Appendix to:
EFSA (European Food Safety Authority), 2019. Conclusion on the peer review of the pesticide risk assessment of the active substance *Phlebiopsis gigantea* strains VRA 1835, VRA 1984 VRA 1984 and FOC PG 410.3. EFSA Journal 2019;17(10):5820, 14 pp. doi:10.2903/j.efsa.2019.5820
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**Appendix A – List of end points for the active substance and the representative formulation**

**FORMAT FOR THE LISTING OF END POINTS FOR A MICROBIAL OR VIRAL PEST CONTROL AGENT (MPCA) USED IN PLANT PROTECTION**

**General remark:**
Testing of microorganisms will often be made using specifically tailored studies. Therefore, e.g. toxicity/effects endpoints may differ from case to case. This endpoint list can therefore be seen as indicative only, to be adapted in order to fit individual cases.

**Identity, Biological properties, Details of uses, Further information, and Proposed Classification and Labelling**

| Active microorganism: | *Phlebiopsis gigantea* |
|-----------------------|------------------------|
| Function (*e.g. control of fungi*): | Forestry biofungicide for the control of root and butt rot in coniferous tree species caused by *Heterobasidion* spp. (*H. annosum, H. parviporum*). |

**Rapporteur Member State:** Estonia

**Co-rapporteur Member State:** France

**Identity of the Microbial or Viral Agent used in plant protection / Active Substance** *(Regulation (EU) N° 283/2013, Annex Part B, point 1; OECD IIM Point 1)*

| Name of the organism: | *Phlebiopsis gigantea* (synonyms *Phlebia gigantea* (Fr) Donk, *Peniophora gigantea* (Fr.) Masssee, *Phanerochaete gigantea* (Fr.;Fr.) Rattan *et al.* |
|-----------------------|----------------------------------------------------------------------------------------------------------------------------------|

**Taxonomy:**
Phylum: Basidiomycota  
Class: Basidiomycetes  
Subclass: Agaricomycetidae  
Order: Polyporales  
Family: Phanerochaetaceae  
Genus: *Phlebiopsis*  
Primary colonizer of fresh conifer wood, common in boreal and temperate forests throughout the world. Causes a typical white rot of coniferous wood.

| Species, subspecies, strain: | Species: *Phlebiopsis gigantea* |
|-----------------------------|--------------------------------|
| 3 strains:  | VRA 1835 |
### Peer review of the pesticide risk assessment of the active substance *Phlebiopsis gigantea* strains VRA 1835, 1984 and FOC PG 410.3

| VRA 1984  
FOC PG 410.3 | Identification / detection: Easily identified as *P. gigantea* through its growth characteristics - growth rate, microscopic appearance of mycelium and conidial structures, enzyme activity etc. Molecular identification of specific strains can be done by microsatellite genotyping with a method developed to differentiate *P. gigantea* to strain level. |
The strains are held within ATCC, DSMZ and IMI collections. |
| Minimum and maximum concentration of the MPCA used for manufacturing of the formulated product (cfu; g/kg): | 2 \( \times \) \( 10^7 \) to 5 \( \times \) \( 10^{10} \) CFU/kg for the representative formulation Rotstop (WP) (average 5 \( \times \) \( 10^{6} \) CFU/g) 10.6 % (w/w) |
| Identity and content of relevant impurities, additives, contaminating organisms in the technical grade of MPCA: | Not relevant – the ‘Technical Grade’ of the MPCA is a hypothetical stage in a continuous production process of end-use products with a strain of *P. gigantea* as active substance. |
| Is the MPCA genetically modified; if so provide type of modification | Wild strains, unmodified |
### Biological properties of the microorganism (Regulation (EU) No 283/2013, Annex Part B, point 2; OECD IIM Point 2)

