BIOPESTICIDES REGISTRATION ACTION DOCUMENT

Bacillus amyloliquefaciens strain D747

Pesticide Chemical (PC) Code: 016482

U.S. Environmental Protection Agency
Office of Pesticide Programs
Biopesticides and Pollution Prevention Division

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I. EXECUTIVE SUMMARY

Certis USA, LLC, ("Certis" or "applicant) submitted data to support the registration of *Bacillus amyloliquefaciens* strain D747 is the active ingredient in two end-use pesticide products, CX-9030 (EPA File Symbol 70051-107) and CX-9032 (EPA File Symbol 70051-108). The products are intended for use to control fungi and bacteria in outdoor agricultural crops, greenhouses, nurseries, shadehouses, ornamentals, and turfgrass. *B. amyloliquefaciens* strain D747 was initially identified by Certis as "*Bacillus subtilis* variant *amyloliquefaciens strain D747,"* since *B. subtilis* and *B. amyloliquefaciens* were originally classified as subtypes or variants of the same species. *B. amyloliquefaciens* is now considered a separate species, and the correct taxonomic designation is used in this Biopesticides Registration Action Document ("BRAD") (Priest et. al., 1987; Logan and de Vos, 2009; and Murray et. al., 2007).

EPA scientists reviewed product analysis, mammalian and nontarget organism toxicity data, and other information submitted by Certis to support the registration of the two aforementioned product registrations. The product analysis data requirements for *B. amyloliquefaciens* strain D747, including product chemistry and composition, analysis of samples, and physical and chemical characteristics, were fulfilled by acceptable studies conducted in accordance with Agency guidelines. Mammalian toxicity data (acute oral, injection, and pulmonary toxicity/pathogenicity) and information from peer-reviewed scientific literature demonstrated that *B. amyloliquefaciens* strain D747 is not toxic, infective or pathogenic in laboratory rats. Acceptable nontarget organism data also demonstrated that *B. amyloliquefaciens* strain D747 is not toxic to estuarine and marine fish and invertebrates, nontarget insects (including honey bees), and nontarget plants.

We have assessed human health and environmental risks of *B. amyloliquefaciens* strain D747, and determined that the pesticide would not cause unreasonable adverse effects to nontarget organisms when used in accordance with the directions on the labels, and in accordance with good agricultural practices. Additional mammalian and nontarget organism toxicity data are not required for the registered uses and application methods.

Bacillus species, including *B. amyloliquefaciens*, are commonly found in soils, including agricultural settings, and are naturally present on fresh produce. The Manual of Clinical Microbiology (9th Edition) states that dried foods, such as spices, milk powder and grains, often contain large amounts of *Bacillus* spores. *B. amyloliquefaciens* is not known to produce any mammalian toxins, and no food-borne disease outbreaks associated with *B. amyloliquefaciens* have been reported. Given that the microorganism occurs naturally in soils, exposure to *B. amyloliquefaciens* from surface and groundwater may occur. No adverse effects have been reported, and none are expected from exposure to *B. amyloliquefaciens* through drinking water.

Despite the low toxicological profile of *B. amyloliquefaciens* strain D747, personal protective equipment (PPE) is required for pesticide handlers that are frequently exposed to the active ingredient for prolonged periods. Handlers will be directed to wear a long-sleeved shirt, long pants, socks, shoes, waterproof gloves, and a dust/mist filtering respirator meeting National Institute for Occupational Safety and Health (NIOSH) standards of at least N-95, R-95, or P-95. EPA may require additional PPE, other than the standard described above, if additional pesticide uses resulting in increased exposures are proposed in the future.

EPA has concluded that, based upon the results of the toxicity tests and lack of adverse incidents,

there is a reasonable certainty that no harm will result to the United States population, including infants and children, from aggregate exposure to residues of *B. amyloliquefaciens* strain D747. No dietary risks (including drinking water) are expected from use of *B. amyloliquefaciens* strain D747 as an active ingredient in pesticide products. The two end-use pesticide products, CX-9030 and CX-9032 meet the standards for registration under Section 3(c)(5) of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA).

On October 1, 2009, EPA announced a new policy to provide a more meaningful opportunity for the public to participate in major registration decisions before they occur. According to this policy, EPA intends to provide a public comment period prior to making a registration decision for, at minimum, the following types of applications: new active ingredients; first food uses; first outdoor uses; first residential uses; or any other registration actions for which EPA believes there may be significant public interest.

Consistent with the policy of making registration actions more transparent, the pesticide products containing *B. amyloliquefaciens* strain D747, a new active ingredient, were subject to a 30-day comment period. . During this comment period, no comments were received on EPA's preliminary decision to register the two end-use pesticide products containing *B. amyloliquefaciens* strain D747, CX-9030 and CX-9032. Therefore, EPA maintained that, based upon the risk assessment and information submitted in support of registration of such pesticide products, it was in the best interest of the public and the environment to issue the end-use pesticide product registrations for *B. amyloliquefaciens* strain D747, CX-9030 and CX-9032. The basis for this preliminary decision can be found in the risk assessment for *B. amyloliquefaciens* strain D747, which is presented in this document.

II. ACTIVE INGREDIENT OVERVIEW

Biological Name:	*Bacillus amyloliquefaciens* strain D747
Culture Deposit:	Agricultural Research Service Culture Collection (also known as the Northern Regional Research Laboratory (NRRL) Collection) in Peoria, Illinois, under Accession Number NRRL B-50405.
OPP Chemical Code:	016482
Type of Pesticide:	Microbial Pesticide – Insecticide See Appendix B for specific information (e.g., use sites, application rates, methods of application, formulation types, and target pests) regarding the registered pesticide products containing this active ingredient.

III. REGULATORY BACKGROUND

A. Applications for Pesticide Product Registration

Certis USA, LLC (address: 9145 Guilford Road, Suite 175, Columbia, MD, 21046), submitted applications to register two end-use pesticide product, CX-9032 and CX-9030, under FIFRA section 3(c)(5) on July 26, 2010. EPA announced receipt of these applications to register pesticide products containing a new active ingredient on February 2, 2011 (76 Federal Register (FR) 5805), and opened a 30-day public comment period, pursuant to the provisions of FIFRA section 3(c)(4). No comments were received following this publication.

B. Food Tolerance Exemption

Concurrent with its registration applications, and under the Federal Food, Drug, and Cosmetic Act (FFDCA) section 408(d), Certis USA, LLC, submitted a petition to establish an exemption from the requirement of a tolerance for *Bacillus subtilis* variant *amyloliquefaciens* strain D747 (Pesticide Petition (PP) 0F7760). In the Federal Register of February 4, 2011 (76 FR 6465), EPA announced that Certis USA, LLC, proposed to establish an exemption from the requirement of a tolerance for residues of the insecticide, *Bacillus subtilis* variant *amyloliquefaciens* strain D747, in or on all food commodities, and opened a 30-day comment period. No comments were received following this publication. The correct taxonomic designation of the microorganism is *Bacillus amyloliquefaciens* strain D747; therefore, the tolerance exemption, when established, will reflect the correct name of the active ingredient.

