US Environmental Protection Agency
Office of Pesticide Programs

BIOPESTICIDES REGISTRATION ACTION DOCUMENT
Bacillus pumilus strain
QST 2808 (PC Code 006485)

November 16, 2004

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U.S. Environmental Protection Agency
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Biopesticides and Pollution Prevention Division

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BIOPESTICIDES REGISTRATION ACTION DOCUMENT TEAM

Office of Pesticide Programs:

Biopesticides and Pollution Prevention Division

Microbial Pesticides Branch

Dennis Szuhay	Botanist, Branch Chief
Shanaz Bacchus	Chemist, Acting Team Leader

Health Effects

John Kough	Biologist, Senior Scientist
Carl Etsitty	Microbiologist

Ecological Effects

Zigfridas Vaituzis	Microbiologist
Hilary Hill	Microbiologist

Regulatory Action Leader

Susanne Cerrelli (May 2000 until May 2004)	Biologist
Barbara Mandula (from May 2004)	Biochemist

I. EXECUTIVE SUMMARY

A. Active Ingredient and Proposed Uses[1]

B. pumilus strain QST 2808 is a naturally occurring bacterium found in soil, water, and other environments. As a pesticide active ingredient, it will be used on a variety of crops to control fungal pests, especially mildews, blights, and molds, and will be used on oaks (Quercus) and other trees to prevent sudden oak death syndrome. *B. pumilus* strain QST 2808 is not toxic or pathogenic to mammals or to other non-target organisms. Use sites include many food and feed crops as well as nurseries, landscapes, greenhouses, and rights-of-way. Two liquid products are being registered. The first is a manufacturing use product called QST 2808 MUP, containing 1.42% by weight of *Bacillus pumilus* strain QST 2808 dried spores, and a minimum of 1 x 10^{10} cfu/g product. The end product, Sonata™ ASO, contains 1.38% by weight *Bacillus pumilus* strain QST 2808 dried spores, and a minimum of 1 x 10^{10} cfu/g product. The end product is mixed with water and applied to foliage by spray application aerially or from the ground, or by chemigation. Applications may be made up to and including the day of harvest. Use of Sonata™ ASO is limited to growing crops.

B. pumilus strain QST 2808 prevents the germination of fungal spores on plants by forming a physical barrier between the spores and the leaf surface; the bacterium then colonizes the fungal spores. There is evidence that *B. pumilus* strain QST 2808 may induce systemic acquired resistance (SAR) in plants, resulting in the plants' becoming more resistant to a variety of pathogens.

B. Human Risk Assessment

1. Mammalian Toxicity

No toxicity to humans is expected from use of *Bacillus pumilus* strain QST 2808 spores as a pesticide active ingredient, based on acceptable studies in laboratory animals. No toxicity or pathogenicity was found in the following acute studies: oral, dermal, pulmonary, inhalation, intravenous, eye irritation, and dermal irritation.

2. Food Tolerances

A permanent exemption from a tolerance for all food commodities is being established in 40 CFR 180.1255 for residues of *Bacillus pumilus* strain QST 2808 when used as

[1] The product referred to in this document as "QST 2808 Technical" is being registered as the manufacturing product "QST 2808 MUP." The word "Technical" is inappropriate, given that the product contains ingredients other than spores. In toxicology studies performed with QST 2808 Technical, the concentration of cfu/g was high enough so the results meet the standards required for registration.

specified on product labels. A temporary exemption from a tolerance was granted on June 18, 2003 for use under an Experimental Use Permit. The granting of an exemption from a tolerance indicates that no risks to humans, including infants and children, are expected from approved uses of the end product on foods.

3. Risk Assessment

With no toxic endpoints identified, *B. pumilus* strain QST 2808 is not expected to pose any risks to the population, including workers, and infants or children. Required Personal Protective Equipment (PPE) will minimize any risks to workers and pesticide handlers. Residential exposure and risk are not expected because the end product is approved only for outdoor, greenhouse, and nursery uses, and because *B. pumilus* strain QST 2808 is not toxic or pathogenic to mammals.

C. Ecological Risk Assessment

No risks to non-target organisms are expected from approved uses of the end product. This conclusion is supported by non-target studies performed for avian species, freshwater fish, aquatic invertebrates, non-target insects, and honey bees. $LC_{50's}$ occurred at much higher concentrations than the organisms will encounter from approved uses of the end product. In addition, because there are no approved aquatic uses of *B. pumilus* strain QST 2808, exposure of aquatic organisms should be no higher than background, further lessening any potential risk. Waivers were granted for studies on plants, wild mammals, and estuarine/marine organisms.

D. Data Gaps and Mitigation Measures

No data gaps exist. Because there are no adverse effects expected on non-target organisms, *B. pumilus* strain QST 2808 is not expected to harm any threatened or endangered species. No mitigation measures are necessary.

II. OVERVIEW

A. Product Overview

- **Microbial Pesticide Name:** *Bacillus pumilus* strain QST 2808
- Trade Names: Sonata™ ASO; QST 2808 MUP
- **OPP Chemical Code**: 006485
- **Basic Manufacturer:** AgraQuest, Inc, 1530 Drew Avenue, Davis, CA 95616

B. Use Profile

Type of Pesticide: Biofungicide

Mechanism of action:

B. pumilus strain QST 2808 prevents the germination of fungal spores on plants by forming a physical barrier between the leaf and the pest spores, and then colonizing the pest spores. There is evidence that *B. pumilus* strain QST 2808 may induce systemic acquired resistance (SAR) in plants, causing an increase in the plants' ability to resist a variety of pathogens.

