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Proposed Registration Decision

Gliocladium Catenulatum strain J1446

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Publications
Pest Management Regulatory Agency
Health Canada
2720 Riverside Drive
A.L. 6605C
Ottawa, Ontario
K1A 0K9

Internet: pmra_publications@hc-sc.gc.ca
www.pmra-arla.gc.ca
Facsimile: 613-736-3758
Information Service:
1-800-267-6315 or 613-736-3799
pmra_infoserv@hc-sc.gc.ca

Canada

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Overview

Proposed Registration Decision for *Gliocladium Catenulatum* Strain J1446

Health Canada's Pest Management Regulatory Agency (PMRA), under the authority of the *Pest Control Products Act* and Regulations, is proposing full registration for the sale and use of *Gliocladium catenulatum* J1446 Dried Cell Mass and Prestop Biofungicide WP containing the technical grade active ingredient *Gliocladium catenulatum* strain J1446 for the suppression of a variety of fungal diseases on the following greenhouse-grown vegetables, herbs and ornamentals: cucumber, tomato, pepper, lettuce, cauliflower, broccoli, oregano, basil, parsley, thyme, dill, alyssum, geranium, pansy, petunia, salvia, snapdragon, tagetes, poinsettia and saintpaulia.

An evaluation of available scientific information found that, under the approved conditions of use, the product has value and does not present an unacceptable risk to human health or the environment.

This Overview describes the key points of the evaluation, while the Science Evaluation Section provides detailed technical information on the human health, environmental and value assessments of *Gliocladium catenulatum* strain J1446 Dried Cell Mass and Prestop Biofungicide WP.

What Does Health Canada Consider When Making a Registration Decision?

The key objective of the *Pest Control Products Act* is to prevent unacceptable risks to people and the environment from the use of pest control products. Health or environmental risk is considered acceptable[1] if there is reasonable certainty that no harm to human health, future generations or the environment will result from use or exposure to the product under its proposed conditions of registration. The Act also requires that products have value[2] when used according to label directions. Conditions of registration may include special precautionary measures on the product label to further reduce risk.

To reach its decisions, the PMRA applies modern, rigorous risk-assessment methods and policies. These methods consider the unique characteristics of sensitive subpopulations in humans (e.g. children) as well as organisms in the environment (e.g. those most sensitive to environmental contaminants). These methods and policies also consider the nature of the effects observed and the uncertainties present when predicting the impact of pesticides. For more

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1 "Acceptable risks" as defined by subsection 2(2) of the *Pest Control Products Act.*

2 "Value" as defined by subsection 2(1) of the *Pest Control Products Act* is "...the product's actual or potential contribution to pest management, taking into account its conditions or proposed conditions of registration, and includes the product's (a) efficacy; (b) effect on host organisms in connection with which it is intended to be used; and (c) health, safety and environmental benefits and social and economic impact."

information on how the PMRA regulates pesticides, the assessment process and risk reduction programs, please visit the PMRA's website at www.pmra-arla.gc.ca.

Before making a final registration decision on *Gliocladium catenulatum* strain J1446 , the PMRA will consider all comments received from the public in response to this consultation document.[3] The PMRA will then publish a Registration Decision document[4] on *G. catenulatum* strain J1446, which will include the decision, the reasons for it, a summary of comments received on the proposed final registration decision and the PMRA's response to these comments.

For more details on the information presented in this Overview, please refer to the Science Evaluation Section of this consultation document.

What is *Gliocladium catenulatum* strain J1446?

Gliocladium catenulatum is a fungus that grows on dead organic matter that can be commonly found in soil worldwide. *G. catenulatum* strain J1446 was originally isolated as a microbial pest control agent (MPCA) because of its ability to suppress soil-borne fungal diseases on plants.

Health Considerations

Can approved uses of *Gliocladium catenulatum* strain J1446 affect human health?

Gliocladium catenulatum strain J1446 is unlikely to affect your health when Prestop Biofungicide WP is used according to label directions.

Exposure to *G. catenulatum* strain J1446 may occur during handling and application of Prestop Biofungicide WP. When assessing health risks, several key factors are considered: the microorganism's biological properties (e.g. production of toxic byproducts); reports of any adverse incidents; its potential to cause disease or toxicity as determined in toxicological studies; and the levels to which people may be exposed relative to exposures already encountered in nature to other isolates of the microorganism.

Toxicology studies in laboratory animals describe potential health effects from large doses in an effort to identify any potential to cause disease or toxicity. When *G. catenulatum* strain J1446 was tested on laboratory animals, there were no signs that it caused any significant toxicity or disease.

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[3] "Consultation statement" as required by subsection 28(2) of the *Pest Control Products Act*

[4] "Decision statement" as required by subsection 28(5) of the *Pest Control Products Act*

Residues in Water and Food

Dietary risks from food and water are not of concern.

The *Food and Drugs Act* prohibits the sale of adulterated food, that is, food containing a pesticide residue that exceeds the established maximum residue limit (MRL). Pesticide MRLs are established for *Food and Drugs Act* purposes through the evaluation of scientific data under the *Pest Control Products Act*. Each MRL value defines the maximum concentration in parts per million (ppm) of a pesticide allowed in or on certain foods. Food containing a pesticide residue that does not exceed the established MRL does not pose an unacceptable health risk. Strains of *G. catenulatum* are common in nature and the use of Prestop Biofungicide WP in greenhouses to suppress fungal plant disease on vegetables, herbs and ornamentals is not expected to significantly increase the natural environmental background levels of this microorganism. Furthermore, when *G. catenulatum* strain J1446 was administered orally to rats, no signs that it caused toxicity or disease were observed. Although secondary metabolites of toxicological significance have been shown to be produced by other isolates of *G. catenulatum*, due to the demonstrated low toxicity and absence of such metabolites in cultures of *G. catenulatum* strain J1446, the risks from secondary metabolites for the general population, including infants/children, are negligible. The establishment of a maximum residue limit (MRL) is therefore not required for *G. catenulatum* strain J1446. As well, the likelihood of residues of *G. catenulatum* strain J1446 contaminating drinking water supplies is negligible to non-existent. Consequently, dietary exposure and risk are minimal to non-existent.

Occupational Risks From Handling Prestop Biofungicide WP

Occupational risks are not of concern when Prestop Biofungicide WP is used according to label directions, which include protective measures.

Growers handling Prestop Biofungicide WP can come into direct contact with *G. catenulatum* strain J1446 on the skin, in the eyes, or by inhalation. For this reason, the label specifies that growers exposed to Prestop Biofungicide WP during handling, mixing/loading, application or clean-up/repair activities must wear waterproof gloves, a long-sleeved shirt, long pants, eye goggles, shoes, socks and a dust/mist filtering respirator (MSH/NIOSH approval number prefix TC-21C) or a NIOSH-approved respirator with any N-95, R-95, P-95 or HE filter. Furthermore, early-entry workers will be restricted from entering areas where Prestop Biofungicide WP has been applied as a foliar spray for a period of four hours unless wearing the indicated personal protective equipment with the exception of eye goggles and a dust/mist filtering respirator, which are required only until the spray mist has settled.

Bystander exposure is expected to be much less than that of applicators, handlers and mixers/loaders and is considered negligible. Therefore, health risks to bystanders are not of concern.

Environmental Considerations

What Happens When Prestop Biofungicide WP Is Introduced Into the Environment?

Environmental risks are not of concern.

Studies designed to examine the effects of *Gliocladium catenulatum* strain J1446 on various non-target organisms were evaluated. Few adverse effects were observed in birds, freshwater fish, terrestrial arthropods (including honeybees), aquatic invertebrates, marine animals or algae.

Gliocladium catenulatum is not generally considered to be a disease-causing agent. Therefore, Prestop Biofungicide WP is expected to present a negligible risk to non-target organisms.

Value Considerations

What is the Value of Prestop Biofungicide WP?

Prestop Biofungicide WP suppresses specific soil and seed-borne pathogens and foliar diseases on greenhouse-grown vegetables, herbs and ornamentals.

The formulated end-use product, Prestop Biofungicide WP, is a biological fungicide. When applied in a 0.5–1.0% solution as a soil media treatment, a soil drench treatment or as a foliar treatment, it will suppress specific soil and seed-borne pathogens and foliar diseases on greenhouse-grown vegetables, herbs and ornamentals. It is best used as a preventative before disease is present, and reapplication is necessary every three to six weeks, depending on the disease pressures in the greenhouse and the method of application.

Prestop Biofungicide WP is considered to be a low risk product and can be used as an integral part of an IPM program to reduce the reliance on chemical alternatives. There are limited fungicide products currently available to greenhouse growers, and therefore Prestop Biofungicide WP is a new product available to the greenhouse sector. It may also be used as a resistance management tool for rotation of fungicides where no other alternative products are available.

Measures to Minimize Risk

Labels of registered pesticide products include specific instructions for use. Directions include risk-reduction measures to protect human and environmental health. These directions must be followed by law.

The key risk-reduction measures being proposed on the label of Prestop Biofungicide WP to address the potential risks identified in this assessment are as follows.

Key Risk-Reduction Measures

- **Human Health**

  Because of concerns with users developing allergic reactions through repeated high exposures to *G. catenulatum* strain J1446, anyone handling, mixing/loading, applying or involved in clean-up/repair activities of Prestop Biofungicide WP must wear waterproof gloves, a long-sleeved shirt, long pants, eye goggles and a dust/mist filtering respirator (MSH/NIOSH approval number prefix TC-21C) or a NIOSH-approved respirator with any N-95, R-95, P-95 or HE filter. Furthermore, early-entry workers will be restricted from entering areas where Prestop Biofungicide WP has been applied as a foliar spray for a period of four hours unless wearing the indicated personal protective equipment with the exception of eye goggles and a dust/mist filtering respirator, which are required only until the spray mist has settled.

- **Environment**

  As a general precaution, handlers are asked not to contaminate irrigation or drinking water or aquatic habitats through equipment cleaning or waste disposal. In addition, growers must not allow effluent or runoff from greenhouses containing this product to enter lakes, streams, ponds or other water bodies.

Next Steps

Before making a final registration decision on *Gliocladium catenulatum* strain J1446 , the PMRA will consider all comments received from the public in response to this consultation document. The PMRA will accept written comments on this proposal up to 45 days from the date of publication of this document. Please forward all comments to Publications (contact information on the cover page of this document). The PMRA will then publish a Registration Decision document, which will include its decision, the reasons for it, a summary of comments received on the proposed final decision and the PMRA's response to these comments.

Other Information

At the time the PMRA makes its registration decision, it will publish a Registration Decision document on *Gliocladium catenulatum* strain J1446 (based on the Science Evaluation Section of this consultation document). In addition, the test data referenced in this consultation document will be available for public inspection, upon application, in the PMRA's Reading Room (located in Ottawa).

Science Evaluation

Gliocladium catenulatum strain J1446

1.0 The Technical Grade Active Ingredient, Its Properties and Uses

1.1 Identity of the Technical Grade Active Ingredient

Active microorganism	*Gliocladium catenulatum* strain J1446
Function	Suppression of soil-borne and seed-borne plant diseases, such as damping-off, root and stem rot, and wilt caused by *Pythium* sp., *Rhizoctonia* sp., *Phytophthora* sp. and *Fusarium* sp., as well as certain foliar diseases caused by *Botrytis* sp. and *Didymella* sp. on greenhouse-grown vegetables, herbs and ornamentals.
Binomial name	*Gliocladium catenulatum* strain J1446
Taxonomic designation	
Kingdom	Fungi
Phylum	Deuteromycotina
Order	Hyphomycetes (syn. Moniliales)
Genus	*Gliocladium*
Species	*catenulatum*
Strain	J1446
Patent status information	A Canadian patent application was submitted in September 1995. The patent is pending.
Nominal purity of the technical grade active ingredient	2.0×10^8 colony forming units/gram
Active microorganism	*Gliocladium catenulatum* strain J1446

Identity of relevant impurities of toxicological and/or environmental significance	The technical grade active ingredient does not contain any impurities or microcontaminants known to be Toxic Substances Management Policy (TSMP) Track 1 substances. In published scientific literature, some isolates of *G. catenulatum* have been reported to produce novel chemotherapeutic compounds as well as metabolites with moderate cytotoxicity to various tumour cell lines. As well, *G. catenulatum* has been shown in vitro to excrete the lytic enzymes chitinase and beta-glucanase, which are known to degrade the cell wall of the fungal organisms. No toxicity was observed as a result of in vivo and in vitro testing with *G catenulatum* strain J1446.

1.2 Physical and Chemical Properties of the Active Ingredient and End-use Product

Technical Product—*Gliocladium catenulatum* J1446 Dried Cell Mass

The physical and chemical properties of the technical product are not required since the technical product is only used as a reference standard and is not formulated into an end-use product.

End-use Product—Prestop Biofungicide WP

Property	Prestop Biofungicide WP
Physical State (presumably room temperature)	Free-flowing fine powder with uniform consistency
Colour	Beige
Odour	A very weak odour
pH at 25°C: 1% aqueous dispersion	6.99
Density	Tap density: 0.49 g/mL
Viscosity	Not applicable
Corrosion Characteristics	No evaluation performed
Suspendability (maximum field application concentration - 5.0% w/v) Suspendability (minimum field application concentration - 2.0% w/v)	101% 90.1%
Moisture Content	6%
Storage Stability	12 months at 4°C

1.3 Directions for Use

Prestop Biofungicide WP is a biological fungicide. When applied in a 0.5%–1.0% solution to soil media, and/or as a soil drench, or as a foliar treatment, it will suppress the following greenhouse diseases on the specified food and ornamental crops: damping-off and crown and root rot caused by *Pythium* spp., and damping-off caused by *Rhizoctonia solani* on cucumber, pepper, tomato, cauliflower, broccoli, lettuce, oregano, basil, parsley, thyme, dill and certain ornamental crops (alyssum, pansy, petunia, poinsettia, tagetes, salvia, saintpaulia, snapdragon, geranium); stem wilt disease caused by *Fusarium oxysporum* on cucumbers and basil; Botrytis stem canker, and Botrytis grey mould (*Botrytis cinerea*) on greenhouse peppers, tomatoes, cucumbers and certain ornamental crops (alyssum, pansy, petunia, poinsettia, tagetes, salvia, saintpaulia, snapdragon and geranium); gummy stem blight disease (*Didymella bryoniae*) on cucumbers and root and basal stem rot caused by *Phytophthora cryptogea* on certain ornamental crops (alyssum, pansy, petunia, poinsettia, tagetes, salvia, saintpaulia, snapdragon and geranium).

Prestop Biofungicide WP has the greatest efficacy when used as a preventative, applied before disease is present, and reapplied every three to six weeks, depending on the disease pressure in the greenhouse and the application method. Higher disease pressures and any foliar application will require the shorter application interval. Prestop Biofungicide WP may not be tank-mixed with any other pesticide or adjuvant, and is not to be applied via drip irrigation or other forms of chemigation.

1.4 Mode of Action

As a biological fungicide, Prestop Biofungicide WP does not fall under any of the fungicide groups classified by the Fungicide Resistance Action Committee (FRAC). The effect of Prestop Biofungicide WP comes from various factors. The first is competitive inhibition, where competing (pathogenic) fungi are deprived of living space and nourishment. Another is that *G. catenulatum* J1446 is a weak hyperparasite of several fungal pathogens and has shown some chitinase activity resulting in lysis of the cell walls of competing fungal pathogens.

2.0 Methods of Analysis

2.1 Methods for Identification of the Microorganism

Morphological methods are employed to confirm the identity of the microbial pest control agent (MPCA) to the species level. The Institute of the Royal Netherlands Academy of Art and Sciences concluded that the MPCA belongs to the species *G. catenulatum* using light microscopy to examine morphological characteristics of conidiophores and conidia. Molecular methods are employed to identify the MPCA to the strain level. A RAPD-PCR (random amplified polymorphic DNA - polymerase chain reaction) method has been developed that is able to distinguish *G. catenulatum* strain J1446 from 41 closely related fungi from the genera *Gliocladium*, *Trichoderma*, *Fusarium*, and *Nectria*.

2.2 Method for Establishment of Purity of Seed Stock

Seed bank ampoules of *G. catenulatum* strain J1446 are stored in liquid nitrogen in order to prevent any genetic changes. All ampoules are from the same origin and are representative of the same generation. To ensure the identity and purity of seed stock, samples are cultivated on agar plates and studied microscopically. As well, random batches are tested for biological efficacy against *Pythium* using cucumber as a test plant.

2.3 Methods to Define the Content of the Microorganism in the Manufactured Material Used for the Production of Formulated Products

Three samples are taken from each production batch during production and tested using a standardized method to determine the viability of the active ingredient. The test is performed to estimate the number of colony forming units (CFUs) of *G. catenulatum* strain J1446 per unit mass of sample (CFU/g dry weight). The test uses colony counts on media platings of serial dilutions of the end-use product to determine the guarantee for the final formulation.

2.4 Determination and Quantification of Residues (viable or non-viable) of the Active Microorganism and Relevant Metabolites

Since the closely related microorganism *Gliocladium virens* (also known as *Trichoderma virens*) is known to produce gliotoxin, analyses were carried out in order to demonstrate that gliotoxin is not produced by *G. catenulatum* strain J1446 nor is present in the formulated end-use product. No gliotoxin was detected in growth media, unformulated cell mass powder or plant rhizosphere treated with *G. catenulatum* strain J1446 (refer to Section 4.2.1 below). *Gliocladium catenulatum* strain J1446 also tested negative in feline fetus lung (FL) cytotoxicity tests as well as standard pathogenicity and toxicity studies (refer to Section 3.1 below).

