and other plant pests.

As evident in the topics identified, the substantive comments received addressed a broad range of topics relevant to the analysis of potential environmental, human health, and socioeconomic impacts. None of the comments, however, required significant revision of the draft EA.

II. Summary of Responses to Comments Opposed to Proposed Field Releases of GE DBMs

Topic 1: EIS versus EA

Several commenters stated that an Environmental Impact Statement (EIS) is needed, rather than the Environmental Assessment (EA) conducted.

APHIS carefully considered and analyzed the possible environmental impacts of the proposed action and determined that none of them met the criteria of significant impacts, which is the NEPA requirement for completing an EIS. The Agency is satisfied that the EA for the requested permit for field release of GE DBM is sufficient. APHIS considered comments and data submitted by those who supported or opposed the proposed permitted field release, the peer-reviewed scientific literature cited in the EA, technical reports, and the standard and supplemental permit conditions developed for the proposed field tests.

APHIS evaluated two alternatives in the EA: (1) No Action Alternative (do not issue a permit); and (2) Preferred Alternative (issue the APHIS the permit consistent with the Agency's statutory authority under the plant pest provisions of the Plant Protection Act, and the regulations at 7 CFR part 340). Based on current peer reviewed literature cited in the EA and permitting requirements developed for the proposed releases, APHIS concluded that the permitted field release is unlikely to result in significant impacts to the quality of the human environment. Consequently, it is unnecessary to prepare an EIS for the proposed field releases of GE DBMs as described in the permit application and consistent with APHIS-imposed permit conditions.

Topic 2: Length of the Comment Period

Some commenters stated that the public comment period for the draft EA should have been longer than 30 days.

APHIS believes that a 30-day comment period is sufficient for public review and comment on the draft EA because the Agency determined that the proposed DBM releases would not pose a plant pest risk greater than any existing one associated with DBMs, and the releases would continue to be regulated under the terms and conditions of an APHIS permit. Following the comment period, the Agency thoroughly reviewed the comments and carefully considered other inputs as it prepared the final EA. Because APHIS did not identify any significant impacts from the proposed field testing of GE DBM, it prepared this FONSI.

Topic 3: Containment of GE DBM – Potential Escape of Transgenic Insects into the Environment

Concerns were expressed regarding containment of GE DBM during field testing, and their potential long-distance dispersal. For instance, some commenters emphasized that windblown moths can be dispersed hundreds to thousands of miles from where they emerge. Some of these comments also expressed concern that GE DBMs might migrate into areas where they may overwinter.

APHIS recognizes and understands these concerns. As described in EA Section 3.2, EA Action Area, and Section 4.2, the Preferred Alternative – Issue the APHIS Permit, conditions imposed by the Agency combined with additional factors to confine field releases of GE DBM (Section 3.2, EA Action Area), ensure that GE DBM will not persist in the environment or cause a significant environmental impact. The proposed releases would be at the New York State Agricultural Experiment Station (NYSAES) in Geneva, NY. The duration for making releases would be limited to 2017 on an experimental field limited to ten acres, where releases would be made at a single point within the field; the 10-acre field and accompanying release point may move to another field within the NYSAES in subsequent growing seasons, if the permit is renewed, due to crop rotation practices. The applicant would release up to 10,000 male GE DBM per release (up to 30,000 males per week). Populations of GE DBM are unlikely to persist and disperse beyond the release site for several reasons.

Dispersal

Dispersal via human clothing or animals was cited as a potential dispersal mechanism. APHIS has addressed these concerns in the EA, "Adult moths do not purposely alight on and use vertebrates for dispersal, and are likely to fly off of/away from any human or wildlife that may come into physical contact with it in the proposed release site. Thus, the dispersal of GE DBM adults through contact with humans or wildlife is unlikely. Furthermore, as an added precaution, permit conditions require staff to visually inspect themselves and their clothing for incidental hitchhiking moths before leaving the release area and field cages."

Dispersal via the food chain was cited as a potential dispersal mechanism. APHIS has accounted for this in the permit conditions: "Permit conditions mandate that any cruciferous crops planted at the proposed release site is intended for research purposes and are prohibited from entering food and feed product streams; furthermore, permit conditions mandate that the planted cruciferous crops will not be harvested or moved." Furthermore, the Supplemental Permit Conditions (SPCs) under Field Test Termination (EA, pg. 27) states: "This is a crop-destruct trial. The host material planted at the release site and in the cages will be treated as regulated material. No plants/plant materials that can function as hosts for DBM can be moved from the proposed release site and isolation perimeter other than in double contained bags transported to the secure laboratory for examination and eventual destruction via freezing and/or autoclaving to render any insects non-viable. No plant/plant materials that can function as hosts for DBMs can be used for food or feed.

On or before the expiration of the permit, the field test must be terminated by treating the release site out to the 10 m buffer and the caged areas with an insecticide to kill any existing DBMs. All

plants within the release site and in the cages must be devitalized by disking into the ground. Cages must not be removed until after insecticide treatment and devitalization of host plants within the cages are completed."

Additional safeguards have been developed by APHIS so as not to attract DBMs to areas that are out of the release area. The second SPC states that: "A 10-meter buffer of bare ground, maintained by weekly disking, must be maintained around the perimeter of the open release site. The buffer must be surrounded by an additional 50 meters that, excepting cages, must not be planted with crops that can act as a host for diamondback moth and any substantial clusters of plants that could serve as hosts must be eliminated. Host plants may be planted in cages used for cage experiments located within the 50 meter area. No caged releases can occur within the 10 meter buffer or additional 50 meter zone at the same time that the open field release is being conducted."

These measures are enacted to limit any odorant sources from host plants or calling (pheromone-emitting) females that could attract GE DBM away from the release area, which is defined as a field of Brassica host plants in which the center of the ~100m radius is the actual release point.

The results of Shirai & Nakamura (1994) are consistent with the SPCs that have been developed. The authors found that, of the DBM recaptured, 0.3-0.5% were recaptured outside of the release field (but inside fields of different host plants); the percentage reported refers to a pooled value that reflects a range of distances from 150m to 800m. Finally, it is worth noting that this study used pheromone traps, which are developed to attract male moths. These data are consistent with Mo et al. (2003), who estimated using pheromone traps that <5% of DBM males disperse to 110m and <1% are expected to disperse past 200m. These estimates likely represent a high-end expectation of dispersal because in their dataset derived from using yellow sticky traps containing no pheromone, the dispersal estimates were approximately halved (Mo et al. 2003).

APHIS recognizes that under extraordinary weather conditions, strong winds may disperse DBMs over long distances. However, as discussed in the EA, this is considered unlikely. In addition, APHIS has implemented a SPC that if a hurricane or similar wind event is predicted, APHIS will be notified and the field trial will be terminated in advance.

Taking all of the above into account, APHIS believes it is highly unlikely that GE DBM will disperse from the field site.