IV. RISK ASSESSMENT

In the Federal Register of October 26, 2007, EPA issued a Final Rule establishing changes to the data requirements that support the registration of microbial pesticides. The rule also updated the definition for microbial pesticides (72 FR 61002), and became effective on December 26, 2007. The data and information evaluated for this BRAD were considered in accordance with these requirements.

The classifications or ratings that are given for each data requirement in this BRAD were assigned by the EPA scientists who reviewed the data, and convey the usefulness of the information for the human health and nontarget risk assessment purposes. “Acceptable” indicates that a study is scientifically sound and is useful for risk assessment. A “supplemental” classification indicates the studies provided some information that can be useful for risk assessment. “Supplemental” ratings are given to studies that are not required for registration, as well as those that are required. In the latter case, the study lacks information that should have been obtained if it was conducted according to the guideline associated with the data requirement. Sometimes the missing information would not add anything necessary to inform a risk assessment, and the study classification remains “supplemental.” A classification of “supplemental:upgradable” indicates the study lacks necessary information, but if it is made available by the applicant, the study may be upgraded to “acceptable.” An “unacceptable” rating indicates that new data must be submitted.

Toxicity categories are assigned to acute toxicity studies, based upon any signs of toxicity (hazards) observed in the test animals during the course of the study, whether the study was

conducted in accordance with test guidelines or comes from other sources, such as peer-reviewed, scientific literature. The active ingredient or particular product is classified into Toxicity Category I, II, III, or IV, where Toxicity Category I indicates the highest toxicity, and Toxicity Category IV indicates the lowest toxicity (see 40 CFR § 156.62).

A. Product Analysis Assessment (40 CFR § 158.2120)

All product analysis data requirements for *B. amyloliquefaciens* strain D747 have been fulfilled for the current product registrations. Refer to Tables 1, 2, 3, and 4 in Appendix A for a brief summary of the data requirements, including both generic and product-specific information.

B. Human Health Assessment (40 CFR § 158.2140)

1. Toxicity

All Tier I mammalian toxicity data requirements for *B. amyloliquefaciens* strain D747 have been fulfilled for the current product registrations. Refer to Tables 5, 6 and 7. Based on the lack of acute toxicity/pathogenicity, Tier II and Tier III studies were not required.

For a comprehensive summary of the generic and product-specific toxicity data requirements described below, refer to Tables 5, 6 and 7 in Appendix A.

a. Acute Toxicity/Pathogenicity – Tier I

Acute Oral Toxicity/Pathogenicity (Office of Chemical Safety and Pollution Prevention (OCSPP) Guideline 885.3050; Master Record Identification Number (MRID No. 481657-04):
B. amyloliquefaciens strain D747 was administered once orally to 14 rats of both sexes (5-weeks old) at a single dosage of 10^8 colony-forming units (CFU) per animals. No deaths occurred, and no abnormalities (clinical signs, body weight) were observed, during the study or at necropsy. The test microbe was detected at 10^3 – 10^5 CFU/g in feces 1 day after administration of the test material, but was not detected on day14. The examination for internal persistence did not detect the test microbe in any organs or tissues, such as the kidney, brain, liver, lung, spleen, stomach, small intestine (duodenum), large intestine (cecum), mesenteric lymph nodes, or blood, throughout the experimental period. Fecal clearance occurred by day 14, and no viable organisms were recovered from blood or other organs or tissues. The results of this acceptable study demonstrated that *B. amyloliquefaciens* strain D747 was not infective, pathogenic, or toxic to rats when orally dosed with $1.0x10^8$ CFU / animal.

Acute Pulmonary Toxicity/Pathogenicity (OCSPP Guideline 885.3150; MRID No.481657-06):
B. amyloliquefaciens strain D747 was administered once intratracheally to 20 male and female Sprague-Dawley rats (5-week old) at a dosage of 10^7 CFU per animal. No deaths occurred, and no abnormalities (clinical signs, body weight) were observed during the study or at necropsy. The examination for internal persistence showed that the test microbe was detected shortly after administration in the lung, trachea, and nasal cavity. The test microbes were not detected in the nasal cavity on day 7, and almost completely cleared from the trachea by day 60, when the study ended. The presence of the test microbes decreased in the lungs by approximately 10^2-10^3 CFU/g from day 0 to day 60. The test microbes were detected in the bronchial lymph nodes on day 3. Since many test microbes remained in the trachea on day 3, their presence in the bronchial lymph nodes on day 3 was attributed to transfer by macrophages from the respiratory tract to the lymph

nodes. This conclusion was supported by the observation that a particle larger than 1 μm in diameter administered into the trachea and bronchi was phagocytized by a macrophage and carried to a lymph node. The presence of the microbes in the lungs, though decreasing throughout the observation period, was attributed to the continuous transfer of viable test organisms to the bronchial lymph nodes. This result is not unusual for spore-forming bacteria, since bacterial spores are extremely tolerant of adverse conditions, and take a longer time for the immune system to clear than bacteria that do not form spores. Since a pattern of clearance was demonstrated, the remaining viable cells were considered to be spores, which take longer for a healthy immune system to clear. This acceptable study demonstrated that *B. amyloliquefaciens* strain D747 was not toxic or pathogenic to rats when dosed intratracheally at 1.0 $x10^7$ CFU/ animal.

Acute Injection Toxicity/Pathogenicity (Intravenous) – Rat (OCSPP Guideline 885.3200; MRID No. 481657-05): In an acute intravenous injection toxicity and pathogenicity study, groups of 17 male and female Sprague-Dawley rats (5-weeks old) were injected with *B. amyloliquefaciens* strain D747 at a dosage of 10^7 CFU per animal, and the influence on the animals was investigated. No deaths occurred, and there were no observed abnormalities (clinical signs, body weight) during the study or at necropsy. The examination for internal persistence of the test microbe showed that the test microbes were mainly detected in the kidney, liver, spleen, and blood shortly after administration. The test microbe decreased after that and was not detected in the blood from day14; clearance from the kidney occurred by day 60. McClintock et al. previously reported that when *B. thuringiensis* and *B. subtilis*, which are both spore-forming bacteria, were administered intravenously to rats, clearance from the liver and spleen is difficult. In this study, the test microbe did not completely disappear from these organs by day 60. No test microbes were detected in the brain, and only a few viable microbes at the limit level were sporadically detected in the small and large intestines, and lymph nodes, but were cleared by day 60. This acceptable study demonstrated that *B. amyloliquefaciens* strain D747 was not toxic, infective, or pathogenic to rats when injected intravenously with 1.0 $x10^7$ CFU/ animal.

Hypersensitivity Incidents (OCSPP Guideline 885.3400; MRID No. 481655-05): No hypersensitivity incidents in humans have been reported, and none occurred during research, development, or testing of *B. amyloliquefaciens* strain D747. Should hypersensitivity or other adverse incidents in humans occur in the future, Certis must report them to EPA, in accordance with FIFRA section 6(a)(2).