| Origin and natural occurrence: | *Phlebiopsis gigantea* is a common and widely distributed saprophytic wood-decay fungus found throughout the coniferous forests of the temperate Northern Hemisphere. It has also been recorded in southern Europe, East Africa, Central America, Australia and New Zealand. Based on morphology and interfertility, all European populations are regarded as a single taxonomic species throughout its geographical distribution. Colonies of *P. gigantea* are naturally formed on untreated coniferous stumps in thinnings, where the fungus contributes to stump decomposition. Fruit bodies produce basidiospores which are abundant in the air, especially during the summer months. The supported strains have been isolated from fruit bodies formed on stumps of spruce or pine in Finland, Sweden and UK, respectively. |
| Background level: | Ambient levels of around 10 spores 100cm⁻² hr⁻¹ are commonly found in coniferous woodland, exact levels depending on temperatures, wind speed, proximity to fruit bodies etc. |
| Target organism(s): | *Heterobasidion annosum* complex (*H. annosum s. lato*) – formerly *H. annosum* S, P and F-type, but now separated into subspecies: *H. annosum sensu stricto* (Fr.) Bref. (host species *Pinus* and many other species) *H. parviporum* Niemelä & Korhonen (host species mostly *Picea abies*) *H. abietinum* Niemelä & Korhonen (host species *Abies*). The pathogen has a wide distribution in the Northern Hemisphere. It causes heart rot, death of young trees, reduced increment, reduced pulping properties and increased risk of wind-throw. Recorded on more than 200 species of woody plants but particularly significant in coniferous forests including (amongst others) *Picea abies* and *P. sitchensis*, *Pinus sylvestris*, *P. cembra*, *P. nigra*, *P. pinaster*, *P. pinea* and *P. peuce* and *Abies alba*, *A. cephalonica*, *A. borisii-regis* (Southern Europe) and *Abies sibirica*. |
| Mode of action: | Efficient competition for substrate and living space on the fresh conifer stump surface and in the stump, based on the unique ability of *P. gigantea* to rapidly colonise freshly exposed conifer sapwood by degrading all compounds of the plant cell wall. Hyphal interference between *P. gigantea* and *H. annosum* has been shown in *vitro*, but is not considered to contribute to the control efficacy. Lack of an inhibition zone and formation of a mycelial barriage zone in dual plate culture indicate that the interaction between *P. gigantea* and *Heterobasidion* spp. is dead-lock or overgrowth rather than antibiosis or parasitism. Induced localised resistance in the host and involvement of molecules such as hydrophobins secreted by *P. gigantea* have also been suggested as mechanisms that further improve the control efficacy of *P. gigantea*. Freshly cut stumps, created in thinnings and at clear-fellings, are the main infection route of *H. annosum* into healthy tree stands. By applying sufficient amounts of spores of *P. gigantea* on the stump surface immediately after felling, the air-borne spores of the pathogen cannot infect the stump and from there spread to healthy living trees via root connections. |
| Host specificity: | Target organism - *P. gigantea* is a natural competitor of *H. annosum*, both occupying the same ecological niche. Generally speaking, colonisation of a stump by *P. gigantea* does not prevent the multiple colonisation of stumps by numerous other fungal species, although artificial inoculation |
through stump treatment may cause transient qualitative changes in species composition of the stump microflora, and sometimes have an effect on species richness.
Host species - *P. gigantea* colonises conifer stumps, especially *Pinus* and *Picea* species.

**Life cycle:**
In coniferous forests *P. gigantea* basidiospores are abundant in the air during the warm season, and can disperse very widely. Spores colonise stumps, fallen trunks and log piles, and characteristic fruit bodies normally form within a year after infection and begin releasing spores.
The mating system of the fungus is bipolar, i.e. the vegetative mycelium can be homo- or heterokaryotic. *In vitro* homokaryotic fruit bodies may form, but in the forest the fruit bodies are heterokaryotic.
In the vegetative mycelium chains of oidial (asexual) spores are formed by segmentation of the hyphae. The oidia can be dispersed e.g. with arthropods feeding on the mycelium. For an infection to develop, the oidial spore must be heterokaryotic, or hypha from two homokaryotic spores (basidiospores or oidia) must mate to become heterokaryotic.