Overwintering

Concerns about dispersal were also tied to concerns about overwintering ability, with regards to off-site dissemination. In particular, a commenter cited the study of (Nguyen et al. 2014), which tested thermal tolerance and that lower lethal temperature for adult moths was -16.5C (~2°F). However, it should be noted that cold treatments were only 2 hr long. (Honda 1990) tested the mortality of DBM after chilling at 0°C for various amounts of time. All immature stages died by 60 days at 0°C (32°F). Though up to 10% of adults survived 60 days at 0°C, none of the female were able to lay any eggs. On the other hand, (Gu 2009) found 20% survival of adult DBM when kept at fluctuating temperatures between 0° and 5°C for 60d and ~30% at -5°C (23°F) for 20d; these moths largely were able to reproduce normally. The same study found that all immature

stages of DBM died after 15d at -5°C (23°F). (Liu et al. 2002) found that no DBM eggs survived more than 55d at 4°-6°C (39°-43°F).

Local climatological data can be found on the National Weather Service website (NOAA 2016). All data reported here are from the years 2000 to 2017. The average low temperature in January for Geneva NY is 16.7°F, or -8.5°C; the average lowest temperature across the month of January is -3°F (-19°C). Examining daily winter temperatures shows that 15 out of the last 18 years have had at least two days at 1°F or below; only 3 out of 18 years did not hit 2°F. Finally, the average first and last freeze for Geneva is October 21 and April 28, respectively. Though temperatures will fluctuate above freezing between late October and late April, during this period very limited, if any, DBM development can occur.

These data indicate that both immature and adult DBM stages are highly unlikely to survive winter conditions at the release site. This is consistent with other reports that DBM are unlikely to overwinter in the action area. In particular, in a lab and field study, Youjun (2010) (abstract only) concluded that DBM cannot overwinter where temperatures are 0°C for 28 days or 5°C for 42d. Butts and McEwen (1981) were not able to recover any surviving DBM (immature or adult stage) in their study of overwintering ability conducted in a field in Ontario Canada. However, Idris (1996) found some overwintering of immature stages, but not adults, in Michigan field conditions. The authors noted that survival of some larvae was likely due to sheltered larvae in crop debris, and in their conclusions, they highlighted the importance of tilling the previous crop in order to destroy potentially overwintering larvae (Idris 1996). As discussed above, one of the permit conditions is that the *Brassica* crop will be tilled (by disking into the ground) and destroyed where the field trial will take place. This measure is commonly done in many other crops to reduce the number of pest insects that could survive the winter.

At least one commenter asserted that DBMs were able to survive winter conditions in western Canada, but this reference was incorrectly cited. Indeed, the preliminary study conducted by (Dosdall 1994) suggests that DBMs can overwinter successfully in Canada, but a later and more comprehensive study by the same author showed no findings of successfully overwintering DBMs over a period of six years (Dosdall et al. 2001).

Hence, based on the data summarized above, no life stage of DBM is likely to survive winter conditions at the release site.

Pest Control

The persistence of any DBM population, genetically engineered or non-genetically engineered, is largely dependent on the presence of host cruciferous plants, whether wild (not planted) or crop plants. It is likely that any cruciferous crop outside the potential field release area would be actively managed, utilizing a variety of integrated pest management (IPM) practices for the purposes of DBM control(if they are present) as well as control of other pests of cruciferous crops (e.g., cabbage looper, cabbageworm, flea beetles). Any GE DBMs or their progeny that potentially disperse to these cruciferous crop fields outside the potential field release area will be subject to these IPM practices, thereby encountering a substantial obstacle to their persistence and spread.

Permit Conditions

Permit conditions require that potential dispersal of regulated GE DBMs within and outside the perimeter of the release site be monitored. If the weekly mark/recapture data indicate that the likelihood of GE DBM dispersal to areas outside boundary of 10 m buffer is greater than anticipated, APHIS must be notified immediately by both phone and in writing. If a hurricane (or similar meteorological event) is projected to affect the release site, no regulated moths may be released within one week prior to the event or the release site must be treated with an EPA-approved insecticide not less than two days before the anticipated weather even to kill any existing regulated moths.

At the conclusion of each experiment, the release site will be devitalized of any remaining DBM by application of an EPA-registered insecticide. Continuous monitoring for GE DBMs after the end of the experiment will be maintained during periods conducive to their survival and development until no GE DBMs are detected for two consecutive months. Based on the permit conditions that are reviewed above, dispersal and persistence of GE DBM in the environment, and adverse ecological impacts from the proposed field releases are considered improbable.

Topic 4: Impacts on Human Health and Non-Target Organisms

Concern was expressed about the potential toxicity of GE DBM when ingested. Some expressed concern regarding the accidental consumption of dead GE DBM adults or larvae that may be present on brassicaceous vegetables harvested for food, and that the death of most female GE moths at the larval stage will significantly increase the number of larvae dying in brassicaceous crops and wild related species including brassicaceous weeds. There was specific concern regarding the safety of DsRed2 and tTAV protein consumption.

Many commenters expressed concern that when animals such as birds, bats, rodents, and other insects consume GE DBMs, introduction of the GE trait will negatively impact them. Several commenters stated that this can alter an animal's digestion, reproduction, general health and longevity, and thereby affect future generations. Some examples of question related to this topic included:
"What is the fate of these dead GE larvae in the environment?"
"Of the trait genes and gene products in soil, what are the effects on the associated biota (soil biota, plants, other insects, birds), and animals that eat the brassicas [sic]?"
"It seems that this groundwork must be completed before open-air tests commence."

Substantial discussion on the potential impact on non-target organisms was presented in the EA in Section 5.3.2 about Wildlife, Plant Communities, and Biological Diversity for the Preferred Alternative; Chapter 6, Cumulative Impacts; and Chapter 7, Threatened and Endangered Species. Discussion of the potential impact on the human health environment can be found in Section 5.4.

APHIS understands these concerns, and clarifies that the risk to human health from the incidental consumption of dead larvae and adults on brassicaceous vegetables is considered negligible. This is because the proposed permitted field testing would be classified by APHIS as a crop-destruct trial. The host material, other brassicaceous species and any other plants within the release site and from within cages will be classified as regulated material. No plants/plant materials that can

function as hosts for DBMs can be moved from the proposed release site and the isolation perimeter unless they are contained in double bags before being transported to the secure laboratory within the APHIS-regulated quarantine facility for examination prior to eventual destruction by freezing and/or autoclaving to render any insects non-viable. No plant/plant materials that can function as hosts for DBMs can be used for food or feed.

When testing is terminated, the release site and surrounding 10 m buffer and the caged areas must be treated with an insecticide to kill any existing DBMs. All plants within the release site and in the cages must be devitalized by disking into the ground. Cages cannot be removed until after insecticide treatment and devitalization of host plants within the cages are completed. The site must be monitored for the presence of regulated moths no less than once every two weeks until two consecutive months, when temperatures are conducive for DBM survival and development, are confirmed to be free of any regulated moths. Therefore, plants at the field site do not pose a risk of entering or contaminating the food supply because no harvesting or movement of plants/plant materials that can function as hosts for DBMs can be moved from the proposed release site and perimeter buffer unless they are contained in double bags prior to transporting to the secure laboratory within the APHIS-regulated quarantine containment facility for examination before eventual destruction. No plant/plant materials that can function as hosts for DBMs can be used for food or feed. Consequently, while inadvertent consumption of dead larvae, or biological material from dead larvae of adults on brassicaceous vegetables is theoretically possible, it is highly improbable.