Cell Culture (OCSPP Guideline 885.3500): This study was not required because *B. amyloliquefaciens* strain D747 is not a virus (refer to test note #4 of 40 CFR § 158.2140(d)).

b. Acute Toxicity and Subchronic Toxicity/Pathogenicity – Tier II; Reproductive Fertility Effects, Carcinogenicity, Immunotoxicity, and Infectivity/Pathogenicity Analysis – Tier III

Tier II and Tier III studies were not required for *B. amyloliquefaciens* strain D747 based on the lack of acute toxicity/pathogenicity in the Tier I studies.

c. Endocrine Disruptors

As required under FFDCA section 408(p), EPA has developed the Endocrine Disruptor Screening Program (EDSP) to determine whether certain substances (including pesticide active

and other ingredients) may have an effect in humans or wildlife similar to an effect produced by a "naturally occurring estrogen, or other such endocrine effects as the Administrator may designate." The EDSP employs a two-tiered approach to making the statutorily required determinations. Tier 1 consists of a battery of 11 screening assays to identify the potential of a chemical substance to interact with the estrogen, androgen, or thyroid (E, A, or T) hormonal systems. Chemicals that go through Tier 1 screening and are found to have the potential to interact with E, A, or T hormonal systems will proceed to the next stage of the EDSP where EPA will determine which, if any, of the Tier 2 tests are necessary based on the available data. Tier 2 testing is designed to identify any adverse endocrine-related effects caused by the substance, and establish a quantitative relationship between the dose and the E, A, or T effect.

Between October 2009 and February 2010, EPA issued test orders/data call-ins for the first group of 67 chemicals, which contains 58 pesticide active ingredients and 9 inert ingredients. This list of chemicals was selected based on the potential for human exposure through pathways such as food and water, residential activity, and certain post-application agricultural scenarios. This list should not be construed as a list of known or likely endocrine disruptors.

B. amyloliquefaciens strain D747 is not among the group of 58 pesticide active ingredients on the initial list to be screened under the EDSP. Under FFDCA section 408(p), EPA must screen all pesticide chemicals. Accordingly, EPA anticipates issuing future EDSP orders/data call-ins for all pesticide active ingredients.

For further information on the status of the EDSP, the policies and procedures, the list of 67 chemicals, the test guidelines and the Tier 1 screening battery, please visit our website: *http://www.epa.gov/endo/*.

2. Federal Food, Drug, and Cosmetic Act (FFDCA) Considerations

Section 408(c)(2)(A)(i) of FFDCA allows EPA to establish an exemption from the requirement for a tolerance (the legal limit for a pesticide chemical residue in or on a food) only if EPA determines that the exemption is "safe." Section 408(c)(2)(A)(ii) of FFDCA defines "safe" to mean that "there is a reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue, including all anticipated dietary exposures and all other exposures for which there is reliable information." This includes exposure through drinking water and in residential settings but does not include occupational exposure. Pursuant to section 408(c)(2)(B) of FFDCA, in establishing or maintaining in effect an exemption from the requirement of a tolerance, EPA must take into account the factors set forth in section 408(b)(2)(C) of FFDCA, which require EPA to give special consideration to exposure of infants and children to the pesticide chemical residue in establishing a tolerance and to "ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure to the pesticide chemical residue...." Additionally, section 408(b)(2)(D) of FFDCA requires that EPA consider "available information concerning the cumulative effects of [a particular pesticide's] . . . residues and other substances that have a common mechanism of toxicity."

EPA performs a number of analyses to determine the risks from aggregate exposure to pesticide residues. First, EPA determines the toxicity of pesticides. Second, EPA examines exposure to the pesticide through food, drinking water, and through other exposures that occur as a result of pesticide use in residential settings.

Consistent with section 408(b)(2)(D) of FFDCA, EPA has reviewed the available scientific data and other relevant information, and considered its validity, completeness, and reliability and the relationship of this information to human risk. EPA has also considered available information concerning the variability of the sensitivities of major identifiable subgroups of consumers, including infants and children. All of the data requirements have been fulfilled, as described previously, and are presented in Table 5 in Appendix A. The following summarizes the results of EPA's dietary risk assessment for *B. amyloliquefaciens* strain D747.

a. Aggregate Exposure

In examining aggregate exposure, section 408 of FFDCA directs EPA to consider available information concerning exposures from the pesticide residue in food and all other non-occupational exposures, including drinking water from ground or surface water, and through pesticide use in residential and other indoor uses.

Food Exposure and Risk Characterization: *Bacillus* species, including *B. amyloliquefaciens*, are commonly found in the soil in agricultural settings, and are present on fresh produce of all kinds with no known adverse effects. The Manual of Clinical Microbiology (9th edition) mentions that dried food such as spices, milk powder and grains often contain large amounts of *B.* spores. *B. amyloliquefaciens* is not known to produce any mammalian toxins, and no foodborne disease outbreaks associated with *B. amyloliquefaciens* have been reported.

Based on the data and other information submitted to satisfy the data requirements for registration of the manufacturing-use and end-use pesticide products containing the active ingredient, *B. amyloliquefaciens* strain D747, no toxicity, infectivity, pathogenicity or other adverse effects from dietary exposure to are expected (see section IV(B)(1)(a), above, and Table 5 in Appendix A).

Drinking Water Exposure and Risk Characterization: *B. amyloliquefaciens* is naturally present in soils (Logan and de Vos, 2009); therefore, *B. amyloliquefaciens* may occur in surface and possibly groundwater. According to the World Health Organization, *Bacillus* species are often detected in drinking water even after going through acceptable water treatment processes, largely because the spores are resistant to these disinfection processes (World Health Organization, 2011). Should this microbial pesticide be present, no adverse effects are expected from exposure to *B. amyloliquefaciens* through drinking water, based on the data and other information submitted to satisfy the data requirements for registration of the manufacturing-use and end-use pesticide products containing the active ingredient, *B. amyloliquefaciens* strain D747 (see section IV(B)(1)(a), above, and Table 5 in Appendix A).

Non-occupational, Residential Risk Characterization: The use sites for these products include residential garden sites and agricultural sites. As stated previously, *B. amyloliquefaciens* is naturally present in soil, and based on the data and other information submitted to satisfy the data requirements for registration of the manufacturing-use and end-use pesticide products containing the active ingredient, *B. amyloliquefaciens* strain D747, no toxicity, infectivity, pathogenicity or other adverse effects from non-occupational exposure are expected (see section IV(B)(1)(a), above, and Table 5 in Appendix A).

b. Cumulative Effects from Substances with a Common Mechanism of Toxicity

Section 408(b)(2)(D)(v) of FFDCA requires that, when considering whether to establish, modify, or revoke a tolerance exemption, EPA consider "… available information concerning the cumulative effects of [a particular pesticide's] … residues and other substances that have a common mechanism of toxicity."