2.5 Methods for Determination of Relevant Impurities in the Manufactured Material

During manufacture, several approaches are used to limit microbial contamination in *G. catenulatum* J1446 Dried Cell Mass and Prestop Biofungicide WP. These approaches include proper handling and storage of starting materials, visual inspection of cultures, normal hygienic standards for bioindustry and monitoring of fermentation conditions.

To ensure that the above quality control procedures limit contaminating microorganisms, frequent checks are performed using standard microbiological procedures. Contaminated cultivations are discarded.

2.6 Methods to Show Absence of Any Human and Mammalian Pathogens

As noted in Section 2.5, several approaches are used to limit microbial contamination in *G. catenulatum* J1446 Dried Cell Mass and Prestop Biofungicide WP. Since these methods do not distinguish human and mammalian pathogens from other contaminating microorganisms, further contaminant testing is performed. Annually, two to three samples from random batches

are analyzed using standard microbiological procedures to determine potentially hazardous contaminants. The levels of pathogenic contaminants are set according to microbiological criteria for foodstuffs in the European Union (Directive 92/46/EEC). Identified contaminating bacteria and their acceptable levels are as follows: *Escherichia coli* $<10^5$ CFU/g and *Salmonella* 0/25 g. These tests are performed only on the end-use product, Prestop Biofungicide WP. Testing on *Gliocladium catenulatum* J1446 Dried Cell Mass is not required because this product is only used as a reference standard and is not formulated into an end-use product.

2.7 Methods to Determine Storage Stability, Shelf Life of the Microorganism

The storage stability of nine batches of Prestop Biofungicide WP was evaluated over a 12-month storage period at two temperatures: 4°C and 28°C. Viability testing demonstrated that the product was stable for a period of up to 12 months at 4°C. Testing also showed that the product was stable at 28°C for a period of up to one month, indicating that the product will remain stable during shipping where the product may experience unrefrigerated temperatures for short periods of time.

3.0 Impact on Human and Animal Health

3.1 Toxicity and Infectivity Summary

The PMRA conducted a detailed review of the toxicological database for *G. catenulatum* strain J1446. The database is complete, consisting of laboratory animal (in vivo) toxicity studies (acute oral toxicity/pathogenicity, acute pulmonary toxicity/pathogenicity, acute intraperitoneal infectivity, acute dermal toxicity/irritation, dermal sensitization, and eye irritation) currently required for health hazard assessment purposes. All studies were carried out in accordance with currently accepted international testing protocols and Good Laboratory Practices. Additional cytotoxicity testing was performed on feline fetus lung cell cultures. Waiver requests were deemed acceptable to address the acute dermal toxicity and dermal irritation of Prestop Biofungicide WP. The scientific quality of the data is high and the database is considered sufficient to characterize the toxicity and infectivity of this pest control agent and end-use product.

In an oral toxicity/pathogenicity study, no significant toxicity was observed in CD rats following oral gavage with 2.0×10^9 CFU/kg body weight of *G. catenulatum* strain J1446. The MPCA was initially detected in the feces of one subject on Day 2 but had subsequently cleared by Day 4. None of the subjects exhibited any treatment-related clinical signs nor were any macroscopic abnormalities observed. In another oral toxicity study, CD rats were given 2000 mg/kg body weight of *G. catenulatum* strain J1446 by oral gavage. No signs of toxicity were observed in any of the subjects. Although this study had some specific deficiencies (i.e. viability check of test substance, inclusion of an untreated control group), the results further demonstrated the low oral toxicity of *G. catenulatum* strain J1446.

In a pulmonary toxicity/pathogenicity study, CD rats were given a single intratracheal instillation of 6.60 or 7.98 × 10^7 CFU/kg body weight. The few mortalities and clinical signs of toxicity that occurred in both the test and control group treated with inactivated test material are considered attributable to the anaesthesia and a result of the procedure. All clinical signs had resolved by Day 2. Viable *G. catenulatum* strain J1446 was recovered only from lung tissue on Days 1 and 3 but had cleared from all subjects by Day 7. No signs of toxicity or pathogenicity attributable to the MPCA were observed in any subject.

In an acute inhalation toxicity study, CD rats were exposed by inhalation to 5.57 mg/L *G. catenulatum* strain J1446 . No signs of toxicity attributable to the MPCA were observed in any subject. Although this study had some specific deficiencies (i.e. viability check of test substance, inclusion of an untreated control group), the results further demonstrated the low pulmonary toxicity of *G. catenulatum* strain J1446.

In an intraperitoneal infectivity study, CD rats were exposed to a single dose of 4.2 × 10^8CFU/kg body weight of *G. catenulatum* strain J1446 administered by intraperitoneal injection. Viable MPCA was recovered from various organs inside the peritoneal cavity up until Day 7. No viable MPCA was recovered from tissue outside of the peritoneal cavity. Clinical signs observed were typical of a response to the trauma of treatment. Macroscopic findings were also attributable to a significant inflammatory response. The clinical and macroscopic observations were similar in the test group and the control group treated with inactivated test material. No signs of pathogenicity from the MPCA were observed during the study.

In a dermal toxicity study, CD rats were dermally exposed to 2000 mg/kg body weight of *G. catenulatum* strain J1446 . No signs of toxicity were observed in any subject. In the dermal irritation study, New Zealand White rabbits were dermally exposed to 0.5 g of *G. catenulatum* strain J1446 . One subject exhibited signs of erythema from Day 2 to 4, which was followed by desquamation on Day 7. A full recovery was made by Day 9. No other subjects exhibited any signs of irritation. *Gliocladium catenulatum* strain J1446 is considered to be minimally irritating to the skin.

In a dermal sensitization study, Dunkin-Hartley guinea pigs were dermally exposed to 75% *G. catenulatum* strain J1446 in a 0.5% carboxymethylcellulose solution via an induction procedure followed by a challenge procedure. Mild positive responses were observed in the test group as a result of the challenge procedure. Therefore *G. catenulatum* strain J1446 is considered to be a sensitizer. To mitigate the risk of sensitization to the MPCA, workers will be required to wear appropriate clothing and equipment to minimize dermal exposure when contact with *G. catenulatum* strain J1446 is likely.

In an eye irritation study on New Zealand White rabbits, 0.1 mL of *G. catenulatum* strain J1446 was instilled into the conjunctival sac of one eye. Moderate to mild redness, chemosis and discharge of the conjunctivae were observed up until 72 hours. According to the Draize method of scoring, this study showed that *G. catenulatum* strain J1446 is minimally irritating to the eye. In another eye irritation study on New Zealand White rabbits, 0.1 g of *G. catenulatum* strain J1446 was instilled into the conjunctival sac of one eye. Slight conjunctival redness was noted in all subjects one hour after instillation and chemosis was observed in one subject for one

hour after instillation. All subjects fully recovered after 24 hours. These studies showed that *G. catenulatum* strain J1446 is minimally irritating to the eye.

In a series of three cytotoxicity studies, cultures of feline fetus lung (FL) cells were exposed to methanol extracts of dried cell mass from four different batches of *G. catenulatum* strain J1446. The cell cultures were observed for toxic effects. No toxic effects were observed in any of the cell cultures. This test is capable of showing toxicity of extracts from cultures of *Stachybotrys* sp., which are known to produce trichothecene mycotoxins, to FL cells. These studies further showed the low toxicity of *G. catenulatum* strain J1446 to mammalian cells in vitro.

Requests to waive acute dermal toxicity and acute dermal irritation studies were accepted for Prestop Biofungicide WP based on the nature and the concentrations of each formulation ingredient. However, one of the formulation ingredients, skim milk powder, is known to be an allergen and must be labelled as such according to PMRA Regulatory Directive, DIR2006-02, *Formulants Policy and Implementation Guidance Document*.

A survey of published literature has revealed that some other isolates of *G. catenulatum* have been shown to produce metabolites with moderate cell toxicity to various tumour cell lines and other types of pharmacological activity. However, the results of the FL cell toxicity tests did not demonstrate any toxicity from extracts of *G. catenulatum* strain J1446.

There are fungi that are closely related to *G. catenulatum* (i.e. *T. virens*, *Gliocladium roseum*) which are known to produce a toxic metabolite called gliotoxin. Specific analyses were performed to show that *G. catenulatum* strain J1446 does not produce gliotoxin (refer to Section 4.2.1 below). Furthermore, the absence of a toxic effect in the FL cell toxicity testing confirms the absence of gliotoxin in *G. catenulatum* strain J1446 cultures.

Two medical certificates written by medical professionals were submitted stating that no workers have developed sensitization or other adverse effects from occupational exposure to the MPCA during the research, development and production of *Gliocladium catenulatum* strain J1446. Exposed workers were assessed via clinical examinations, laboratory and X-ray examinations and respiratory functions tests.

Higher tier subchronic and chronic toxicity studies were not required due to the low acute toxicity of the MPCA and due to the fact that there were no indications of infectivity, toxicity or pathogenicity in the test animals treated in the Tier I acute oral and pulmonary toxicity/infectivity tests.

Within the available scientific literature, there are no reports that suggest *G. catenulatum* has the potential to cause adverse effects on the endocrine system of animals. The submitted toxicity/infectivity studies in the rodent indicate that, following oral and pulmonary routes of exposure, the immune system is still intact and able to process and clear the MPCA. Based on the weight of evidence of available data, no adverse effects to the endocrine or immune systems are anticipated for *G. catenulatum* strain J1446.

3.2 Occupational/Bystander Exposure and Risk Assessment

3.2.1 Occupational

When handled according to label instructions, the potential routes of handler exposure to *G. catenulatum* strain J1446 are pulmonary, dermal and to some extent ocular.

The potential for dermal, eye and inhalation exposure for applicators, mixers/loaders, handlers and early-entry workers exists, with the primary source of exposure to workers being dermal. Since unbroken skin is a natural barrier to microbial invasion of the human body, dermal absorption could occur only if the skin were cut, if the microbe were a pathogen equipped with mechanisms for entry through or infection of the skin, or if metabolites were produced that could be dermally absorbed. *G. catenulatum* has not been identified as a wound pathogen and there is no indication that it could penetrate intact skin of healthy individuals.

Although the risk of toxicity is low in individuals exposed to large quantities of *G. catenulatum* strain J1446, respiratory hypersensitivity could possibly develop upon repeated exposure to the product. Specific label wording to minimize exposure to dusts or mists generated while handling or applying the product are required. Exposure to applicators, mixers/loaders, handlers and early-entry workers will be mitigated by a restricted-entry interval (REI) and a label requirement for personal protective equipment (PPE), including a dust/mist filtering respirator. Although no dermal toxicity and little dermal irritation are expected based on toxicological studies of the MPCA and toxicological characteristics of the formulation ingredients present in end-use formulations, all MPCAs are considered potential sensitizers. The PMRA assumes that all microorganisms contain substances that can elicit positive hypersensitivity reactions. In addition, a dermal sensitization study found that *G. catenulatum* strain J1446 was a sensitizing agent. Label restrictions and risk mitigation measures are required to protect workers that are likely to be exposed to the products. Such exposure to applicators, mixers/loaders, handlers and early-entry workers can be minimized if they wear waterproof gloves, a long-sleeved shirt, long pants, shoes and socks.

No eye irritation studies were submitted for the end-use product, Prestop Biofungicide WP. However, an eye irritation using the active ingredient *G. catenulatum* strain J1446 showed that the MPCA was minimally irritating. Consequently, some label restrictions are required to protect workers that are likely to be exposed to the products. Such exposure can be minimized if applicators, mixers/loaders, handlers and early-entry workers wear eye goggles.

3.2.2 Bystander

Overall the PMRA does not expect that bystander exposure will pose an undue risk on the basis of the low toxicity/pathogenicity profile for *G. catenulatum* strain J1446 and the assumption that precautionary label statements will be followed in the use of Prestop Biofungicide WP.

The label does not permit applications outside of commercial greenhouses. Therefore, non-occupational dermal exposure and risk to adults, infants and children are low. Due to the fact that the use sites are agricultural, exposure to infants and children in school and residential and daycare facilities is likely to be minimal to non-existent. Consequently, the health risk to infants and children is expected to be negligible.

3.3 Dietary Exposure and Risk Assessment

3.3.1 Food

While the use pattern may result in some dietary exposure with possible residues in or on agricultural commodities, negligible to no risk is expected for the general population, including infants and children because *G. catenulatum* strain J1446 demonstrated no pathogenicity, infectivity or oral toxicity at the maximum dose tested in the Tier I acute oral toxicity/infectivity study. Although secondary metabolites of toxicological significance have been shown to be produced by other isolates of *G. catenulatum*, due to the demonstrated low toxicity of the MPCA, the risks from secondary metabolites to the general population, including infants and children, are negligible. Furthermore, higher tier subchronic and chronic dietary exposure studies were not required because of the low toxicity of the MPCA, and there were no indications of infectivity, toxicity or pathogenicity in the test animals treated in the Tier I acute oral and pulmonary toxicity/infectivity studies. Therefore, there are no concerns for chronic risks posed by dietary exposure of the general population and sensitive subpopulations, such as infants and children.

3.3.2 Drinking Water

The likelihood that *G. catenulatum* strain J1446 could enter neighbouring aquatic environments as a result of greenhouse use is negligible. No risks are expected from exposure to this microorganism via drinking water because exposure will be minimal and because there were no harmful effects observed in Tier I acute oral toxicity and infectivity testing. The Prestop Biofungicide WP label instructs users not to contaminate irrigation or drinking water supplies or aquatic habitats through equipment cleaning or waste disposal. Users are also requested not to allow effluent or runoff from greenhouses containing this product to enter lakes, streams, ponds or other waters. Furthermore, municipal treatment of drinking water is expected to remove the transfer of residues to drinking water. Therefore, potential exposure to *G. catenulatum* strain J1446 in surface and drinking water is negligible.

3.3.3 Acute and Chronic Dietary Risks for Sensitive Subpopulations

Calculations of acute reference doses (ARfDs) and acceptable daily intakes (ADIs) are not usually possible for predicting acute and long-term effects of microbial agents in the general population or to potentially sensitive subpopulations, particularly infants and children. The single (maximum hazard) dose approach to testing MPCAs is sufficient for conducting a reasonable general assessment of risk if no significant adverse effects (i.e. no acute toxicity, infectivity or pathogenicity endpoints of concern) are noted in acute toxicity and infectivity tests. Based on all the available information and hazard data, the PMRA concludes that

G. catenulatum strain J1446 is of low toxicity, that it is not pathogenic or infective to mammals and that infants and children are likely to be no more sensitive to the MPCA than the general population. Thus, there are no threshold effects of concern and, as a result, there is no need to require definitive (multiple-dose) testing or to apply uncertainty factors to account for intra- and interspecies variability, safety factors or margins of exposure. Further factoring of consumption patterns among infants and children, special susceptibility in these subpopulations to the effects of the MPCA, including neurological effects from pre- or post-natal exposures, and cumulative effects on infants and children of the MPCA and other registered micro-organisms that have a common mechanism of toxicity, does not apply to this MPCA. As a result, the PMRA has not used a margin of exposure (safety) approach to assess the risks of *G. catenulatum* strain J1446 to human health.

3.4 Maximum Residue Limits

The *Food and Drugs Act* prohibits the sale of adulterated food, that is, food containing a pesticide residue that exceeds the established maximum residue limit (MRL). Pesticide MRLs are established for *Food and Drugs Act* purposes through the evaluation of scientific data under the *Pest Control Products Act*. Each MRL value defines the maximum concentration in parts per million (ppm) of a pesticide allowed in or on certain foods. Food containing a pesticide residue that does not exceed the established MRL does not pose an unacceptable health risk.

There were no signs of toxicity and no signs of pathogenicity observed when *G. catenulatum* strain J1446 was administered orally to rats. In addition, no metabolic byproducts of toxicological concern were produced by this microorganism. The establishment of an MRL is therefore not required for *G. catenulatum* strain J1446 under section 4(*d*) of the *Food and Drugs Act* (adulteration of food) as defined under Division 15, section B.15.002 of the Food and Drugs Regulations.

3.5 Aggregate Exposure

Based on the toxicity and infectivity test data submitted and other relevant information in the PMRA's files, there is reasonable certainty that no harm will result from aggregate exposure of residues of *G. catenulatum* strain J1446 to the general population, including infants and children, when the microbial pest control product is used as labelled. This includes all anticipated dietary (food and drinking water) exposures and all other non-occupational exposures (dermal and inhalation) for which there is reliable information. As the product is to be used at greenhouse sites and is not allowed for use on turf, residential or recreational areas, dermal and inhalation exposure to the general public will be very low. Furthermore, few adverse effects from exposure to other isolates of *G. catenulatum* encountered in the environment have been reported. Even if there is an increase in exposure to this microorganism from the use of Prestop Biofungicide WP, it is anticipated that there will be no increase in potential human health risk.