DsRed2 is a reporter gene/gene product that was previously examined in a prior APHIS EA (USDA-APHIS 2011b). The similar DsRed gene/gene product (DsRed2 is derived from DsRed) was examined by APHIS in a prior EIS (USDA-APHIS 2008b). Based on these analyses APHIS determined that the DsRed2 gene/gene product and its parent sequence, DsRed, did not resemble an allergen or toxin, and posed no detectable health hazards to animals or humans. This determination was supported by the FDA's evaluation, which concluded that the DsRed2 gene/gene product posed no food or feed risk (e.g., see (US-FDA 2016). The tTAV gene/gene product is responsible for the female-specific lethality in GE DBM. Like DsRed2, tTAV was previously examined by APHIS in a prior EIS (USDA-APHIS 2008b) and was determined to have no resemblance or similarity to any known allergens or toxins, nor pose an unreasonable risk to animal and human health. (Nordin et al. 2013) examined the effect of DsRed2 and tTAV genes/gene products on two predatory mosquito species, *Toxorhynchites splendens* and *Tx. amboinensis*, after they had consumed mosquito larvae transformed with DsRed2 and tTAV. No significant negative effect was observed in either insect predator in this study.

Studies were referenced in the public comments that tTAV expression could have adverse effects in humans or wildlife that may consume GE DBM. However, these articles are about transgenic mice that are used as models for human disease. It should be noted that the transgenic mice in these studies cannot be directly used to assess potential toxicity from a route of exposure due to consumption. If a human or animal were to ingest GE DBM, the tTAV would be subjected to the gastric digestion system. Numerous studies of proteins produced by GE crops have been submitted to the U.S. EPA using a protocol that tests degradation in simulated gastric fluid (Metcalfe et al. 1996; Thomas et al. 2004). All studies have indicated that the protein is readily degraded by incubation in simulated gastric fluid. The probability of the protein surviving the digestive system is therefore extremely low. Based upon the rapid break-down of protein in

studies submitted to the U.S. EPA, and that no allergen or toxin similarity of the introduced gene in DBM is present, APHIS does not anticipate affects upon non-target organisms that would eat brassica or ingest the GE insects. As stated the EA, the intentional or unintentional consumption of introduced gene and gene products is not likely to have any significant impact on wildlife or human health.

In addition to the protein surviving the digestive system, the tTAV would also need to cross the gut barrier, enter the bloodstream and finally cross the cell membrane. However, proteins are not generally membrane permeable (Biswas 2011). Furthermore, components of the tTAV system do not readily cross the cell membrane either (Ye 2002; Mortlock 2003). Therefore, given the low probability of tTAV to survive the digestion in gastric fluid, in addition to barriers to membrane permeability, APHIS concludes that tTAV is highly unlikely to affect human or animal health.

Any transient increase and subsequent decrease in the DBM population is not anticipated to substantially affect insectivore populations or predator-prey dynamics among populations at the test site. Most insectivores are generalist predators, with few exhibiting a strong preference for DBM as prey (e.g., (Clare et al. 2009)). Hence, effects on insectivorous non-target species as a result of transient increases and decreases in DBM populations are highly unlikely. Dead DBM larvae, pupae, and adults would simply decompose as other Lepidoptera do. Decomposers (saprotrophs) such as bacteria, fungi, and earthworms would contribute to the degradation of proteins and genetic material, and recycling elemental nutrients like carbon and nitrogen.

As part of issuing any permit, APHIS requires reporting of any unintended effects. Pursuant to 7 CFR § 340.4(f)(10)(ii), APHIS shall be notified in writing as soon as possible if the regulated article or associated host organism is found to have characteristics substantially different from those listed in the permit application or suffers any unusual occurrence (excessive mortality or morbidity, or unanticipated effect on non-target organisms).

Based on the above factors, adverse effects of DsRed2 and tTAV genes/gene products on predators of DBM is considered improbable. Substantial effects on biological control agents exposed to GE DBMs at the experimental site are also unlikely. DBM are actively controlled worldwide on annual basis using synthetic chemicals and organic control methods. Control of DBM pest populations at the test site via GE DBMs would be equivalent (if effective) to control DBM pest populations (and other lepidopteran pests) using synthetic chemicals and organic methods by suppressing DBM numbers to a level that is below the threshold that causes economically significant crop damage. Populations of parasitic wasps, ladybugs, lacewings, spiders, and birds, all natural predators of DBMs, are unlikely to be negatively affected by the 10-acre field tests.

Topic 5: Antibiotic Resistance

Several commenters expressed concern that field release of GE DBM could promote the spread of tetracycline-resistant bacteria within and around release environments, and present a risk to human health. Some comments, in particular, expressed concern that the field trial would "endanger the human population with resistant bacterial infections whether the genetically modified insects are mosquitoes or moths."

Other specific comments on this topic were:

"The use of tetracycline to breed the GE diamondback moths in the lab carries the risk of spreading antibiotic resistance, which could pose a major risk to human and animal health. This is because insect guts are reservoirs for antibiotic resistance genes which can be spread into the environment."

"GE insect production in factories exposed to antibiotics could lead to drug resistance in bacteria in their guts so that the insects disseminate antibiotic resistance when released into the environment."

One commenter listed several questions:

"What are the implications of using tetracycline in this research?"

"How will the materials used in breeding the insects be disposed of?"

"What are the implications of releasing tetracycline-fed insects into the field?"

"Will resistance be spread through gut bacteria? These questions must be answered before open-air trials commence."

Antibiotic-producing and antibiotic-resistant bacteria occur naturally in soils, independent of human activities (Allen et al. 2010; Durso et al. 2016). For example, it was found that among naturally occurring enterobacterial strains collected from around the world before therapeutic antibiotics were widely used (between 1917 and 1952, known as the Murray collection), 11 strains were resistant to ampicillin or tetracycline (Hughes and Datta 1983; Allen et al. 2010). Soil bacteria naturally resistant to tetracycline have also been identified in areas removed from human activities (Allen et al. 2010; Qichun et al. 2016).

These and other studies demonstrate that, soils are a natural reservoir of antibiotic-producing bacteria; bacteria that are also comprised of transferable antibiotic-resistance genes and mechanisms (e.g., efflux pumps) (Allen et al. 2010). For instance, approximately 50% of bacteria in the order Actinomycetales isolated from soil and are capable of synthesizing antibiotics (Souza et al. 1999; Allen et al. 2010). Of the more common antibiotics in use, more than 90% originate from actinomycetes, which also account for about two-thirds of all known bioactive substances of microbial origin (Hamaki et al. 2005). Hence, soils are a reservoir of a natural, ancient antibiotic pool of biota with genes that confer antibiotic-resistance mechanisms that occur independently of human activities.

Intrinsic tetracycline resistance, including multidrug resistance to various antibiotics, has also been described in cave dwelling bacteria (Walsh and Duffy 2013). The antibiotic susceptibility of 93 bacterial strains isolated from the Lechuguilla Cave, New Mexico, was surveyed. This was a genetically diverse collection of organisms highly adapted to survive in a nutrient-limited environment. Like soil dwelling organisms, the majority of these strains were multidrug resistant indicating that antibiotic resistance is a common and widespread phenotype in pristine environments that are not-impacted by human activities, with bacteria exhibiting differences in the pattern of resistance (Walsh and Duffy 2013).