EPA has found that *B. amyloliquefaciens* strain D747 does not share a common mechanism of toxicity with any other microorganism, pesticidal or toxic substance. EPA concludes that there are no cumulative effects associated with *B. amyloliquefaciens* strain D747 that need to be considered. For information regarding how EPA determines common mechanisms of toxicity and evaluates cumulative effects, see EPA's website at: *http://www.epa.gov/pesticides/cumulative*.

c. Determination of Safety for the United States (U.S.) Population, Infants and Children

FFDCA section 408(b)(2)(C) provides that EPA shall assess the available information about consumption patterns among infants and children, special susceptibility of infants and children to pesticide chemical residues, and the cumulative effects on infants and children of the residues and other substances with a common mechanism of toxicity. In addition, FFDCA section 408(b)(2)(C) provides that EPA shall apply an additional tenfold (10X) margin of safety for infants and children in the case of threshold effects to account for prenatal and postnatal toxicity and the completeness of the database on toxicity and exposure, unless EPA determines that a different margin of safety will be safe for infants and children. This additional margin of safety is commonly referred to as the Food Quality Protection Act Safety Factor. In applying this provision, EPA either retains the default value of 10X or uses a different additional safety factor when reliable data available to EPA support the choice of a different factor.

EPA concludes that, based upon the results of the toxicity data and other information considered and described in this document, there is a reasonable certainty that no harm will result to the U.S. population, including infants and children, from aggregate exposure to the residues of *B. amyloliquefaciens* strain D747. Such exposure includes all anticipated dietary and other exposures for which there is reliable information. With no threshold effects of concern, an additional margin of safety is not required for infants and children.

3. Occupational Exposure and Risk Characterization

Occupational exposure to *B. amyloliquefaciens* strain D747 is not expected to undue risks to pesticide handlers (mixer/loader/applicators), but EPA requires appropriate personal protective equipment and precautionary statements to mitigate any potential risks (e.g., respiratory allergenicity) to pesticide handlers from prolonged or repeated exposures. Handlers applying *B. amyloliquefaciens* strain D747 end-use products in agricultural settings must wear a long-sleeved shirt, long pants, socks, shoes, waterproof gloves, and a dust/mist filtering respirator meeting NIOSH standards of at least N-95, R-95, or P-95.

4. Human Health Risk Characterization

EPA considered human exposure to *B. amyloliquefaciens* strain D747 in light of the registration standards of FIFRA and the relevant FFDCA safety factors for allowable pesticide residues in

food and animal feed commodities. EPA has determined that no unreasonable adverse effects to the U.S. population in general, and to infants and children in particular, will result when the pesticide products containing *B. amyloliquefaciens* strain D747 as the active ingredient are used in accordance with EPA-approved labeling.

C. Environmental Assessment (40 CFR § 158.2150)

The primary habitat of *Bacillus* species is soil, although they have also been isolated from a wide variety of other habitats. *B. amyloliquefaciens* has a wide-spread distribution, owed in part to its ability to form endospores that are resistant to greater variation in environmental conditions than the vegetative cells, should transfer to other climates occur. *B. amyloliquefaciens* has been isolated from internal tissues of healthy plants, and is known to promote plant growth. It is not recognized as a pathogen among *Bacillus* species (Logan and de Vos, 2009).

The data, literature citations, and data waiver rationale submitted by the applicant to support the pesticide products containing *B. amyloliquefaciens* strain D747 fulfilled the Tier I nontarget organism data requirements, and were sufficient for risk assessment purposes. Further testing of nontarget organisms at higher tier levels (i.e., Tiers II, III, and IV) is not required for the EPA-approved use sites. EPA performed an environmental risk assessment, and has determined that the use of *B. amyloliquefaciens* strain D747 is not expected to cause unreasonable adverse effects to nontarget organisms.

For a comprehensive summary of the generic data requirements described in sections IV(C)(1) of this BRAD, refer to Table 8 in Appendix A.

1. Ecological Exposure and Risk Characterization

a. Terrestrial Animals and Plants

The end-use products include a water-dispersible granular formulation and an aqueous suspension, for application to agricultural crops, nurseries, ornamental plants, turfgrass, greenhouses, and shadehouses. Applications can be made to both foliar surfaces and soil, so exposure to nontarget organisms is possible. The maximum application rates from the labels of registered products were used in the assessment of nontarget risk.

Data on the naturally occurring levels of *B. amyloliquefaciens* are not available. Many factors influence the environmental fate of microbial pesticides, and resulting population levels in the environment cannot be predicted. EPA expects that *B. amyloliquefaciens* strain D747 may survive after application if conditions are favorable, but the strain would not significantly add to the overall levels of *B. amyloliquefaciens* already present in the environment.

Birds (OCSPP Guideline 885.4050) and Mammals (OCSPP Guideline 885.4150):
A supplemental study showed that the acute oral LD_{50} for *B. amyloliquefaciens* strain D747 is > 4.5×10^{11} spores/kg BW or > 8×10^{9} spores/bird in Northern bobwhite (*Colinus virginianus*; MRID 48165712). Additionally, *B. amyloliquefaciens* is intentionally included in some domestic avian food diets as a nutritional additive (e.g., European Food Safety Authority, 2010; Wizna et al., 2009), and it is not known to be pathogenic to animals (Logan and de Vos, 2009). An extensive literature search in several databases returned no reports of toxicity or pathogenicity of *B. amyloliquefaciens* in birds. Based on these lines of evidence, *B.*

amyloliquefaciens strain D747 is not expected to pose risk of adverse effects in birds.

A study with laboratory rats (MRID 48165704) also showed that *B. amyloliquefaciens* strain D747 is not toxic, infective, or pathogenic at the maximum hazard dose of 1.0×10^8 CFU/animal. Therefore, adverse effects to wild mammals are also not expected as a result of the applications of *B. amyloliquefaciens* strain D747 in accordance with label instructions.

Nontarget Insects (OCSPP Guideline 885.4340) and Honey Bees (OCSPP Guideline 885.4380): Studies with *Orius stricollis*, *Crysoperla carnea*, and *Phytoseiulus persimilis* were submitted for the nontarget insect data requirement (MRID 48165716). While no effects of *B. amyloliquefaciens* strain D747 were observed in these studies, they were not acceptable for use in the ecological risk assessment. Two honey bee studies were also submitted that showed no adverse effects of *B. amyloliquefaciens* D747 after 48 hours (MRID 48165717) and 17 days (no MRID currently assigned). These studies were rated Supplemental because they were not of sufficient duration (30 days); however, except in rare cases, bacteria that are pathogenic to insects typically produce toxins that kill the insect within a few days (Tanada and Kaya, 1993). Adverse effects resulting from exposure to *B. amyloliquefaciens* strain D747, therefore, would likely have been evident in the bee studies, especially after 17 days. Scientific rationale was submitted to show that adverse effects are not expected to nontarget insects (MRIDs 48621502 and 48621503). Entomopathogenic *B.* species (e.g., *B. thuringiensis*, *B. sphaericus*) have been extensively studied, and their pathobiology is well-known. *B. amyloliquefaciens* is not among the *B.* species recognized as frank pathogens to insects or other animals (Logan and de Vos, 2009). There are some accounts in the literature of effects of *B. subtilis* on insects; however, none of these were associated with *B. amyloliquefaciens* or *B. subtilis* var. *amyloliquefaciens*. Therefore, based on the studies and other information provided, *B. amyloliquefaciens* strain D747 is not expected to pose risk to honey bees and other nontarget insects as a result when applied in accordance with label instructions..

