List of end points

Rapporteur Member State	Month and year	Microbial or Viral Agent (Name)
The Netherlands	***July 2018***	***Bacillus thuringiensis* subsp. *aizawai* strain GC-91**

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:	*Bacillus thuringiensis* subsp. *aizawai* (Bta) strain GC-91
Function (*e.g.* control of fungi):	Biological insecticide
Rapporteur Member State:	The Netherlands
Co-rapporteur Member State:	Germany
Identity of the Microbial or Viral Agent used in plant protection / Active Substance) (Regulation (EU) N° 283/2013, Annex Part B, point 1)	
Name of the organism:	*Bacillus thuringiensis* subsp. *aizawai* (Bta) strain GC-91
Taxonomy:	Domain: Bacteria Phylum: Firmicutes Class: Bacilli Order: Bacilliales Family: Bacillaceae Genus: *Bacillus*
Species, subspecies, strain:	Species: *Bacillus thuringiensis* Subspecies: *aizawai* Strain: GC-91
Identification / detection:	Morphological and biochemical characterization, serotyping, plasmid profiling, activity spectrum, fatty acid analysis, DNA fingerprinting AFLP, cry toxin analysis, strain specific marker
Culture collection:	National Collection of Type Cultures (NCTC), at the Health Protection Agency, Centre for Emergency Preparedness and Response, Porton Down, Salisbury, Wiltshire, SP4 0JG (formerly Central Public Health Laboratory, Colindale Avenue, London NW9 5HT). Reference number: NCTC 11821
Minimum and maximum concentration of the MPCA used for manufacturing of the formulated product (cfu; g/kg):	Min: 6.2×10^{10} CFU/g Max: 7.9×10^{10} CFU/g
Identity and content of relevant impurities, additives, contaminating organisms in the technical grade of MPCA:	No additives, no impurities expected Microbial contaminant screening: Coliforms: < 10 CFU/g *E. coli*: Absence in 10 g

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	Listeria: Absence in 25 g *Salmonella*: Absence in 10 g *Shigella*: Absence in 25 g *Staphylococcus aureus*: Absence in 10 g *Vibrio cholera*: Absence in 10 g Yeast and Mold: < 1000 CFU/g
Is the MPCA genetically modified; if so provide type of modification	Bta GC-91 is not a genetically modified but a transconjugant strain.

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Biological properties of the microorganism (Regulation (EU) N° 283/2013, Annex Part B, point 2)

Origin and natural occurrence, Background level:	Bt as a species (including Bta) occurs naturally in a range of environmental compartments such as soils, plant surfaces and infected insects. Background populations of Bt in the environment were found in the range from 10^4 to 10^8 CFU/g in soil and 0 – 10^4 CFU/g on plants in areas not previously treated with Bt.
Target organism(s):	Lepidopteran pests (GAP: *Cydia pomonella*, *Spodoptera littoralis, Lobesia botrana, Eupoecilia ambiguella, Tuta absoluta*)
Mode of action:	The insecticidal activity of Bta is mainly attributed to spore bound insecticidal pro-toxins (Cry toxins) which are ingested by the target pests (lepidopteran larvae) and activated under alkaline conditions in the midgut of the larvae.
Host specificity:	It is generally agreed that Bta acts highly specific against members of the insect family of Lepidoptera. Some are also active against Diptera or Coleoptera. The activity spectrum of a certain strain is defined by the production of *cry* toxins. Bta GC-91 was shown to be active against lepidopteran species only.
Life cycle:	*Bacillus thuringiensis* is a ubiquitous micro-organism that colonizes a range of habitats and environments and can be found in two different stages. Under favourable conditions regarding moisture, temperature and nutrients, the basic metabolizing cell type is the vegetative cell that is actively growing and dividing. When a population of vegetative cells passes out of the exponential phase of growth, usually as a result of nutrient depletion, the differentiation of endospores begins. Endospores are formed intracellularly and are liberated after lysis of the parent cells. The transformation of dormant spores into vegetative cells can be described in three stages: (i) Activation: a reversible process that prepares the spore for germination and usually results from treatments like heating or exposure to certain chemical stimuli; (ii) Germination: the breaking of the spore stage involves the swelling, rupture of the spore coat, loss of resistance to deleterious environmental factors and increase of metabolic activity; (iii) Outgrowth: development into a vegetative cell by remerging new components from the spore coat.
Infectivity, dispersal and colonisation ability:	Spores are the form of Bt that assures survival. They can survive in soil for months and it was showed that cells and spores of Bt can also survive for 10 days in water, without altering their number. Neither cells nor spores of Bt are mobile, so their dispersal is limited. It is generally agreed that Bt is a poor competitor and does not germinate and grow extensively in the environment. Except for target insects, Bta GC-91 is not expected to colonize any non-target organism and is not infective in humans.
Relationships to known plant, animal or human pathogens:	As a member of the *B. cereus*-group, Bta is closely related to *B. anthracis* and *B. cereus*. Btk strains are however phylogenetically distinguishable from *B. cereus* and *B. anthracis.*
Genetic stability:	Culture maintenance programs ensure that only genetically unchanged and pure cultures of Bta GC-91 are used for manufacturing of the strain and the end-use product. After field or greenhouse application genetic exchange is unlikely to occur and will not lead to any adverse effects.