It is also true that there has been an increase in the relative abundance of antibiotic resistance genes for major antibiotic families (ß-lactams, tetracyclines, and macrolides) in contemporary

soil samples, when compared to those from the pre-antibiotic era, i.e., soils sampled before 1950 (Knapp et al. 2010). Development of antibiotic resistance is a modern phenomenon linked to the widespread human uses of antibiotics. For example, some of this is associated with the use of manures or other effluent from swine or poultry treated with antibiotics.

One study found that while chlortetracycline and chlortetracycline resistant bacteria could be detected in swine effluent, fertilization of agricultural fields with swine effluent at agronomically relevant rates did not necessarily lead to the development of antibiotic resistance in soil (Popova et al. 2017). Under dry semi-arid conditions, the survival rates of tetracycline resistant intestinal bacteria originating from swine effluent may be a critical factor in limiting transfer of tetracycline resistant genes to soil ecosystems (Popova et al. 2017). In contrast, bioavailability of chlortetracycline introduced into soil in swine effluent is hindered by its strong sorption to soil and chlortetracycline's low mobility within the soil profile (Popova et al. 2017). (Popova et al. 2017) concluded that in semi-arid ecosystems, the use of chlortetracycline containing swine effluent for fertilization of agricultural fields does not necessarily lead to the increase in the tetracycline resistance level in soils, when agronomically relevant amounts of swine effluent are used.

In a similar study, chlortetracycline-resistant bacteria were characterized in soils exposed to the manure of animals fed sub-therapeutic concentrations of antibiotics. These bacteria were compared to those chlortetracycline resistant bacteria from soils at farms with restricted antibiotic use (dairy farms) and from non-agricultural soils. No significant differences were observed at nine different study sites with respect to the numbers and types of cultivated chlortetracycline-resistant bacteria (Ghosh and LaPara 2007). The findings by (Popova et al. 2017) and (Ghosh and LaPara 2007) are consistent with the fact that tetracycline-producing bacteria (e.g., *Streptomyces* spp.) occur naturally in soils, independent of human activities (Allen et al. 2010; Durso et al. 2016), and consequently, other species of soil bacteria have naturally evolved tetracycline resistance (Durso et al. 2016).

The occurrence of antibiotic resistance is a serious problem when it occurs in human and animal pathogens and the resistance genes conveying the trait spread among populations of both pathogenic and non-pathogenic bacteria. This prompts an important question with regard to DBMs: do the gut bacteria of larvae and adult moths include pathogens or species closely related to pathogenic bacteria? The microbiota in DBM larvae, pupae, and adult has been characterized, and among the normal flora are *Enterobacter* spp., *Carnobacterium* spp., and *Escherichia coli* are the most common (Xia et al. 2017). Among these, *Enterobacter* spp. and *E. coli* are enteric bacteria common to many organisms, and some species are considered opportunistic pathogens in humans, usually associated with nosocomial and similar secondary infections.

Variations in the gut microbiome of *Plutella xylostella* can result from the development of resistance gut microbiota to pesticides (Li et al. 2017; Xia et al. 2017). Considering there are various mechanisms by which bacteria evolve resistance to synthetic chemical agents, it is in theory possible that the gut bacteria of *P. xylostella* may evolve (develop natural) tetracycline resistance if exposed to tetracycline, this relative to the dose, duration, and frequency of gut microflora exposure to the antibiotic. As soil bacteria produce tetracycline, and exhibit natural resistance to tetracycline (Souza et al. 1999; Allen et al. 2010; Durso et al. 2016; Qichun et al. 2016), any environmental introduction of tetracycline-resistant gut microbiota from dead DBM

would not necessarily present a novel challenge to soil communities, nor any novel mechanism of antibiotic resistance that could be transferred by horizontal gene transfer (HGT), e.g., genes encoding for efflux pumps; particularly in consideration of the relative contribution of bacterial mass from the moth gut to the variety and number of bacteria species present in soils. Relative to human risk, any concern about presumed tetracycline-resistant microbiota associated with DBM would be more reasonably limited to consumption of raw insects. This is not a common practice in the United States, and accidental ingestion is unlikely to occur. In comparing all possible risks from GE DBM, further theoretical assessments of these human risks would be highly speculative and beyond the practical scope of analysis required for the ten-acre field test being considered.

The field testing of male GE DBMs for control of wild DBM populations is not a feasible or foreseeable pathway for induction of tetracycline resistance into human or animal pathogenic bacteria. Antibiotic resistance is mainly associated with hospital/clinical environments that have utilized antibiotics for disease treatment, human populations that have been exposed to antibiotics and antibiotic-resistant pathogens, and food animal rearing operations that have utilized antibiotics for growth enhancement and therapeutic purposes. These primary sources, or pools, of antibiotic resistant bacteria developed over an extended periods (many years to decades). Crop production, which includes small scale and commercial operations, apart from use of manures containing antibiotic residue and antibiotic-resistant bacteria, is not a significant reservoir for or source of antibiotic-resistant bacteria that derive from human activities. This would be particularly true for the proposed NYSAES tests, because of their limited duration and small geographic scale (10 acres).

Considering these factors, the risk that GE DBM would exacerbate tetracycline resistance in bacteria of pathogenic relevance to humans and other animal populations is considered to be low. The proposed field releases are designed to test the effectiveness and safety of a method of controlling DBMs by releasing males genetically engineered to have a female autocidal trait. It remains to be determined if this technique for reducing the population of this pest species will prove effective and economically feasible. That is why the realistic testing under this application has been requested. For the one-year study proposed, the risk to human health as result of fostering the development of tetracycline resistance in bacteria is considered negligible.

Topic 6: Horizontal Gene Transfer to Other Species

Several commenters expressed concern regarding horizontal gene transfer (HGT) from GE DBM to other species, in general, and microorganisms in particular. HGT was also expressed as a concern associated with antibiotic resistance in microorganisms. This is discussed in more detail under Topic 6, above.

Horizontal gene transfer has arisen as one of the more controversial and theoretical risks regarding GE insects developed for crop protection and human disease vector control (e.g., the control of mosquitos that can transmit the Zika virus). While there is evidence for gene transfer from parasitic insects to vertebrates, these events have taken place over evolutionary timescales involving hundreds of thousands to millions of years (Lacroix and Citovsky 2016). Bioinformatic analyses indicate that autonomous transposons, which carry their own transposases (enzymes necessary for movement in or among genome(s)), have occasionally (in evolutionary terms) been able to move from one lineage to another. Autonomous transposons

differ significantly from the non-autonomous transposons used as gene vectors in insect genetic transformation in that non-autonomous transposons do not encode their own transposase and are, consequently, incapable of driving their own transposition. In other words, the difference between autonomous and non-autonomous elements is the ability of the former to catalyze their own transposition and, thereby, spread through a single species. In GE DBM, the transposase-encoding sequence required for production of transposase necessary for movement of the piggyBac-derived transposable element has been disabled. Hence, movement of the piggyBac-derived transposable element used to introduce the DsRed2 and tTAV traits among species of insects is biologically improbable (see more detailed discussion in (USDA-APHIS 2008b)).

Lastly, considering the theoretical acquisition of the DsRed2 and tTAV traits by another species, these will most likely provide a selective disadvantage as most natural mutational changes in nature do. Many mutations occur in the genomes of insects. However, very few provide a selective advantage. A disadvantage is particularly likely for an autocidal trait such as tTAV, which by design is intended to confer a significant disadvantage. Hence, the gene would not successfully propagate and introgress into the genome of another species in the natural environment. It is only sustained in this GE line of DBM (*P. xylostella*) under laboratory conditions.