Nontarget Plants (OCSPP Guideline 885.4300):
Studies with plants exposed to *B. amyloliquefaciens* strain D747 were unacceptable, based on several deficiencies (MRID 48165715). *B. amyloliquefaciens* is not taxonomically related to any known plant pathogens. As discussed previously, however, the microorganism has been isolated from tissues of healthy plants and is known as a plant growth-promoting rhizobacterium. It is, therefore, not expected to pose risk to nontarget plants as a result of t applications made in accordance with label instructions.

b. Aquatic Animals and Plants

B. amyloliquefaciens strain D747 is not intended to be applied directly to water, but some of the applied product may reach aquatic habitats through runoff or spray drift. Spray drift at application is the primary mechanism by which the pesticide is expected to reach water. A spray drift analysis was included in the aquatic risk assessment to determine exposure, and further detail is provided in the environmental risk assessment for *B. amyloliquefaciens* strain D747.

Freshwater Fish (OCSPP 885.4200) Guideline and Invertebrates (OCSPP Guideline 885.4240): In a 30-day study with rainbow trout (*Oncorhynchus mykiss*), the LC_{50} for *B. amyloliquefaciens* strain D747 was 8.1×10^{10} CFU/L, and the NOEC based on sub-lethal effects was 1.44×10^{10} CFU/L. If the maximum broadcast application rate (4 fluid ounces/1000 ft^2 or 2.55×10^6 CFU/cm^2) is applied directly to a 1-ha body of water 15 cm deep (the EPA Standard Wetland),

the resulting concentration would be 1.7 x 10^8 CFU/L. This would be the maximum possible aquatic concentration at this application rate, but this direct application to water is not on the EP labels. The NOEC and LC_{50} are approximately 85X and 476X higher than this concentration, respectively. Exposure in freshwater environments will be well below the concentrations that would produce adverse effects, and the applications of *B. amyloliquefaciens* strain D747 as presented on EPA- approved labels are not expected to pose risks to freshwater fish.

A study with *Daphnia magna* provided an EC_{50} based on mortality/immobility of 3.7 x 10^{10} CFU/L, and a NOEC for sub-lethal effects of 2.84 x 10^8 CFU/L. Based on the spray drift analysis, the NOEC would be approximately 20X the expected environmental concentration (EEC), and the EC_{50} would be 218X the EEC; therefore, the applications of *B. amyloliquefaciens* strain D747 D747 as presented on EPA- approved labels are not expected to result in adverse effects to freshwater invertebrates.

Marine/Estuarine Fish (OCSPP 885. 4280) and Invertebrates (OCSPP 885.4240):
Concentrations reaching marine or estuarine areas are expected to be less than those calculated above for freshwater animals, due to further dilution in deeper water. *B. amyloliquefaciens* strain D747 is not expected to reach marine or estuarine environments in significant concentrations, and risk to animals in these environments is not anticipated.

Aquatic Plants (OCSPP Guideline 885.4300):
B. amyloliquefaciens strain D747 is not related to known plant pathogens; therefore, adverse effects to aquatic plants are not anticipated.

2. Environmental Fate Data

The information provided to support uses and application methods that are presented on EPA-approved labels was sufficient to satisfy the Tier I nontarget organism data requirements and for nontarget organism risk assessment for *B. amyloliquefaciens* strain D747; further testing at higher tier levels (i.e., Tiers II, III, and IV) is not required.

3. Threatened and Endangered Species Assessment

Since EPA has determined that no effects are anticipated for any nontarget species exposed to *B. amyloliquefaciens* strain D747 as a result of applications made in accordance with EPA-approved labels, effects to federally listed threatened and endangered species and their designated critical habitats are also not expected. Therefore, a "No Effect" determination is made for direct and indirect effects to listed species and their designated critical habitats resulting from the approved uses of *B. amyloliquefaciens* strain D747.

V. ENVIRONMENTAL JUSTICE

EPA seeks to achieve environmental justice—the fair treatment and meaningful involvement of all people regardless of race, color, national origin, or income—with respect to the development, implementation, and enforcement of environmental laws, regulations, and policies. Fair treatment means that no group of people, including racial, ethnic, or socioeconomic groups, should bear a disproportionate share of the negative environmental consequences resulting from industrial, municipal, and commercial operations or the execution of federal, state, local, and tribal environmental programs and policies. Meaningful involvement means that (1) potentially

affected community residents have an appropriate opportunity to participate in decisions about a proposed activity that will affect their environment and/or health; (2) the public's contribution can influence the regulatory agency's decision; (3) the concerns of all participants involved will be considered in the decision-making process; and (4) the decision-makers seek out and facilitate the involvement of those potentially affected. EPA has this goal for all communities and persons across the United States.

EPA sought information on any groups or segments of the population who, as a result their location, cultural practices, or other factors, may have atypical, unusually high exposure to *B. amyloliquefaciens* strain D747, compared to the general population. No public comments were received on this particular matter.

For additional information regarding environmental justice issues, please visit EPA's web site at *http://www.epa.gov/compliance/environmentaljustice/index.html*.

VI. RISK MANAGEMENT DECISION

Section 3(c)(5) of FIFRA permits for the registration of a pesticide provided that all the following determinations are made:

(1) Its composition is such as to warrant the proposed claims for it;
(2) Its labeling and other material required to be submitted comply with the requirements of FIFRA;
(3) It will perform its intended function without unreasonable adverse effects on the environment; AND
(4) When used in accordance with widespread and commonly recognized practice, it will not generally cause unreasonable adverse effects on the environment.

To satisfy criterion 1, the *B. amyloliquefaciens* strain D747 pesticide products have well-known properties. EPA has no knowledge that would contradict the claims made for these products, the CX-9030 and CX-9032 EP labels, and we have concluded that such products are not expected to cause unreasonable adverse effects on the environment when used according to the label instructions. Criterion 2 is satisfied by the current product labels, as well as the data and information presented in this document. It is believed that the *B. amyloliquefaciens* strain D747 pesticide products will not cause any unreasonable adverse effects on the environment, and CX-9030 and CX-9032 (end-use pesticide product), in particular, are likely to provide protection against fungal and bacterial pests as claimed, satisfying criterion 3. Criterion 4 is satisfied in that the *B. amyloliquefaciens* strain D747 pesticide products are not expected to cause unreasonable adverse effects when used according to label instructions. Therefore the end-use products, CX-9030 and CX-9032, containing *B. amyloliquefaciens* strain D747 as a new active ingredient, are eligible for registration under FIFRA section 3(c)(5) for the labeled uses. Should uses that are more extensive be proposed in the future (e.g., aquatic uses), EPA will likely require that additional data be submitted.