3.6 Cumulative Effects

The PMRA has considered available information on the cumulative effects of residues and other substances that have a common mechanism of toxicity. These considerations included the cumulative effects of such residues and other substances with a common mechanism of toxicity on infants and children. Besides naturally occurring strains of *G. catenulatum* in the environment, the PMRA is not aware of any other microorganisms or other substances that share a common mechanism of toxicity with this active ingredient. No cumulative effects are anticipated if the residues of *G. catenulatum* strain J1446 interact with related strains of this microbial species.

4.0 Impact on the Environment

4.1 Fate and Behaviour in the Environment

Environmental fate testing is intended to demonstrate whether a microbial pest control agent (MPCA) is capable of surviving or replicating in the environment to which it is applied, and could provide an indication of which non-target organisms may be exposed to the MPCA as well as provide an indication of the extent of exposure. Environmental fate data (Tier II/III) are not normally required at Tier I, and are only triggered if significant toxicological effects in non-target organisms are noted in Tier I testing.

For *G. catenulatum* strain J1446, several studies were submitted on the survival of *G. catenulatum* strain J1446 in water and peat under greenhouse conditions as well as on the leaf surface after foliar application. The possible transfer of the MPCA to the soil as well as mobility of conidia within the soil, as a result of horticultural peat-based growth substrate treatment, were discussed as well.

The persistence of *G. catenulatum* strain J1446 in sterilized distilled water, tap water, sea water and lake water was studied at two different temperatures, at room temperature (~23 °C) and in cold storage (8 °C). Persistence in test waters was evaluated by regularly counting the number of viable colonies over a period of up to 28 weeks. The results showed that *G. catenulatum* strain J1446 remained viable in all test waters until study termination at 28 weeks, but it did not proliferate under these conditions.

The survival of *G. catenulatum* strain J1446 in a commercial peat-based growing medium after its application either by direct incorporation into the growing medium or by drench treatment was studied under greenhouse conditions (15–25 °C). The persistence of the MPCA in peat was evaluated biweekly by plating samples onto agar medium. In this study, the population of *G. catenulatum* gradually decreased from 10^5 CFU/g to 10^4 CFU/g within the first four weeks. Between weeks 4 and 6, the population continued to decline to 10^2 CFU/g, and by week 8, *G. catenulatum* was no longer detected in any of the samples. Furthermore, the persistence of *G. catenulatum* strain J1446 in the peat-based growing medium was not affected by the method of application.

The survival of *G. catenulatum* strain J1446 on leaves was studied under greenhouse conditions (18–20°C) by spraying cucumber plants with a 0.5% aqueous suspension. Following its foliar application to plants, the viability of the MPCA was assayed a day after the treatments, and at weekly intervals by plating aqueous extracts and leaf pieces directly onto agar medium. In this study, *G. catenulatum* strain J1446 persisted on the treated cucumber leaves for the duration of the study (i.e. 4 weeks). However, the amount of the MPCA decreased steadily throughout the study period: 44% of the original amount after seven days, and only 1% of the original amount after three weeks.

The survival of *G. catenulatum* strain J1446 was similarly studied on *Pelargonium* plantlets. In this study, the plantlets were sprayed with an aqueous suspension of *G. catenulatum* strain J1446 at a rate of 10 g of unformulated powder/m^2 (equivalent to a nominal concentration of 1×10^{10} CFU/m^2). The MPCA was monitored at weekly intervals by cutting small pieces of leaves and plating them onto agar medium, then observing them using a stereomicroscope. *Gliocladium catenulatum* strain J1446 persisted on leaves of *Pelargonium* plants but its frequency of detection declined slightly during the study period (4 weeks). In addition, *G. catenulatum* strain J1446 did not appear to spread onto the new leaves of the treated plant.

Gliocladium catenulatum is a naturally occurring and relatively common soil saprophyte that is distributed worldwide. This isolate was originally isolated from Finnish agricultural soil, and grows well on roots, or in very close proximity to them, but to a lesser extent in the surrounding soil matrix. Since this MPCA spreads in the environment through mycelial growth, chlamydospore production and the release of conidia (phialospores) that collect in mucilaginous droplets, its mobility in soil is limited to rainwater runoff, soil movements due to human or animal activity and vectors such as insects. On foliar surfaces, *G. catenulatum* strain J1446's potential for mobility increases through the dispersion of plant detritus by wind, animals and insects. In Canada, the uses of Prestop Biofungicide WP include greenhouse food and greenhouse non-food crops only. Consequently, the dispersal of strain J1446 from greenhouses to outside soil environments is expected through human activities, i.e. removal and composting of spent crop growing media. Once outside, the dispersal of *G. catenulatum* strain J1446 should be limited to mostly runoff and vectors, and based on all the available information on the fate of this organism in the environment, the populations of *G. catenulatum* strain J1446 should return to naturally occurring levels for this species. The exact amount of time that is required to return to naturally occurring levels, however, is difficult to predict since it will depend on many factors, including the growth medium (i.e. on soil or plants, or in water), temperature, pH and light conditions.

4.2 Effects on Non-Target Species

4.2.1 Effects on Terrestrial Organisms

Several studies were submitted to address the hazards of *G. catenulatum* strain J1446 to terrestrial non-target organisms. These studies included non-target avian species, terrestrial athropods and non-athropod invertebrates.

The acute avian oral toxicity of *G. catenulatum* strain J1446 to 21-day-old northern bobwhite quail (*Colinus virginianus*) was assessed over a period of 30 days. *Gliocladium catenulatum* strain J1446 was administered to 30 birds as an aqueous suspension by oral gavage at the limit dose of 1.4×10^6 colony forming units per gram of body weight per day for five consecutive days. A negative control group consisting of 10 birds was similarly treated with reverse-osmosis water. No treatment-related toxicity or pathogenicity was observed. The 30-day acute oral LD_{50} was determined to be greater than 1.4×10^6 colony forming units per gram of body weight per day for five consecutive days. The 30-day NOEL of *G. catenulatum* strain J1446 to the northern bobwhite, based on the absence of adverse effects, was determined to be 1.4×10^6 CFU/g bw/day for five consecutive days.

Four terrestrial arthropod studies were submitted for review. The test species included honeybees (*Apis mellifera*), ladybird beetle (*Hippodamia convergens*), green lacewing larvae (*Chrysoperla carnea*) and parasitic Hymenoptora (*Nasonia vitripennis*). Three of these four studies (honeybee, ladybird beetle and parasitic Hymenoptera) were conducted using honey as the suspension medium for *G. catenulatum* strain J1446. Honey naturally contains antimicrobial agents, including peroxide and proteinaceous components, which may have interfered with the pathogenicity results by killing or inhibiting the MPCA before it was administered to insects. As a result, the pathogenicity portions of these studies were ignored.

Honeybees (75 bees/group) were exposed to honey containing 52 ppm (3.6×10^4 CFU/mL), 520 ppm (3.6×10^5 CFU/mL) and 5200 ppm (3.6×10^6 CFU/mL) *G. catenulatum* strain J1446 powder. Another group of bees (75 bees) was similarly treated with honey and deionized water, and served as a negative control. The study was terminated on Day 10 when mortality in the negative control group exceeded 20%. Mortality in the 52, 520 and 5200 ppm treatment groups was 37% (28 of 75), 37% (28 of 75) and 36% (27 of 75), respectively. A small number of bees exhibited lethargy or loss of equilibrium. All other surviving bees were normal in appearance and behaviour throughout the study period. Based on the results of this study, the 10-day LC_{50} was greater than 5200 ppm.

Ladybird beetles (75 beetles/group) were exposed to honey containing 52 ppm (3.6×10^4 CFU/mL), 520 ppm (3.6×10^5 CFU/mL) and 5200 ppm (3.6×10^6 CFU/mL) *G. catenulatum* strain J1446 powder. Another group of beetles (75 beetles) was similarly treated with honey and deionized water, and served as a negative control. The study was terminated on Day 16 when mortality in the negative control group exceeded 20%. Mortality in the 52, 520 and 5200 ppm treatment groups was 17%, 19% and 17%, respectively. A small number of lethargic and immobile beetles were observed during the study, including the negative control group. All other surviving beetles were normal in appearance and behaviour throughout the study period. Based on the results of this study, the 16-day LC_{50} was greater than 5200 ppm.

Green lacewing larvae (30 larvae/group) were exposed to moth egg meal containing 3.6×10^4 CFU/mL (52 ppm), 3.6×10^5 CFU/mL (520 ppm) and 3.6×10^6 CFU/mL (5200 ppm) *G. catenulatum* strain J1446 powder. Another group of larvae (30 larvae) was similarly treated with moth egg meal only and served as the negative control. The study was terminated on Day 12 when mortality in the negative control group exceeded 20% and pupation exceeded 50%. Mortality in the 3.6×10^4, 3.6×10^5 and 3.6×10^6 CFU/mL treatment groups was 20%, 13% and

13%, respectively, with 67%, 70% and 70% pupation, respectively. No overt signs of toxicity were noted among the surviving larvae in the control and treatment groups during the study. Based on the results of this study, the 12-day LC_{50} was greater than 3.6×10^6 CFU/mL (5200 ppm) and the no observed effect concentration (NOEC) was 3.6×10^6 CFU/mL (5200 ppm). In this study, *G. catenulatum* strain J1446 was not pathogenic to green lacewing larvae.

Parasitic Hymenoptera (75 larvae/group) were exposed to honey containing 52 ppm (3.6×10^4 CFU/mL), 520 ppm (3.6×10^5 CFU/mL) and 5200 ppm (3.6×10^6 CFU/mL) *G. catenulatum* strain J1446 powder. Another group of wasps (75 wasps) was similarly treated with honey and deionized water, and served as a negative control. This study was terminated on Day 8 when mortality in the negative control group exceeded 20%. Mortality in the 52, 520 and 5200 ppm treatment groups was 35%, 25% and 68%, respectively. A small number of wasps exhibited lethargy, immobility or loss of equilibrium. All other surviving wasps were normal in appearance and behaviour throughout the study. Based on the results of this study, the 8-day LC_{50} was 1992 ppm and the no adverse effect concentration (NOEC) was 520 ppm.

The toxicity and pathogenicity of *G. catenulatum* strain J1446 to earthworms (*Eisenia fetida*) was assessed over a period of 14 days. In this study, earthworms (40/group) were exposed to artificial soil containing 1.1×10^8, 1.9×10^8, 3.2×10^8, 5.3×10^8 and 8.8×10^8 CFU of *G. catenulatum* strain J1446 powder per kilogram of dry soil (equivalent to 0, 162, 270, 459, 750 and 1250 mg/kg dry soil). Another group of earthworms (40 earthworms) was similarly treated with deionized water and served as a negative control. A single mortality was noted on Day 14 in the test group treated with 8.8×10^8 CFU/kg dry soil. This earthworm could not be located in the soil and was presumed dead and decomposed. A treatment-related effect could not be precluded. All other earthworms were normal in appearance and behaviour. Based on these results, the 14-day LC_{50} was greater than 8.8×10^8 CFU/kg dry soil. The NOEC value was 5.3×10^8 CFU/kg dry soil.

In addition to the above studies, two scientific rationales were submitted to waive testing on soil microorganisms and terrestrial plants.

To address potential hazard to soil microorganisms, several studies and reports were submitted that evaluated and discussed *G. catenulatum* strain J1446's mode of action, and its potential to produce toxic metabolites as well as its direct effects on other microbial populations in peat-based growing media. These studies included dual culture investigations against phytopathogens, a gliotoxin bioassay in *Escherichia coli*, high performance liquid chromatography separations to detect gliotoxin in various samples as well as studies that directly measured the effects of *G. catenulatum* strain J1446 on various populations of non-target soil microorganisms in peat-based growing medium. None of the studies were conducted in accordance with the principles of Good Laboratory Practice and, more importantly, no raw data were submitted. Consequently, none of these studies were adequate to address the PMRA's underlying concerns on their own. However, the weight of evidence suggests that *G. catenulatum* strain J1446's mode of action is due to hyperparasitism and the production of lytic enzymes rather than toxic antimicrobial agents such as gliotoxin. No inhibition zones were observed in dual cultures against phytopathogens and no gliotoxin was detected in any of the studies. Furthermore, *G. catenulatum* strain J1446 did not significantly affect other microbial

populations in a commercial peat-based growing medium. These results are consistent with this species' natural occurrence in plant debris and soils throughout the world and it is unlikely that this microorganism would completely destabilize non-target soil or plant-associated microbial populations given that they have evolved in close proximity to each other. Based on all the available information, the hazard of *G. catenulatum* strain J1446 to non-target environmentally or economically important microbial species or microbiologically mediated biogeochemical processes was considered to be low. No additional microorganism testing is required to support the use of Prestop Biofungicide WP in greenhouse food and greenhouse non-food crops.

The potential hazard to non-target terrestrial plants was addressed with data and a scientific rationale as well as results of extensive literature searches. A study was submitted that evaluated the effect of high concentrations of *G. catenulatum* strain J1446 on 31 selected plant species. The MPCA was mixed with horticultural growing medium (steamed peat) at rates of 8×10^8, 8×10^9 and 8×10^{10} CFU per litre of growing medium prior to sowing surface-sterilized seeds. In general, few phytotoxic or phytopathogenic effects were observed in most plant species. In some plant species, the lowest dose had no effect, but the highest doses did cause slight growth reductions. The emergence of rye and spinach seemed to be inhibited by all three test concentrations, but no typical dose response was observed in these plants. Apparent dose responses were observed in the emergence of corn and oat, and the percent dry weights of carrot, cauliflower, dill, parsley, radish, Swedish turnip, turnip rape as well as celery (possibly), but the significance of these effects could not be tested since no raw data were provided and no standard deviations were reported. The scientific rationale also cited the results of an algal inhibition study (see Section 4.2.2 for additional details) where no toxic effects were noted at a limit concentration of 100 mg/L *G. catenulatum* strain J1446 powder.

An extensive literature search using several comprehensive databases found five articles reporting the isolation of *G. catenulatum* from diseased or dead plants as a potential causal agent of the infection, including damping-off on coffee in Cuba, part of pea root rot pathogen complex in China and dead grafted ornamental trees. It was noted that the isolation of *G. catenulatum* from diseased plant tissues does not necessarily indicate that it was the causal agent of the disease. It is likely that *G. catenulatum* appeared on the diseased plant material as a secondary organism. It was also noted that no phytotoxic or phytopathogenic effects were observed in any of the numerous efficacy studies conducted on this MPCA. The uses of Prestop Biofungicide WP on greenhouse food and greenhouse non-food crops are not expected to result in significant increased exposures or adverse effects to non-target plants. Therefore, no additional terrestrial plant testing is required to support the use of Prestop Biofungicide WP.

In mammals, no reports of adverse effects were found in the databases of PubMed and TOXNET using the keywords "gliocladium catenulatum". Furthermore, the laboratory animal studies on the rat, submitted in support of this registration and reviewed in Section 3.1, indicate that there is no pathogenicity and little toxicity from most routes of exposure at the maximum hazard dose level tested.

Based on all the available data and information on the effects of *G. catenulatum* strain J1446 to terrestrial organisms, there is reasonable certainty that no harm will be caused to birds, wild mammals, arthropods, non-arthropod invertebrates, plants or to other non-target microorganisms from the use of Prestop Biofungicide WP in greenhouses.

4.2.2 Effects on Aquatic Organisms

Several studies were submitted to address the hazards of *G. catenulatum* strain J1446 to aquatic non-target organisms. These studies included non-target freshwater fish, aquatic arthropods and freshwater algae.

In a toxicity and pathogenicity study, juvenile rainbow trout (*Onchorynchus mykiss*, 10/group) were exposed to *G. catenulatum* strain J1446 powder at nominal concentrations of 3.15×10^7, 6.23×10^7, 1.25×10^8, 2.50×10^8 and 5.00×10^8 CFU/L (equivalent to 0, 45, 89, 179, 357 and 714 mg/L) under static-renewal conditions. Test fish were also exposed to a diet of trout chow containing 6.6 mg test material per 100 g of trout food (equivalent to a nominal concentration of 4.62×10^7 CFU/kg food). A separate group of fish (10) remained untreated and served as a negative control. Mortalities were only noted in the 5.00×10^8 CFU/L treatment group, where all of the fish died within eight days of study initiation. Clinical signs included lethargy and lying on the bottom of the test chamber with only gill movements. External and internal examinations of surviving fish at study termination found all fish to be healthy in appearance and free from signs of infection. No external or internal examinations were conducted on dead fish and no cause of death was provided. It is clear that the effect was treatment-related; however, it is not clear if the fish died as a result of toxicity or pathogenicity. This study did not incorporate sterile filtrate (spent growth medium) and infectivity (inactivated MPCA) controls. Thus the PMRA's ability to distinguish the two possible effects is greatly hindered. The steep response to the moderate increment in dose, however, suggests that these effects are likely due to toxicity rather than infectivity or pathogenicity. Also, few colonies of *G. catenulatum* were recovered from tissues of fish in the 1.25×10^8 CFU/L and 2.50×10^8 CFU/L treatment groups. Assays were not performed on fish tissues collected from the 5.00×10^8 CFU/L treatment group. The isolated colonies were likely due to contamination during the collection of fish tissues rather than infection of tissues. *Gliocladium* spp. are generally recognized as prolific producers of secondary metabolites and it is possible that an unknown secondary metabolite may be responsible for the observed toxicity, even though *G. catenulatum* strain J1446 tested negative for gliotoxin in various studies as well as in a mammalian cell culture assay. Alternatively, the observed toxicity may be attributed to an inflammatory response in the gill lamellar tissues due to the high levels of active ingredient and/or other suspended solids in the highest test concentration. Such responses impair oxygen uptake despite the availability of dissolved oxygen in the dilution water. The exact cause of death could not be determined without additional necropsy information on fish exposed to the highest test concentration. Based on these results, the 30-day LC_{50} was determined to be approximately 3.5×10^8 CFU/L (equivalent to 504 mg/L). The NOEC and LOEC values, based on sublethal effects, were 1.25×10^8 and 2.5×10^8 CFU/L, respectively.