There was also expressed concern that horizontal gene transfer may present a risk to the spread of antibiotic resistance genes and development of resistance in bacteria arising from bacterial transformation, transduction, or conjugation. There is no antibiotic-resistant gene extant in the genome of the GE DBM. Hence, there is no inherent hazard presented. Theoretically, microbiota in the gut of GE DBM may evolve antibiotic resistance. However, this scenario is highly speculative and any hazard it may present is ambiguous, as discussed above under the Topic 6, Antibiotic Resistance.

Considering that gene introgression among eukaryotes is a sexual reproductive process (vertical inheritance), that integration of genes by HGT is almost exclusively limited to prokaryotes (Lacroix and Citovsky 2016); and that HGT is an extremely rare event among prokaryotes and eukaryotes observed over timescales involving millions of years, the likelihood of a negative outcome on the environment or human health as result of horizontal gene transfer of the DsRed2 and tTAV traits during field testing of GE DBM is considered highly improbable. Further discussion on this topic can be found in prior analysis by APHIS for pink bollworm (USDA-APHIS 2008b).

Topic 7: Unintentional Survival of Female GE DBM

Several commenters expressed concern regarding the potential failure of the autocidal trait, and consequences for DBM population dynamics. There were also concerns that GE DBM might encounter sufficient tetracycline in the environment to allow them to survive and breed.

Unintended survival due to resistance to the autocidal trait

APHIS acknowledges that evolution of resistance is possible, as it is with any other pest control method. Indeed, this is the reason for existence of a scientific field devoted to Insect Resistance Management (Onstad 2014). As described in the EA Section 5.3.2, under the review of Wildlife, Plant Communities, and Biological Diversity for the Preferred Alternative, GE DBM exhibits reduced fitness, as the transgene imposes a fitness cost (Harvey-Samuel et al. 2014). Resistance, were it to occur, would render non-GE DBM less susceptible to control via the GE DBM being released. However, in the absence of a selective pressure to maintain GE DBM that are able to survive in the absence of tetracycline, the GE DBM will have to compete for survival with the larger number of non-GE DBM present. In this regard, APHIS refers to the Harvey-Samuel et al. (2014) paper, which tested persistence of the release of insects with dominant lethality (RIDL) trait in conditions with and without tetracycline. Even in conditions of 100ug/ml of chlortetracycline, the allele frequency declined in replicate populations, which means that there is a fitness cost to the introduced trait. Fitness costs mean that the trait is unlikely to persist in wild populations when there is no benefit to the trait (i.e. larvae are not feeding on tetracycline). Therefore, any resistance that emerges in GE DBM is unlikely to lead to the persistence of GE DBM in the environment, particularly after the onset of winter conditions.

Unintended survival due to tetracycline in the environment

Concern was also expressed about the availability of environmental sources of tetracycline that could result in establishment of female GE DBM. APHIS understands that tetracycline or other similar chemicals with a similar mode of action (MOA) may be present in agricultural soils (Hughes and Datta 1983; Allen et al. 2010; Durso et al. 2016), although not at concentrations sufficient to promote survival of female offspring. Female GE DBM larvae must feed on tetracycline in order to repress the female lethality trait (Jin et al. 2013), not simply be exposed to tetracycline through contact. Moreover, they must consume tetracycline in sufficient quantities for repression to occur. Exposure through ingestion could occur from consumption of Brassica plants that have taken up tetracycline from the soil (due to manure that may be used as fertilizer) or that have surface tetracycline as a result of spray drift.

Several studies have examined uptake of tetracycline-like antibiotics in plants. Cabbage plants were found to have a maximum concentration of 10 ng/g (ppb) of chlortetracycline after the soil was directly dosed at a level of 20 µg/L (ppb); the uptake in plants after adding manure to the soil was less than 5 ng/g (Kumar et al. 2005). (Kang et al. 2013) also examined antibiotic uptake by cabbage and other vegetable crops. They found that 90% of the plant tissues had chlortetracycline concentrations less than the limit of quantification; concentration in cabbage was always under 10ng/g (ppb) (Kang et al. 2013). Finally, another study by (Chowdhury et al. 2016) showed no detectable amounts of chlortetracycline in red cabbage plants after receiving treatments of manure spiked with antibiotic.

APHIS notes there is a < 1 acre plot of apple trees at NYSAES that receives antibiotic treatment sprays (which could include tetracycline) on an as-needed basis only. This factor is considered of *de minimis* risk for several reasons. The apple plot is at least 200m from the release point, and approximately 100m from the outer edge of the release area radius. Hence, GE DBMs are highly unlikely to be present near the apple trees (see discussion of dissemination under Topic 3). The location of the plot of apple trees is north and east of the release site, which is generally downwind of the release site. The use of pesticide on windy days is highly unlikely, per EPA

guidance and use requirements.[6] Tetracycline loses activity in soils (Subbiah et al. 2011) and is sensitive to photo-degradation. For example, (Christiano et al. 2010) evaluated oxytetracycline residues on peach leaves and found 78% degradation after two days of natural sunlight conditions, and 92% after four days. Therefore, tetracycline has limited persistence in the environment.

Considering that various plants may uptake antibiotics from soils and/or the potential of spray drift from antibiotics landing on the host plants, it is not implausible that consumption of tetracyclines in the plant tissues of host species could have limiting effects on lethality of GE DBM females. However, these concentrations are insufficient to support survival of GE female DBM. Survival of GE DBM females is generally observed at concentrations exceeding 1 µg/ml (1 ppm) of tetracycline, with no survival at 10 ng/ml (~ng/g) (Jin et al. 2013), which was the highest level found in the studies cited above. Taking into account the concentration of possible uptake of tetracycline in plants and the low persistence of tetracycline in the environment, it is considered highly improbable that GE DBM at the release site will receive a dose of tetracycline sufficient to cause survival of GE DBM females.

APHIS has therefore concluded it is highly unlikely that released GE DBMs and their progeny would be exposed to exogenous tetracycline and its derivatives at environmental concentrations that would suppress tTAV expression and allow GE DBM females to establish at the proposed trial site.

Topic 8: Response of Non-GE DBM and Non-Target Plant Pests to Release of GE DBM

Some expressed concern in regards to the effects of GE DBM on the non-GE DBM in the surrounding area of the release site, and/or that if the autocidal trait is effective in reducing DBM populations, that an increase in other pest species may occur.

The release of GE diamondback moth is not anticipated to substantially affect non-GE DBM in the action area, other than a transient decrease in population numbers due to release of the RIDL trait. As stated above, GE DBM are unlikely to persist and disperse out of the action area, but it should be noted that the presence of any diamondback moth strain, GE or non-GE, is largely dependent on the commercial planting of cruciferous crops by farmers. It is likely that any cruciferous crop outside the potential field release area would be actively managed, utilizing a variety of best management practices, for the purposes of diamondback moth control. Any genetically engineered diamondback moth or its progeny that potentially disperses to these cruciferous crop fields outside the potential field release area will be subject to these best management practices, thereby encountering a substantial obstacle to the development of further genetically engineered diamondback moths and its establishment/invasiveness.

Reference was made in the public comments to a mathematical model by Yakob et al. (2008) that predicts an increase in pest populations as a result of sterile insect release. APHIS would like to point out that this model compares sterile insect technique (SIT) with GE-produced RIDL, and the results indicate that RIDL was more effective in general at reducing pest populations.