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	From the literature search for Bta GC-91 it can be concluded, that transfer of genetic material cannot be completely ruled out upon use of the strain as pest control agent in agricultural settings but the likelihood is rather low because the event requires germination and growth of the applied GC-91 spores at a high level and the presence of competent recipient vegetative cells at a high level. Even under these conditions, rates of genetic exchange were shown to be extremely low. In addition, Bta GC-91 is a wild type strain and does not have the capacity to produce any other compounds than indigenous Bt´s already present in the environment and it is not multi-resistant. Hence, in the unlikely case that genetic material would be transferred from GC-91 to indigenous bacteria, there is no risk that any unwanted properties are spread in the environment.
Information on the production of relevant metabolites (especially toxins):	Bta GC-91 produces Cry1Ac, Cry1C, Cry1D and Cry2A insecticidal proteins. Apart from the Cry proteins several other insecticidal proteins are produced by Bt (vegetative insecticidal proteins VIP, cytolytic proteins Cyt etc.). Absence of toxicity to humans and mammals from all metabolites involved in the mode of action was confirmed by a literature search. Beta-exotoxins, are considered to have toxic properties but were shown not to be produced by commercial Bta strains. The ability to produce *B. cereus*-enterotoxins and possible consequences for consumers is discussed since first evaluation of the strain. However, based on available knowledge on Bta GC-91, there is no hint that the strain has the ability to cause foodborne disease as it will not fulfil all prerequisites required for pathogenic action in humans. In fact, it was demonstrated that the strain has a low toxigenic potential.
Resistance/ sensitivity to antibiotics / anti-microbial agents used in human or veterinary medicine:	Bta GC-91 has been shown to be sensitive to a broad range of antibiotics commonly used in human and veterinary medicine.

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Summary of uses supported by available data (Regulation (EU) N° 283/2013, Annex Part B, point 3)

Summary of representative uses evaluated, for which all risk assessments needed to be completed (*name of active substance or the respective variant*) (Regulation (EU) N° 284/2013, Annex Part A, points 3, 4)

The supported uses of the representative product Agree 50 WG a Water Dispersible Granules (WG) formulation containing 500 g/kg *Bacillus thuringiensis* ssp. *aizawai* (strain GC-91), are summarised in the following table.

1	2	3	4	5	6	7	8	9	10	11	12	13
Use-No.	**Member state(s)**	**Crop and/ or situation** **(crop destination / purpose of crop)**	**F G or I**	**Pests or Group of pests controlled** (additionally: developmental stages of the pest or pest group)	**Application**			**Application rate**			**PHI** (days)	**Remarks:** e.g. g safener/synergist per ha
					Method / Kind	Timing / Growth stage of crop & season	Max. number (min. interval between applications) a) per use b) per crop/ season	Kg product / ha a) max. rate per appl. b) max. total rate per crop/season	g as/ha IU/ha CFU/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha min / max		