Use Sites:

Terrestrial Food. Brassica, bulb vegetables, cereal grains, cucurbits, fruiting vegetables, grapes, hops, leafy vegetables, legume vegetables, peanuts, pome fruits, root/tuber vegetables, stone fruits, strawberries, sweet corn, artichokes, asparagus, avocado, bananas, citrus, kiwi, mango, olive, papaya, tree nuts, and herbs and spices

Terrestrial Feed. Legume vegetables, peanuts, cereal grains

Terrestrial Non-Food. Fields of roses, grass seed, and tobacco; arbor culture in nurseries, landscapes, and rights-of-way

Forestry. Quercus species (oak), California bay laurel, big leaf maple, huckleberry, and honeysuckle

Target Pests for Active Ingredient: *Alaternia spp., Botytis spp. Bremia lactucae, Cercospora* spp*., Cercopshoridium personatum, Erysiphe* spp., *Erwinia amylovora*, *Oidiopsis taurica*, *Peronospora spp., Phoma cucurbitacearum, Phytophthora* spp*, Plasmopara viticola, Podosphaera leuchotricha, Pseudoperonospara cubensis, Puccinia* spp., *Sclerotinia* spp., *Sphaerotheca spp., Uncinula necator, Uromyces spp.*, *Venturia spp*., *Xanthomonas spp*. (Common names of target pests: Bacterial Spot, Blue Mold, Downy Mildew, Early Blight, Fire Blight, Gray Mold, Gummy Stem Blight, Late Blight, Powdery Mildew, Rust, Sclerotinia Leaf Drop, Scab, White Mold)

Formulation Types Registered:

Types: 1) Manufacturing Use Product; 2) End Product
Form:aqueous solution

Method and Rates of Application:

Types of Treatment

Timing Varies depending on crop. For most crops, users begin application when conditions are conducive to disease development, and repeat application at 7 to 10 day intervals or as needed.

Method of Application: Ground, aerial, or chemigation spray

Application: Application rates of Sonata™ ASO range from 2-4 quarts/acre for crops listed above under "Use sites." For treatment of ornamentals, shrubs, and trees (including treatment for sudden oak death syndrome), 2-4 quarts of product should be applied in 100 gallons of water to runoff. Sonata™ ASO contains a minimum of 1 x 10^{10} cfu per gram. The product will be applied as a foliar spray alone, in alternating spray programs, or in tank mixes with other registered pesticide products. Application instructions vary by crop.

C. Regulatory History

AgraQuest, Inc submitted an application on May 31, 2000 for registration of the product QST 2808 Technical (later renamed QST 2808 MUP), to be used for manufacturing an end product. The notice announcing this submission was published on May 8, 2002 (67 FR 30917-8) (FRL 6829-7).

In the Federal Register of May 3, 2001 (66 FR 22225) (FRL-6773-9), EPA issued a notice pursuant to section 408 of the FFDCA, 21 U.S.C. 346a, as amended by FQPA (Public Law 104-170), announcing the filing of a pesticide tolerance petition (PP-1G6240) by AgraQuest, Inc, 1530 Drew Avenue, Davis, CA 95616 asking for a temporary exemption from the requirement of a tolerance. This notice included a summary of the petition prepared by the petitioner AgraQuest, Inc. There were no comments received in response to the notice of filing. On the same day, a notice in the Federal Register announced receipt of a submission for an Experimental Use Permit for use of QST 2808 Technical on 4000 acres in 22 states on a variety of crops (May 3, 2001 (66 FR 22228) (FRL-6774-1)). The notice granting the temporary exemption from a tolerance was published in the Federal Register on June 18, 2003 (68 FR 36476-80)(FRL 7301-1). The granting of a 2-year EUP, expiring June 30, 2005, was announced in the Federal Register on August 20, 2003 (68 FR 50143)(FRL 7316-3).

Agraquest submitted a petition (PP 4F6826) for a permanent exemption from the requirement of a tolerance, which was announced and published in the Federal Register on May 5, 2004 (69 FR 25092-5) (FRL 7354-4). The announcement of the tolerance

exemption was published in the Federal Register on November 3, 2004 (69 FR 63950-54) (FRL 7684-4). The tolerance exemption at 40 CFR 180.1255, exempts *B. pumilus* QST 2808 from the requirements of a tolerance in or on all agricultural commodities when label directions are followed.

On October 14, 2004 EPA registered the first two pesticide products containing *Bacillus pumilus* strain QST 2808 as the active ingredient. These were:

QST 2808 MUP (EPA Reg # 69592-6) for manufacturing use only; and
Sonata™ ASO (EPA Reg # 69592-13) as an end product.

III. SCIENCE ASSESSMENT

A. Physical and Chemical Properties Assessment

1. Characteristics of *Bacillus pumilus* strain QST 2808.

Bacillus pumilus is a ubiquitous bacterium belonging to the *B. subtilis* group of bacilli commonly found in soil, water, air, and decomposing plant tissue that contributes to nutrient cycling when not in spore form. The amount of *B. pumilus* QST 2808 in the environment has not been determined, but bacilli generally occur in population levels of 10^6 to 10^7 per gram of soil. Cited studies show that the vast majority of soil bacilli populations are likely to exist in an inactive endospore stage unless soil is amended with organic matter providing readily utilizable nutrients.

The agency has classified *Bacillus pumilus* strain QST 2808 as a microbial pesticide active ingredient. Sonata™ ASO and QST 2808 MUP contain this active ingredient. Product chemistry data that support the registration of *Bacillus pumilus* strain QST 2808 are summarized in Table 1.

Table 1. Physical and Chemical Properties for *Bacillus pumilus* strain QST 2808

OPPTS GUIDELINE Number	STUDY	RESULT	MRID#
885.1100	Product Identity and Disclosure of Ingredients	Acceptable	452572-01
885.1200	Manufacturing Process	Acceptable	452572-01 456643-01
885.1300	Formation of Unintentional Ingredients	Acceptable	452572-01
885.1400	Analysis of Samples	Acceptable	452572-01
885.1500	Certification of Limits	Acceptable	452572-01 452572-02
830.6302, 830.6303, 830.6304, 830.7000, 885.7300	Product Chemistry	Acceptable	452572-01 456643-01

B. Human Risk Assessment

There is a reasonable certainty that no harm will result from exposure to *Bacillus pumilus* strain QST 2808. This includes all anticipated dietary exposures and all other exposures with reliable information.