The aquatic toxicity and pathogenicity of *G. catenulatum* strain J1446 to *Daphnia magna* was studied under static-renewal conditions over a period of 21 days. Daphnids (40/group) were exposed to the test material at nominal aqueous concentrations of 2.0×10^6, 3.9×10^6, 7.7×10^6,

1.6×10^7 and 3.2×10^7 CFU/L (equivalent to 2.8, 5.6, 11, 23 and 45 mg/L). Another group (20 daphnids) remained untreated and served as a negative control. In this study, mortality and other sublethal effects, including reproduction and growth, were observed during the study starting on Day 7. After 21 days of exposure, cumulative mortality in the negative control group was 20% and there were no statistically significant (p >0.05) differences in survival of the daphnids in the 2.0×10^6 and 3.9×10^6 CFU/L treatment groups when compared to the negative control group. However, cumulative mortality in the 7.7×10^6, 1.6×10^7 and 3.2×10^7 CFU/L treatment groups was 70%, 75% and 95% by study termination and was statistically different (p >0.05) from the negative control group. The cause of death was not provided, but it was clearly related to treatment. It was not apparent, however, if daphnids died as a result of toxicity or pathogenicity, or other factors. As discussed above for freshwater fish, the study did not incorporate sterile filtrate and infectivity controls. The exact cause of death could not be determined without these additional control groups. Effects on daphnid growth were observed in the 7.7×10^6, 1.6×10^7 and 3.2×10^7 CFU/L treatment groups, but the significance was not analyzed statistically since a statistically significant (p >0.05) effect on survival was already established. The mean number of young per adult in the 3.9×10^6 CFU/L treatment group was significantly reduced (p >0.05) compared to the negative control group. Reproduction in the 7.7×10^6, 1.6×10^7 and 3.2×10^7 CFU/L treatment groups was also significantly reduced, but was not analyzed statistically due to a significant effect (p >0.05) on survival. Based on the results of this study, the 21-day LC_{50} was calculated at 5.5×10^6 CFU/L (or 7.8 mg/L). The 21-day NOEC and LOEC based on adverse effects on reproduction were 2.0×10^6 CFU/L (or 2.8 mg/L) and 3.9×10^6 CFU/L (or 5.6 mg/L), respectively.

The toxicity of *G. catenulatum* strain J1446 powder to the freshwater green alga (*Scenedesmus subspicatus*) was studied at a nominal limit concentration of 100 mg/L under static conditions over a period of 72 hours. An untreated group served as a negative control. There were no adverse effects noted on the growth of *S. subspicatus*. Significant increases in algal growth were observed in replicates treated with *G. catenulatum* strain J1446 powder. However, this observation was likely due to nutrients present in the test material. Based on the results, the EC_{50} and NOEC values were determined to be >100 mg/L and 100 mg/L, respectively.

Various data and information were submitted as part of a scientific rationale to waive pathogenicity testing to non-target aquatic plants. *Gliocladium catenulatum* is a common saprophyte that is widely distributed in soils and on plants worldwide with few reports of adverse effects despite its ubiquitous nature. As noted above for terrestrial plants, *G. catenulatum* was isolated from diseased or dead plants as a potential causal agent of the infection. However, this isolation from diseased plant tissues does not necessarily indicate that it was the causal agent of the disease. It is likely that *G. catenulatum* appeared on the diseased plant material as a secondary organism. In non-target testing, some adverse effects were reported on several terrestrial plant species following exposure to large concentrations of *G. catenulatum* strain J1446. However, no toxicity was observed in the algal inhibition study on the green alga, *S. subspicatus*. In addition, the uses of Prestop Biofungicide WP on greenhouse food and greenhouse non-food crops are not expected to result in significant increased exposures to non-target aquatic plants. Based on all the available information on *G. catenulatum* and the limited degree of exposure as a result of the use of Prestop Biofungicide WP in greenhouses, no additional hazard testing is required for aquatic plants.

The biological properties of this microorganism suggest that it could survive in aquatic ecosystems for a significant amount of time (see Section 4.1). However, no harm to aquatic organisms is expected despite the noted adverse effects in freshwater fish and daphnid studies. The use of Prestop Biofungicide WP is in greenhouses, and direct exposure to non-target aquatic organisms should be limited to surface water runoff from spent growing medium and treated plants. As a precaution, handlers are required not to contaminate aquatic habitats through equipment cleaning or waste disposal. In addition, growers must not allow effluent or runoff from greenhouses containing this product to enter lakes, streams, ponds or other water bodies.

5.0 Value

5.1 Effectiveness Against Pests

5.1.1 Suppression of *Pythium ultimum* and *P. aphanidermatum* as a Soil Media Application or Drench Application

A total of 12 studies were reviewed for the soil media application treatment. Trials tested Prestop Biofungicide WP at rates ranging between 0.1 and 1.0 g product per litre of peat (growing media), with the majority of the studies testing the 0.5% rate. Crops tested include cucumbers, lettuce, basil, parsley, thyme and dill.

Results showed that the lower rate of 0.1% was too low to provide acceptable disease control. However, consistent levels of acceptable disease reduction were achieved at the 0.5 g /L rate. While higher rates of 1.0% also provided good disease control, there was little advantage with respect to disease control over the 0.5% rate. When various application methods and timings (soil mix, soil drench, applied before planting and after) were compared in a side-by-side trial, the data showed a consistent trend that the greatest disease reduction was achieved when Prestop Biofungicide WP was applied first as a soil media application just before planting, followed by a soil drench application made one week after planting. Prestop Biofungicide WP was also tested at rates ranging between 0.05 and 0.5% as a drench to the soil surface prior to transplanting. Results were similar to the soil media application trials for *Pythium*, in that the 0.5% rate provided consistent suppression. When compared to another biofungicide registered for suppression of *Pythium* diseases, Prestop Biofungicide WP performed as well or better with regards to reduction in percent disease severity and percent mortality.

In general, Prestop Biofungicide WP provided good disease reduction. However, the disease control levels may not meet industry standards of control compared to standard chemical commercial fungicides, and therefore, the claim of "suppression" rather than "control" can be supported for Prestop Biofungicide WP. With regards to the actual *Pythium* diseases that can be listed on the product label, the following claims can be supported: suppression of damping-off and crown and root rot caused by *P. ultimum* and *P. aphanidermatum* (*Pythium* spp.) on cucumbers, lettuce, basil, parsley, thyme and dill.

5.1.2 Suppression of Stem Canker and Grey Mould (*Botrytis cinerea*) as a Foliar Spray Application

Three leaf bioassay studies assessed leaf sections only, while the six greenhouse trials assessed plant stems (not the fruits) for the presence of Botrytis stem canker. Prestop Biofungicide WP was tested at 0.1 to 0.5%.

Leaf Bioassay Trials
The three trials tested cucumbers, peppers and tomatoes separately, with the response variables assessed being the percent healthy disks, the percent slightly infected disks and the percent severely infected disks. The cucumber trial did not show any differences at all between treatments. For the pepper trial, all rates of Prestop Biofungicide WP demonstrated greater disease reduction than the control treatments. It was established that only the 0.5% provided consistently fewer severely diseased leaf disks, and the greatest percentage of undiseased leaf disks. The tomato trial just tested Prestop Biofungicide WP at one rate, so there was no comparison possible, except to demonstrate that there was a reduction in the level of disease compared to the inoculated control treatment.

Greenhouse Trials
With the exception of one trial, the greenhouse trials on cucumber plants each assessed only one Prestop Biofungicide WP rate, so no direct comparison can be made to other Prestop Biofungicide WP rates. Results support that the 0.5% Prestop Biofungicide WP rate provided acceptable disease severity control, as assessed through a reduction in stem lesion length and the number of stem lesions per plant. These results are consistent with the bioassay trials. Therefore, the claim of suppression of Botrytis stem canker can be supported.

A general claim of control of Botrytis grey mould was made on the proposed label. While only data assessing the effectiveness of Prestop Biofungicide WP for suppression of Botrytis stem canker was made, it is believed that the same rate, applied directly to the foliage and fruit, will also suppress general Botrytis grey mould infections. Therefore, the claim of suppression of Botrytis grey mould disease can be supported based on evidence extrapolated from the Botrytis stem canker data.

For the stem canker claim, supported crops include greenhouse peppers, tomatoes and cucumbers. Botrytis stem canker is not a problem disease for other greenhouse vegetables and herbs; therefore, this claim is not extended to other crops.

The suppression of Botrytis grey mould disease (*Botrytis cinerea*) on greenhouse peppers, tomatoes and cucumbers is also supported. Since Botrytis grey mould is a common disease of greenhouse lettuce, oregano, basil, parsley, thyme and dill, this claim can be extended to include these crops, since they have not shown any signs of phytotoxicity in any of the studies reviewed.

5.1.3 Suppression of *Rhizoctonia solani* as a Soil Media and Drench Application

A total of seven studies were assessed for this claim (four on lettuce, three on cauliflower). Results from the four lettuce trials demonstrated that Prestop Biofungicide WP did not meet acceptable levels of disease suppression at the low rates tested.

For the cauliflower trials, one application of Prestop Biofungicide WP was made, and it was incorporated into the peat before sowing in all studies. Prestop Biofungicide WP was tested at 0.001, 0.01, 0.05, 0.1, 0.5, 1.0 g product per L peat. Results suggest that there is a rate effect with Prestop Biofungicide WP, with a higher rate resulting in higher emergence counts and a greater percentage of healthy seedlings. Based on the consistency between trials, the 0.5% rate provided acceptable disease suppression when incorporated into the peat mixture before sowing. Since only early season *R. solani* disease assessments were made, only the disease claim of suppression of damping-off on can be supported. Since it was demonstrated that Prestop Biofungicide WP is effective in suppressing early-season *R. solani* when applied to cauliflower as a soil treatment, it is believed that application as a soil drench just after sowing would also be effective. For label consistency with the *Pythium* claim, Prestop Biofungicide WP applied as a 0.5% solution, a soil drench just after sowing is supported.

Since there was no evidence of phytotoxicity to lettuce, this crop can also be added to the label for these claims. However, the claim for suppression of lettuce bottom rot caused by *R. solani* cannot be supported as this disease occurs later in the growth and development of the plant (when leaves are touching the soil).

5.1.4 Suppression of Fusarium Root and Stem Wilt Disease Caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum* (on Cucumber) and *Fusarium oxysporum* f. sp. *basilica* (on Basil)

Three studies were assessed for this claim (two on cucumber and one on basil). The cucumber applications tested a single drench application in rockwool at seeding and the basil study tested Prestop Biofungicide WP incorporated into the soil before planting. Two *Fusarium* species were assessed in the trials: *Fusarium oxysporum* f. sp. *radicis-cucumerinum*, which is host-specific to cucumbers, and *Fusarium oxysporum* f. sp. *basilica,* which is host-specific to basil only.

The response variables assessed include disease incidence, disease severity and the percentage of dead plants at the time of assessment. Prestop Biofungicide WP was tested at the 1.0% rate (soil drench) and 0.5% rate (soil media incorporation). Results show that when the 1.0% Prestop Biofungicide WP solution was applied once at seeding, there was a significant reduction in disease incidence and disease severity compared to the untreated, inoculated control. For the soil media application, Prestop Biofungicide WP applied once resulted in a significantly lower percentage of dead plants at the time of assessment (three assessments made seven days apart) compared to the inoculated control plants. Again, no other Prestop Biofungicide WP rates were directly tested, so a comparison of lower rates could not be made.

It is noted that the *Fusarium* species identified in the trials are host-specific and are attributed to a very specific disease of cucumbers and basil. It should not be confused with other *Fusarium* species (*F. solani, F. graminearum, F. avenaceum*) that cause various generic seedling diseases (seed rot, damping-off, seedling blight, etc.) on vegetables. Based on these results, the claim of suppression of *Fusarium* root and stem wilt disease caused by *Fusarium oxysporum* f.sp. *radicis-cucumerinum* on cucumbers, and *Fusarium oxysporum* f.sp. *basilica* on basil, when applied as a growing media treatment, or a soil drench application at 1.0% is supported.

5.1.5 Suppression of Cucumber Gummy Stem Blight (*Didymella bryoniae*) When Applied as a Spray Application

Four trials on cucumbers were reviewed. Between two and four applications of Prestop Biofungicide WP were made, with the plant stems sprayed with 0.1% to 2% concentrations. The majority of the trials assessed at the 1% rate.

Results show that Prestop Biofungicide WP at the rate of 0.1% was too low to provide consistent, acceptable reduction in disease levels. Increasing the rate to 1% resulted in greater disease control longer into the season. A rate of just 0.5% was not tested directly, as one study tested an initial application on 0.5%, then made the subsequent applications at 1.0 %. Prestop Biofungicide WP was assessed against other biological products as well as a chemical fungicide, and the results demonstrated that Prestop Biofungicide WP provided better disease control than the other biologicals, but not to the disease control levels of the chemical fungicide. Therefore, Prestop Biofungicide WP is supported with a claim of suppression, not control.

5.1.6 Crop Extrapolations

5.1.6.1 Crop Extrapolations for the Claim of Damping-off Caused by *Rhizoctonia solani*

Certain cole crop vegetables, including cauliflower and broccoli, are grown in the greenhouse until the seedling stage, and then transplanted into the field for production and sale to the general public as bedding plants. Based on this information, and on the fact that (a) there is direct Prestop Biofungicide WP efficacy data to support suppression of damping-off caused by *R. solani* on cauliflower, (b) no phytotoxicity on cauliflower was noted, and (c) cauliflower and broccoli are very closely related crops and are susceptible to the same seedling diseases, the claim can be extended to include broccoli. The following claim is supported: Prestop Biofungicide WP will suppress damping-off caused by *Rhizoctonia solani* on cauliflower and broccoli when applied at 0.5% solution, incorporated into the growing media prior to sowing, or else applied as a drench to the surface of the growing media.

5.1.6.2 Crop Extrapolations for the Claim of Damping-off Caused by *Pythium ultimum* and *P. aphanidermatum*

Crops currently supported for this claim include cucumbers, lettuce, basil, parsley, thyme and dill. Trials assessed Prestop Biofungicide WP when applied on cauliflower, and no adverse effects were reported. Since *Pythium ultimum* and *P. aphanidermatum* can infect cauliflower and broccoli seedlings, and no adverse effects are expected on these crops, they are among the crops listed on the label for this disease claim.

5.1.6.3 Crop Extrapolations from Botrytis Stem Canker to Botrytis Grey Mould

The extrapolation of suppression of Botrytis grey mould from the Botrytis stem canker data has already been discussed in the Botrytis section above. Since Botrytis grey mould is a non-host specific disease, the claim is extended to include any greenhouse crops tested in the efficacy trials that did not show any adverse phytotoxic response. Therefore, Prestop Biofungicide WP, when applied as a 0.5% solution foliar spray to the plant leaves and fruits to runoff, will suppress Botrytis grey mould (*Botrytis cinerea*) on greenhouse peppers, tomatoes, cucumbers, lettuce, oregano, basil, parsley, thyme and dill.

5.1.6.4 Crop Extrapolation for Ubiquitous Fungal Pathogens

It is noted that certain pathogens are ubiquitous opportunists, and may infect any plant at the seed to seedling stage, including *Pythium* spp. (damping-off, crown and root rot), *Rhizoctonia solani* (damping-off), and *Botrytis cinerea* (grey mould). Based on the fact that these claims have been reviewed for their efficacy, that they are not host-specific pathogens and that the crops tested in any of the trials did not display any adverse effects (phytotoxicity), these generic seedling disease claims can be extended to include additional crops. Based on the rationale above, the following claims are supported: suppression of seedling damping-off and crown and root rot caused by *Pythium* spp., damping-off caused by *Rhizoctonia solani,* and Botrytis grey mould (*Botrytis cinerea*) on cucumbers, peppers, tomatoes, peppers, lettuce, basil, oregano, parsley, thyme, dill, cauliflower and broccoli.

5.2 Effectiveness Against Pests on Ornamental Crops

5.2.1 Suppression of Root and Basal Stem Rot Caused by *Phytophthora cryptogea* as a Soil Media Application or Drench Application

Two efficacy trials conducted on petunia and tagetes were reviewed. The results of these trials showed that Prestop Biofungicide WP applied by incorporation at the rate of 1.0 g of product/L of growing medium resulted in up to a seven-fold increase of total plant dry weight of petunia and tagetes compared to the untreated inoculated control. No direct assessment of *P. cryptogea* disease presence was made. The tested rate was twice the proposed rate. However, based on the result of Prestop Biofungicide WP applied at 1.0 g of product/L of growing medium, it is expected that the rate of 0.5 g of product/L of growing medium will provide suppression of root and basal stem rot on petunia and tagetes. This claim is extended to include alyssum, geranium, pansy, poinsettia, saintpaulia, salvia and snapdragon, the ornamentals tested to support the

proposed efficacy claims for Prestop Biofungicide WP and that are susceptible to *P. cryptogea.* Since the genus *Phytophthora* contains various species that are responsible for many economically important diseases in Canada, the claim cannot be extrapolated to include all species of *Phytophthora*, just *P. cryptogea*.