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1	2	3	4	5	6	7	8	9	10	11	12	13
Use-No.	**Member state(s)**	**Crop and/ or situation** **(crop destination / purpose of crop)**	**F G or I**	**Pests or Group of pests controlled** (additionally: developmental stages of the pest or pest group)	**Application**			**Application rate**			**PHI** (days)	**Remarks:** e.g. g safener/synergist per ha
					Method / Kind	Timing / Growth stage of crop & season	Max. number (min. interval between applications) a) per use b) per crop/ season	Kg product / ha a) max. rate per appl. b) max. total rate per crop/season	g as/ha IU/ha CFU/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha min / max		
1	EU	Pome fruits (apple, pear)	F	*Cydia pomonella*	Foliar spray	BBCH 53-99 (April-October)	a) 6 (7) b) 6 (7)	a) 2.0 b) 12.0	a) 1000 5×10^{10} IU/ha min $1,7*10^{13}$- max $6,6 \times 10^{13}$ CFU/ha b) 6000 3×10^{11} IU/ha Min $1,02 \times 10^{14}$ - max $3,96 \times 10^{14}$ CFU/kg	1000-1500	-	Maximum spray concentration (0.4 %)-400 g product/HL
2	EU	Grapes	F	*Lobesia botrana, Eupoecilia ambiguella*	Foliar spray	BBCH 53-99 (April-October)	a) 6 (7) b) 6 (7)	a) 2.0 b) 12.0	a) 1000 g/ha 5×10^{10}IU/ha min $1,7*10^{13}$- max 6.6×10^{13} CFU/ha b) 6000 3×10^{11}IU/ha Min $1,02 \times 10^{14}$ - max $3,96 \times 10^{14}$ CFU/kg	200-1200	-	Maximum spray concentration (0.4 %)-400 g product/HL -
3	EU	Tomato	G	*Tuta absoluta*	Foliar spray	BBCH 12-89 (all seasons, January-December)	a) 6 (7) b) 6 (7)	a) 2.0 b) 12.0	a) 1000 g/ha 5×10^{10} IU/ha min $1,7*10^{13}$- max $6.6\ 10^{13}$ CFU/ha b) 6000 3×10^{11}IU/ha Min $1,02 \times 10^{14}$ - max $3,96 \times 10^{14}$ CFU/kg	500-1500	.	Maximum spray concentration (0.4 %)-400 g product/HL -

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1	2	3	4	5	6	7	8	9	10	11	12	13
Use-No.	**Member state(s)**	**Crop and/ or situation** **(crop destination / purpose of crop)**	**F G or I**	**Pests or Group of pests controlled** (additionally: developmental stages of the pest or pest group)	**Application**			**Application rate**			**PHI** (days)	**Remarks:** e.g. g safener/synergist per ha
					Method / Kind	Timing / Growth stage of crop & season	Max. number (min. interval between applications) a) per use b) per crop/ season	Kg product / ha a) max. rate per appl. b) max. total rate per crop/season	g as/ha IU/ha CFU/ha a) max. rate per appl. b) max. total rate per crop/season	Water L/ha min / max		
4	EU	Turf, Sports	F	*Spodoptera* spp.	Foliar spray	BBCH 12-89 (all seasons, January-December)	a) 6 (7) b) 6 (7)	a) 2.0 b) 12.0	a) 1000 g/ha 5×10^{10} IU/ha min $1,7*10^{13}$- max 6.6×10^{13} CFU/ha b) 6000 3×10^{11} IU/ha Min $1,02 \times 10^{14}$ - max $3,96 \times 10^{14}$ CFU/kg	1000-1500	.	-

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Further information, Efficacy

Effectiveness (Regulation (EU) N° 284/2013, Annex Part A, point 6.2)

According to the latest guidance on the preparation of dossiers for the renewal of active substances, information on efficacy is not required (SANCO/10181/2013 – rev. 2.1, 13 May 2013). The representative products have all been authorised at Member State level for > 10 years and have therefore been assessed in line with Uniform Principles.The GAP for the representative uses is realistic.

Adverse effects on field crops (Regulation (EU) N° 284/2013, Annex Part A, point 6.4)

The representative products have all been authorised at Member State level for > 10 years and have therefore been assessed in line with Uniform Principles. No unacceptable adverse effects are known.

Observations on other undesirable or unintended side-effects (Regulation (EU) N° 284/2013, Annex Part A, point 6.5)

The representative products have all been authorised at Member State level for > 10 years and have therefore been assessed in line with Uniform Principles. No unacceptable side effects are known.