VII. ACTIONS REQUIRED BY THE REGISTRANT

A. Final Printed Labeling

Before releasing pesticide products containing *B. amyloliquefaciens* strain D747 for shipment, the registrant is required to provide appropriate final printed labeling to EPA prior to shipment of product.

B. Terms of Registration

As a term of the registration CX-9032 EP, the registrant must submit the following data within one year of this product's registration:

(1) Storage Stability (OCSPP Guideline 830.6317) and Corrosion Characteristics (OCSPP Guideline 830.6320): The results of a one-year storage stability and corrosion characteristics study.

C. Reporting of Adverse Effects and Hypersensitivity Incidents

Notwithstanding the information stated previously, it should be clearly understood that certain specific data are required to be reported to EPA as a requirement for maintaining the federal registration for a pesticide product. A brief summary of these types of data are described below.

Reports of all incidents of adverse effects to the environment must be submitted to EPA under the provisions stated in FIFRA section 6(a)(2). Additionally, all incidents of hypersensitivity (including both suspected and confirmed incidents) must be reported to EPA under the provisions of 40 CFR § 158.2140(d).

VIII. GLOSSARY OF ACRONYMS AND ABBREVIATIONS

ASAE	American Society of Agricultural Engineers
BPPD	Biopesticides and Pollution Prevention Division
BRAD	Biopesticides Registration Action Document
CFR	Code of Federal Regulations
CFU	colony-forming unit(s)
cfu/kg	colony-forming units per kilogram
cfu/mL	colony-forming units per milliliter
cP	centipoise
EDSP	Endocrine Disruptor Screening Program
EP	end-use product
EPA	Environmental Protection Agency (the "Agency")
FFDCA	Federal Food, Drug, and Cosmetic Act
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
FQPA	Food Quality Protection Act
FR	Federal Register
g/mL	gram per milliliter
LC_{50}	median lethal concentration. A statistically derived concentration of a substance that can be expected to cause death in 50% of test animals. It is

	usually expressed as the weight of substance per weight or volume of water, air, or feed (e.g., mg/L, mg/kg, or ppm).
LD_{50}	median lethal dose. A statistically derived single dose that can be expected to cause death in 50% of the test animals when administered by the route indicated (oral, dermal, or inhalation). It is expressed as a weight of substance per unit weight of animal (e.g., mg/kg).
mg/kg	milligrams per kilogram
mg/L	milligrams per liter
MP	manufacturing-use product
mPa·s	milliPascal-second, term used as the unit of dynamic viscosity.
MRID No.	Master Record Identification Number
NIOSH	National Institute for Occupational Safety and Health
NRRL	Northern Regional Research Laboratory
OCSPP	Office of Chemical Safety and Pollution Prevention
OPP	Office of Pesticide Programs
PC Code	Pesticide Chemical Code
PP	Pesticide Petition
PPE	personal protective equipment
ppm	parts per million
TGAI	technical grade of the active ingredient
U.S.	United States

IX. BIBLIOGRAPHY

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APPENDIX A. MICROBIAL PESTICIDES DATA REQUIREMENTS (40 CFR PART 158 – SUBPART V)

TABLE 1. Product Analysis Data Requirements for the End-Use Product (EP), CX-9030 (40 CFR § 158.2120)

OCSPP Guideline Number	Data Requirement	Results	MRID No.
885.1100	Product Identity	Submitted data fulfill the requirement for product identity. CX-9030 contains 25.0% by weight *Bacillus amyloliquefaciens strain* strain D747 (minimum of $5x\ 10^{10}$ CFU/g)	481655-01 CSF dated 10/18/2011
885.1200	Manufacturing Process	Submitted data fulfill the requirement for manufacturing process.	481655-01
Not applicable	Deposition of a Sample in a Nationally Recognized Culture Collection	Submitted data fulfill the requirement for deposition. Culture on deposit under Accession Number NRRL B-50405.	481655-01
885.1300	Discussion of Formation of Unintentional Ingredients	Submitted data fulfill the requirement for discussion of formation of unintentional ingredients.	481655-01
885.1400	Analysis of Samples	Submitted date fulfill the requirement for analysis of samples.	481655-01
885.1500	Certification of Limits	Limits listed on the confidential statement of formula are adequate/acceptable	CSF dated 10/18/2011
Additional Studies			
830.1800	Enforcement Analytical Method	Submitted data fulfill the requirement for an enforcement analytical method	481655-01

TABLE 2. Product Analysis Data Requirements for the End-Use Product (EP), CX-9032 (40 CFR § 158.2120)

OCSPP Guideline Number	Data Requirement	Results	MRID No.
885.1100	Product Identity	Submitted data fulfill the requirement for product identity. CX-9030 contains 98.95% by weight *Bacillus amyloliquefaciens strain* strain D747 (minimum of $1x\ 10^{10}$ CFU/g)	481655-01 CSF dated 10/18/2011
885.1200	Manufacturing Process	Submitted data fulfill the requirement for manufacturing process.	481657-01 481655-01
Not applicable	Deposition of a Sample in a Nationally Recognized Culture Collection	Submitted data fulfill the requirement for deposition. Culture on deposit under Accession Number NRRL B-50405.	481657-01 481655-01
885.1300	Discussion of Formation of Unintentional Ingredients	Submitted data fulfill the requirement for discussion of formation of unintentional ingredients.	481657-01 481655-01
885.1400	Analysis of Samples	Submitted date fulfill the requirement for analysis of samples.	481655-01 481657-01
885.1500	Certification of Limits	Limits listed on the confidential statement of formula are adequate/acceptable	CSF dated 10/18/2011
Additional Studies			
830.1800	Enforcement Analytical Method	Submitted data fulfill the requirement for an enforcement analytical method	481655-01

TABLE 3. Physical and Chemical Characteristics for the Technical Grade of the Active Ingredient (TGAI) *Bacillus. amyloliquefaciens strain* strain D747 / CX-9030 (EP). (40 CFR § 158.2120)

OCSPP Guideline Number	Data Requirement	Results		MRID No.
		TGAI	CX-9030 (EP)	
830.6302	Color	Beige	Not Applicable	481657-02
830.6303	Physical State	Fine powder	Not applicable	481657-02
830.6304	Odor	Yeast odor	Not applicable	481657-02
830.6313	Stability to Normal and Elevated Temperatures, Metals, and Metal Ions	Spores inactivated at 54°C	Not applicable	481657-02
830.6314	Oxidation/Reduction: Chemical Incompatibility	Not applicable, the product does not contain oxidizing or reducing agents		481657-02
830.6315	Flammability	Not applicable, the product does not contain flammable ingredients		481657-02
830.6316	Explodability	Not applicable, the product does not contain explosive ingredients		481657-02
830.6317	Storage Stability	Stable up to one year at 25°C without loss of viability .		481657-02 481657-03
830.6319	Miscibility	Not applicable, the product is not an emulsifiable liquid. (refer to test note #2 of 40 CFR § 158.2120(d)).		481657-02
830.6320	Corrosion Characteristics	Not applicable, the product is a powder.		481657-02
830.6321e	Dielectric Breakdown Voltage	Not applicable, the product is not for use around electrical equipment		481657-02
830.7000	pH	6.5 – 7.0 (1% w/w)	7.5- 8.0	481657-02
830.7100	Viscosity	Not applicable, the product is a powder.	Not Applicable.CX-9030 is not a liquid.	481657-02
830.7300	Density/Relative Density/Bulk Density (Specific Gravity)	0.307- 0.375 g/ml	0.60-.0.78 g/ cm^3	481657-02