5.2.2 Suppression of Damping-off Caused by *Pythium* spp. as a Soil Media Application or Drench Application

Three efficacy trials on pansy (two) and snapdragon (one) were reviewed. When applied via media incorporation at the rate of 0.5 g of product per L of growing medium, Prestop Biofungicide WP increased seedling emergence after 10, 14, 21 and 28 days. The claim that Prestop Biofungicide WP will suppress damping-off caused by *Pythium* spp. as a soil media application or drench application at 0.5 g of product per L of growing medium is supported. This claim is extended to include alyssum, geranium, petunia, poinsettia, saintpaulia, salvia and tagetes, the ornamentals tested to support the proposed efficacy claims for Prestop Biofungicide WP that are susceptible to *Pythium* spp.

5.2.3 Suppression of Damping-off Caused by *Rhizoctonia solani* as a Soil Media Application or Drench Application

Two efficacy trials (one on alyssum and one on salvia) were reviewed. Prestop Biofungicide WP applied by media incorporation at 0.5 g of product/L of growing medium provided significant increased seedling emergence after 14, 21 and 28 days. When applied by incorporation at the rate of 0.5 g of product/L of growing medium for salvia, Prestop Biofungicide WP provided 47% control of damping-off caused by *R. solani.* The claim that Prestop Biofungicide WP will suppress damping-off caused *R. solani* as a soil media application or drench application at the rate of 0.5 g of product per L of growing medium is supported. This claim is extended to include geranium, pansy, petunia, poinsettia, saintpaulia, snapdragon and tagetes, the ornamentals tested in the current submission that are susceptible to *R. solani.*

5.2.4 Extrapolation of disease claims from food crops to the tested ornamental crops

5.2.4.1 Suppression of grey mould caused by *Botrytis cinerea* as a foliar spray application

Botrytis cinerea is a non-specific pathogen that attacks a wide range of ornamentals. Submitted data for food crops showed that Prestop Biofungicide WP applied at 0.5% (0.5 g/L) will suppress botrytis stem rot on greenhouse peppers, tomatoes and cucumbers. Based on the food crop results and the fact that there are no concerns with regard to phytotoxicity on the tested ornamental crops, the claim that Prestop Biofungicide WP will suppress *Botrytis cinerea* when applied at 0.5% is extrapolated from greenhouse peppers, tomatoes, and cucumbers to include the tested ornamentals: alyssum, geranium, pansy, petunia, poinsettia, salvia, saintpaulia, snapdragon and tagetes.

5.3 Phytotoxicity to Host Plants

Prestop Biofungicide WP has been tested at very high rates on 30 plant species, and the only adverse effects reported were on rye and spinach, where the growth was shown to be slightly inhibited. Therefore, Prestop Biofungicide WP is not expected to adversely affect the supported plants when applied as per the approved directions for use.

5.3.1 Acceptable Label Claims for Food and Ornamental Plants

Disease (Pathogen)	Crop	Method of Application; Rate	Application Timing and Interval
Damping-off, crown and root rot (caused by *Pythium ultimum, P. aphanidermatum)* Damping-off (caused by *Rhizoctonia solani)*	Vegetables: cucumbers, peppers, tomatoes, cauliflower, broccoli, lettuce Herbs: oregano, basil, parsley, thyme, dill	Soil media treatment: 0.5% Prestop Biofungicide WP Solution	Apply once, before crops are seeded, planted or transplanted.
		Drench application: 0.5% Prestop Biofungicide WP Solution	Apply immediately after seeding, planting or transplanting. Repeat applications every 3–6 weeks.*
Damping-off (caused by *Pythium ultimum, P. aphanidermatum, Rhizoctonia solani)*	Ornamentals: alyssum, pansy, petunia, poinsettia, tagetes, salvia, saintpaulia, snapdragon, geranium	Soil media treatment: 0.5% Prestop Biofungicide WP Solution	Apply once, before crops are seeded, planted or transplanted.
		Drench application: 0.5% Prestop Biofungicide WP Solution	Apply immediately after seeding, planting or transplanting. Repeat applications every 3–6 weeks.*
Root and basal stem rot (caused by *Phytophthora cryptogea*)	Ornamentals: alyssum, pansy, petunia, poinsettia, tagetes, salvia, saintpaulia, snapdragon, geranium	Soil media treatment: 0.5% Prestop Biofungicide WP Solution	Apply once, before crops are seeded, planted or transplanted.
		Drench application: 0.5% Prestop Biofungicide WP Solution	Apply immediately after seeding, planting or transplanting. Repeat applications every 3–6 weeks.

Disease (Pathogen)	Crop	Method of Application; Rate	Application Timing and Interval
Root and stem wilt (caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum*, *Fusarium oxysporum* f. sp. *basilica)*	Cucumbers, basil	Soil media treatment: 1.0% Prestop Biofungicide WP Solution	Apply once before crops are seeded, planted or transplanted.
		Drench application: 1.0% Prestop Biofungicide WP Solution	Apply immediately after seeding, planting or transplanting. Repeat applications every 3–6 weeks.
Botrytis grey mould (caused by *Botrytis cinerea*)	Vegetables: cucumbers, peppers, tomatoes, cauliflower, broccoli, lettuce Herbs: oregano, basil, parsley, thyme, dill Ornamentals: alyssum, pansy, petunia, poinsettia, tagetes, salvia, saintpaulia, snapdragon, geranium	Foliar spray application: 0.5% Prestop Biofungicide WP Solution	Apply preventatively. Apply as a foliar spray to wet. Repeat applications every 3–4 weeks.
Stem canker (caused by *Botrytis cinerea*)	Vegetables: cucumbers, peppers tomatoes	Foliar spray application: 0.5% Prestop Biofungicide WP Solution	Apply preventatively. Apply as a foliar spray to wet. Repeat applications every 3–4 weeks.
Gummy stem blight (*Didymella bryoniae*)	Vegetables: cucumbers only	Foliar spray application: 1.0% Prestop Biofungicide WP Solution	Apply preventatively. Apply as a foliar spray to wet. Repeat applications every 3–4 weeks.

***** Apply the shorter application interval when disease pressures are moderate to high.

5.4 Impact on Succeeding Crops

Not assessed.

5.5 Economics

Not assessed.

5.6 Sustainability

5.6.1 Survey of Alternatives

5.6.1.1 Summary Table of Alternative Actives Registered in Canada for Control of the Greenhouse-grown Edible Crops and Related Disease, as Listed on the Prestop Biofungicide WP Label

Disease claim	Crop	Technical Grade Active Ingredient
Damping-off, and/or crown and root rot (caused by *Pythium)*	cucumbers	metalaxyl-m, propamocarb hydrochloride, *Trichoderma harzianum*, *Bacillus subtilis*
	peppers, lettuce, cauliflower	*Bacillus subtilis*
	tomatoes	*Trichoderma harzianum*, *Bacillus subtilis*
Gummy stem blight	cucumbers	myclobutanil, mancozeb, iprodione
Damping-off (caused by *Rhizoctonia solani*)	cucumbers, broccoli, peppers, tomatoes, lettuce, parsley, dill, thyme, basil, oregano	fludioxonil (seed treatment)
Botrytis grey mould (caused by *Botrytis cinerea*)	lettuce	ferbam, iprodione, fenhexamid
	cucumber	iprodione
	tomato	iprodione, fenhexamid, *Bacillus subtilis*
	peppers	*Bacillus subtilis*
Botrytis stem canker (caused by *Botrytis cinerea*)	tomato	dichloran

5.6.1.2 Summary Table of Alternative Actives Registered in Canada for Control of the Ornamental Crops and Related Disease Listed on the Prestop Biofungicide WP Label

Crop	Disease claim	Technical Grade Active Ingredient
alyssum, pansy, poinsettia, petunia, salvia, snapdragon	Root and basal stem rot caused by *Phytophthora cryptogea*	etridiazole
	Damping-off caused by *Pythium* spp.	etridiazole
	Root rot caused by *Rhizoctonia*	thiophanate-methyl
alyssum, pansy, petunia, poinsettia	*Rhizoctonia* root rot	tryfloxystrobin
poinsettia	*Pythium* root rot	folpet
pansy, poinsettia, petunia, salvia, snapdragon	Damping-off caused by *Pythium* spp.	metalaxyl-M
pansy, poinsettia, petunia, salvia, snapdragon	Root and basal stem rot *Phytophthora cryptogea*	metalaxyl-M

5.6.2 Compatibility with Current Management Practices Including Integrated Pest Management (IPM)

Prestop Biofungicide WP can easily be incorporated into current IPM practices. There is no need for new equipment or techniques to be used when applying the product. In addition, since Prestop Biofungicide WP is to be used as a preventative, it may contribute to reducing the reliance on chemical fungicides during later growing stages, which is a cornerstone of IPM practices.

5.6.3 Information on the Occurrence or Possible Occurrence of the Development of Resistance

Due to the fact that Prestop Biofungicide WP is a biofungicide, and the mode of action is competitive inhibition along with some chitinase activity, there is very little concern for resistance developing in pathogenic fungi populations. The Fungicide Resistance Action Committee (FRAC) does not assign a group code to biofungicide active ingredients.

5.6.4 Contribution to Risk Reduction and Sustainability

Registration of Prestop Biofungicide WP will contribute to risk reduction because the nature of the mode of action is such that there is little chance of resistance developing in pathogenic fungal populations. In addition, it is another product available to growers that, when incorporated into current growing practices, may lead to a reduced need for chemical fungicide alternatives.

6.0 Pest Control Product Policy Considerations

6.1 Toxic Substances Management Policy Considerations

The management of toxic substances is guided by the federal government's Toxic Substances Management Policy (TSMP), which puts forward a preventive and precautionary approach to deal with substances that enter the environment and could harm the environment or human health. The policy provides decision makers with direction and sets out a science-based management framework to ensure that federal programs are consistent with its objectives. One of the key management objectives is virtual elimination from the environment of toxic substances that result predominantly from human activity and that are persistent and bioaccumulative. These substances are referred to in the policy as Track 1 substances.

In its review, the PMRA took into account the federal Toxic Substances Management Policy and followed its Regulatory Directive DIR99-03, *The Pest Management Regulatory Agency's Strategy for Implementing the Toxic Substances Management Policy*. Substances associated with its use were also considered, including microcontaminants in the technical product, *Gliocladium catenulatum* strain J1446, and formulants in the end-use product, Prestop Biofungicide WP. The PMRA has reached the following conclusions.

Gliocladium catenulatum strain J1446 does not meet the Track 1 criteria because the active ingredient is a biological organism and hence is not subject to the criteria used to define persistence, bioaccumulation and toxicity properties of chemical control products. There are also no formulants, contaminants or impurities present in the end-use product that would meet the TSMP Track 1 criteria.

Therefore, the use of *Gliocladium catenulatum* strain J1446 and Prestop Biofungicide WP is not expected to result in the entry of Track 1 substances into the environment.

6.2 Formulants and Contaminants of Health or Environmental Concern

Gliocladium catenulatum strain J1446 does not contain any contaminants of health or environmental concern identified in the *Canada Gazette*, Part II, Volume 139, Number 24, pages 2641-2643: *List of Pest Control Product Formulants and Contaminants of Health or Environmental Concern*.

The end-use product, Prestop Biofungicide WP, contains the formulant skim milk powder, which is identified in the *Canada Gazette,* Part II, Volume 139, Number 24, pages 2641-2643: *List of Pest Control Product Formulants of Health or Environmental Concern* as an allergen known to cause anaphylactic-type reactions. Therefore, the label for Prestop Biofungicide WP will include the precautionary statement "Warning: this product contains the allergen skim milk powder (56% weight/weight)"on the principal display panel.

7.0 Summary

7.1 Characterization and Methods for Analysis

The product characterization data for *G. catenulatum* strain J1446 (*Gliocladium catenulatum* J1446 Dried Cell Mass) and Prestop Biofungicide WP were judged to be adequate to assess their potential human health and environmental risks. The technical material was fully characterized and the specifications were supported by the analyses of a sufficient number of batches. Although the quality assurance program lacked microbe-specific screens for specific pathogens, the absence of contaminating microorganisms in a number of representative batches indicates that the manufacturer's quality assurance program is successful at limiting contaminating microorganisms. No additional microbe-specific testing is required to assure product quality.

Storage stability data were sufficient to support a shelf life of 12 months for Prestop Biofungicide WP when the product is stored at temperatures below 4°C.

7.2 Human Health and Safety

The acute toxicity and infectivity studies submitted in support of *G. catenulatum* strain J1446 were determined to be sufficiently complete to permit a decision on registration. *Gliocladium catenulatum* strain J1446 was of low toxicity in the rat when administered via the oral, dermal, pulmonary and intraperitoneal routes. *Gliocladium catenulatum* strain J1446 was not pathogenic or infective via the oral, pulmonary and intraperitoneal routes. Slight dermal irritation was observed in the dermal toxicity study and slight ocular irritation was observed in the eye irritation study. Waiver requests were submitted to address all toxicology requirements (acute dermal toxicity and dermal irritation) for Prestop Biofungicide WP. These waiver requests were accepted based on the nature and concentration of the formulation ingredients.

As a result of a dermal sensitization study, *G. catenulatum* strain J1446 was determined to be a sensitizing agent. All microbial pesticides are generally considered to be potential sensitizers. Exposure to allergens, including *G. catenulatum* strain J1446, may cause allergies following repeated exposures to high concentrations. As a result, the signal words "POTENTIAL SENSITIZER" are required on the principal display panels of all labels, *Gliocladium catenulatum* J1446 Dried Cell Mass, and Prestop Biofungicide WP. No eye irritation studies were submitted for Prestop Biofungicide WP since the formulation ingredients are not expected

to cause eye irritation. Therefore, no signal words are required for Prestop Biofungicide WP. However, one of the formulation ingredients, skim milk powder, is known to be an allergen and must be labelled as such according to PMRA Regulatory Directive DIR2006-02, *Formulants Policy and Implementation Guidance Document*.

When handled according to the label instructions, the potential routes of exposure to applicators, mixers/loaders, handlers and early-entry workers are pulmonary, dermal and ocular. While submitted studies on *G. catenulatum* strain J1446 indicated a potential for sensitization, inhalation and dermal exposure are not a concern if the required dust/mist filtering respirator and appropriate personal protective equipment (PPE) to be stipulated on the end-use product label is worn by applicators, mixers/loaders and handlers. Early-entry workers will be restricted from entering areas where Prestop Biofungicide WP has been applied as a foliar spray for a period of four hours unless wearing the indicated personal protective equipment, with the exception of eye goggles and a dust/mist filtering respirator, which are required only until the spray mist has settled. The label does not allow applications to turf or residential and recreational areas. Because the use sites are agricultural, exposure to infants and children in school, residential and daycare facilities is likely to be minimal to non-existent. Consequently, the health risk to infants and children is expected to be negligible.

No significant toxicity or signs of pathogenicity were observed when *G. catenulatum* strain J1446 was administered orally to rats, and no metabolic byproducts of toxicological concern are produced by this microorganism. The establishment of a maximum residue limit (MRL) is therefore not required for *G. catenulatum* strain J1446 under section 4(*d*) of the *Food and Drugs Act* (adulteration of food) as defined under Division 15, section B.15.002 of the Food and Drugs Regulations.

7.3 Environmental Risk

The non-target studies, scientific rationales and published scientific literature submitted in support of *G. catenulatum* strain J1446 were determined to be sufficiently complete to permit a decision on registration.

Several environmental fate and behaviour studies were submitted on the survival of *G. catenulatum* strain J1446 in water and peat under greenhouse conditions as well as on the leaf surface after foliar application. These studies showed that the MPCA will persist to varying degrees in different environments, including peat-based growing media, aqueous environments and on foliage, then return to naturally occurring levels for this species. The exact amount of time that is required, however, depends on many factors, including its growth medium, temperature, pH and light conditions. No additional studies were required to address the environmental fate and behaviour of *G. catenulatum* strain J1446. Environmental fate data (Tier II/III) are not normally required in the absence of significant toxicological effects in non-target organisms in Tier I testing.

Environmental effects studies and published literature were submitted to address risks of *G. catenulatum* strain J1446 to non-target organisms. These studies and other published information showed that the use of Prestop Biofungicide WP containing *G. catenulatum* strain J1446 would not harm birds, mammals, terrestrial arthropods (including honeybees), non-athropod invertebrates, algae or soil microogranisms. *Gliocadium catenulatum* strain J1446 could however pose a mild hazard to freshwater fish and aquatic arthropods as well as some terrestrial plants following exposure to large concentrations of *G. catenulatum* strain J1446. The biological properties of this microorganism suggest that this MPCA could survive in aquatic ecosystems. However, no harm to organisms is expected based on the limited degree of exposure resulting from the use of Prestop Biofungicide WP in commercial greenhouses. As a precaution, handlers are required not to contaminate aquatic habitats through equipment cleaning or waste disposal. In addition, growers must not allow effluent or runoff from greenhouses containing this product to enter lakes, streams, ponds or other water bodies.