Classification and proposed labelling (Symbol, Indication of danger, Risk phrases, Safety phrases)

with regard to physical/chemical data:	-
with regard to toxicological data:	-
with regard to fate and behaviour:	-
with regard to ecotoxicological data:	-

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)

Manufactured microorganism (principle of method):	For original approval a set of methods have been applied for characterisation of the strain including morpholog-ical and biochemical characterization, serotyping, plasmid profiling, activity spectrum, fatty acid analysis, DNA fingerprinting AFLP, cry toxin analysis. For renewal, the strain was sequenced and specific markers were de-veloped and validated allowing an unequivocal

	identification of Bta GC-91. Moreover a biopotency (bioassay with *T. n)* is available.
Impurities and contaminating microorganisms in manufactured material (principle of method):	There are no relevant impurities, additives and metabolites. Standard microbiological methods for detection of microbial contaminants are available.
Microbial Pest Control Product (principle of method):	See above

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

of the active microorganism (principle of method):	Not required as Bta GC-91 is proposed for inclusion into Annex IV of Regulation (EC) No 396/2005. However, specific markers are available to monitor the strain in agricultural fields. Soil , water and air: not required
of relevant metabolites (principle of method):	Cry1Ab Soil: extraction with phosphate buffered saline Tween, quantification with commercial ELISA kit. LOQ 0.25 ng/mL. Water: processing via lyophilization and filter centrifugation, quantification with ELISA. Method detection limit 2.1 ng/L

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Impact on Human and Animal Health (Regulation (EU) N° 283/2013, Annex Part B, point 5 and Regulation (EU) N° 284/2013, Annex Part B, point 7)

Medical data: (including medical surveillance on manufacturing plant personnel) (MA 5.1.1))	Only very few and mild cases of human infections have been reported for Btk. No data have been published or reported about possible infection or pathogenicity on humans due to Bta. No incidents related to adverse health effects such as toxicological effects, allergic response, or irritation, to employees, resulting from exposure to Bta GC-91 during production and packaging of the product have been reported.
Sensitisation: (MA 5.2.1 & MP 7.2.3)	Not sensitizing
Acute oral infectivity, toxicity and pathogenicity: (MA 5.2.2.1 & MP 7.1.1)	There is no evidence that Bta GC-91 may cause acute oral toxicity, pathogenicity or infectivity in mammals. $LD_{50} > 9.4 \times 10^8$ CFU per kg bw, $LD_{50} > 5050$ mg/kg bw
Acute intratracheal/inhalation infectivity, toxicity and pathogenicity: (MA 5.2.2.2 & MP 7.1.2)	There is no evidence that Bta GC-91 may cause acute respiratory toxicity, pathogenicity or infectivity in mammals. $LC_{50} > 3.16$ mg/L (corresponding to 3.77×10^7 CFU/kg bw)
Acute intravenous/intraperitonial infectivity: (MA 5.2.2.3)	There is no evidence that Bta GC-91 acts toxic or pathogenic following intravenous or intraperitoneal administration.
Genotoxicity: (MA 5.2.3)	No validated methods available for microorganisms.
Cell culture study: (MA 5.2.4)	Bta/Btk is not an intracellular replicating micro-organisms, cell culture studies are not required.
Information on short-term toxicity and pathogenicity: (MA 5.2.5)	Acute toxicity studies did not reveal any signs of toxicity or pathogenicity, thus, there is no evidence that Bta GC-91 acts toxic or pathogenic following short-term exposure.
Dermal toxicity: (MP 7.1.3)	No adverse effects observed
Specific toxicity, pathogenicity and infectivity: (MA 5.3)	There is no evidence that Bta GC-91 may cause acute dermal toxicity. Bta GC-91 is neither a skin nor an eye irritant.
Genotoxicity – *in vivo* studies in germ cells: (MA 5.5)	No indications of genotoxicity are known for Bta/Btk.

Reference values

AOEL:	Not applicable
ADI:	Not applicable
ARfD:	Not applicable

Exposure (operator, workers, bystander, consumer): (MA 6.1 & MP 7.3, 8.0)	No reference values are derived for Bta GC-91 and metabolites. As a consequence no exposure calculations are necessary. As a consequence there is no health risk

	for operators, bystanders or workers.

Residues (Regulation (EU) N° 283/2013, Annex Part B, point 6 and Regulation (EU) N° 284/2013, Annex Part B, point 8)

Viable residues:	*B. thuringiensis ssp. aizawai* spores and crystal proteins, are not toxic or pathogenic to humans, plants, and most animals. Spores are not persistent on crop, half-life less than 1 day.
Non-viable residues:	Crystal proteins are unstable when exposed to light. half-life for insecticidal activity on leaves was 34 to 47 hours following application DT_{50} of crystal protein 24 hours after exposure to sunlight

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)

Persistence and multiplication (competitiveness) in soil, water and air:	*B. thuringiensis* and its secondary metabolites are not persistent in soil water and air.
Mobility:	The mobility of *B. thuringiensis* and the spores can be considered limited.