**Infectivity, dispersal and colonisation ability:**
*P. gigantea* is highly adapted to survival on moribund wood and is a rapid coloniser of recently cut stumps, forming fruit bodies within a year. Basidiospores released by fruit bodies are highly mobile in air currents, and have been trapped up to 250 miles from likely sources of inoculum.
Oidia (asexual spores) are formed by segmentation of vegetative hyphae. They are not long-lived *in vivo* and have only limited ability for dispersal, although there is some evidence that they can be passively transferred between stumps by arthropods (e.g. bark beetles). When applied on stumps during stump treatment operations they rapidly colonise the woody substrate.

**Relationships to known plant, animal or human pathogens:**
No relationship to any known animal or human pathogens. Two distantly related plant pathogens are found within the same family (Phanerochaetaceae), but both occur in ecological niches and climatic conditions very different from those inhabited by *P. gigantea*.

**Genetic stability:**
*P. gigantea* acts against *H. annosum* via direct competition. The traits governing this, which are a combination of characters such as spore germination, growth rate and wood-colonising ability, are all under continuous, stable, polygenic genetic control, and are not controlled by only a few major genes. This means that these traits are not subject to breakdown or loss of action via mutation, which can be the case for traits controlled by one or a few major genes, and so the ability of *P. gigantea* to control *H. annosum* can be considered genetically stable.
In support of this, isolates of *P. gigantea* have been used within biofungicides for over a decade, with no discernible change in appearance or efficacy.

**Information on the production of relevant metabolites (especially toxins):**
Metabolites produced by *P. gigantea* strain VRA 1835: phlebiopsin A, B and C, glycosylated p-terphenyl and o-orsellinaldehyde
*P. gigantea* has not been reported to produce toxins. Genomic analysis has identified genes with diverse functions in fungal growth and morphologic development, including genes coding for hydrophobins and other genes with potential functional relevance during the competitive interaction between *P. gigantea* and *Heterobasidion* spp.
Secondary metabolites of *Phlebiopsis gigantea* strains VRA 1984 and FOC PG 410.3 need to be further identified and quantified, in particular hydrophobins for which a toxicological concern cannot be excluded. Once characterised, the toxicological relevance of the metabolites needs to be investigated (e.g. considering QSAR analysis where possible, natural background levels, published data, etc.)

**Resistance/sensitivity to**
Sensitivity to typical antibiotics used against dermatophytes was tested:
| antibiotics / anti-microbial agents used in human or veterinary medicine: | *P. gigantea* is sensitive, or highly sensitive, to a range of antifungal drugs used in the treatment against dermatophytes. |
Summary of uses supported by available data (Regulation (EU) N° 283/2013, Annex Part B, point 3; OECD IIM Point 3)

Details of representative uses

| Crop and/or situation | Member State or Country | Product name | F G or I | Pests or Group of pests controlled | Formulation | Application | Application rate per treatment | PHI (days) | Remarks: |
|-----------------------|-------------------------|--------------|---------|-----------------------------------|-------------|------------|-------------------------------|------------|---------|
| Conifer forests:      |                         | Rotstop F    | (a)     | (b)                               | WP          |            |                               |            |         |
| Pine (Pinus) Spruce (Picea), Larch (Larix) | Northern, Central and Southern EU zones | Heterobasidion spp.: (H. annosum, H. parviporum) | (c) | (d-f) | 5 x 10^9 CFU/kg, (106 g/kg) strain VRA 1835 | Mechanised or manual spraying of freshly cut stumps | First thinning to final cutting, all year at temp’s above 5 °C | Once per harvesting time | Minimum 10-15 years in the same stand | 106 mg/L | 1 L/m² stump surface in manual treatment, 2 L/m² stump surface in mechanised treatment equivalent to 8-16 L/ha in first thinnings and 34-68 L/ha in final cutting | Rotstop: 1 g/m² stump surface in manual treatment, 2 g/m² stump surface in mechanised treatment, equivalent to 8-16 g/ha in first thinnings and 34-68 g/ha in final cutting | NA | Spraying of the stump surface only, with minimized application around the stump |