7.4 Value

Prestop Biofungicide WP, when applied as per the supported directions for use as a soil media treatment, a soil drench treatment or as a foliar treatment, will effectively suppress the listed soil-borne pathogens, and root and foliar diseases on specific greenhouse-grown vegetables, herbs and ornamentals. It is best used as a preventative before disease is present, and reapplication is necessary every three to six weeks, depending on the disease pressures in the greenhouse and the method of application. When applied as directed, Prestop Biofungicide WP will not result in phytotoxicity to the target plants.

7.5 Unsupported Uses

Certain claims and methods of application originally proposed with this application, including diseases caused by various other species of *Phytophthora*, dip application of Prestop Biofungicide, and drip irrigation (chemigation) methods, are not supported by the PMRA because supporting evidence has not been adequately provided. In addition, lettuce bottom rot disease (*R. solani*) was not supported because only early season disease assessments were made on this crop, and generally, the disease does not become a problem until the crop has become well established.

8.0 Proposed Regulatory Decision

Health Canada's PMRA, under the authority of the *Pest Control Products Act*, is proposing full registration for the sale and use of the technical grade active ingredient *Gliocladium catenulatum* J1446 Dried Cell Mass and the end-use product Prestop Biofungicide WP for the suppression of a variety of fungal diseases on the following greenhouse-grown vegetables, herbs and ornamentals: cucumber, tomato, pepper, lettuce, cauliflower, broccoli, oregano, basil, parsley, thyme, dill, alyssum, geranium, pansy, petunia, salvia, snapdragon, tagetes, poinsettia and saintpaulia. An evaluation of current scientific data from the applicant and scientific reports has resulted in the determination that, under the proposed conditions of use, the end-use product has value and does not present an unacceptable risk to human health or the environment.

List of Abbreviations

°C	degree(s) Celsius
µg	microgram
a.i.	active ingredient
ADI	acceptable daily intake
ARfD	acute reference dose
bw	body weight
CAS	chemical abstracts service
CFU	colony forming units
DNA	deoxyribonucleic acid
EC_{50}	effective concentration on 50% of the population
EEC	environmental effects concentration
g	gram
IPM	integrated pest management
IUPAC	International Union of Pure and Applied Chemistry
kg	kilogram
km	kilometre
K_{ow}	*n*-octanol–water partition coefficient
L	litre
LC_{50}	lethal concentration 50%
LD_{50}	lethal dose 50%
LOAEL	lowest observed adverse effect level
LOEC	low observed effect concentration
LOQ	limit of quantitation
mg	milligram
mL	millilitre
MAS	maximum average score
MOE	margin of exposure
MPCA	microbial pest control agent
MRL	maximum residue limit
N/A	not applicable
NIOSH	National Institute for Occupational Safety and Health
NOAEL	no observed adverse effect level
NOEC	no observed effect concentration
NOEL	no observed effect level
NOER	no observed effect rate
N/R	not required
NZW	New Zealand white
pKa	dissociation constant
PMRA	Pest Management Regulatory Agency
ppm	parts per million
RSD	relative standard deviation
$t_{1/2}$	half-life
T3	tri-iodothyronine
T4	thyroxine

TGAI	technical grade active ingredient
TSMP	Toxic Substances Management Policy
UF	uncertainty factor
v/v	volume per volume dilution
WP	wettable powder

Appendix I Tables and Figures

Table 1 Toxicity and Infectivity of *Gliocladium catenulatum* strain J1446 and its Associated Products (Prestop Biofungicide WP)

Study Type	Species, Strain, and Doses	Result	Significant Effects and Comments	Reference(s)
Acute Toxicity/Infectivity of *G. catenulatum* strain J1446				
Acute Oral Toxicity and Infectivity	Rat - CD 5 groups of 3/sex treated with dose of MPCA at 2.0×10^9 CFU in 20 mL saline/kg body weight. Sacrificed (in groups of 3/sex) on Days 2, 4, 8, 15 and 22. 3/sex treated with autoclaved MPCA (equivalent amount as above). Sacrificed on Day 22. 2 groups of 3/sex untreated shelf and non-shelf control.	$LD_{50} > \sim 2.0 \times 10^9$ CFU/kg body weight	- No mortalities and no abnormalities upon necropsy. - All animals displayed piloerection but resolved by Day 2. No other clinical observations noted. - Viable *Gliocladium catenulatum* recovered from feces of 2 subjects on Day 2. No other viable MPCA was recovered from the brain, kidneys, spleen, liver, heart, lungs, mesenteric lymph nodes, or blood of any other subject. LOW TOXICITY, NOT INFECTIVE	1187993
Acute Oral Toxicity	Rat - CD 5/sex treated with dose of 2000 mg/kg body weight of MPCA. Observed daily and sacrificed on Day 14.	$LD_{50} > 2000$ mg/kg body weight	- No mortalities and no abnormalities upon necropsy. - Reduced activity noted 4 hours after dosing. No other clinical observations noted. LOW TOXICITY	1187994

Study Type	Species, Strain, and Doses	Result	Significant Effects and Comments	Reference(s)
Acute Pulmonary Toxicity and Infectivity	Rat - CD 2 groups of 10/sex (sacrificed on Days 1 and 22) and 4 groups of 5/sex (sacrificed on Days 2, 4, 8 and 15) were treated with intratracheal instillation dose of MPCA at 6.60 or 7.98×10^7 CFU/kg body weight. 10/sex treated with autoclaved MPCA (equivalent amount as above). Sacrificed on Day 22. 2 groups of 10/sex untreated shelf and non-shelf control.	LC_{50} > ~6.60×10^7 CFU/kg body weight	- 3 mortalities occurred within 24 hours of dosing. - Piloerection, hunched posture, waddling gait, lethargy, partially closed eyelids, respiratory disturbances and pallor of extremities in many of subjects treated with viable and autoclaved MPCA. All symptoms resolved by Day 3. - Viable MPCA recovered from lung tissue on Days 1 and 3. Not recovered from brain, kidneys, spleen, liver, heart, lungs, mesenteric lymph nodes or blood. - No macroscopic observations noted at necropsy. LOW TOXICITY, NOT INFECTIVE	1187996
Acute Pulmonary Toxicity	Rat - CD 5/sex exposed to 5.57 mg/L MPCA. Observed daily until sacrifice and necropsy on Day 14.	LC_{50} > 5.57 mg/L	- No mortalities and no abnormalities upon necropsy. - Reduced activity and hunched posture resolving after 24 hours. LOW TOXICITY	1187997

Study Type	Species, Strain, and Doses	Result	Significant Effects and Comments	Reference(s)
Intraperitoneal Infectivity	Rat - CD 6 groups of 3/sex treated with dose of MPCA at 4.2×10^8 CFU in 3mL saline/kg body weight. Sacrificed (in groups of 3/sex) at 1 hour, and Days 2, 4, 8, 15 and 22. 3/sex treated with autoclaved MPCA (equivalent amount as above). Sacrificed on Day 22. 3/sex untreated shelf and 5/sex untreated non-shelf control.		- No mortalities. - Macroscopic findings include generalized congestion (enlarged organs, pus-filled nodules, pallor, spongy tissue, granulomatous tissue and adhesion of organs/tissue in abdominal cavity) typical of a strong inflammatory response. - Piloerection, hunched posture and waddling gait were noted. Less common were increased respiration, swollen abdomen, fecal disturbances, soiled fur, cold body surfaces, ungroomed and thin appearance. - Viable MPCA was recovered from the kidneys, spleen, liver, mesenteric lymph nodes, heart, lungs and caecum up to Day 3 but none were recovered from brain or blood.	1187999
Acute Dermal Toxicity	Rat - CD 5/sex treated with a dose of 2000 mg/kg body weight of MPCA. Observed daily and sacrificed on Day 14.	LD_{50} > 2000 mg/kg body weight	- No mortalities and no abnormalities upon necropsy. - No clinical observations noted. LOW TOXICITY	1188001
Acute Dermal Irritation	Rabbit - New Zealand White 3 males given 0.5 g dose of MPCA for 4 hours. Skin reaction observed at 1, 24, 48 and 72 hours and further observations made up to 9 days.	MINIMAL IRRITANT	- Slight erythema in one subject from 48 hours to 4 days, which turned to desquamation up to 7 days, fully recovering by Day 9. MINIMALLY IRRITATING	1188006

Study Type	Species, Strain, and Doses	Result	Significant Effects and Comments	Reference(s)
Dermal Sensitization	Guinea Pigs - Dunkin-Hartley Induction: 20 females (test) treated with 75% MPCA preparation in suspension with 0.5% carboxymethylcellulose vehicle for 6-hour period one day/week for 3 weeks. 10 females (control) treated with vehicle only. Challenge: 2 weeks after 3rd application, all subjects were treated with both MPCA preparation and vehicle on two separate sites. Irritation assessments performed at 24 and 48 hours.	SENSITIZER	Induction: - Slight irritation in 2 test group subjects after 1st application and in one subject after 3rd application. - No reactions observed in control group. Challenge: - 11 positive responses (55%) in test group. - 2 slight reactions (20%) in control group.	1188012
Primary Eye Irritation	Rabbit - New Zealand White 2 males and one female were treated with 0.1 mL of MPCA. Subjects were observed at 1, 24, 48 and 72 hours, with one subject observed at Day 7.	MAS[1] = 5.77/110 MIS[2] = 12/110	- Redness, chemosis and discharge were observed in all subjects at 1, 24 and 48 hours. The female subject had redness at 72 hours. MINIMALLY IRRITATING	1188013
Primary Eye Irritation	Rabbit - New Zealand White 3 males were treated with 0.1g of MPCA. Subjects were observed at 1, 24, 48, and 72 hours.		- Redness and discharge observed in all subjects at 1 hour. Chemosis observed in 1 subject at 1 hour. MINIMALLY IRRITATING	1188014
Cell Toxicity	Feline fetus lung cells Cells were exposed to extracts of 4 batches of MPCA and observed for cytotoxic effects.		- No toxic effects observed. LOW TOXICITY	1.187967e+27

Study Type	Species, Strain, and Doses	Result	Significant Effects and Comments	Reference(s)
Acute Toxicity/Irritation of Prestop Biofungicide WP				
All Acute Toxicity Testing			Based on the nature and concentration of each formulation ingredient, data waiver requests were found to be acceptable to fully assess the risks associated with the end-use formulations. WAIVERS ACCEPTED	1.188000e+13

[1] MAS = Maximum Average Score
[2] MIS = Maximum Irritation Score

Table 2 Toxicity to Non-Target Species

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Terrestrial Organisms					
Vertebrates					
Birds (*Colinus virginianus*; Northern bobwhite quail)	Oral (Acute)	i. *Gliocladium catenulatum* cell mass (Technical) in reverse osmosis water ii. reverse-osmosis water	$LD_{50} > 1.4\times10^6$ CFU/g bw/day for 5 consecutive days (equivalent to 10 mL/kg bw/day for 5 consecutive days NOEL 1.4×10^6 CFU/g bw/day for 5 consecutive days	- No mortalities - One bird in the treatment group was observed with subcutaneous emphysema (a pocket of air under the skin) from Day 25 to test termination. Also on Day 25, a bird in the control group suffered a fractured wing during body weight procedures. These observations were not considered to be related to treatment. - No differences in body weight, body weight gain or feed consumption. - At necropsy, one bird in the negative control group was noted with a fractured right humerus. A bird in the treatment group was noted with subcutaneous emphysema in the abdominal and thoracic regions and pale breast muscle with petechial hemorrhaging. These findings were not considered to be related to treatment. **ACCEPTABLE**	PMRA 1188027

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Wild Mammals	No study or waiver submitted. No reports of adverse effects were found in the databases of PubMed and TOXNET using the keywords "gliocladium catenulatum". Furthermore, the laboratory animal studies on the rat submitted in support of this registration and reviewed in Section 3.1 indicate that there is no pathogenicity and little toxicity from most routes of exposure at maximum hazard dose levels. **WAIVER ACCEPTED**				PMRA 1187993 1187994 1187996 1187997 1187999 1188000 1188001 1188005 1188006 1188009 1188011 1188012 1188013 1188014 1187967 1187969 1187970 1187972
Invertebrates					
Honeybees (*Apis mellifera*)	Oral (Dietary)	i.*Gliocladium catenulatum* cell mass (TGAI) in honey and water ii. honey and water	LC_{50} > 5200 ppm NOEC 5200 ppm	- The test was terminated on Day 10 when mortality in the negative control group exceeded 20%. - On Day 10, mortality in the 52, 520, and 5200 ppm treatment groups was 37% (28 of 75), 37% (28 of 75) and 36% (27 of 75), respectively. - A small number of bees exhibited lethargy or loss of equilibrium in the control and/or treatment groups during the test. - Pathogenicity could not be assessed since the test material was administered in honey, which naturally contains antimicrobial agents. **ACCEPTABLE**	PMRA 1188031

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Ladybird beetles (*Hippodamia convergens*)	Oral (Dietary)	i. *Gliocladium catenulatum* cell mass (TGAI) in honey and water ii. honey and water	LC_{50} > 5200 ppm NOEC 5200 ppm	- The test was terminated on Day 16 when mortality in the negative control group exceeded 20%. - On Day 16, mortality in the 52, 520, and 5200 ppm treatment groups was 17% (13 of 75), 19% (14 of 75) and 17% (13 of 75), respectively. - Starting on Day 9, small number of immobile beetles were observed in both control and treated beetles. - Pathogenicity could not be assessed since the test material was administered in honey, which naturally contains antimicrobial agents. **ACCEPTABLE**	PMRA 1188032
Green lacewing larvae (*Chrysoperla carnea*)	Oral (Dietary)	i. *Gliocladium catenulatum* cell mass (TGAI) in moth egg meal ii. moth egg meal	LC_{50} > 3.6×10^6 CFU/mL diet (equivalent to 5200 ppm) NOEC 3.6×10^6 CFU/mL diet	- The study was terminated on Day 12 when mortality in the negative control group exceeded 20% and pupation exceeded 50%. - On Day 12, mortality in the 3.6×10^4, 3.6×10^5 and 3.6×10^6 CFU/mL treatment groups was 20% (6 of 30), 13% (4 of 30) and 13% (4 of 30), respectively, with 67% (20 of 30), 70% (21 of 30) and 70% (21 of 30) pupation, respectively. - No signs of toxicity were noted among the surviving larvae in the treatment groups during the study. **ACCEPTABLE**	PMRA 1188033

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Parasitic Hymenopteran (*Nasonia vitripennis*)	Oral (diet)	i. *Gliocladium catenulatum* cell mass (Technical) in honey and water ii. honey and water	LC_{50} 1992 ppm NOEC 520 ppm	- The study was terminated on Day 8 when mortality in the negative control group exceeded 20%. - On Day 8, mortality in the 52, 520, and 5200 ppm treatment groups was 35% (26 of 75), 25% (19 of 75) and 68% (51 of 75), respectively. - A small number of wasps in the treated and control groups exhibited lethargy, a loss of equilibrium or were immobile. - All other surviving wasps were normal in appearance and behaviour. - Pathogenicity could not be assessed since the test material was administered in honey, which naturally contains antimicrobial agents. **ACCEPTABLE**	PMRA 1188034
Earthworms	Acute	i. *Gliocladium catenulatum* cell mass (Technical) ii. untreated control	$LC_{50} > 8.8\times10^8$ CFU/kg dry soil NOEC 5.3×10^8 CFU/kg dry soil	- On Day 14, an earthworm died in the test group treated with 8.8×10^8 CFU/kg dry soil. This earthworm could not be located in the soil and was presumed dead and decomposed. A treatment-related effect could not be precluded. - All other surviving worms in the treatment groups were normal in appearance and behaviour. - No soil aversions were noted throughout the study period. **ACCEPTABLE**	1188036

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Soil microbes	Acute	A waiver was requested based on data and other information. The weight of evidence suggests that *G. catenulatum* strain J1446's mode of action is due to hyperparasitism and the production of lytic enzymes rather than toxic antimicrobial agents such as gliotoxin. Furthermore, *G. catenulatum* is a normal component of the soil, and the organism is not expected to affect environmentally or economically important microbial species or microbiologically mediated biogeochemical processes. **WAIVER ACCEPTED**			1.188e+62
Plants					
Terrestrial Plants	Acute	A waiver was requested based on data and extensive literature searches. Few adverse effects were noted in some terrestrial plants following exposure to large concentrations of *G. catenulatum* strain J1446, and no adverse effects were observed in an algal inhibition study at a limit concentration of 100 mg/L. Extensive literature searches using several comprehensive databases found five articles that reported the isolation of *G. catenulatum* from diseased or dead plants but the isolation of *G. catenulatum* from diseased plant tissues does not necessarily indicate that it was the causal agent of the disease. It is likely that *G. catenulatum* appeared on the diseased plant material as a secondary organism. The use of Prestop Biofungicide WP on greenhouse food and greenhouse non-food crops is not expected to result in significant increased exposures or adverse effects to non-target plants. **WAIVER ACCEPTED**			1.188e+41

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Aquatic Organisms					
Vertebrates					
Rainbow trout (*Oncorhynchus mykiss*)	Aqueous and oral (diet)	i. *Gliocladium catenulatum* cell mass (Technical) ii. dilution water (negative control)	30-day LC_{50} 3.5×10^8 CFU/L (equivalent to 504 mg/L) NOEC 1.3×10^8 CFU/L (equivalent to 179 mg/L) LOEC 2.5×10^8 CFU/L (equivalent to 357 mg/L)	- Test suspensions were very cloudy in 2.5×10^8 CFU/L and 5.0×10^8 CFU/L treatment groups, i.e. two highest test concentrations. - All rainbow trout treated with 5.00×10^8 CFU/L died within 8 days. - Fish in the 3.15×10^7, 6.23×10^7 and 1.25×10^8 CFU/L treatments survived and were normal in appearance. - Fish in the 2.50×10^8 CFU/L treatment survived, but some exhibited lethargy and others laid on the bottom of the test chamber with only gill movements being observed. - There were no necropsy findings in fish that survived. Fish that died during the course of the study were not necropsied. - Viable colonies (up to 14) were recovered from the kidney, heart, liver and brains of fish treated with 1.25×10^8 CFU/L and 2.5×10^8 CFU/L. **ACCEPTABLE**	1188028, 1188029
Estuarine/ marine fish	Acute	No studies were submitted. Effects data not required as minimal exposure is expected based on the uses.			