TABLE 4. Physical and Chemical Characteristics for CX-9032 (EP). (40 CFR § 158.2120)				
OCSPP Guideline Number	**Data Requirement**	**Results**		**MRID No.**
		TGAI	**CX-9030 (EP)**	
830.6302	Color	Beige	Light brown	481655-02
830.6303	Physical State	Fine powder	Liquid	481655-02
830.6304	Odor	Yeast odor	Yeast odor	481655-02
830.6313	Stability to Normal & Elevated Temperatures, Metals, & Metal Ions	Spores inactivated at 54°C..		481655-02
830.6317	Storage Stability	Stable up to one year at 25°C without loss of viability	Stable up to 78 days at 25°C without loss of viability. **As a term of the registration, EPA is requiring submission of the results of a one-year storage stability and corrosion characteristics study be submitted within one year.**	481655-02
830.6319	Miscibility	Not applicable; product is not an emulsifiable liquid. (refer to test note #2 of 40 CFR § 158.2120(d)).		481655-02
830.6320	Corrosion Characteristics	Not applicable; product is a powder.	None evident to polyethylene packaging after 78 days. **As a term of the CX-9032 (EP) registration, EPA is requiring submission of the results of a one-year storage stability & corrosion characteristics within one year.**	481655-02
830.7000	pH	6.5 – 7.0 (1% w/w)	4.2 - 4.3 (1% w/w).	481655-02
830.7100	Viscosity	Not applicable, the product is a powder.	4.6 - 16.0 milliPascal-second (mPa s) at 25°C	481655-02
830.7300	Density/Relative Density/Bulk Density (Specific Gravity)	0.307- 0.375 g/ml	1.02 – 1.03 g/mL	481655-02

TABLE 5. Toxicology Data Requirements for the Technical Grade of the Active Ingredient (TGAI) (40 CFR § 158.2140)			
OCSPP Guideline Number	**Data Requirement**	**Results**	**MRID No.**
		TGAI	
Tier I			
885.3050	Acute Oral Toxicity/Pathogenicity	No evidence of infectivity, pathogenicity or toxicity was found from oral administration of $1.0x10^8$ CFU *Bacillus amyloliquefaciens* strain D747 to rats. Clearance from fecal material occurred by day 14 and no viable organisms were recovered from blood or any other organ or tissue. **Classification: Acceptable**	481657-04
885.3150	Acute Pulmonary Toxicity/Pathogenicity	No evidence of infectivity, pathogenicity or toxicity was found from intratracheal administration of $1.0x10^7$ spores *B. amyloliquefaciens* strain D747 to rats. **Classification: Acceptable**	481657-06
885.3200	Acute Injection Toxicity/Pathogenicity (Intravenous)	Not toxic, infective, and/or pathogenic to rats when dosed intravenously at $1.0x10^7$ spores per animal. **Classification: Acceptable**	481657-05
885.3400	Hypersensitivity Incidents	No hypersensitivity incidents, including immediate-type or delayed-type reactions of humans and domestic animals that occurred during research, development, or testing of the TGAI/MP, were reported. Future hypersensitivity incidents must be reported (For reporting format: OCSPP Guideline 885.3400).	479450-23
885.3500	Cell Culture	Not required. *B. amyloliquefaciens* strain D747 is not a virus (Test note #4 of 40 CFR § 158.2140(d)).	
Tiers II and III			
Not required for *Bacillus amyloliquefaciens* strain D747 based on the lack of acute toxicity/pathogenicity in the Tier I studies.			

TABLE 6. Toxicology Data Requirements for the End-Use Product (EP), CX-9030 (40 CFR § 158.2140)			
OCSPP Guideline Number	**Data Requirement**	**Results**	**MRID No.**
885.3400	Hypersensitivity Incidents	No hypersensitivity incidents, including immediate-type or delayed-type reactions of humans and domestic animals that occurred during research, development, or testing of the EP, were reported by the applicant. Any future hypersensitivity incidents must be reported per OCSPP Guideline 885.3400.	
870.1100	Acute Oral Toxicity	Oral LD_{50} > 5000 mg/Kg **Classification: Acceptable** **TOXICITY CATEGORY IV**	481657-07
870.1200	Acute Dermal Toxicity	Dermal LD_{50} > 5050 mg/Kg **Classification: Acceptable** **TOXICITY CATEGORY IV**	481657-08
870.1300	Acute Inhalation Toxicity	Inhalation LC_{50} > 2.18 mg/L **Classification: Acceptable** **TOXICITY CATEGORY IV**	481657-09
870.2400	Acute Eye Irritation	The maximum average irritation score of 18.3 obtained 1 hour after treatment declined to 17.33 after 24 hours, 2 after 48 hours and 0 after 72 hours following ocular administration of 0.1 mL *B. amyloliquefaciens* strain D747 to New Zealand White rabbits in a 72 hour observation period. **Classification: Acceptable** **TOXICITY CATEGORY III**	481657-10
870.2500	Primary Dermal Irritation	No evidence of irritation was found from dermal administration of 500 mg *B. amyloliquefaciens* strain D747 CX-9030 to rabbits during the 4 hour exposure and 72 observation periods. The dermal irritation score for *B. amyloliquefaciens* strain D747 CX-9030 was 0.00. **Classification: Acceptable** **TOXICITY CATEGORY IV**	481657-11

TABLE 7. Toxicology Data Requirements for the End-Use Product (EP), CX-9032 (40 CFR § 158.2140)			
OCSPP Guideline Number	**Data Requirement**	**Results**	**MRID No.**
885.3400	Hypersensitivity Incidents	No hypersensitivity incidents including immediate-type or delayed-type reactions of humans and domestic animals that occurred during research, development, or testing of the EP were observed or reported. All hypersensitivity incidents must be reported per OCSPP Guideline 885.3400.	
870.1100	Acute Oral Toxicity	Waiver request submitted. Requirement satisfied by submitted MP data. Oral exposure to the MP showed no adverse effects including infectivity, pathogenicity and toxicity up to the limit dose. Inerts are exempt from tolerance. No additional oral toxicity is expected from inerts. **Classification: Acceptable**	481655-04
870.1200	Acute Dermal Toxicity	Waiver request submitted. Requirement adequately addressed by CX-9030 EP data as well as, dermal irritation testing on CX-9032-EP. Dermal toxicity and irritation testing for the CX-9030 EP and dermal irritation data on CX-9032 EP showed no adverse effects up to the limit doses. No additional dermal toxicity is expected from inerts. **Classification: Acceptable**	481655-04 481655-06 481655-11 481657-08
870.1300	Acute Inhalation Toxicity	Waiver request submitted. Pulmonary exposure to the MP showed no adverse effects, including infectivity, pathogenicity or toxicity though slight toxicity lasting 2 days from a 4-hour aerosol inhalation administration of 2.18 mg/L, where inert ingredients were also present, was noted in a CX-9030 EP study. No additional toxicity is expected from this EP's inert ingredients. **Classification: Acceptable**	481655-04 481657-06 481657-09
870.2400	Acute Eye Irritation	The maximum average irritation score of 0.667 at 1 hour after treatment declined to 0 after 24 hours following ocular administration of 0.1 mL *B. amyloliquefaciens* strain D747 CX-9032 to New Zealand White rabbits (72-hour observation period). **Classification: Acceptable** **TOXICITY CATEGORY IV**	481655-05