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)

Effects on birds (MA 8.1 & MP 10.1)

Species	**Test duration**	**Dose range**	**Results/ Endpoint**	**Observations**
TOXICITY				
Bobwhite quail *(Colinus virginianus)*	30 days (5 days of treatment)	LC_{50}: >3333 mg/kg >3.53 x 10^{11} CFU/kg b.w./day	LC_{50}: >3333 mg/kg >3.53 x 10^{11} CFU/kg b.w./day	No signs of mortality.
Mallard duck *(Anas platyrhynchos)*	30 days (5 days of treatment)	3333 mg/kg or approximately 3.53 x 10^{11} CFU/kg b.w./day	3333 mg/kg or approximately 3.53 x 10^{11} CFU/kg b.w./day	No signs of mortality.

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Species	**Test duration**	**Dose range**	**Results/ Endpoint**	**Observations**
INFECTIVENESS				
PATHO-GENICITY				
Bobwhite quail *(Colinus virginianus)*	30 days	LC_{50}: >3333 mg/kg >3.53 x 10^{11} CFU/kg b.w./day	LC_{50}: >3333 mg/kg >3.53 x 10^{11} CFU/kg b.w./day	No signs of pathogenicity.
Mallard duck *(Anas platyrhynchos)*	30 days	3333 mg/kg or approximately 3.53 x 10^{11} CFU/kg b.w./day	3333 mg/kg or approximately 3.53 x 10^{11} CFU/kg b.w./day	No signs of pathogenicity.

Effects on mammals
There is no evidence that Bta GC-91 may cause acute oral toxicity, pathogenicity or infectivity in mammals.

LD_{50} > 9.4 × 10^8 CFU per kg bw, LD_{50} > 5050 mg/kg bw

Effects on other terrestrial vertebrates

No information provided.

Effects on aquatic organisms (MA 8.2 & 10.2)

Species	**Test duration**	**Dose range**	**Results/ Endpoint**	**Observations**
TOXICITY				
Rainbow trout (*Onchorhyncus mykiss*)	32 d	3.90 x 10^{10} CFU/L, 3.90 x 10^9 CFU/L and 1.0 x 10^9 CFU/L.	LC_{50} > 2.0 x 10^{10} CFU/L (mean measured concentration).	No signs of toxicity, pathogenicity or infectivity. Increased concentration of cfu were measured in the gills after necropsy, but did not contstitute

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Rapporteur Member State	Month and year	Microbial or Viral Agent (Name)
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Species	**Test duration**	**Dose range**	**Results/ Endpoint**	**Observations**
				infection
Sheepshead minnow (*Cyprinodon variegatus*)	30 d	3.90 x 10^{10} CFU/L, 3.90 x 10^{9} CFU/L and 1.0 x 10^{9} CFU/L.	LC_{50} > 2.1 x 10^{9} CFU/L (mean measured concnetration)	No signs of pathogenicity or infectivity.
Daphnia magna	10 d	Not applicable	Not applicable	No endpoint calculated, due to different purpose of study. It concluded that heat labile components from the fermentation broth are carreied over into the technical material. These components elicit the toxicity of GC-91 observed for Daphnids.
Daphnia magna	21 d	1.17 x 10^{10}, 3.51 x 10^{9}, 1.05 x 10^{9}, 3.16 x 10^{8}, 9.50 x 10^{7} CFU/L	21-day EC_{50} (survival): 3.24 x 10^{8} CFU/L. NOEC(length, offspring and survival): 1.57 x 10^{8} CFU/L	Signs of infectivity were not observed when appying the normal test procedures.
Daphnia magna	21 d	8.5 x 10^{6}, 2.8 x 10^{7}, 9.5 x 10^{7}, 3.15 x 10^{8} and 1.05 x 10^{9} CFU/L	21-day EC50 > 6.2 x 10^{8} CFU/L NOEC ≥ 6.2 x 10^{8} CFU/L	No effects on pathogenicity or infectivity observed
Palaemonetes vulgaris	30 d	1.58 x 10^{10} CFU/g (water exposure)	21-d NOEC 1.9 x 10^{9} CFU/g (diet	No signs of treatment related

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Species	Test duration	Dose range	Results/ Endpoint	Observations
			exposure)	pathogenicity or infectivity were observed.
Scenedesmus subspicatus	72 h	4.2 X 10^7 14.1 X 10^7 46.5 X 10^7 125.5 X 10^7 356 X 10^7 CFU/L	$E_bC_{50:}$ > 3.6 x 10^9 CFU/L NOE_bC: 3.6 x 10^9 CFU/L	No signs of infectivity or pathogenicity found