[6] Improving Labels to Reduce Pesticide Drift: https://www.epa.gov/reducing-pesticide-drift/improving-labels-reduce-pesticide-drift

Furthermore, the increase in wild-type insects that was reported in this paper was found only with radiation-induced SIT, and not with RIDL.

The release of GE diamondback moth is not anticipated to substantially affect the pest species composition outside the action area. The release is a small scale field trial and thus is unlikely to have any effect beyond the field site, or any lasting effect at the field site, on the numbers or species of non-target pests.

Topic 9: Potential Socioeconomic Impacts

Among several comments received, there was expressed concern that large releases of GE DBMs could make it economically and agriculturally challenging to grow crucifers in New York State (NYS) for both conventional and organic farmers. There was concern about direct harm to conventional and organic growers from herbivory and contamination by GE DBM larvae, which may include reputational damage for conventional and organic growers of cruciferous crops grown in the area. There was concern that "Organic farms and methods of control could be overwhelmed in the face of the infestation created, making organic brassicas a thing of the past in NYS."

Field releases will be limited to the 10-acre test site at the NYSAES (EA Section 3.2 – Action Area). Releases of adults will be made at a single point in the middle of a large circular field of cabbage, and their movement patterns will be assessed. The permit conditions would impose confinement, monitoring, and reporting requirements, which are expected to be effective in containing GE DBMs to the release site.

In the unlikely event there is dispersal of GE DBMs beyond the action area (NYSAES), growers of cruciferous crops have various tools available to them to control DBMs and limit any potential damage that may be caused by the insect.[7] Organic and IPM methods when properly timed and used appropriately provide excellent control of diamondback moths.

As noted by the Northeast Organic Farming Association (NOFA-NY),[8] some of the more effective organic control methods include exclusion like insect netting or row cover. An IPM approach, where sprays or other applications of materials are made once a threshold population level is observed, is often effective. At low population levels, naturally occurring predators and parasitoids may be sufficient to control DBM populations. Heavy rainfall has also been observed to reduce larva, and even well timed use of sprinklers simulating a heavy rainfall can reduce larval populations.

Several varieties of parasitic wasps and flies have also been found to be effective. In addition, several Organic Materials Review Institute approved products have been found to work well if spraying is needed. Entrust and Dipel DF are both effective against DBMs and other larvae that could be present on the affected crop. These materials are also very specific and affect larvae feeding on the crop, but do not kill the beneficial insects which feed on the larvae. Pyganic is

[7] http://ipm.ucanr.edu/PMG/r280300611.html

[8] https://nofanewyork.wordpress.com/2015/10/02/organic-management-of-diamondback-moth-and-similar-insects/

also labeled for DBM larvae, imported cabbageworms, and several other larvae which may feed on cabbage and related crops, but it is a broad spectrum insecticide and will also kill many of the beneficial insects feeding upon the larvae.

DBM larvae are not the only pest that feed on cabbage and related crucifers. In New York, cabbage loopers, cabbageworms, thrips, and flea beetles are also present and affect cruciferous crops. Several species may be present at any time in the field and with early prevention and monitoring, they can be successfully controlled using organic methods.

Considering the permit conditions imposed on the field test (Section 4.2 of the EA) that it is probable that all, or at least the vast majority, of released GE DBM will remain in the 10 acre test site; that most organic and conventional farmers will already be implementing controls for various extant cruciferous plant pests (e.g., cabbage looper, cabbageworm, flea beetles); and that there are a variety of options for control of diamondback moth, adverse economic impacts to conventional and organic crop producers in the area as a result of the proposed field test are not likely to occur. In the unlikely event of dispersal of GE DBM beyond the authorized field site, the required monitoring and reporting requirements; APHIS' ability to respond appropriately to any unusual occurrence reported during field testing; and available control methods for DBMs, all are expected to preclude or reduce effects, so that there are no significant adverse economic impacts on conventional or organic crop producers.

References

Allen HK, Donato J, Wang HH, Cloud-Hansen KA, Davies J, and Handelsman J. 2010. *Call of the wild: antibiotic resistance genes in natural environments*. Nat Rev Micro 8, pp. 251-259. Retrieved from http://dx.doi.org/10.1038/nrmicro2312

Bahar MH, Soroka JJ, Grenkow L, and Dosdall L. 2014. *New Threshold Temperatures for the Development of a North American Diamondback Moth (Lepidoptera: Plutellidae) Population and its Larval Parasitoid, Diadegma insulare (Hymenoptera: Ichneumonidae)*. Environmental Entomology 43, pp. 1443-1452.

Biswas AJ, Kye-Il; Liu,Jing; Zhao,Muxun; Fan, Guoping; Wang,Pin; Gu,Zhen; Tang,Yi 2011. *Endoprotease-Mediated Intracellular Protein Delivery Using Nanocapsules*. ACS Nano 5, pp. 1385–1394.

Butts RA and McEwen FL. 1981. *Seasonal Populations of the Diamondback Moth, Plutella Xylostella (Lepidoptera: Plutellidae) in Relation to Day-Degree Accumulation*. The Canadian Entomologist 113, pp. 127-131.

Chapman JR, DR; Smith, AD; Riley, JR; Pedgley, DE; Woiwod, IP. 2002. *High-altitude migration of the diamondback moth Plutella xylostella to the U.K.: a study using radar, aerial netting, and ground trapping*. Ecological Entomology 27, pp. 641-650.

Chowdhury F, Langenkämper G, and Grote M. 2016. *Studies on uptake and distribution of antibiotics in red cabbage*. Journal für Verbraucherschutz und Lebensmittelsicherheit 11, pp. 61-69. Retrieved from http://dx.doi.org/10.1007/s00003-015-1008-y

Christiano RSC, Reilly CC, Miller WP, and Scherm H. 2010. *Oxytetracycline Dynamics on Peach Leaves in Relation to Temperature, Sunlight, and Simulated Rain*. Plant Disease 94, pp. 1213-1218. Retrieved from https://doi.org/10.1094/PDIS-04-10-0282 Last accessed 2017/06/05.

Clare EL, Fraser EE, Braid HE, Fenton MB, and Hebert PDN. 2009. *Species on the menu of a generalist predator, the eastern red bat (Lasiurus borealis): using a molecular approach to detect arthropod prey*. Molecular Ecology 18, pp. 2532-2542. Retrieved from http://dx.doi.org/10.1111/j.1365-294X.2009.04184.x

Data USC. 2016. *Climate Ithaca-New York*. Retrieved from http://www.usclimatedata.com/climate/ithaca/new-york/united-states/usny0717; Last accessed April, 2016.

Dosdall L. 1994. *Evidence for Successful Overwintering of Diamondback Moth, Plutella xylostella (L.) (Lepidoptera: Plutellidae) in Alberta*. Canadian Entomologist 126, pp. 183-185.