(a) For crops, the EU and Codex classifications (both) should be used; where relevant, the use situation should be described (e.g. fumigation of a structure)
(b) Outdoor or field use (F), glasshouse application (G) or indoor application (I)
(c) e.g. biting and sucking insects, soil born insects, foliar fungi, weeds
(d) e.g. wettable powder (WP), emulsifiable concentrate (EC), granule (GR)
(e) CropLife International Technical Monograph no 2, 6th Edition.
(h) Kind, e.g. overall, broadcast, aerial spraying, row, individual plant, between the plant - type of equipment used must be indicated
(i) cfu = colony forming units and g/kg or g/L
(j) Growth stage at last treatment (BBCH Monograph, Growth Stages of Plants, 1997, Blackwell, ISBN 3-8263-3152-4), including where relevant, information on season at time of application
(k) Indicate the minimum and maximum number of applications possible under practical conditions of use
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Revised May 2008. Catalog of pesticide.

(f) All abbreviations used must be explained

(g) Method, e.g. high volume spraying, low volume spraying, spreading, dusting, drench

(l) PHI - minimum pre-harvest interval

(m) Remarks may include: Extent of use/economic importance/restrictions
Classification and proposed labelling (Symbol, Indication of danger, Risk phrases, Safety phrases)

| with regard to physical/chemical data: | No classification |
|--------------------------------------|-------------------|
| with regard to toxicological data:   | No classification. The following labelling phrase is proposed: “Micro-organisms may have the potential to provoke sensitising reactions”. |
| with regard to fate and behaviour:   | No classification |
| with regard to ecotoxicological data:| No classification |

**Methods of analysis** (Regulation (EU) N° 283/2013, Annex Part B, point 4 and Regulation (EU) N° 284/2013, Annex Part B, point 5)

**Analytical methods for the microorganism** (MA 4.1 & MP 5.1; OECD IIM 4.3 & IIIM 5.1)

| Manufactured microorganism (principle of method): | Standard microbiological methods (dilution, plate culturing and counting, microscopy) are used to isolate, identify and quantify the active ingredient and possible contaminants in the product, and the active ingredient in treated stumps and harvested timber, in soil, water and air and in animal body tissue and fluids. |
|--------------------------------------------------|---------------------------------------------------------------|
| Impurities and contaminating microorganisms in manufactured material (principle of method): | There are no harmful impurities originating from the raw materials used in product manufacture since they are of food grade. Standard dilution-plate counting methods or Most Probable Number (MPN) methods are used to detect contamination at various stages of the manufacturing process. |
| Microbial Pest Control Product (principle of method): | Specified viability and microbiological purity are the main quality criteria for the end-product: -viability is checked using a Most Probable Number (MPN) dilution-plate counting method. -storage stability is determined by analysing viability after storage at different temperatures - short-term storage stability at 28°C after 1 week and 1 month, and long-term storage stability at 4°C at regular intervals during 1 year; based on the data provided the shelf life is at least 6 months at -18°C and +4°C and two weeks at room temperature. -contaminating microbes present in samples taken at various stages of production are detected and quantified by using standard dilution-plate counting methods or MPN methods. Colonies appearing on the quality control agar plates are identified based on gross morphology of the colonies and with standard taxonomic identification methods. -ISO standard methods are used to detect pathogens in production batches at limits set according to SANCO/12116/2012 rev.0. -methodology exists for analysing the content of organic
material (spores and mycelium), inert formulants and water in the formulated end-product.