1. Human Toxicity Assessment

a. Acute Toxicity

All mammalian toxicology data requirements have been submitted and adequately satisfy data requirements to support registration. Following are the toxicity categories for five of the acute studies:

Toxicity Category	Acute Study
IV	Oral
IV	Eye irritation
IV	Dermal toxicity (end product)
III	Inhalation
III	Dermal irritation
Yes	Dermal sensitization (end product)

Table 2, below, describes the toxicity studies. Studies were performed on the Technical (MUP) product unless endproduct is specified.

Table 2. Toxicity Data Requirements and Results

OPPTS GUIDELINE NUMBER	STUDY	RESULT	MRID#
885.3050	Acute Oral Toxicity/ Pathogenicity	Fifteen male and fifteen female rats each were administered 4.1×10^9 cfu of *B. pumilus* strain QST 2808 Technical and observed for 14 days. Based on the data, *B. pumilus* strain QST 2808 does not appear to be toxic, infective, or pathogenic in rats under these conditions. Classification: ACCEPTABLE. **Toxicity Category IV.**	**451366-04**
885.3100	Acute Dermal Toxicity/ Pathogenicity	The acute lethal dose (LD_{50}) of *Bacillus pumilus* strain QST 2808 Technical is greater than 2000 mg/kg. Classification: ACCEPTABLE. **Toxicity Category III.**	451366-05

OPPTS GUIDELINE NUMBER	STUDY	RESULT	MRID#
885.3150	Acute Pulmonary Toxicity/ Pathogenicity	Eighteen male and eighteen female rats each received 1.6 x 10^8 cfu *Bacillus pumilus* strain QST 2808 Technical by a single intratracheal administration. Rats were monitored for 35 days for clinical signs of toxicity. Necropsy studies showed no significant signs of abnormalities due to the test organism. Based on the data, *B. pumilus* strain QST 2808 was not toxic, infective, or pathogenic to rats when administered at 1.6 x 10^8 cfu/animal. Classification: ACCEPTABLE.	451366-06
870.1100	Acute Oral Toxicity (endproduct)	Rats were dosed with Sonata™ AS containing 1.7 x 10^{10} cfu/mL at 5000mg/kg body wt. The oral LD50 was >5000mg/kg. ACCEPTABLE. **Toxicity category IV.**	460295-04
870.1200	Acute Dermal Toxicity (endproduct)	The dermal LD50 for rats was > 5000 mg/kg body weight of Sonata™ AS containing 1.7 x 10^{10} cfu/mL. ACCEPTABLE. **Toxicity Category IV.**	460295-05
870.1300	Acute Inhalation (Endproduct)	Rats were exposed nose only for 4 hours, and observed for the following 14 days. No significant clinical signs were noted. The LC_{50} of Sonata™ AS for rats is greater than 1.06 mg/L. ACCEPTABLE. **Toxicity Category III.**	460295-06

OPPTS GUIDELINE NUMBER	STUDY	RESULT	MRID#
885.3200	Acute Intravenous Toxicity/Pathogenicity	Eighteen male and eighteen female rats each were dosed at 1.6 x 10^8 cfu *Bacillus pumilus* strain QST 2808 Technical intravenously and monitored over a period of 28 days. A gross necropsy was performed on all rats. Based on the data, the test organism was not toxic, infective, or pathogenic to rats. Classification: ACCEPTABLE.	451366-07
870.2400	Primary Eye Irritation	Three male rabbits each were administered 0.1 mL of QST 2808 Technical in the everted lower lid of one eye and then observed for 72 hours. Based on the data, QST 2808 Technical showed minimal effects to the eye. Classification: ACCEPTABLE. **Toxicity Category IV.**	452679-01
870.2400	Primary Eye Irritation (endproduct)	Six rabbits each received 1.7 x 10^9 cfu in their right eyes. Sonata™ AS was minimally irritating. ACCEPTABLE. **Toxicity Category IV**	460295-07
870.2500	Primary Dermal Irritation	When dosed with QST 2808 Technical at 0.5 mL/rabbit, QST 2808 Technical was essentially non-irritating. Classification: ACCEPTABLE. **Toxicity Category IV.**	452679-02

OPPTS GUIDELINE NUMBER	STUDY	RESULT	MRID#
870.2500	Primary Dermal Irritation (endproduct)	0.5ml product (containing 1.7 x 10^{10} CFU/mL) was administered to 6 rabbits on a 1x1- inch patch of clipped skin. Sonata™ AS was slightly irritating. ACCEPTABLE. **Toxicity Category III**	460295-08
870.2600	Delayed Contact Hypersensitivity in Guinea Pigs (endproduct)	Female guinea pigs were dosed on clipped skin once a week for 3 weeks with 0.4mL of Sonata™ AS containing 1.7 x 10^{10} cfu/mL. When challenged 14 days after the last induction, 1/19 guinea pigs showed sensitization. Sonata™ AS is a dermal sensitizer. ACCEPTABLE.	460295-10
870.2600	Delayed Contact Hypersensitivity in Guinea Pigs	Female guinea pigs were dosed on shaved skin once a week for 3 weeks with 0.4mL of QST 2808 Technical (total 2.9 x 10^{9} cfu). When challenged 14 days after the last induction, no signs of sensitization appeared. ACCEPTABLE.	460295-09

b. Subchronic Toxicity and Chronic Toxicity

Subchronic and chronic toxicity studies were not required because survival, replication, infectivity, toxicity, or persistence of the microbial agent was not observed in the test animals treated in the acute oral infectivity test.

c. Effects on the Endocrine and Immune Systems

EPA is required under section 408(p) of the FFDCA, as amended by FQPA, to develop a screening program to determine whether certain substances (including all pesticide active and other ingredients) "may have an effect in humans that is similar to an effect produced by a naturally-occurring estrogen, or other such endocrine effects as the

Administrator may designate." Following the recommendations of its Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC), EPA determined that there is no scientific basis for including, as part of the screening program, the androgen and thyroid hormone systems in addition to the estrogen hormone system. EPA also adopted EDSTAC's recommendation that the program include evaluations of potential effects in wildlife. For pesticide chemicals, EPA will use FIFRA and, to the extent that effects in wildlife may help determine whether a substance may have an effect in humans, FFDCA authority to require wildlife evaluations. As the science develops and resources allow, screening of additional hormone systems may be added to the Endocrine Disruptor Screening Program (EDSP). When the appropriate screening and/or testing protocols being considered under the Agency's EDSP have been developed, *Bacillus pumilus* strain QST 2808 may be subjected to additional screening and/or testing to better characterize effects related to endocrine disruption. To date, the Agency has no information to suggest that *Bacillus pumilus* strain QST 2808 has an effect on the endocrine systems.