Durso LM, Wedin DA, Gilley JE, Miller DN, and Marx DB. 2016. *Assessment of Selected Antibiotic Resistances in Ungrazed Native Nebraska Prairie Soils*. Journal of Environmental Quality 45, pp. 454-462. Retrieved from http://dx.doi.org/10.2134/jeq2015.06.0280

Extension CUC and Markets NYSDoAa. 2015. *2015 Organic Production and IPM Guide for Cole Crops*. Retrieved from http://nysipm.cornell.edu/organic_guide/cole_crops.pdf

Fu X, Xing Z, Liu Z, Ali A, and Wu K. 2014. *Migration of the Diamondback Moth, Plutella xylostella, across the Bohai Sea in Northern China*. Crop Protection 64, pp. 143-149.

Furlong M, Wright D, and Dosdall L. 2013. *Diamondback Moth Ecology and Management: Problems, Progress, and Prospects*. Annual Review of Entomology 58, pp. 517-541.

Ghosh S and LaPara TM. 2007. *The effects of subtherapeutic antibiotic use in farm animals on the proliferation and persistence of antibiotic resistance among soil bacteria*. The ISME journal 1, pp. 191-203. Retrieved from http://www.nature.com/ismej/journal/v1/n3/full/ismej200731a.html

Gu H. 2009. *Cold tolerance and overwintering of the diamondback moth (Lepidoptera: Plutellidae) in southeastern Australia*. Environ Entomol 38, pp. 524-529.

Hamaki T, Suzuki M, Fudou R, Jojima Y, Kajiura T, Tabuchi A, Sen K, and Shibai H. 2005. *Isolation of novel bacteria and actinomycetes using soil-extract agar medium*. Journal of Bioscience and Bioengineering 99, pp. 485-492. Retrieved from http://www.sciencedirect.com/science/article/pii/S1389172305703984

Harvey-Samuel T, Ant T, Gong H, Morrison NI, and Alphey L. 2014. *Population-level effects of fitness costs associated with repressible female-lethal transgene insertions in two pest insects*. Evolutionary Applications 7, pp. 597-606. Retrieved from http://dx.doi.org/10.1111/eva.12159

Honda K. 1990. Hibernation and migration of diamondback moth in northern Japan In: *Diamondback Moth and Other Crucifer Pests : Proceedings of the Second International Workshop; Tainan, Taiwan*, pp. 43-50.

Hopkinson RF and Soroka JJ. 2010. *Air trajectory model applied to an in-depth diagnosis of potential diamondback moth infestations on the Canadian Prairies*. Agricultural and Forest Meteorology 150, pp. 1-11. Retrieved from http://www.sciencedirect.com/science/article/pii/S0168192309001932

Hughes VM and Datta N. 1983. *Conjugative plasmids in bacteria of the 'pre-antibiotic' era*. Nature 302, pp. 725-726.

Idris AG, EJ. 1996. *Evidence of pre-imaginal overwintering of diamondback moth, Plulella xylostella (Lepidoptera: Plutellidae) in Michigan*. Great Lakes Entomologist 29, pp. 25-30.

Jin L, Walker AS, Fu G, Harvey-Samuel T, Dafa'alla T, and Miles A. 2013. *Engineered female-specific lethality for control of pest Lepidoptera*. ACS Syn Biol. 2. Retrieved from http://dx.doi.org/10.1021/sb300123m

Kang DH, Gupta S, Rosen C, Fritz V, Singh A, Chander Y, Murray H, and Rohwer C. 2013. *Antibiotic uptake by vegetable crops from manure-applied soils*. J Agric Food Chem 61, pp. 9992-10001. Retrieved from https://dx.doi.org/10.1021/jf404045m

Knapp CW, Dolfing J, Ehlert PA, and Graham DW. 2010. *Evidence of increasing antibiotic resistance gene abundances in archived soils since 1940*. Environmental science & technology 44, pp. 580-587. Retrieved from http://pubs.acs.org/doi/abs/10.1021/es901221x

Lacroix B and Citovsky V. 2016. *Transfer of DNA from Bacteria to Eukaryotes*. mBio 7, pp. e00863-00816. Retrieved from http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4958254/

Leskinen M, Markkula I, Koistinen J, Pylkkö P, Ooperi S, Siljamo P, Ojanen H, Raiskio S, and Tiilikkala K. 2011. *Pest insect immigration warning by an atmospheric dispersion model, weather radars and traps*. Journal of Applied Entomology 135, pp. 55-67.

Li W, Jin D, Shi C, and Li F. 2017. *Midgut bacteria in deltamethrin-resistant, deltamethrin-susceptible, and field-caught populations of Plutella xylostella, and phenomics of the predominant midgut bacterium Enterococcus mundtii*. Scientific Reports 7, pp. 1947. Retrieved from http://dx.doi.org/10.1038/s41598-017-02138-9

Liu S-S, Chen F-Z, and Zalucki MP. 2002. *Development and Survival of the Diamondback Moth (Lepidoptera: Plutellidae) at Constant and Alternating Temperatures*. Environmental Entomology 31, pp. 221-231. Retrieved from http://dx.doi.org/10.1603/0046-225X-31.2.221 Last accessed 2017/06/05.

Metcalfe DD, Astwood JD, Townsend R, Sampson HA, Taylor SL, and Fuchs RL. 1996. *Assessment of the allergenic potential of foods derived from genetically engineered crop plants**. Critical Reviews in Food Science and Nutrition 36, pp. 165-186.

Mo J, Baker G, Keller M, and Roush R. 2003. *Local Dispersal of the Diamondback Moth (Plutella xylostella (L.)) (Lepidoptera: Plutellidae)*. Environmental Entomology 32, pp. 71-79.

Mortlock AL, Walter; Crisanti, Andrea 2003. *Suppression of gene expression by a cell-permeable Tet repressor*. Nucleic Acids Research 31, pp. e152.

Nguyen C, Bahar MH, Baker G, and Andrew NR. 2014. *Thermal Tolerance Limits of Diamondback Moth in Ramping and Plunging Assays*. PLoS ONE 9, pp. e87535. Retrieved from http://dx.doi.org/10.1371%2Fjournal.pone.0087535

NOAA-NCEI. 1998. *Climatic Wind Data for the United States*. National Centers for Environmental Information, National Oceanic and Atmospheric Administration. Retrieved from https://www.ncdc.noaa.gov/sites/default/files/attachments/wind1996.pdf Last accessed April, 2016.

NOAA-NCEI. 2016. Quality Controlled Local Climatological Data: *Quality Controlled Local Climatological Data, Penn Yan Airport, 2015*. Retrieved from http://www.ncdc.noaa.gov/qclcd/QCLCD Last accessed April, 2016.

NOAA. 2016. *Three-month outlook probability (Dec, Jan, Feb 2017)*. National Oceanic and Atmospheric Administration. Retrieved from http://www.cpc.ncep.noaa.gov/products/predictions/multi_season/13_seasonal_outlooks/color/t.gif Last accessed April, 2016.

Nordin O, Donald W, Ming WH, Ney TG, Mohamed KA, Halim NAA, Winskill P, Hadi AA, Muhammad ZSi, Lacroix R, Scaife S, McKemey AR, Beech C, Shahnaz M, Alphey L, Nimmo DD, Nazni WA, and Lee HL. 2013. *Oral Ingestion of Transgenic RIDL Ae. aegypti Larvae Has No Negative Effect on Two Predator Toxorhynchites Species*. PLOS ONE 8, pp. e58805. Retrieved from https://doi.org/10.1371/journal.pone.0058805

NYSAES. 2014. *New York State Agricultural Experiment Station website*. Cornell University, College of Agriculture and Life Sciences

New York State Agricultural Experiment Station. Retrieved from http://www.nysaes.cornell.edu/ Last accessed March, 2014.