Analytical methods for residues (viable and non-viable) in exposed compartments and organisms (MA 4.2 & MP 5.2; OECD IIIM 4.5 & IIIM 5.2)

| of the active microorganism (principle of method): | P. gigantea is used only for stump treatment in coniferous forests, where it is already naturally abundant as spores in the air during the summer months and as fruit bodies on rotting wood. The fungus itself is non-toxic and no increased consumer exposure or risks due to stump treatment can be foreseen. Therefore, no methods for analysis of residues are needed. |
**Impact on Human and Animal Health** (Regulation (EU) No 283/2013, Annex Part B, point 5 and Regulation (EU) No 284/2013, Annex Part B, point 7)

| Medical data: (including medical surveillance on manufacturing plant personnel) (MA 5.1.1) | Limited data: no adverse effects were observed among researches, production workers and field technicians when appropriate protective equipment was used. No adverse effects have been observed in persons involved in research and development, production and use of *P. gigantea* products during 50 years. |
|---|---|
| Sensitisation: (MA 5.2.1 & MP 7.2.3) | Negative result in Buehler test (*P. gigantea* VRA 1835). (Buehler test is not acceptable.) |
| Acute oral infectivity, toxicity and pathogenicity: (MA 5.2.2.1 & MP 7.1.1) | Rat LD$_{50}$ > 4.26 x 10$^7$ CFU of *P. gigantea* VRA 1835/ kg bw Non infective Non pathogenic |
| Acute intratracheal/inhalation infectivity, toxicity and pathogenicity: (MA 5.2.2.2 & MP 7.1.2) | Rat LC$_{50}$ > 1.12 x 10$^6$ CFU of *P. gigantea* VRA 1835 /kg bw Non infective Non pathogenic Occurrence of treatment-related white nodules observed upon i.p. administration of *Phlebiopsis gigantea* on organs (liver, spleen, kidneys and intestines) is a potential concern justifying further the data gap set on secondary metabolites/toxins |
| Acute intravenous/intraperitoneal infectivity: (MA 5.2.2.3) | Rat LD$_{50}$ = 9.31x10$^4$ – 1.27x10$^5$ CFU of *P. gigantea* VRA 1835/animal Non infective Non pathogenic |
| Genotoxicity: (MA 5.2.3) | Open, pending on the identification and quantification of secondary metabolites/toxins |
| Cell culture study: (MA 5.2.4) | No data required |
| Information on short-term toxicity and pathogenicity: (MA 5.2.5) | No data required |
| Dermal toxicity: (MP 7.1.3) | Rat LD$_{50}$ > 2000 mg/kg bw |
| Specific toxicity, pathogenicity and infectivity: (MA 5.3) | No *in vitro* cytotoxicity was observed in liquid culture broth of *P. gigantea* strains VRA 1835, VRA 1984 and FOC PG 410.3. There were some indications of cytotoxicity in extracts of solid culture medium of *P. gigantea* VRA 1835 and VRA 1984, but due to interference of the test substance (solid growth medium) with the test method, it cannot unequivocally be concluded that *P. gigantea* has the potential to produce cytotoxic substances in solid culture. |
| Genotoxicity – *in vivo* studies in germ cells: (MA 5.5) | No studies provided. *In vivo* genotoxicity studies are not required based on the absence of concerns arising from the above tests. |

**Reference values**
AOEL: Not applicable since *Phlebiopsis gigantea* was not found to be toxic, infective or pathogenic.

Acute Acceptable Operator Exposure Level (AAOEL) Not applicable since *Phlebiopsis gigantea* was not found to be toxic, infective or pathogenic.

ADI: Not applicable since *Phlebiopsis gigantea* was not found to be toxic, infective or pathogenic.

ARfD: Not applicable since *Phlebiopsis gigantea* was not found to be toxic, infective or pathogenic.

**Exposure (operator, workers, bystander, consumer):**

(MA 6.1 & MP 7.3, 8.0) Based on the submitted data, risk evaluation was not possible. The sensitising potential of Rotstop suggests that exposure control measures are necessary. Rotstop is mainly used in mechanical timber harvesting, where operator exposure may occur during preparation of the working solution but not much during stump treatment. A field survey of harvesting operations in Finland indicated that respiratory and dermal exposure to Rotstop was insignificant, and that exposure can be controlled with adequate working methods and by the use of proper personal protective equipment for skin, eye, and respiratory protection.

