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(54) **Titre : CONIOTHYRIUM MINITANS POUR UNE UTILISATION CONTRE LA CROISSANCE DE MOUSSE**
(54) **Title: CONIOTHYRIUM MINITANS FOR USE AGAINST MOSS GROWTH**

(57) **Abrégé/Abstract:**

The invention relates to the use of Coniothyrium minitans for controlling moss growth and to a method of controlling moss growth, comprising applying Coniothyrium minitans to an area to be kept free from mosses.



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Coniothyrium minitans for use against moss growth

Moss growth is in a number of places considered to be a nuisance and in many places unwelcome, for example on turfed areas or on trees and on asphalted or cobbled ground or otherwise paved ground and on the roofs of
5 houses.

Methods for controlling moss growth include physical removal and chemical treatment or a combination of these methods. Chemical methods, in particular, frequently have side effects such as, for example, damage to other plants, staining, corrosion or pollution of the air and/or water (Davison and Byther, 1999; Algae, Lichens and Mosses on Plants. Pacific Northwest Plant Disease Control Handbook. p. 39). Moreover, the efficacy of many
10 herbicides against mosses has not been studied thoroughly. Finally, the safety regulations to be adhered to also make the handling, dosing and storing of chemical products more complex.

The fungus *Coniothyrium minitans* was identified firstly in 1947 and can be found in soils worldwide. It attacks sclerotia (overwintering structures or survival structures) of *Sclerotinia sclerotiorum* and *Sclerotinia minor*, other
15 *Sclerotinia* species and *Sclerotium cepivorum* and destroys them. These plant pathogens have a range of host plants, especially including useful plants, where they cause considerable damage.

In view of the possibilities for controlling moss, which are not optimal, and the intended reduction of water and air pollution by herbicides, alternative and less harmful methods for controlling mosses should be found.

20 This technical problem is solved by the present invention as defined in the claims.

The present invention relates to the use of *Coniothyrium minitans* for controlling or preventing moss growth, for example in crops of useful plants or in places such as paved places where moss growth is undesired.

25 *Coniothyrium minitans* has as yet only been known as a biological fungicide which acts against *Sclerotinia* species. In the context of the present invention, however, it has been found that the fungus also inhibits moss growth and/or causes existing moss to die. It is to be noted that the effect can indeed be attributed to the fungus and not, for example, to the presence of formulation aids (see example).

30 In accordance with the invention, all strains belonging to the species *Coniothyrium minitans* can be used which have moss-growth-inhibiting or moss-controlling properties.

An especially preferred strain of *Coniothyrium minitans* is CON/M/91-08 (described in WO 96/21358), which
35 has been deposited at the Deutsche Sammlung für Mikroorganismen und Zellkulturen in Braunschweig under the accession number DSM9660. The strain is used as biological plant protection agent against the fungi *Sclerotinia*

sclerotiorum and *Sclerotinia minor*, which are causative organisms of white mould, and is commercially available under the trade name Contans®.

Coniothyrium minitans strain CON/M/91-08, like other *Coniothyrium minitans* strains, may be cultured as described for example in WO 96/21358, which is herewith incorporated by reference. For example, *Coniothyrium minitans* can be cultured on suitable substrates such as, for example, seeds of cereals, bran, straw or other plant matter, or else with the aid of agar culture medium conventionally used in fungal culture, for example potato dextrose agar or malt peptone agar, or on suitable support materials to which a culture medium has been added; and also in liquid nutrient media without the use of agar.

Usually, the strain is formulated on a carrier, preferably a water-soluble sugar carrier. The water-soluble carrier is preferably glucose or dextrose.

Besides *Coniothyrium minitans* CON/M/91-08, mutants of this strain which have the same properties in respect of moss growth as CON/M/91-08 are also encompassed.

Mosses are green terrestrial plants which can be divided into hornworts, liverworts and Bryophyta. *Coniothyrium minitans* is active against mosses in general, but in particular against Bryophyta such as, for example, haircap mosses.

Mosses sometimes grow in places where useful plants or ornamentals are being grown. To be mentioned here in particular is turf as found in parks, golf courses, sports grounds or in gardens. Turf is also frequently sold as turf rolls or turf sods, likewise susceptible to moss growth.

Other places where moss growth is undesired are certain areas or places, including paved areas or places, such as, for example, tiled, cobbled or asphalted areas, where mosses colonize joints or undesired gaps.