TABLE 7. Toxicology Data Requirements for the End-Use Product (EP), CX-9032 (40 CFR § 158.2140)			
OCSPP Guideline Number	**Data Requirement**	**Results**	**MRID No.**
870.2500	Primary Dermal Irritation	No irritation occurred from dermal administration of 0.5 mL undiluted *B. amyloliquefaciens* strain D747 CX-9032 to shaved skin of rabbits during the 4 hour exposure and 72 observation periods. The dermal irritation score for *B. amyloliquefaciens* strain D747 CX-9032 was 0.00. **Classification: Acceptable** **TOXICITY CATEGORY IV**	481655-06

TABLE 8. Nontarget Organism Toxicity and Environmental Fate Data Requirements for the Technical Grade of the Active Ingredient (TGAI), *Bacillus amyloliquefaciens* strain D747 **(40 CFR § 158.2150)**			
OCSPP Guideline Number	**Data Requirement**	**Results**	**MRID No.**
Tier I			
885.4050	Avian Oral Toxicity	A study showed that *B. amyloliquefaciens* D747 is not toxic to birds at 8.9 x 10^9 spores/bird. **Classification: Supplemental** Scientific rationale is sufficient to conclude that *B. amyloliquefaciens* D747 is not expected to pose a hazard to birds. **Classification: Acceptable**	48165712 48621501
885.4100	Avian Inhalation Toxicity/Pathogenicity	Not required. *B. amyloliquefaciens* D747 is not considered to be pathogenic to birds	
885.4150	Wild Mammal Toxicity/Pathogenicity	Tests required by 40 CFR § 158.2140 are adequate/ appropriate for assessment of hazards to wild mammals. *B. amyloliquefaciens* D747 was not infective, toxic or pathogenic to laboratory rats 1.0 x 10^8 CFU/animal	48165704
885.4200	Freshwater Fish Toxicity/Pathogenicity	A 30-day study shows that the LC_{50} to rainbow trout is 8 x 10^{10} CFU/L. The NOEC for sublethal effects is 1.44 x 10^{10} CFU/L. **Classification: Acceptable**	48165713
885.4240	Freshwater Invertebrate Toxicity/Pathogenicity	A 21-day study shows that the EC_{50} to *Daphnia magna* based on mortality is 3.7 x 10^{10} CFU/L. The NOEC for sublethal effects is 2.84 x 10^8 CFU/L. **Classification: Acceptable**	48165714
885.4280	Estuarine/Marine Fish and Invertebrate Testing	Not required. *B. amyloliquefaciens* D747 is not to be applied directly to water and is not expected to reach estuarine or marine environments in significant quantities.	
885.4300	Nontarget Plant Testing	A study submitted was determined to be **unacceptable**; however, testing is not required because. *B. amyloliquefaciens* is not related to known plant pathogens	48165715

TABLE 8. Nontarget Organism Toxicity and Environmental Fate Data Requirements for the Technical Grade of the Active Ingredient (TGAI), *Bacillus amyloliquefaciens* strain D747 **(40 CFR § 158.2150)**			
OCSPP Guideline Number	**Data Requirement**	**Results**	**MRID No.**
885.4340	Nontarget Insect Testing	Studies with three species of arthropods were determined to be **unacceptable**. Additional scientific rationale was submitted to show that *B. amyloliquefaciens* is not expected to be toxic or pathogenic to nontarget insects. **Classification: Acceptable**	48165716 48621502
885.4380	Honey Bee Testing	Two studies showed that *B. amyloliquefaciens* D747 is not toxic to honey bees. **Classification: Supplemental** Additional rationale was sufficient to show that pathogenicity to honey bees is not expected as a result of exposure to *B. amyloliquefaciens* D747. **Classification: Acceptable**	48165717 48621503
Tiers II, III, and IV			
Not required for *Bacillus amyloliquefaciens* strain D747 based on the current uses and application methods.			

APPENDIX B. PESTICIDE PRODUCTS

TABLE 9. Table Title?

EPA Registration Number	Registration Name	Percentage Active Ingredient	Formulation Type	Use Site(s)	Method(s) of Application	Application Rate	Target Pest
70051-107	CX-9032	98.35%	End Use – Aqueous Suspension	Various agricultural and greenhouse crops (e.g., vegetables, tree fruits, berries, grapes and tropical fruit, tree nuts, herbs and spices, coffee, tobacco, hops, forestry seedlings ornamentals, and turf)	Tractor mounted boom, airblast, hose-end, backpack and other pressurized sprayers; foggers or mist blowers; water wheel and other drench applicators; soil injection; aerial; and chemigation with drip or sprinkler irrigation and cutting or root dip	Rate listed on label varies depending on application method from O.5 pints /acre to 6 quarts/acre	Various fungal and bacterial pests listed on the label including: *Alternaria, Botrytis cinerea Didymella bryoniae Phoma cucurbitacearum Erisphe, Fusarium, Macrophomina phaseoli Monosporascus cannonballus Peronospora , Phytophthora Pseudomonas syringae* pv. *tomato Pseudoperonospora* spp. *Puccinia* spp. *Pythium, Rhizoctonia, Sphaerotheca* spp. *Verticillium*, spp. *Xanthomonas* spp.
70051-108	CX-9030	25.0%	Water Dispersible Granule	Various agricultural and greenhouse crops (e.g., vegetables, ornamentals, and turf)	Tractor mounted boom, airblast, hose-end, backpack and other pressurized sprayers; foggers or mist blowers; water wheel and other drench applicators; shank or other soil injection equipment; aerial; and chemigation with drip or sprinkler irrigation and cutting or root dip	0.25 –3 pounds per acre	Various fungal and bacterial pests listed on the label including: *Alternaria, Botrytis cinerea Didymella bryoniae Phoma cucurbitacearum Erisphe Fusarium, Macrophomina phaseoli Monosporascus cannonballus Peronospora, Phytophthora Pseudomonas syringae* pv. *tomato Pseudoperonospora* spp. *Puccinia* spp. *Pythium, Rhizoctonia, Sphaerotheca* spp. *Verticillium*, spp., *Xanthomonas* spp.