Moreover, as is expected from a non-pathogenic microorganism that is practically non-toxic to mammals, the submitted toxicity/pathogenicity studies in rodents indicated that following several routes of exposure, the immune system remains intact and able to process and clear the active ingredient after exposure. No evidence of a compromised immune system was found after oral, dermal, or pulmonary exposure. (BPPD Review-1/7/02).

2. Dose Response Assessment

No toxicological endpoints are identified from oral, dermal, or pulmonary exposure to this microbial agent. (*See* Table 2. Toxicity Data Requirements). Therefore, a dose-response assessment cannot be performed.

3. Aggregate Exposure and Risk Characterization

In examining aggregate exposure, section 408 of the 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 water or surface water and exposure through pesticide use in gardens, lawns, or buildings (residential and other indoor uses). There is no evidence of adverse effects from oral, dermal, or pulmonary exposure to this microbial agent. (*See* **Table 2. Toxicity Data Requirements**)

a. Dietary. Humans and animals are commonly exposed to *B. pumilus* strain QST 2808, a ubiquitous microorganism that inhabits soil. No toxicological endpoints were identified for *B. pumilus* strain QST 2808. The low toxicity and non-pathogenicity/infectivity of *B. pumilus* strain QST 2808 is demonstrated by the data summarized in Table 2. Toxicity Data Requirements of this document..

i. Food While the proposed use pattern may result in dietary exposure with possible residues in or on agricultural commodities, negligible to no risk is expected for the general population, including infants and children, or animals because *B. pumilus* strain QST 2808 demonstrated no pathogenicity or oral toxicity at the maximum doses tested, as noted above (Table 2. Toxicity Data Requirements).

ii. Drinking Water

Exposure from drinking water is not expected to cause adverse effects. The potential for transfer of *B. pumilus* strain QST 2808 to surface or ground water during run-off after pesticide application is considered minimal to non-existent, due in part to the microbe's percolation through and resulting capture in soil. Accordingly, the use of this microbial pest control agent on terrestrial plants is not expected to affect the quality of drinking water.

b. Other Non-occupational Exposure

Based on the proposed use patterns, the potential for non-dietary exposures to *B. pumilus* strain QST 2808 pesticide residues for the general population, including infants and children, is very low. Accordingly, the Agency believes that the potential aggregate non-occupational exposure through dermal and inhalation exposure will be too low to harm the general population, including infants and children.

1. *Dermal exposure*.

Dermal exposure to *B. pumilus* strain QST 2808 pesticide residues for the general population, including infants and children, is unlikely because the end product is applied to foliage at outdoor agricultural and horticultural sites, and dermal exposure to the general public will be very low. Because *B. pumilus* strain QST 2808 occurs in soil, there is a great likelihood of prior exposure for most, if not all, individuals. Any increase in dermal exposure due to uses of pesticidal products containing *B. pumilus* strain QST 2808 would be negligible. Furthermore, as demonstrated in Unit III of this document, the organism is of low dermal toxicity; the acute lethal dose (LD_{50}) is greater

than 2000 mg/kg; the QST 2808 Technical was essentially non-irritating (Toxicity Category IV); and only a weak hypersensitivity reaction was seen at the high dose tested in guinea pigs. Accordingly, the risks anticipated for the dermal route of exposure are considered minimal.

2. *Inhalation exposure*. The potential for inhalation exposure to *B. pumilus* strain QST 2808 pesticide residues for the general population, including infants and children, is low because potential use sites are agricultural and horticultural. Furthermore, no significant clinical signs were observed in rats exposed to Sonata™ AS by inhalation 4 hours/day for two weeks, as demonstrated in Table 2. Toxicity Data Requirements of this document. Therefore, the risks anticipated from inhalation exposure are considered minimal.

4. Dietary Exposure and Risk Characterization

Humans and animals are commonly exposed to *B. pumilus* strain QST 2808, an organism found in soil. No toxicological or pathological endpoints were identified from oral exposure to this microbe, as demonstrated in **Table 2. Toxicity Data Requirements** of this document. Furthermore, the granting of a tolerance exemption to this active ingredient indicates that residues are not expected to cause adverse effects when people, including infants and children, are exposed via food or drinking water.

5. Residential, School, Day Care, and Occupational Exposure and Risk Characterization

There is a reasonable certainty that no harm to the U.S. population, including infants and children, will result from aggregate exposure to residues of *B. pumilus* strain QST 2808 due to its use in pesticide products. This includes all anticipated dietary exposures and all other exposures for which there is reliable information. As discussed previously, *B. pumilus* strain QST 2808 is not pathogenic or infective and is practically non-toxic to mammals. Accordingly, exempting *Bacillus pumilus* strain QST 2808 from the requirement of a tolerance should pose no significant risk.