In manual harvesting, the use of a chain-saw requires appropriate protective clothing which reduces the risk for dermal exposure. Adequate working methods and use of protective equipment to prevent respiratory exposure will further reduce the risk. Considering the data gap identified for secondary metabolites/toxins, the operator, worker and residential exposure assessment could not be finalised. Considering the representative uses (application in areas where trees are being cut), bystanders are expected not to be allowed in the vicinity of the product application.

**Residues** (Regulation (EU) N° 283/2013, Annex Part B, point 6 and Regulation (EU) N° 284/2013, Annex Part B, point 8; OECD IIM Point 6 & IIIM Point 8)

Viable residues: Not relevant considering the nature of the fungus and its intended use.

Non-viable residues: Not relevant considering the nature of the fungus and its intended use.

**Fate and Behaviour in the Environment** (Regulation (EU) N° 283/2013, Annex Part B, point 7 and Regulation (EU) N° 284/2013, Annex Part B, point 9; OECD IIM Point 7 & IIIM Point 9)

Persistence and multiplication (competitiveness) in soil

*P. gigantea*, the species populations generally do not multiply within water or soil.

Information on *Phlebiopsis gigantea* strains VRA 1835, VRA 1984 and FOC PG 410.3 in soil was not available

Assumptions:

- According to the LoEP max. content of microorganisms = 5 x
10^{10} \text{ CFU/kg}

- dose rate, considering application: 7200 mg MPCA/ha
- incorporation into the top 5 cm layer (= 50 L soil/m²)
- soil density of 1.5 g/cm³ (= 75 kg soil/m³) (=750,000 kg/ha)
- plant interception: not considered in order to create a worst case scenario that covers all uses.

\[
\text{PEC}_{\text{soil}} (\text{mg/kg}) = \frac{\text{dose of active substance}}{\text{mass of soil}} = \frac{7200 \text{ mg}}{75000 \text{ kg}} = 9.6 \text{ µg/kg}
\]

equivalent to 4.8 × 10^6 CFU/kg (considering max. content of microorganisms = 5 × 10^{10} CFU/kg)

Thus PECsoil under worst case conditions is assumed to be 9.6 µg/kg, which is equivalent to 4.8 × 10^6 CFU/kg dry weight soil.

### Persistence and multiplication (competitiveness) in air

Basidiospores of *P. gigantea* are naturally present in the air in relatively high numbers. They are robust, lightweight structures adapted for passive dispersal in air currents. However, stump treatment products utilise oidia which are relatively fragile spores with little capacity to travel, although they may be passively carried from stump to stump by arthropods. Both spore types will mainly colonise suitable media such as woody stump tissue.

### Mobility:

*P. gigantea* hyphae and oidia have been demonstrated to exhibit low mobility via air, but basidiospores have the potential to move long distances in air currents.

**Effects on non-target organisms** (Regulation (EU) N° 283/2013, Annex Part B, point 8 and Regulation (EU) N° 284/2013, Annex Part B, point 10; OECD IIM Point 8 & IIIM Point 10)
Phlebiopsis gigantea is adapted to grow in moribund wood and it is a natural component of forest ecosystems. The Phlebiopsis gigantea strains VRA 1835, VRA 1984 and FOC PG 410.3 are native to northern and central Europe. Non-target organisms are naturally exposed to the fungus and its spores. The representative use of the plant protection product will result in oidia (asexual spores) concentrations which are lower than the natural deposition of basidiospores of Phlebiopsis gigantea. Taking into consideration all evidence from the submitted information it can be concluded that the risk to non-target organisms from the representative uses of Phlebiopsis gigantea strains VRA 1835, VRA 1984 and FOC PG 410.3 is likely to be low. However, the risk assessment for secondary metabolites was not finalised. Data on the potential for Phlebiopsis gigantea strains VRA 1835, VRA 1984 and FOC PG 410.3 to produce metabolites in relation to the potential for the Phlebiopsis gigantea species generally to produce the metabolites, was not available. Information on this is needed for a definitive conclusion on the need for an environmental exposure and risk assessment for metabolites.