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Invertebrates					
Daphnia magna	21-day life cycle	i. *Gliocladium catenulatum* Cell Mass (Technical) ii. dilution water (negative control)	LC_{50} 5.5×10^6 CFU/L (equivalent to 7.8 mg/L) NOEC 2.0×10^6 CFU/L (equivalent to 2.8 mg/L) LOEC 3.9×10^6 CFU/L (equivalent to 5.8 mg/L), due to effects on reproduction	- None of the daphnids were immobile. - On Day 21, cumulative mortality in daphnids treated with 2.0×10^6, 3.9×10^6, 7.7×10^6, 1.6×10^7 and 3.2×10^7 CFU/L was 20%, 35%, 70%, 75% and 95%, respectively. - One daphnid in each of the 3.9×10^6 and 1.6×10^7 CFU/L treatment groups appeared lethargic on Days 14 and 9, respectively. - The mean length and mean dry weight in the 7.7×10^6, 1.6×10^7 and 3.2×10^7 CFU/L treatment groups were reduced but the statistical significance was not evaluated due to the statistically significant effect on survival. - The mean number of young per adult in the 3.9×10^6 CFU/L treatment group was significantly reduced. Reproduction in the 7.7×10^6, 1.6×10^7 and 3.2×10^7 CFU/L treatment groups was also reduced but not analyzed statistically due to a significant effect on survival. - It is not obvious whether daphnids died as a result of toxicity, or pathogenicity due to a lack of sterile filtrate and infectivity controls. **ACCEPTABLE**	1188035

Organism	Exposure	Test Substance(s)	Endpoint Value	Significant Effects, Comments	Reference
Plants					
Green Alga (*Scenedesmus subspicatus*)	Aqueous	i. *Gliocladium catenulatum* cell mass (technical product) ii. dilution water (negative control)	LC_{50} > 100 mg/L NOEC 100 mg/L	- No significant inhibitory effects observed at concentrations. - Significant increases in algal growth were observed in replicates treated with *G. catenulatum* cell mass, which was likely due to nutrients present in the test material. **ACCEPTABLE**	1188057
Aquatic Plants	Acute	A waiver was requested based on data and extensive literature searches. Few adverse effects were noted in some terrestrial plants following exposure to large concentrations of *G. catenulatum* strain J1446, and no adverse effects were observed in an algal inhibition study at a limit concentration of 100 mg/L. Extensive literature searches using several comprehensive databases found few reports of adverse effects. This microorganism can survive in aquatic ecosystems for a significant amount of time, but the use of Prestop Biofungicide WP in greenhouses is likely to limit direct exposure to non-target aquatic plant. Since exposure can occur from surface water runoff from spent growing medium and treated plants, growers must not allow effluent or runoff from greenhouses containing this product to enter lakes, streams, ponds or other water bodies. Handlers are also required not to contaminate aquatic habitats through equipment cleaning or waste disposal. **WAIVER ACCEPTED**			1188056

References

A. LIST OF STUDIES/INFORMATION SUBMITTED BY REGISTRANT

1.0 The Active Ingredient, Its Properties and Uses

PMRA 1187813 1997, History of the strain *Gliocladium catenulatum* J1446., DACO: M1.2, M2.7.1

PMRA 1187814 *Gliocladium* Corda 1840., DACO: M1.2, M2.7.1

PMRA 1187815 1996, Report of identification of the strain J1446., det 225 (1996), DACO: M1.2, M2.7.1

PMRA 1187816 2000, Identification of the strain J1446., DACO: M1.2, M2.7.1

PMRA 1187817 2000, Description of *Gliocladium catenulatum* J1446, DACO: M1.2, M2.7.1

PMRA 1187818 Paavanen-Huhtala, S. et al, 1999, Development of strain-specific primers for a strain of *Gliocladium catenulatum* used in biological control., European Journal of Plant Pathology 106: 187-198, 2000., DACO: M1.2, M2.7.1

PMRA 1187819 2000, Identification of J1446 strain by RAPD-PCR, UP-PCR and by specific primers., DACO: M1.2, M2.7.1

PMRA 1187820 A discussion on the relationship of *Gliocladium catenulatum* J1446 to known pathogenic / toxigenic strains or species., 152-1., DACO: M1.2, M2.7.2

PMRA 1187821 1996, The role of antibiosis, mycoparasitism and lytic enzymes in the control efficacy of *Gliocladium catenulatum* strain J1446., DACO: M1.2, M2.7.2

2.0 Methods of Analysis

PMRA 1187764 2003, Material safety data sheet. Prestop WP, DACO: 0.9,M2.9.1

PMRA 1187932 2006, Prestop WP. Comprehensive Data Summary, DACO: M12.7

PMRA 1187933 Registrant's name and address, DACO: M2.1

PMRA 1187934 Name and address of manufacturing plant, DACO: M2.2

PMRA 1187935 Name and address of formulating plant (Prestop (EP), DACO: M2.3

PMRA 1187936 Trade name: Prestop, DACO: M2.4

PMRA 1187937 Binominal name (MPCA), DACO: M2.5

PMRA 1187938 Canadian patent status information, DACO: M2.6

PMRA 1187939 2006, *Gliocladium catenulatum* J1446: Summary of the characterization of the MPCA., DACO: M2.7.1, M2.7.2

PMRA 1187941 2003, Summary of available information on the taxonomy of *Gliocladium catenulatum* J1446., DACO: M2.7.1

PMRA 1187942 2004, Maintenance and subculturing of microbial strain used in *Gliocladium catenulatum* - formulations., SOP-137-1, DACO: M2.7.1

PMRA 1187943 1996, Identification of J1446 strain by RAPD-PCR, UP-PCR and by specific primers., DACO: M2.7.1

PMRA 1187944 1999, Current status of taxonomy of *Trichoderma* and *Gliocladium*. 6th International *Trichoderma* and *Gliocladium* Workshop, Espoo, Finland. 11-13.6 1999., DACO: M2.7.1

PMRA 1187945 1994, Identification of the microorganism. Accession number given by the International Depositary Authority: DSM 9212. Budapest Treaty on the International Recognition of the Deposit of Microorganisms for the Purpose of Patent Procedure., DACO: M2.7.1

PMRA 1187946 Yli-Mattila, T. et al, 1997, Molecular identification of *Gliocladium* and *Nectria* isolates in Finland. Fifth European Fusarium Seminar, Szeged, Hungary Cereal Research Communications, Vol. 25 No. 3/1, pp. 293-297., Fifth European Fusarium Seminar, DACO: M2

PMRA 1187947 Park, J. et al, 1999, Phylogenetic analysis of the genus *Gliocladium* and its related taxa by comparing the sequences of internal transcribed spacers and 5.8S r-DNA., The Korean Journal of Mycology, Vol. 27 No 3, pp. 191-197., DACO: M2.7.1

PMRA 1187948 1996, Production of chitinase (chitobiosidase) by *Gliocladium* strain J1446., DACO: M2.7.2

PMRA 1187949 1996, Production of ß-1,3-glucanase (laminarinase) by *Gliocladium* strain J1446., DACO: M2.7.2

PMRA 1187950 1995, Study of the minimum and maximum temperatures for the growth of *Gliocladium catenulatum* strain J1446., DACO: M2.7.2

PMRA 1187951 1997, Effect of temperature on the control efficacy of *Gliocladium catenulatum* J1446., kas 12-94, DACO: M2.7.2

PMRA 1187952 1997, Effect of pH on the control efficacy of *Gliocladium catenulatum* J1446: cauliflower/*Rhizoctonia*., Rhil14-94, DACO: M2.7.2

PMRA 1187954 1997, *Gliocladium catenulatum* J1446: cauliflower/*Alternaria* I., 3-93, DACO: M2.7.2

PMRA 1187955 1997, Effect of pH on the control efficacy of *Gliocladium catenulatum* J1446: cauliflower/*Alternaria* II., 5-93., DACO: M2.7.2

PMRA 1187956 1998, The persistence of *Gliocladium catenulatum* J1446 on leaf surface., 16-98, 24-98 and 25-98, DACO: M2.7.2

PMRA 1187957 1998, Survival of *Gliocladium catenulatum* J1446 on cucumber leaves., 060698JO, DACO: M2.7.2

PMRA 1187958 2004, Literature search *Gliocladium* (s) (phytotox? or pathog?)., 060698JO, DACO: M2.7.2

PMRA 1187959 2006, Mycology of dermatophyte infections. Dermnet NZ. http://dermnetnz.org/fungak/mycology.html, DACO: M2.7.2

PMRA 1187960 Classification based on 9th edition of the Dictionary of the Fungi. CABI Bioscience Databases. http://www.indexfungorum.org/Names/fundic.asp, DACO: M2.7.2

PMRA 1187961 2004, Summary of the literature searches on the production of gliotoxin and/or genotoxins by *Gliocladium catenulatum*., DACO: M2.7.2

PMRA 1187962 2000, Summary of the literature search on production of toxins/metabolites by *Gliocladium catenulatum*., DACO: M2.7.2

PMRA 1187963 2001, Analysis of gliotoxins in culture broth., Bel 78/2001, DACO: M2.7.2

PMRA 1187964 2001, Analysis of gliotoxins in unformulated cell mass., Bel 103/2001, DACO: M2.7.2

PMRA 1187965 2001, Analysis of gliotoxins in mineral wool cultivation pot samples., Bel 184/2001, DACO: M2.7.2

PMRA 1187966 2000, Growing of *Gliocladium* treated cucumber seedlings in rockwool., 29-00, DACO: M2.7.2

PMRA 1187967 1995, Cytotoxicity test on *Gliocladium catenulatum* J1446 product 53/94., 1608/TT., DACO: M2.7.2

PMRA 1187969 1996, Cytotoxicity test on *Gliocladium catenulatum* J1446 cell mass batches 37/95 and 44/95., 1661/TT., DACO: M2.7.2

PMRA 1187970 1996, Cytotoxicity test on *Gliocladium catenulatum* J1446 formulation batch 41/95., DACO: M2.7.2

PMRA 1187971 Pasanen, A. et al, 1992, Laboratory experiments on membrane filter sampling of airborne mycotoxins produced by *Stachybotrys atra* Corda. Atmospheric Environment, Vol. 27A, No. 1: 9-13., Atmosphere Environment Vol. 22A, No. 1 pp. 9-12, 1993, DACO: M2.7.2

PMRA 1187972 2000, *Gliocladium catenulatum* J1446/ cell toxicity test., DACO: M2.7.2

PMRA 1187973 2003, Summary of available information on the secondary metabolite profile of *Gliocladium catenulatum* J1446., DACO: M2.7.2

PMRA 1187974 2000, Medical certificate., DACO: M2.7.2

PMRA 1187975 2005, Medical certificate., DACO: M2.7.2

PMRA 1187976 2006, *Gliocladium catenulatum* J1446: Summary of the Dacos M2.8-M2.12., DACO: M2.10, M2.11, M2.12, M2.8, M2.9

PMRA 1187977 2004, Quality control of Prestop WP production., SOP- 105-2., DACO: M2.8

PMRA 1187978 2004, Production of Prestop WP., SOP-106-2., DACO: M2.8

PMRA 1187979 2004, Production of *Gliocladium catenulatum* Technical Grade Active Ingredient (TGAI)., SOP- 136-1, DACO: M2.8

PMRA 1187980 2004, Product Specification Prestop, DACO: M2.9.1, M2.9.2

PMRA 1187981 2004, Five batch analysis of Prestop., REP-102-2, DACO: M2.9.2

PMRA 1187983 2004, Correlation between the viability and the control efficacy of Prestop WP., Based on kas-40-01, DACO: M2.9.2

PMRA 1187984 2005, Discussion of formation of unintentional ingredients Prestop WP - *Gliocladium catenulatum*, biofungicide sprayable formulation., REP-104-2, DACO: M2.9.3

PMRA 1187985 1998, The isolation of *Gliocladium catenulatum* J1446 from roots and growth substrates., REP-104-2, DACO: M2.10.1

PMRA 1187986 2004, Viability determination of biological pesticides., DACO: M2.10.1

PMRA 1187987 2004, Analysis of pathogenic contaminants in Prestop WP., REP-130-1, DACO: M2.10.2

PMRA 1496779 2006, Analyysitodistus (Analysis Certificate), 2006-00946-001, MRID: n.a., DACO: M2.10.2

PMRA 1496780 2006, Analyysitodistus (Analysis certificate), 2006-00945-001, MRID: n.a., DACO: M2.10.2

PMRA 1187988 2005, Storage stability of Prestop WP., REP-108-3, DACO: M2.11

PMRA 1187989 2006, Tap density of Prestop WP., REP-168-1, DACO: M2.12

PMRA 1187990 2006, *Gliocladium catenulatum* biofungicide (Prestop WP): Determination of long-term storage stability and physico-chemical characteristics., 1841/0009, DACO: M2.11

PMRA 1188054 2004, Summary of the literature searches on the phytotoxicity and/or pathogenicity of Gliocladium catenulatum., DACO: M2.7.2,M9.8.1

PMRA 1188055 2000, Summary of the literature search on Gliocladium catenulatum regarding pathogenicity or other interactions with plants, DACO: M2.7.2,M9.8.1

PMRA 1465323 2005, Quality control in Prestop production facilities, SOP-3.2, MRID: N/A, DACO: M2.9.3

PMRA 1465324 1999, Isolation and structure determination of TMC-151s: Novel polyketide antibiotics from Gliocladium catenulatum Gilman & Abbot TC 1280, N/A, MRID: N/A, DACO: M2.7.2

PMRA 1465325 Wong, S.-M. et al., 1993, Anthrotainin, an inhibitor of substance P binding produced by Gliocladium catenulatum, N/A, MRID: N/A, DACO: M2.7.2

PMRA 1465326 2007, Storage stability of Prestop WP, 108-4, MRID: N/A, DACO: M2.11

PMRA 1474679 2006, Specification of Prestop Mix, RE-179-1, DACO: M2.9.1

PMRA 1188046 2001, Discussion on analysis of gliotoxin in mineral wool; culture broth and unformulated cell mass samples., DACO: M9.7

3.0 Impact on Human and Animal Health

PMRA 1187767 U.S. EPA, Registration Eligibility Decision *Gliocladium catenulatum* strain J1446 (PC Code 021009)., DACO: 12.5.4

PMRA 1187768 U.S. EPA, US EPA Pesticide Fact Sheet: *Gliocladium catenulatum* strain J1446, DACO: 12.5.4

PMRA 1187822 1998, The spread and persistence of *Gliocladium catenulatum* J1446 in the air. Part I., 20/98, DACO: M1.2,M5.0,M8.3

PMRA 1187823 1998, The spread and persistence of *Gliocladium catenulatum* J1446 in the air. Part II., 24/98 and 26/98, DACO: M1.2,M5.0,M8.3

PMRA 1187824 1998, The spread and persistence of *Gliocladium catenulatum* J1446 in the air. Part III., 28/98, DACO: M1.2,M5.0,M8.3

PMRA 1187991 2005, Summary of Human Health and Safety Testing., DACO: M4.1

PMRA 1187992 2006, *Gliocladium catenulatum* J1446: Summary of the infectivity and toxicity studies., DACO: M4.2.1

PMRA 1187993 1996, *Gliocladium catenulatum* cell mass: Acute oral toxicity and pathogenicity to the rat., RKY 135, DACO: M4.2.2

PMRA 1187994 1996, *Gliocladium* (J1446) preparation: Acute oral toxicity (limit) test in rats., 556576; 10919, DACO: M4.2.2

PMRA 1187996 1996, *Gliocladium catenulatum* cell mass: Acute pulmonary toxicity and pathogenicity to rat., RKY 137, DACO: M4.2.3

PMRA 1187997 1996, *Gliocladium* (J1446) preparation: Acute inhalation toxicity study in rats., 654570;10916, DACO: M4.2.3

PMRA 1187998 2006, *Gliocladium catenulatum* J1446: Summary of the acute infectivity study (IP), DACO: M4.3.1

PMRA 1187999 1996, *Gliocladium catenulatum* cell mass: Acute intra-peritoneal toxicity and pathogenicity to the rat, RKY 136, DACO: M4.3.3

PMRA 1188000 2004, Data waiver for the acute dermal toxicity study on Prestop., DACO: M4.4