FFDCA section 408(b)(2)(C) provides that EPA shall apply an additional tenfold margin of exposure (safety) for infants and children in the case of threshold effects to account for prenatal and postnatal toxicity and the completeness of the data base on toxicity and exposure, unless EPA determines that a different margin of exposure (safety) will be safe for infants and children. Margins of exposure (safety) are incorporated into EPA risk assessments either directly through the use of a margin of exposure analysis or by using uncertainty (safety) factors in calculating a dose level that poses no appreciable risk to humans. Due to the ubiquitous nature of *B. pumilus*

strain QST 2808, residues of this microbial pesticide in or on agricultural commodities are not expected to significantly increase exposure to the U.S. population, including infants and children. Here, EPA concludes that the toxicity and exposure data are sufficiently complete to adequately address the potential for additional sensitivity of infants and children to residues of *B. pumilus* strain QST 2808 and that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure to *B. pumilus* strain QST 2808 residues.

a. Occupational Exposure and Risk Characterization

Occupational exposure is minimized by use of PPE. In addition, lack of toxic or pathogenic endpoints by any route of exposure indicates that occupational risk is minimal to non-existent.

6. Drinking Water Exposure and Risk Characterization

No risks are expected from exposure to *B. pumilus* strain QST 2808 via drinking water because exposure will be minimum and the organism showed no harmful effects on animals that were exposed orally. The potential for transfer of *B. pumilus* strain QST 2808 to surface or ground water during run-off is considered minimal to non-existent, due in part to its slow movement in soil, where the organism is normally found. Product labels instruct users not to allow *B. pumilus* strain QST 2808 to enter bodies of water during use or disposal. Therefore, potential exposure to *B. pumilus* strain QST 2808 in surface and drinking water is negligible.

7. Acute and Chronic Dietary Risks for Sensitive Subpopulations Particularly Infants and Children

Based on the acute toxicity information discussed above, there is a reasonable certainty that no harm to the U.S. population, including infants and children, will result from dietary exposure to residues of *B. pumilus* strain QST 2808 due to its use as a microbial pest control agent.

FFDCA section 408 provides that EPA shall apply an additional ten-fold margin of exposure (safety) for infants and children in the case of threshold effects to account for pre- and post-natal toxicity and the completeness of the database, unless EPA determines that a different margin of exposure (safety) will be safe for infants and children. Margins of exposure (safety) are often referred to as uncertainty (safety) factors. In this instance, the Agency believes there is reliable data to support the

conclusion that *Bacillus pumilus* strain QST 2808 is practically non-toxic to mammals, including infants and children, and that there are no threshold effects; therefore, EPA has not used a margin of exposure (safety) approach to assess the safety of *Bacillus pumilus* strain QST 2808 . As a result, the provision requiring an additional margin of exposure (safety) does not apply.

8. Aggregate Exposure from Multiple Routes Including Dermal, Oral, and Inhalation

Because no toxic endpoints for mammals have been identified, and because no toxic effects have been reported from limited human exposure, no toxicity or pathogenicity is expected from aggregate exposure of the public via inhalation, dermal, and oral routes of exposure. Worker exposure via inhalation and dermal routes will be minimized by the use of personal protective equipment.

Based on the available information, EPA concludes that there is a reasonable certainty that no harm will result from aggregate exposure to the United States population, including infants and children, to residues of *Bacillus pumilus* strain QST 2808. This includes all anticipated dietary exposures and all other exposures for which there is reliable information.

9. Cumulative Effects

The Agency has considered the potential for cumulative effects of *B. pumilus* strain QST 2808 and other substances in relation to a common mechanism of toxicity. These considerations include the possible cumulative effects of such residues on infants and children. *B. pumilus* strain QST 2808 is practically non-toxic to mammals. Because no mechanism of pathogenicity or toxicity in mammals has been identified for this organism (see above), no cumulative effects are anticipated if the residues of a product with *B. pumilus* strain QST 2808 interact with related microbial pesticides.

C. Environmental Assessment

The purpose of this section is to determine if the submitted non-target studies and written data waiver justifications for several of the OPPTS Microbial Pesticide Guideline requirements are sufficient to establish that "no unreasonable adverse effects" are expected from exposure of non-target organisms to *Bacillus pumilus* QST 2808. 40 CFR part 158.45

contains provisions for granting waivers for data requirements in response to specific written requests by applicants.

1. Environmental Fate

Bacillus pumilus is a ubiquitous bacterium belonging to the *subtilis* group of bacilli commonly found in soil, water, air, and decomposing plant tissue. Bacilli typically occur at 10^6 to 10^7 cfu/g of soil. *Bacillus pumilus,* under most conditions, is inactive and exists in spore form. *Bacillus pumilus* is not known to be pathogenic or toxic to animal or plant species, or to produce toxins. *Bacillus pumilus* produces cytolytic enzymes, proteases, and other enzymes that degrade a variety of natural substrates and contribute to nutrient recycling. It also produces antibiotics. The microbe prevents fungal spore germination by forming a physical barrier between the leaf surface and the fungal spores, and then colonizing the fungal spores. *Bacillus pumilus* is used as a hay preservative in animal feed and in food and chemical fermentation processes.

2. Ecological Toxicity

a. Overview of toxic effects on non-target species

The registrant has submitted studies for avian species, freshwater fish, aquatic invertebrates, non-target insects, and honey bees. *The registrant also submitted written data waiver justifications for the other OPPTS Microbial Pesticide Testing Guideline* requirements (non-target plant, wild mammal toxicity/pathogenicity, and estuarine/marine organisms) based on the following arguments: 1) *Bacillus pumilus* is ubiquitous in soil, air, and water and its level in the environment will not significantly increase from the proposed usage; 2) no adverse effects have been observed in greenhouse and field trials; 3) an extensive literature search did not reveal reports of adverse effects in plants (aquatic or terrestrial); 4) the lack of infectivity or toxicity of *B. pumilus* to mammals in the laboratory (MRID No. 451366-04, -06, -07); 5) the presence of *B. pumilus* in aquatic environments with no reports of adverse effects on marine or estuarine organisms; 6) minimal effects observed in submitted non-target freshwater organism studies (MRID Nos. 460295-12, -13, and -14).