Effects on bees (MA 8.3 & MP 10.3)

Species	Test type	Micro-organism tested	Endpoints and observations
Bumble bee *(Bombus terrestris)*	Oral and contact Exposure (11 weeks)	*Bta* (Xentari)	100% mortality orally via sugar water at 1.5 x 10^4 IU/mL 0% mortality orally via sugar water at 1.5 x 10^3 IU/mL 100% reduction in reproduction orally via sugar water at 1.5 x 10^4 IU/mL 0% reduction in reproduction oraglly via sugar water at 1.5 x 10^3 IU/mL 31% reduction in reproduction orally via pollen at 1.5 x 10^4 IU/mL. No effects via dermal exposure.
Bumble bee *(Bombus terrestris)*	Oral and contact Exposure (11 weeks)	*Btk* (Dipel)	3% mortality via contact at 1.6 x10^4 IU/mL

			5% mortality via sugar water at 1.6 $x10^4$ IU/mL 0% mortality via treated pollen at 1.6 $x10^4$ IU/mL No effects on reproduction at 1.6 $x10^4$ IU/mL
Honey bee *(Apis mellifera)*	Oral and contact (19 days)	*Btk* (Delfin WG)	96 h and 19-day LC_{50} > 41 kg/L 96 h and 15-day LC_{50} > 5 kg/L
Honey bee *(Apis mellifera)*	Oral (14 days)	*Bta* Technical grade material CGA 237218 (GC 91), 9.9% delta endotoxin	5-day LC_{50}: 3656 ppm (3652 mg/L, 897 µg/bee) 10-day LC_{50}: 170 ppm (169.8 mg/L, 91 µg/bee) NOEC: 50 ppm Delta-endotoxin (9.9%): 5-day LC_{50}: 88.8 µg/bee 10-dayLC_{50}: 9.0 µg/bee
Honey bee *(Apis mellifera)*	Oral, respiratory, direct and indirect contact (72 h)	CGD 97220 I (Agree 50 WP)	LD50 is > 2 g a.s./L. No behavioural anomaly was observed.

Effects on terrestrial arthropods other than bees (MA 8.4 & MP 10.4)

Species	Test type	Test substance	Endpoints and observations
Aphidius rhopalosiphi	Tier I	Turex 50 WP	No adverse effects on mortality of *Aphidius rhopalosiphi* while a significant reduction in the reproduction rate was observed up to the tested application rate of 4.5 kg product/ha (i.e. 2.25 kg a.s./ha). Infectivity and pathogenicity was not tested.

Typhlodromus pyri			No adverse effects on *Typhlodromus pyri* up to the tested application rate of 4.5 kg/ha (i.e. 2.25 kg a.s./ha). Infectivity and pathogenicity was not tested
Uga menoni	Tier I	Bta Technical grade material CGA 237218 (GC 91)	30 day dietary LC50: 4.4 x 10^7 CFU/g feed Infectivity not tested.

Effects on other terrestrial invertebrates (MA 8.5 & MP 10.5)

Species	Test type	Test substance	Endpoints and observations
Eisenia fetida	Laboratory (acute test)	CGD 97220 I (Agree 50 WP)	The test substance CGD 97220 I caused no mortality with the concentrations of 500 and 1000 mg/kg d.w. At both concentrations an increase of weight was observed. This was significant at the concentration of 1000 mg/kg d.w. test duration was too short to investigate infectivity and pathogenicity.

Effects on soil microorganisms (MA 8.6 & MP 10.6)

CGD97220 I (Agree 50 WP) had no negative influence neither to the nitrogen turnover nor to the dehydrogenase activity in both soils. In conclusion, the test substance CGD 97220 I represents no hazard to soil microflora.

Additional studies (MA 8.7 & MP 10.7)

Evidence from over fifty phytotoxicity trials performed on twelve different crops in the USA with Javelin Biological Insecticide and SAN 415I SC 353 support the lack of toxicity expected on terrestrial plants following application of Btk containing formulations in the fields. The highest mean toxicity was observed in trials (n = 3) with Bok choy (*Brassica chinensis*) at 4.3%, the next highest mean toxicity was in trials (n = 10) with Sugar beet (*Beta vulgaris*) at 1.7% (Anonymous, year unknown). These results may be extrapolated to Bta due to their family relationship.