PMRA 1188001 1996, *Gliocladium* (J1446) preparation: Acute dermal toxicity (limit) test in rats., 556576;10920, DACO: M4.4

PMRA 1188002 2002, Safety Data Sheet. Valio Skimmed Milk Powder Medium Heat, DACO: 0.9.1,M2.9.1,M4.4

PMRA 1188003 1999, Material safety data sheet. C-Sorbidex S 16603, DACO: 0.9.1,M2.9.1,M4.4

PMRA 1188004 2006, *Gliocladium catenulatum* J1446: Summary of the dermal irritation study., DACO: M4.5.1

PMRA 1188005 2004, Data waiver for a dermal irritation study on Prestop., DACO: M4.5.2

PMRA 1188006 1996, *Gliocladium* (J1446) preparation: Acute dermal irritation test in rabbits., 556576;10921, DACO: M4.5.2

PMRA 1188007 2006, *Gliocladium catenulatum* J1446: Summary of the skin sensitization study, DACO: M4.6

PMRA 1188009 2004, Prestop Mix: *Gliocladium catenulatum* J1446 biofungicide hypersensitivity reporting., DACO: M4.6

PMRA 1188011 2004, Prestop WP: *Gliocladium catenulatum* J1446 biofungicide hypersensitivity reporting., DACO: M4.6

PMRA 1188012 1996, *Gliocladium* (J1446) preparation: Buehler sensitisation test in guinea pigs., 556576;10923, DACO: M4.6

PMRA 1188013 1999, *Gliocladium* J1446 biopesticide: Acute eye irritation test in the rabbit., 245/187, DACO: M4.9

PMRA 1188014 1996, *Gliocladium* (J1446) preparation: Acute eye irritation test in rabbits., 556576;10922, DACO: M4.9

PMRA 1188015 2004, Assessment of operator, bystander and worker exposure to Prestop Mix (*Gliocladium catenulatum*)., DACO: M5.0

PMRA 1188016 1997, Viable residues of *Gliocladium catenulatum* on cucumber fruits., 11/97, DACO: M7.0

PMRA 1188017 2006, Data waiver for the food and feed residue studies., DACO: M7.0

PMRA 1188018 1997, Viable residues of *Gliocladium catenulatum* J1446 on cucumber fruits one day after the treatment., 11/97, DACO: M7.0

PMRA 1187967 1995, Cytotoxicity test on *Gliocladium catenulatum* J1446 product 53/94., 1608/TT., DACO: M2.7.2

PMRA 1187969 1996, Cytotoxicity test on *Gliocladium catenulatum* J1446 cell mass batches 37/95 and 44/95., 1661/TT., DACO: M2.7.2

PMRA 1187970 1996, Cytotoxicity test on *Gliocladium catenulatum* J1446 formulation batch 41/95., DACO: M2.7.2

PMRA 1187971 Pasanen, A. et al, 1992, Laboratory experiments on membrane filter sampling of airborne mycotoxins produced by *Stachybotrys atra* Corda. Atmospheric Environment, Vol. 27A, No. 1: 9-13., Atmosphere Environment Vol. 22A, No. 1 pp. 9-12, 1993, DACO: M2.7.2

4.0 Impact on the Environment

PMRA #	Reference
1187948	1996, Production of chitinase (chitobiosidase) by *Gliocladium* strain J1446., DACO: M2.7.2
1187949	1996, Production of β-1,3-glucanase (laminarinase) by *Gliocladium* strain J1446., DACO: M2.7.2
1187950	1995, Study of the minimum and maximum temperatures for the growth of *Gliocladium catenulatum* strain J1446., DACO: M2.7.2
1187966	2000, Growing of *Gliocladium* treated cucumber seedlings in rockwool., 29-00, DACO: M2.7.2
1188020	2005, Summary on the environmental fate of *Gliocladium catenulatum* J1446, DACO: M8.1
1188021	1997, The survival of *Gliocladium catenulatum* J1446 in water., Lab-48-96, DACO: M8.2.2

1188022 1998, Survival of *Gliocladium catenulatum* J1446 on cucumber leaves., 060698Jo., DACO: M8.3

1188023 1998, The persistence of *Gliocladium catenulatum* J1446 on leaf surface., 16-98, 24-98 and 25-98., DACO: M8.3

1188024 1996, Confidential report on the survival of J1446 in a commercial peat-based growing medium for Kemira Agro Oy., DACO: M8.3

1188025 2001, Possible mobility of the conidia into the soil., DACO: M8.5

1188026 2006, *Gliocladium catenulatum* J1446 Summary of the environmental toxicity studies., DACO: M9.1

1188027 1997, *Gliocladium catenulatum* cell mass: an avian oral pathogenicity and toxicity study in the northern bobwhite., 293-109, DACO: M9.2.1

1188028 2004, Response to PMRA data evaluation for *Gliocladium catenulatum* cell mass (technical product): a five-concentration toxicity and pathogenicity test with the rainbow trout (*Oncorhynchus mykiss*)., 293A-104, DACO: M9.4.1

1188029 1998, *Gliocladium catenulatum* cell mass (technical product): a five-concentration toxicity and pathogenicity test with the rainbow trout (*Oncorhynchus mykiss*)., 293A-104, DACO: M9.4.1

1188031 1998, *Gliocladium catenulatum* cell mass (technical product): a dietary pathogenicity and toxicity study with the honey bee (*Apis mellifera*)., 293-110, DACO: M9.5.1

1188032 1998, *Gliocladium catenulatum* cell mass (technical product): a dietary pathogenicity and toxicity study with the ladybird beetle (*Hippodamia convergens*)., 293-111, DACO: M9.5.1

1188033 1998, *Gliocladium catenulatum* cell mass (technical product): a dietary pathogenicity and toxicity study with green lacewing larvae (*Chrysoperla carnea*)., 293-113A, DACO: M9.5.1

1188034 1998, *Gliocladium catenulatum* cell mass (technical product): a dietary pathogenicity and toxicity study with parasitic hymenoptera (*Nasonia vitripennis*)., 293-112, DACO: M9.5.1

1188035 1998, *Gliocladium catenulatum* cell mass (technical product): a 21-day life-cycle toxicity and pathogenicity test with the cladoceran (*Daphnia magna*)., 293A-103, DACO: M9.5.2

1188036 1997, *Gliocladium catenulatum* cell mass: a pathogenicity study with the earthworm in an artificial soil substrate., 293A-105, DACO: M9.6

1188037 1997, Effect of *Gliocladium catenulatum* J1446 biofungicide powder on other soil microbes., DACO: M9.7

1188038 1998, Effect of *Gliocladium catenulatum* J1446 incorporation on peat microflora., 061797JO, DACO: M9.7

1188039 2001, Analysis of gliotoxin in culture broth., BEL78/2001, DACO: M9.7

1188040 2001, Analysis of gliotoxin in unformulated cell mass, BEL103/2001, DACO: M9.7

1188042 2001, Analysis of gliotoxin in mineral wool cultivation in pot samples., BEL184/2001, DACO: M9.7

1188043 1996, The role of antibiosis, mycoparasitism and lytic enzymes in the control efficacy of *Gliocladium catenulatum* strain 1446., DACO: M9.7

1188044 2003, Summary of available information on the secondary metabolite profile of *Gliocladium catenulatum* J1446, DACO: M9.7

1188046 2001, Discussion on analysis of gliotoxin in mineral wool; culture broth and unformulated cell mass samples., DACO: M9.7

1188047 Bioassay of gliotoxin in *Gliocladium* extracts based on *Escherichia coli* CM871., DACO: M9.7

1188049 2004, Literature searches conducted in 2000 and 2004 on the pathogenicity or phytotoxicity of *Gliocladium catenulatum*., DACO: M9.8.1

1188050 2005, Data waiver for terrestrial plant toxicity/pathogenicity study., DACO: M9.8.1

1188052 1997, Effect of high dosages of *Gliocladium catenulatum* J1446 on different crops., DACO: M9.8.1

1188054 2004, Summary of the literature searches on the phytotoxicity and/or pathogenicity of *Gliocladium catenulatum*., DACO: M2.7.2,M9.8.1

1188055 2000, Summary of the literature search on *Gliocladium catenulatum* regarding pathogenicity or other interactions with plants, DACO: M2.7.2,M9.8.1

1188056 2005, Data waiver for the aquatic plant infectivity study., DACO: M9.8.2

1188057 2001, *Gliocladium catenulatum* cell mass (technical product): Algal inhibition test., 245/205, DACO: M9.8.2

5.0 Value

PMRA 1187842 1998. The effect of *Gliocladium catenulatum* J1446 on grey mold (*Botrytis cinerea*) on cucumber leaves. DACO M10.2.1

PMRA 1187843 1998. The effect of *Gliocladium catenulatum* J1446 on grey mold (*Botrytis cinerea*) on pepper leaves. DACO M10.2.1

PMRA 1187844 1998. The effect of *Gliocladium catenulatum* J1446 fungal suspension on grey mold (*Botrytis cinerea*) on pepper leaves. DACO M10.2.1

PMRA 1187845 1998. Control of bottom rot on lettuce caused by *Rhizoctonia solani*. DACO M10.2.1

PMRA 1187846 2002. Biological control agents and composts suppress Fusarium and Pythium root rots on greenhouse cucumbers. DACO M10.2.1, M10.2.2

PMRA 1187847 2003. Biological control of damping-off and root rot caused by *Pythium aphanidermatum* on greenhouse cucumbers. DACO M10.2.1, M10.2.2

PMRA 1187848 2003. Efficacy of biological and chemical treatments for control of *Fusarium* root and stem rot on greenhouse cucumber. DACO M10.2.1, M10.2.2

PMRA 1187849 2006. Efficacy of Prestop WP (*Gliocladium catenulatum* J1446) for the control of *Fusarium solani* in greenhouse grown peppers. DACO M10.2.2

PMRA 1187851 1998. The effect of *Gliocladium catenulatum* J1446 on *Botrytis cinerea* on tomato plants. DACO M10.2.2

PMRA 1187852 1998. The effect of *Gliocladium catenulatum* J1446 fungal suspension on grey mold (*Botrytis cinerea*) on pepper plants. DACO M10.2.2

PMRA 1187853 1998. *Gliocladium catenulatum* J1446 on the cuttings of Fuchsia. DACO M10.2.2

PMRA 1187854 1998. The effect of *Gliocladium catenulatum* J1446 on grey mold (*Botrytis cinerea*) on tomato plants. DACO M10.2.1

PMRA 1187855 1997. The effect of *Gliocladium catenulatum* J1446 on grey mold (*Botrytis cinerea*) on cucumber leaves. DACO M10.2.1

PMRA 1187856 1994. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in cucumber. DACO M10.2.2

PMRA 1187857 1994. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in cucumber. DACO M10.2.2

PMRA 1187858 1994. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in cucumber. DACO M10.2.2

PMRA 1187859 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium in lettuce. DACO M10.2.2

PMRA 1187860 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium in lettuce. DACO M10.2.2

PMRA 1187861 1995. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium in lettuce. DACO M10.2.2

PMRA 1187862 1995. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium in lettuce. DACO M10.2.2

PMRA 1187863 1995. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium in lettuce. DACO M10.2.2

PMRA 1187864 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in oregano. DACO M10.2.2

PMRA 1187865 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in basilica. DACO M10.2.2

PMRA 1187867 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in parsley. DACO M10.2.2

PMRA 1187868 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in thyme. DACO M10.2.2

PMRA 1187869 1995. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Pythium damping-off in oregano. DACO M10.2.2

PMRA 1187870 1997. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Rhizoctonia damping-off in cauliflower. DACO M10.2.2

PMRA 1187871 1994. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Rhizoctonia damping-off in cauliflower. DACO M10.2.2

PMRA 1187872 1996. The effect of *Gliocladium catenulatum* J1446 biofungicide powder on Rhizoctonia damping-off in cauliflower. DACO M10.2.2

PMRA 1187873 1998. The effect of *Gliocladium catenulatum* J1446 fungal suspension on grey mold (*Botrytis cinerea*) on pepper plants. DACO M10.2.2

PMRA 1187874 1998. *Gliocladium catenulatum* J1446 on the cuttings of Fuchsia. DACO M10.2.2

PMRA 1187875 Use of Prestop mix for control of Pythium root rot in poinsettia. DACO M10.2.2

PMRA 1187876 2000. Microbial products for prevention of poinsettia root rot diseases. IR-4 report. DACO M10.2.2

PMRA 1187877 Prestop and Fusarium crown and root rot of cucumber. DACO M10.2.2

PMRA 1187878 Control of bottom rot of lettuce caused by *Rhizoctonia solani*. DACO M10.2.1

PMRA 1187879 1998. The effect of *Gliocladium catenulatum* J1446 on *Botrytis cinerea* on tomato plants. DACO M10.2.2

PMRA 1187880 1998. The effect of *Gliocladium catenulatum* J1446 fungal suspension on grey mold (*Botrytis cinerea*) on pepper plants. DACO M10.2.2

PMRA 1187881 1997. The effect of *Gliocladium catenulatum* J1446 against damping-off of cucumber on rockwool. DACO M10.2.2

PMRA 1187883 1996. The effect of *Gliocladium catenulatum* J1446 against damping-off of cucumber on rockwool. DACO M10.2.2

PMRA 1187885 1997. The effect of *Gliocladium catenulatum* J1446 on *Pythium ultimum* with normal and sub irrigation. DACO M10.2.2

PMRA 1187886 1997. The use of *Gliocladium catenulatum* J1446 biofungicide powder in the control of *Phytophthora* in poinsettia. DACO M10.2.2

PMRA 1187887 1998. Control of *Didymella* by *Gliocladium catenulatum* J1446 in commercial cultivation. DACO M10.2.2

PMRA 1187888 1998. The effect of *Gliocladium catenulatum* J1446 on *Didymella* of cucumber. DACO M10.2.2

PMRA 1187889 1998. The effect of *Gliocladium catenulatum* J1446 on *Didymella* of cucumber. DACO M10.2.2

PMRA 1187890 1998. The effect of *Gliocladium catenulatum* J1446 on *Didymella* of cucumber. DACO M10.2.2

PMRA 1187891 1995. Rhizoctonia damping-off of alyssum and salvia. Pythium damping-off of pansy, tomato and snapdragon. Confidential report on the evaluation of J1446 for Kemira Agro Oy. DACO M10.2.2

PMRA 1187892 1996. Phytophthora root and basal stem rot of petunia and tagetes. DACO M10.2.2

PMRA 1187893 1996. Phytophthora root and basal stem rot of petunia. DACO M10.2.2

PMRA 1187894 2001. Control of soil borne diseases of greenhouse tomato. DACO M10.2.2

PMRA 1187895 2002. Trial against *Pythium ultimum* on basil (cv *Genovese gigante*). DACO M10.2.2

PMRA 1187896 2002. Trial against *Fusarium oxysporum* f. sp. *basilici* on basil (cv *Genovese gigante*). DACO M10.2.2

PMRA 1187898 2002. Trial against *Fusarium oxysporum* f. sp. *basilici* on basil (cv *Fine verde*). DACO M10.2.2

PMRA 1187899 2005. Comparing the efficacy of the two Prestop formulations: Prestop and Prestop mix. DACO M10.2.2

PMRA 1187900 2006. The effect of Prestop WP biofungicide on basil artificially inoculated with *Pythium.* DACO M10.2.2

PMRA 1187901 2004. The effect of Prestop WP biofungicide on saintpaulia artificially inoculated with *Pythium.* DACO M10.2.2

PMRA 1187902 2004. The effect of Prestop WP biofungicide on geranium artificially inoculated with *Pythium.* DACO M10.2.2

PMRA 1187903 2003. Mycostop and Prestop WP on sweet pepper in Finland. DACO M10.2.2

PMRA 1187904 1998. Control of Didymella by *Gliocladium catenulatum* J1446 in commercial cultivation. DACO M10.2.2

PMRA 1187905 1998. The effect of *Gliocladium catenulatum* J1446 on *Didymella* of cucumber. DACO M10.2.2

PMRA 1187906 1998. The effect of *Gliocladium catenulatum* J1446 on *Didymella* of cucumber. DACO M10.2.2

PMRA 1187907 2002. Botrytis stem canker of greenhouse tomato managing the disease through cultural, biological and chemical controls. DACO M10.2.2

PMRA 1187908 Evaluation of Prestop for control of gummy stem blight on naturally infected cucumber plants in a commercial greenhouse. DACO M10.2.2

PMRA 1187909 2002. Evaluation of biological and chemical treatments for control of gummy stem blight on cucumber plants grown hydroponically in greenhouses. Pacific Agrifood Research Centre, Agriculture and Agrifood Canada Biocontrol 49:109-117. DACO M10.2.2

PMRA 1187910 2001. Biological control of stem canker of greenhouse tomatoes caused by *Botrytis cinerea* Can. J. Microbiol. 48:550-554. DACO M10.2.2

PMRA 1187911 2005. Effect of *Gliocladium catenulatum* J1446 on lettuce at a high dose. DACO M10.3.1

PMRA 1187913 1997. Effect of high doses *of Gliocladium catenulatum* on different crops. DACO M10.3.1

PMRA 1187931 2006. Efficacy of Prestop WP strain J1446 for the control of *Fusarium solani* in greenhouse grown peppers. . DACO M10.3.1