The Agency has performed an assessment of submitted data and justifications of waiver requests, and has determined that the proposed uses of *Bacillus pumilus* strain QST 2808 are not expected to have adverse effects on avian species, wild mammals, non-target insects (including the honey bee), freshwater fish and aquatic invertebrates, marine/estuarine organisms, and non-target aquatic and terrestrial plants. Because *Bacillus pumilus* strain QST 2808 does not appear to pose a risk to wildlife at the proposed use rates, there is also no "may affect" finding to any endangered/threatened species listed by the U.S. Fish and Wildlife Service. (See Table 3, below, for details.)

Table 3. Toxicity Data for Non-target Organisms

Guideline No	Study	Results	MRID NO
USEPA OPPTS 885.4050	*Avian Oral Testing, Tier I*	The dietary LC_{50} value for *B. pumilus* QST 2808 administered to immature northern bobwhite quail was >7.0 x 10^{10} cfu/kg of body weight per day for five days. Observation for 30 days did not reveal treatment-related signs of illness or abnormal behavior. There was no evidence of pathogenicity due to treatment during gross necropsy at test termination. Body weight and feed consumption of the treatment birds were comparable to those of the negative control. ACCEPTABLE.	460295-11
885.4150	Wild Mammal Toxicity/Pathogenicity	No hazards from *B. pumilus* QST 2808 for wild mammalian species are anticipated for this use. The rat laboratory studies submitted in support of this registration indicate that there is no toxicity or pathogenicity to rodents from testing at the maximum hazard dose (MRID Nos. 451366-04, -06, -07). See Table 2 of this document for details. The rodent results support a waiver for testing under Guideline No 885.4150.	Data waived
885.4340	Non-target Insect Testing, Tier I (Green Lacewing Larvae)	*The LC_{50} for green lacewing larvae exposed to 1X-100X the expected field dose of B. Pumilus QST 2808 in a moth egg diet over 12 days was 12,300 ppm (1.32 x 10^{11} cfu per kg of diet). The LC_{50} is approximately 25X higher than the expected field level. Based on the Kenaga nomogram, the expected residues on vegetation at this rate of application fall well below 12,300 ppm.* No risk is expected from approved uses. ACCEPTABLE.	460295-15
885.4340	Non-target Insect Testing, Tier I (Parasitic Hymenoptera)	*The LC_{50} for Nasonia vitripennis* exposed in its diet to 1X-100X the expected field level of *B. pumilus* QST 2808 in the diet over ten days was >50,000 ppm. The LC_{50} is >100x the expected field level. *No risk is expected from approved uses.* ACCEPTABLE.	460295-16
885.4340	Non-target Insect Testing, Tier I (Ladybird Beetle)	*The LC_{50} for adult ladybird beetles exposed to 1X-100X expected field levels of B pumilus QST 2808 in the diet over seven days was >50,000 ppm. The LC_{50} is >100x the expected field use rate.* ACCEPTABLE.	460295-17

Guideline No	Study	Results	MRID NO
885.4380	Honeybee Testing, Tier I	Toxicity and pathogenicity of *QST 2808 strain of B. pumilus to honey bees was determined by a semi-field, dietary study in standard-sized colonies for 30 days. Various toxicity/pathogenicity measures were the same for negative control and treated groups. Test material contained* 1.1 x 10^{10} cfu/mL *The Johansen Mayer hazard rating for QST 2808 strain of B. pumilus was non-hazardous (≤ 100 dead bees found/day).* No risk is expected from approved uses. ACCEPTABLE.	460295-18
885.4200	Freshwater Fish Testing	The LC_{50} for juvenile rainbow trout was 2.6 x 10^7 cfu/mL (~440x the EEC in a 6-inch layer of water immediately after a direct application) in a 30-day dietary study. Assuming direct application of QST 2808 strain of *B. pumilus* to the aquatic environment, the risk quotient (RQ) for rainbow trout based on the LC_{50} is 0.0001 with a standard application rate of 1 gallon/acre (4 x 10^{13} cfu/acre). This is a worst case RQ because the product is not intended for direct application to water. This RQ is well below any levels of concern. Results from necropsies suggest that QST 2808 is not pathogenic to rainbow trout. High mortality in a control with the highest dose of sterile filtrate suggests that mortality in the test groups may have been due to the physical nature of the test substance, rather than the active microbial ingredient. These data show that QST 2808 strain of *B. pumilus* is highly unlikely to cause adverse effects to freshwater fish at field use rates. ACCEPTABLE.	460295-12
850.1010	Freshwater Aquatic Invertebrate Acute Toxicity Testing, Tier I	Acute toxicity of *B. pumilus* QST 2808 to *Daphnia magna* neonates was determined in a 48-hour static bioassay with an estimated 48-hour EC_{50} >2.7 x 10^7 cfu/mL. The data show that QST 2808 strain of *B. pumilus* is not acutely toxic to aquatic invertebrates at expected field use rates. This study is not required for registration.	460295-13

Guideline No	Study	Results	MRID NO
885.4240	Freshwater Aquatic Invertebrate Testing, Tier I	The 21-day EC_{50} for daphnids was 6.2 x 10^5 cfu/mL in a 21-day static renewal bioassay test. Assuming direct application of QST 2808 strain of *B. pumilus* to the aquatic environment, the RQ for daphnids based on the EC_{50} is 0.1 with an application rate of one gallon/acre (4 x 10^{13} cfu/acre). This is a worst case RQ because the product is not intended for direct application to water. Assuming 5% drift from terrestrial application into the aquatic environment lowers the RQ to 0.005, which is well below the level of concern for aquatic organisms. These data show that QST 2808 strain of *B. pumilus* is not likely to cause adverse effects in aquatic invertebrates at expected field use rates. ACCEPTABLE.	460295-14
885.4280	Estuarine/Marine Organisms Toxicity/Pathogenicity	The data submitted for freshwater fish and *Daphnia* (MRID Nos. 460295-12, -13, and -14; see above) are sufficient to demonstrate that *B. pumilus* QST 2808 is not likely to pose a hazard to estuarine or marine organisms. Exposure will be very low. Therefore, this study requirement was waived.	Data waived
885.4300	*Non-target Plant Studies, Tier I*	*Bacillus pumilus* is ubiquitous and the proposed uses are not expected to result in increased exposure or adverse effects in non-target plants; no adverse effects have been observed in greenhouse and field trials conducted to determine the efficacy of Sonata™ ASO; and the majority of cited literature demonstrates that *B. pumilus* has no adverse effects on non-target plants. *Bacillus pumilus* is not listed as a plant pathogen in *Bergey's Manual of Systematic Bacteriology* (Claus & Berkeley 1986) or in the USDA's Widely Prevalent Plant Pathogenic Bacteria in the United States (USDA 1998). Based on these findings, non-target plant studies were waived.	Data waived
None	Endangered Species Impact Assessment	The Agency performed an Endangered Species Act (ESA) assessment and determined that no adverse effects are expected to endangered/threatened species. This conclusion is justified because no risks to animals or plants are expected from approved uses of *B. pumilus* QST 2808, and therefore there is no "may affect" finding for endangered species.	None assigned