Onstad D. 2014. *Insect Resistance Management: Biology, Economics, and Prediction*. Academic Press. Retrieved from http://www.sciencedirect.com/science/book/9780123969552

Popova IE, Josue RDR, Deng S, and Hattey JA. 2017. *Tetracycline resistance in semi-arid agricultural soils under long-term swine effluent application*. Journal of Environmental Science and Health, Part B 52, pp. 298-305. Retrieved from http://dx.doi.org/10.1080/03601234.2017.1281639

Qichun Z, Gu C, Shamsi IH, Asgher Z, Abid AA, and Kong Z. 2016. *Antibiotic resistance profile and genetic traits of soil isolated bacteria and their subsistence on Penicillin or Tetracycline*. Research & Reviews: Journal of Microbiology and Biotechnology 5. Retrieved from https://www.rroij.com/open-access/antibiotic-resistance-profile-and-

genetic-traits-of-soil-isolated-bacteriaand-their-subsistence-on-penicillin-or-tetracycline-.php?aid=72598
Schellhorn N, Bellati J, Paull C, and Maratos L. 2008. *Parasitoid and Moth Movement from Refuge to Crop*. Basic and Applied Ecology 9, pp. 691-700.
Shelton A. 2001. *Management of the diamondback moth: deja vu all over again?* Retrieved from http://www.regional.org.au/au/esa/2001/02/0201shelton.htm?print=1 Last accessed March, 2014.
Shirai Y and Nakamura A. 1994. *Dispersal Movement of Male Adults of the Diamondback Moth, Plutella xylostella (Lepidoptera: Yponomeutidae), on Cruciferous Vegetable Fields, Studied Using the Mark-Recapture Method*. Applied Entomology and Zoology 29, pp. 339-348. Retrieved from https://www.jstage.jst.go.jp/article/aez1966/29/3/29_3_339/_article Last accessed April, 2016.
Souza V, Rocha M, Valera A, and Eguiarte LE. 1999. *Genetic structure of natural populations of Escherichia coli in wild hosts on different continents*. Appl Environ Microbiol 65, pp. 3373-3385. Retrieved from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC91507/
Subbiah M, Mitchell SM, Ullman JL, and Call DR. 2011. *Beta-lactams and florfenicol antibiotics remain bioactive in soils while ciprofloxacin, neomycin, and tetracycline are neutralized*. Applied and environmental microbiology 77, pp. 7255-7260. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/21856822
Talekar NS and Shelton AM. 1993. *Biology, Ecology, and Management of the Diamondback Moth*. Annual Review of Entomology 38, pp. 275-301. Retrieved from http://www.annualreviews.org/doi/abs/10.1146/annurev.en.38.010193.001423
Thomas K, Aalbers M, Bannon GA, Bartels M, Dearman RJ, Esdaile DJ, Fu TJ, Glatt CM, Hadfield N, Hatzos C, Hefle SL, Heylings JR, Goodman RE, Henry B, Herouet C, Holsapple M, Ladics GS, Landry TD, MacIntosh SC, Rice EA, Privalle LS, Steiner HY, Teshima R, Van Ree R, Woolhiser M, and Zawodny J. 2004. *A multi-laboratory evaluation of a common in vitro pepsin digestion assay protocol used in assessing the safety of novel proteins*. Regulatory toxicology and pharmacology : RTP 39, pp. 87-98. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/15041142
US-FDA. 2016. *Environmental Assessment for Investigational Use of Aedes aegypti OX513A: In support of a proposed field trial of genetically engineered (GE) male Ae. aegypti mosquitoes of the line OX513A in Key Haven, Monroe County, Florida under an investigational new animal drug exemption*. Retrieved from https://www.fda.gov/downloads/AnimalVeterinary/DevelopmentApprovalProcess/GeneticEngineering/GeneticallyEngineeredAnimals/UCM514698.pdf
USDA-APHIS. 2008a. *Use of Genetically Engineered Fruit Fly and Pink Bollworm in APHIS Plant Pest Control Programs: Final Environmental Impact Statement*. Riverdale, MD.
USDA-APHIS. 2008b. *Use of Genetically Engineered Fruit Fly and Pink Bollworm in APHIS Plant Pest Control Programs: Final Environmental Impact Statement*. Retrieved from https://www.aphis.usda.gov/plant_health/ea/downloads/eis-gen-pbw-ff.pdf
USDA-APHIS. 2009. *Light Brown Apple Moth Sterile Insect Field Evaluation Projects in Sanoma and Napa, California: Environmental Assessment*. Riverdale, MD.
USDA-APHIS. 2011a. *Light Brown Apple Moth Sterile Insect Field Evaluation Projects in Long Beach and San Diego, California: Environmental Assessment*. Riverdale, MD.

USDA-APHIS. 2011b. *Pioneer Hi-Bred International, Inc. Seed Production Technology (SPT) Process DP-32138-1 Corn: Final Environmental Assessment*. Retrieved from https://www.aphis.usda.gov/brs/aphisdocs/08_33801p_fea.pdf

USDA-APHIS. 2017. *Petitions for Determination of Nonregulated Status*. United States Department of Agriculture - Animal and Plant Health Inspection Service. Retrieved from http://www.aphis.usda.gov/biotechnology/petitions_table_pending.shtml Last accessed March, 2014.

Walsh F and Duffy B. 2013. *The Culturable Soil Antibiotic Resistome: A Community of Multi-Drug Resistant Bacteria*. PLOS ONE 8, pp. e65567. Retrieved from https://doi.org/10.1371/journal.pone.0065567

Wei S-J, Shi B-C, Gong Y-J, Jin G-H, Chen X-X, and Meng X-F. 2013. *Genetic Structure and Demographic History Reveal Migration of the Diamondback Moth Plutella xylostella (Lepidoptera: Plutellidae) from the Southern to the Northern Regions of China*. PLoS ONE 8, pp. e59654.

Xia X, Gurr GM, Vasseur L, Zheng D, Zhong H, Qin B, Lin J, Wang Y, Song F, Li Y, Lin H, and You M. 2017. *Metagenomic Sequencing of Diamondback Moth Gut Microbiome Unveils Key Holobiont Adaptations for Herbivory*. Frontiers in Microbiology 8, pp. 663. Retrieved from http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5405146/

Yakob L, Alphey L, and Bonsall MB. 2008. *Aedes aegypti control: the concomitant role of competition, space and transgenic technologies*. Journal of Applied Ecology 45, pp. 1258-1265.

Yang J, Tian X, Xu B, Xie W, Wang S, Zhang Y, Wang X, and Wu Q. 2015. *Insight into the Migration Routes of Plutella xylostella in China Using mtCOI and ISSR Markers*. PLoS ONE 10, pp. e0130905.

Ye DX, Dong; Singer, AU ; Juliano, RL. 2002. *Evaluation of Strategies for the Intracellular Delivery of Proteins*. Pharmaceutical Research 19, pp. 1302-1309.

Youjun XLWQWSXBZGZ. 2010. *Biological characteristic of overwintering in the diamondback moth, Plutella xylostella*. Plant Protection. Retrieved from http://en.cnki.com.cn/Article_en/CJFDTOTAL-ZWBH201002023.htm