In a preferred embodiment, spores of *Coniothyrium minitans* are used.

Spores formed by *Coniothyrium minitans* are conidia (asexually formed spores), which are formed in pycnidia and are also referred to as pycnidiospores or pycnospores.

In a further preferred embodiment, the use takes place on turf.

In a further preferred embodiment, the use according to the invention takes place in gardens, parks, sport grounds, terraces, pavements and otherwise plant-free places.

In yet another preferred embodiment, the use takes place in combination with at least one herbicide which is preferably active against mosses.

In a further preferred embodiment, the herbicide is selected from the group consisting of dodecyldimethylammonium chloride, ACN (2-amino-3-chloro-1,4-naphthoquinone), carfentrazone-ethyl, lauroyl methyl-[beta]-alanine-sodium, ferrous ammonium sulphate, copper hydroxide, mancozeb, propineb, oxadiazon, formaldehyde and chlorothalonil.

In a further especially preferred embodiment, the use according to the invention takes place with at least one herbicide, simultaneously or alternately.

In a preferred embodiment, the application rate is between 2 and 2000 kg/ha. Preferred ranges comprise an application rate of between approximately 10 and approximately 500 kg/ha, furthermore preferred between approximately 20 and 200 kg/ha, for example 30, 40, 50, 100 or 150 kg/ha.

In a further preferred embodiment, the spore concentration in the formulation applied is between 1×10^8 and 1×10^{10} /g.

The spores may be present in a concentration of between about 1×10^7 and about 1×10^{15} spores/g carrier, preferably between about 1×10^8 and about 1×10^{13} spores/g carrier. The most preferred concentration is between about 1×10^8 and about 1×10^{10} spores/g carrier, for example about 1×10^9 spores/g carrier.

In another preferred embodiment, the use according to the invention is preventive.

In a further aspect, the invention relates to a method of controlling mosses, comprising applying *Coniothyrium minitans* to an area to be kept free from mosses, such as, for example, a crop of useful plants or places such as paved places.

To describe the preferred embodiments of this aspect of the invention, those of the first aspect of the invention may be used.

In a preferred embodiment, *Coniothyrium minitans* strain CON/M/91-08 is used.

In a preferred embodiment of the method according to the invention, spores of *Coniothyrium minitans* are used.

In a further preferred embodiment, the application takes place onto and/or into the soil or the area to be treated.

In a further preferred embodiment the application is effected on turf.

In a further preferred embodiment, the method according to the invention is applied in gardens, parks, sports grounds, terraces, pavements and otherwise plant-free places.

- 5 In a further preferred embodiment, the application of *Coniothyrium minitans* takes place in combination with at least one herbicide which is active against mosses.

In an especially preferred embodiment, the herbicide is selected from the group consisting of dodecyldimethylammonium chloride, ACN (2-amino-3-chloro-1,4-naphthoquinone), carfentrazone-ethyl, lauroyl
10 methyl-[beta]-alanine-sodium, ferrous ammonium sulphate, copper hydroxide, mancozeb, propineb, oxadiazon, formaldehyde and chlorothalonil.

In a further especially preferred embodiment, the application of *Coniothyrium minitans* and at least one herbicide takes place simultaneously or alternately.

- 15 In a preferred embodiment of the method according to the invention, the application rate is between 2 and 2000 kg/ha.

In a further preferred embodiment, the spore concentration in the formulation applied is between 1×10^7 and
20 1×10^{10} /g.

In another preferred embodiment, the method according to the invention is preventive.

The examples which follow describe the invention in non-limiting form.

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Example 1: Drench test

The test was carried out under greenhouse conditions.

120 ml of a mixture of steamed field soil and sand (1:1) was filled into transparent cylindrical vessels 5.5 cm in size.

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The test formulation, the product Contans®, comprising 1×10^9 spores of the strain *Coniothyrium minitans* CON/M/91-08, was dissolved in water and diluted with water to the desired concentrations. 20 ml of the made-up solutions were pipetted onto the soil in the vessels.

The vessels were covered with a transparent lid.

10 The vessels remained in the greenhouse for the next 19 weeks, at 15°C and at natural light conditions. The lids were opened slightly after approximately 2 weeks, and the soil was watered every 3 weeks and kept moist.