a. *B. pumulis* May Induce Systemic Acquired Resistance

Cited studies provide evidence that *B. pumilus* induces in some plants a response known as "systemic acquired resistance (SAR)," which enhances a plant's ability to resist numerous pathogens. Reported examples of this induced resistance include: resistance against bacterial wilt (*Erwinia tracheiphilia*) in cucumbers; mycelial growth inhibition of multiple species of *Aspergillus*, *Penicillium*, and *Fusarium*; inhibition of *Botrytis cinerea*; suppression of take-all disease of wheat caused by *Gaumannomyces graminis*; postharvest control of *Penicillium digitatum* decay on citrus fruit; efficacy in the control of plant pathogens like *Botrytis cinerea* in strawberries; and suppression of *Phytophthora vignae* on greenhouse cowpea. *B. pumilus* has been reported as the most common isolate in the soybean phylloplane, and as a common endophyte of stems in healthy, field-grown cotton. So, in addition to not causing adverse effects in non-target plants, *B pumilus* strain QST 2808 may enhance the natural systems that plants use to defend themselves against pathogenic organisms.

3. ENVIRONMENTAL ASSESSMENT SUMMARY

The data waiver justifications and studies submitted by the registrant do not show a risk *from B. pumilus* QST 2808 to non-target organisms.

The registrant sufficiently demonstrated that additional mammalian toxicity/pathogenicity studies (USEPA OPPTS 885.4150) are not necessary, primarily due to the lack of toxicity and pathogenicity findings when *B. pumilus* QST 2808 was tested in laboratory rats (MRID Nos. 451366-04, -06, -07). The freshwater fish and daphnid data (MRID Nos. 460295-12, -13, and -14) show that no adverse effects on non-target marine or estuarine organisms are expected from the proposed uses of *B. pumilus* QST 2808 .

Proposed uses of Sonata™ ASO are limited to foliar applications to growing plants by ground, aerial, or chemigation spray equipment on terrestrial agricultural crops. During various studies, the registrant reports that no significant adverse effects on non-target plants were observed. Several citations (see "Non-target Plant Risk Assessment" section above) demonstrate that *B. pumilus* may induce SAR against several plant pathogens. The citations also show that *B. pumilus* is a common isolate in the soybean phylloplane and a common endophyte of stems in healthy, field-grown cotton. Thus, the weight of evidence provided in the literature citations suggests that there will be no hazard to non-target plants from the proposed uses of *B. pumilus* QST 2808.

The submitted literature citations and studies show that the proposed uses of Sonata™ ASO are not likely to result in adverse effects to non-target organisms, including avian species, insects, freshwater fish and invertebrates, estuarine and marine animals, and non-target plant

species. Since a review of the submitted studies and citations shows no evidence that Sonata™ ASO poses a risk to wildlife at the proposed use rates, there is also a no "may affect" finding to endangered/threatened species listed by the USFWS.

References

Claus, D., and R.C.W. Berkeley. 1986. Genus *Bacillus* Cohn 1872, pp. 1105-1139. In P.H.A. Sneath, et al. (eds.), Bergey's Manual of Systematic Bacteriology, Vol. 2. Williams and Wilkins Co., Baltimore, MD.

USDA. 1998. Widely prevalent plant pathogenic bacteria in the United States.

IV. RISK MANAGEMENT AND REGISTRATION DECISION

A. Determination of Eligibility-

Section 3(c)(5) of FIFRA provides for the registration of new active ingredients if it is determined that (A) its composition is such as to warrant the proposed claims for it; (B) its labeling and other materials required to be submitted comply with the requirements of FIFRA; (C) it will perform its intended function without unreasonable adverse effects on the environment; and (D) when used in accordance with widespread and commonly recognized practice, it will not generally cause unreasonable adverse effects on the environment.

To satisfy criterion "A" above, *Bacillus pumilus* strain QST 2808 has well known properties. The Agency has no knowledge that would contradict the claims made on the label of this product. Criterion "B" is satisfied by the current label and by the data presented in this document. It is believed that this new pesticidal active ingredient will not cause any unreasonable adverse effects, is a broad spectrum microbial fungicide, and does provide protection as claimed satisfying criterion "C". Criterion "D" is satisfied in that *Bacillus pumilus* strain QST 2808 is not expected to cause unreasonable adverse effects when used according to label instructions.

Therefore, *Bacillus pumilus* strain QST 2808 is eligible for registration. The uses are listed in the **Section II, B. Use Profile**. There are no ineligible uses.

B. Regulatory Position

1. Unconditional Registration

The data requirements are fulfilled and the Biopesticides and Pollution Prevention Division recommends unconditional registration of products that contain *Bacillus*

pumilus strain QST 2808 as the sole active ingredient (QST 2808 MUP; Sonata™ ASO).