**Effects on birds and other terrestrial vertebrates** (MA 8.1 & MP 10.1; OECD IIM 8.1 & IIM 10.1)

| Effects on mammals: | Rat, acute LD₅₀ > 4.26 x 10⁷ CFU of *P. gigantea* kg bw |
|---------------------|------------------------------------------------------|
| Effects on birds:   | There is no evidence in the literature to indicate that *P. gigantea* is toxic, infective or pathogenic to birds. |
| Risk assessment for birds and mammals: | *P. gigantea* is a specialised fungus which lives within moribund wood, such as fallen branches and recently cut stumps. It is a natural component of forest ecosystems, and its spores will be present in the air and on most exposed surfaces within a forest environment. Levels of spores applied during treatment are small in comparison to natural spore loads, treatment is targeted onto the stump surface, and treatment operations do not increase the ambient spores levels within the forest. Wild mammals are unlikely to be affected by stump treatment operations, and in any case the toxicity tests indicate that LD₅₀ values for vertebrates are too high to be properly measured. In conclusion, non-target organisms will not be exposed to higher levels of the fungus than those already naturally present in the forest environment. |

**Effects on aquatic organisms** (MA 8.2 & 10.2; OECD IIM 8.2, 8.3& IIM 10.2)

There is no evidence in the literature to indicate that *P. gigantea* is toxic, infective or pathogenic to aquatic organisms.

**Effects on bees** (MA 8.3 & MP 10.3; OECD IIM 8.7 & IIM 10.3)

| Species           | Crop | Test Substance | Route/time-scale | Toxicity, infectivity and pathogenicity (endpoint, value or other description of effects) |
|-------------------|------|----------------|------------------|---------------------------------------------------------------------------------|
| Laboratory Tests  |      |                |                  |                                                                                  |
| Worker honey bee  | n/a  | Formulated product | Oral and contact, 24 | Oral and contact LD₅₀ values at 24 and 48 hours were estimated to be >100 µg product/bee, |
| *Aphis*           |      |                |                  |                                                                                  |
**Effects on terrestrial arthropods other than bees** (MA 8.4 & MP 10.4; OECD IIM 8.8 & IIIM 10.4)

*P. gigantea* co-exists with many arthropods within stumps, and is associated with bark- and wood-boring insects such as bark beetles. Although there is some indication that *P. gigantea* may compete with certain arthropods (e.g. *Hylobius abietis*) which share the same ecological niche, there is no indication from published literature that *P. gigantea* is toxic, infective or pathogenic to arthropods.

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**Effects on other terrestrial invertebrates** (MA 8.5 & MP 10.5; OECD IIM 8.9.1 and IIM 8.9.2 & IIIM 10.5)

There is no evidence in the literature to indicate that *P. gigantea* is toxic, infective or pathogenic to other terrestrial invertebrates.

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**Effects on soil microorganisms** (MA 8.6 & MP 10.6; OECD IIM 8.10 & IIIM 10.6)

*P. gigantea* does not persist or multiply within soil, although it has been shown to occasionally occur below-ground in forest ecosystems. In addition, there is no evidence in the literature to indicate that *P. gigantea* is toxic, infective or pathogenic to soil microorganisms.

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**Additional studies** (MA 8.7 & MP 10.7; OECD IIM 8.11 & IIIM 10.7)

**Effects on terrestrial plants:**

*P. gigantea* has a limited ability to colonise living terrestrial trees through inoculated and naturally infected wounds in the bark. It has been shown to be able to colonise fine roots of spruce in a mycorrhiza-like manner. There is no indication in the literature that *P. gigantea* can infect unwounded trees, on the contrary it has been shown to have limited necrotrophic capability. *P. gigantea* caused no deleterious effects on other terrestrial plants on the forest floor.

**Effects on stump microflora:**

Colonisation of a stump by natural or artificially inoculated *P. gigantea* does not prevent the multiple colonisation of stumps by numerous other fungal species, and stump treatment does not affect the diversity of resident populations of *P. gigantea*. However, there can be qualitative, usually short-term, differences in fungal and bacterial species composition, and sometimes an effect on species richness.