The evaluation was effected by estimating, per vessel, the soil surface area which had in the meantime been covered by growth, predominantly haircup moss growth. 0% means an efficacy which corresponds to that of the untreated control, while an efficacy of 100% means that no moss growth was discernible.

15 In this test, the following formulations display an efficacy of up to 100% at a dose of 200kg/ha (see table 1).

	Concentration	Efficacy %
Control		0*
Contans **	2kg/ha = 0.024mg/ml	50
Contans**	20kg/ha = 0.24mg/ml	38
Contans**	200kg/ha = 2.4mg/ml	100
Glucose	200kg/ha = 2.4mg/ml	13
DDAC-C10***	6.25kg/ha = 20µl/ml	100

Table 1

20 The *Coniothyrium minitans* spores used are bound to glucose. To rule out a potential effect of glucose on the moss growth, an experiment with glucose was carried out, and this experiment shows that the effect cannot be attributed to the glucose.

*Moss growth: 80% of area

** *Coniothyrium minitans* CON/M/91-08; 5×10^8 viable spores/g

***Didecyltrimethylammonium chloride with alkyl chain length C10

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1-2	line	1-3
1-3	Identification of deposit	
1-3-1	Name of depositary institution	DSMZ Leibniz-Institut DSMZ - Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH (DSMZ)
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CLAIMS:

1. Use of a strain of *Coniothyrium minitans* for controlling or preventing moss growth.
2. Use according to Claim 1, wherein the strain is CON/M/91-08, deposited at the Deutsche Sammlung für Mikroorganismen und Zellkulturen in Braunschweig under the accession number
5 DSM9660.
3. Use according to Claim 1 or 2, which takes place on turf.
4. Use according to Claim 1 or 2, which takes place in gardens, parks, sports grounds, terraces, pavements and otherwise plant-free places.
5. Use according to any one of Claims 1 to 4, wherein the use takes place in combination
10 with at least one herbicide.
6. Use according to Claim 5, wherein the herbicide is active against mosses.
7. Use according to Claim 5 or 6, wherein the herbicide is selected from the group consisting of dodecyldimethylammonium chloride, ACN (2-amino-3-chloro-1,4-naphtoquinone), carfentrazone-ethyl, lauroyl methyl-[beta]-alanine-sodium, ferrous ammonium sulphate, copper
15 hydroxide, mancozeb, propineb, oxadiazon, formaldehyde and chlorothalonil.
8. Use according to any one of Claims 5 to 7, wherein the use takes place with at least one herbicide, simultaneously or alternately.
9. Use according to any one of Claims 1 to 8, wherein spores of *Coniothyrium minitans* are used.
- 20 10. Use according to Claim 9, wherein the spores are applied in a formulation at a rate of between 2 and 2000 kg/ha.
11. Use according to Claim 10, wherein the spore concentration in the formulation applied is between 1×10^7 and 1×10^{10} /g.
12. Use according to any one of Claims 1 to 11, which is preventive.
- 25 13. Method of controlling moss growth, comprising applying *Coniothyrium minitans* to an area to be kept free from mosses.
14. Method according to Claim 13, wherein the application takes place onto and/or into soil.
15. Method according to Claim 13 or 14, wherein the application is effected on turf.

16. Method according to any one of Claims 13 to 15, wherein the application of *Coniothyrium minitans* takes place in combination with at least one herbicide which is active against mosses.
17. Method according to Claim 16, wherein the herbicide is selected from the group consisting of dodecyldimethylammonium chloride, ACN (2-amino-3-chloro-1,4-naphtoquinone),
5 carfentrazone-ethyl, lauroyl methyl-[beta]-alanine-sodium, ferrous ammonium sulphate, copper hydroxide, mancozeb, propineb, oxadiazon, formaldehyde and chlorothalonil.
18. Method according to Claim 16 or 17, wherein the application of *Coniothyrium minitans* and at least one herbicide takes place simultaneously or alternately.
19. Method according to any one of Claims 13 to 18, wherein spores of *Coniothyrium*
10 *minitans* are used.
20. Method according to Claim 19, wherein the spores are applied in a formulation at a rate of between 2 and 2000 kg/ha.
21. Method according to Claim 20, wherein the spore concentration in the formulation applied is between 1×10^7 and 1×10^{10} /g.
- 15 22. Method according to any one of Claims 13 to 21, wherein the application takes place as a preventive measure.