2. Tolerances for Food Uses and /or exemptions

EPA received a pesticide petition (PP 4F6826) from AgraQuest, Inc, Davis, CA, proposing [pursuant to section 408(d) of the Federal Food, Drug and Cosmetic Act, 21 U.S.C. section 346a(d)], to amend 40 CFR part 180 by establishing an exemption from the requirement of a tolerance for residues of the microbial pesticide, *Bacillus pumilus* strain QST 2808.

EPA is issuing a notice establishing an exemption from the requirements of a tolerance for residues of *Bacillus pumilus* strain QST 2808 in or on all food commodities.

3. CODEX Harmonization

There is no Codex Alimentarius Commission Maximum Residue Level (MRL) for *Bacillus pumilus* strain QST 2808.

4. Risk Mitigation

There are no significant risk issues relating to dietary risk, residential risk, or ground and surface water contamination. Therefore, mitigation measures for them are not required. Occupational risk will be mitigated by appropriate label precautions and required PPE.

5. Endangered Species Statement

The submitted data and waiver request justifications show no adverse effects from the proposed uses of Sonata™ ASO to avian species, mammals, non-target insects (including the honey bee), freshwater fish and aquatic and non-aquatic invertebrates, marine/estuarine organisms, and non-target plants. Given that *B. pumilus* strain QST 2808 has not been shown to pose a risk to non-target organisms at the approved use rates, there is also no "may affect" finding to any endangered/threatened species listed by the U.S. Fish and Wildlife Service.

Therefore. the Agency has determined that this registration action will have no adverse effects on currently listed endangered and threatened species.

C. Labeling Rational

1. Human Health Hazard (WPS and non-WPS)

Products containing *Bacillus pumilus* strain QST 2808 for use on commercial use sites are subject to the Worker Protection Standard. Because of the low toxicity of *Bacillus pumilus* strain QST 2808, the Re-Entry Interval for uses within the scope of WPS is 4 hr. Precautionary statements and personal protective equipment as specified below are required based on the acute toxicity categories of this organism.

2. Environmental Hazard

Precautionary labeling is required as indicated below.

V. ACTIONS REQUIRED BY REGISTRANTS

A. Precautionary Labeling

Bacillus pumilus strain QST 2808 products must state the following under the heading "Precautionary Statements":

Personal Protective Equipment required for Applicators and other Handlers:

Long sleeved shirt and long pants.
Waterproof gloves.
Shoes plus socks.
NIOSH approved respirator with any N, P, R, or HE filter.

WPS labels must state the following under the heading "User Safety Recommendations"

Wash hands before eating, drinking, chewing gum, using tobacco, or using the toilet.

Remove clothing immediately if pesticide gets inside. Then wash skin thoroughly and put on clean clothing.

Remove PPE immediately after handling this product. Wash the outside of gloves before removing. As soon as possible, wash skin thoroughly and change into clean clothing.

B. Environmental Hazards Labeling

Provided the following statement is placed into the environmental hazards statement, the risk of *Bacillus pumilus* strain QST 2808 is minimal to nonexistent to non-target organisms including endangered species.

1. End-Use Product Environmental Hazards Labeling

"Do not apply directly to water, or to areas where surface water is present or to intertidal areas below the mean high water mark. Do not contaminate water by cleaning of equipment or disposal of equipment washwaters. "

2. Manufacturing-Use Product Environmental Hazards Labeling

"Do not discharge effluent containing this product into lakes, streams, ponds, estuaries, oceans, or public water unless this product is specifically identified and addressed in an NPDES permit. Do not discharge effluent containing this product to sewer systems without previously notifying the sewage treatment plant authority. For guidance, contact your State Water Board or Regional Office of the EPA."

3. Application Rate

It is the Agency's position that the labeling for the pesticide products containing *Bacillus pumilus* strain QST 2808 as the active ingredient complies with the current pesticide labeling requirements. The Agency has not set a maximum number of applications per season of this active ingredient.

C. Labeling

The labels for QST 2808 MUP (EPA File # 69592-6) and for Sonata™ ASO (EPA File # 69592-13) conform with the labeling requirements for products containing *Bacillus pumilus* strain QST 2808 as the active ingredient. See ingredient statements below.

Manufacturing Use Product:

QST 2808 MUP

ACTIVE INGREDIENT

Bacillus pumilus QST strain 2808.....1.42.%

OTHER INGREDIENTS.....98.58%

Total 100.00%

Contains a minimum of 1×10^{10} cfu/g

End Use Product:

Sonata ASO

ACTIVE INGREDIENT

Bacillus pumilus QST strain 2808.....1.38.%

OTHER INGREDIENTS.....98.62%

Total 100.00%

Contains a minimum of 1 x 10^{10} cfu/g.

For both products, signal word is "Caution," based on studies showing toxicity category III. The label will include all the information required according to the Label Review Manual, such as:

- Product Name
- Ingredient Statement
- Registration Number
- "Keep Out of Reach of Children"
- Directions for Use

VI. Use Sites

Table 3. End Product Use Sites

Terrestrial Food. Brassica, bulb vegetables, cereal grains, cucurbits, fruiting vegetables, grapes, hops, leafy vegetables, legume vegetables, peanuts, pome fruits, root/tuber vegetables, stone fruits, strawberries, sweet corn, artichokes, asparagus, avocado, bananas, citrus, kiwi, mango, olive, papaya, tree nuts, and herbs and spices Terrestrial Feed. Legume vegetables, peanuts, cereal grains Terrestrial Non-Food. Fields of roses, grass seed, and tobacco; arbor culture in nurseries, landscapes, and rights-of-way Forestry. Quercus species (oak), California bay laurel, big leaf maple, huckleberry, and honeysuckle	Registered: October 14, 2004

BIBLIOGRAPHY

General References

Claus, D., and R.C.W. Berkeley. 1986. Genus *Bacillus* Cohn 1872, pp. 1105-1139. In P.H.A. Sneath, et al. (eds.), Bergey's Manual of Systematic Bacteriology, Vol. 2. Williams and Wilkins Co., Baltimore, MD.

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