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CONTANS®

TECHNICAL DESCRIPTION

BIOLOGICAL FUNGICIDE AGAINST SCLEROTINIA SPP.

General Information

Contans® is a new biological fungicide consisting of highly active spores of the soil fungus *Coniothyrium minitans* which has specific antagonistic action against the survival structures (sclerotia) of the plant pathogens *Sclerotinia sclerotiorum* and *Sclerotinia minor*. Therefore the product can be used for the control of soil infestations of both pathogens. Contans® may also be effective to *Sclerotium cepivorum*.

Since Contans® is a microbiological agent of high activity which specially attacks the sclerotia of the pathogens within the soil, there are no adverse effects on the soil ecosystem in general. In addition there are no toxic or allergic effects on mammals, fish or insects (see toxicity information below).

Application

The target plants for treatment with Contans® are high value agricultural and vegetable crops susceptible to *Sclerotinia* infection. These include peanuts, oilseeds (sunflowers, oilseed rape, soja), vegetable crops including lettuce, celery, beans, cucumbers, tomatoes etc. as well as ornamentals e.g. chrysanthemum. There are even indications in the literature that *C. minitans* could be active against *Sclerotium cepivorum*, a major disease agent in onion and garlic.

The application method is to apply and incorporate the agent three to four months before planting (for example in Autumn for Spring planting) in order to allow Contans® to destroy the sclerotia in the soil. The soil should not then be ploughed before planting the susceptible crop. Otherwise untreated sclerotia from lower soil layers could be transferred to the top soil

and cause disease. The deep Contans® has to be incorporated to the soil should lay between 5 and 15 cm.

The second application method aims to stop soil infestation after harvesting a heavily infested crop. This application would take place prior to ploughing-in infested harvest residues. In this way, soil infestation will be reduced step by step.

The active agent is effective at rates of 2 to 8 kg per hectare, with the higher concentration being used at a higher deep of incorporation.

Physical Properties of the Pure Agent (fungus spores)

Name of the agent:	<i>Coniothyrium minitans</i> (strain CON/M/91-08)
Colour:	dark brown
Consistency:	solid
Smell:	pleasant, like mushrooms
Stability:	in pure form resistant to decomposition by light, not resistant to high temperatures (above 40 degrees Centigrade, 104 degrees F.), not resistant to degradation by chemicals. Contans® will degrade slowly in time. Shelf life studies indicate that the activity of fungus spores is reduced by 25 percent within 18 months. Current plans for distribution assume a shelf life of at least six months from completion of preparation until application.
Solubility:	insoluble, but suitable for suspension in water. The active agent is estimated to consist of 1×10^{12} spores per kilogram, and is diluted to 1 : 200 up to 1 : 1000 in water for application.

Application would be made of anywhere from 2 to 6 kg per hectare of active ingredient.

Larger concentrations of active ingredient would be mixed at a smaller ratio of water.

Toxicity

Acute oral LD50 (rat)	Relatively non-toxic (>2500 mg/kg OECD method 401)
Acute dermal LD50 (rat)	Relatively non-toxic (>2500 mg/kg OECD method 402)
Acute intraperitoneal LD50 (rat)	Relatively non-toxic (>2000 mg/kg OECD method 401)
Acute inhalation toxicity (rat)	Relatively non-toxic (>12.74 mg/litre of air for 4 hrs)
Eye irritation (rabbit)	None
Skin irritation (rabbit)	None

Environmental Toxicology

Acute toxicity (fish)	>100 mg of ai per litre 96h (OECD method 203)
Acute toxicity (Daphnia)	>100 mg of ai per litre 48h (OECD method 202)
Acute toxicity (Algae)	>100 mg of ai per litre 72h (OECD method 201)

Storage and Disposal

Storage of the dry active agent should be at temperatures at below 5 degrees Centigrade (75 degrees Fahrenheit).

Residues of the active agent can be disposed of by spraying onto waste land or onto soil together with organic manure or with liquid nitrogen fertilizers. Empty containers have no toxic residue and can be disposed with other waste products.

Effectiveness

In the summers 1994 and 1995 batches of 10 litres of soil were mixed with 80 sclerotia of *S. sclerotiorum*. Contans® solution (8 ml) was then mixed into the soil which is equivalent to 2 kg Contans® per hectare. The soil was refilled into microplots with a size of 40 x 50 cm and a depth of 5 cm. To ensure recovery of the sclerotia in the following year, the bottom of the plots were covered with gauze. In the beginning of May 1995 and 1996 the soil was removed from the plots and the sclerotia were recovered by sieving and washing. The sclerotia were counted and viability evaluated (Fig. 1).

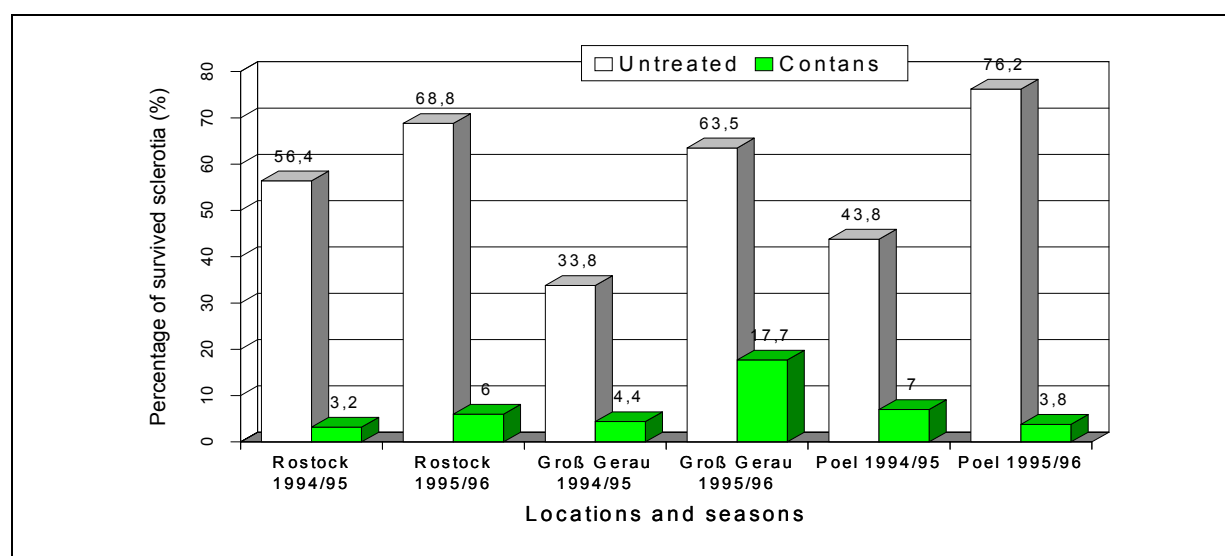


Fig. 1 Survival of sclerotia of *S. sclerotiorum* in different field soils after addition of Contans® to the soil and incubation from August 1994/95 to May 1995/96 under natural conditions

In the season 1996/97 official tests on oilseed rape were carried out by different plant protection offices. Soil was artificially infested with 50 sclerotia per m² and an equivalent of 2 kilogram per hectare of Contans® applied to the soil. The treatment was carried out in August 1996 just before sowing. The disease rate was evaluated in July 1997.

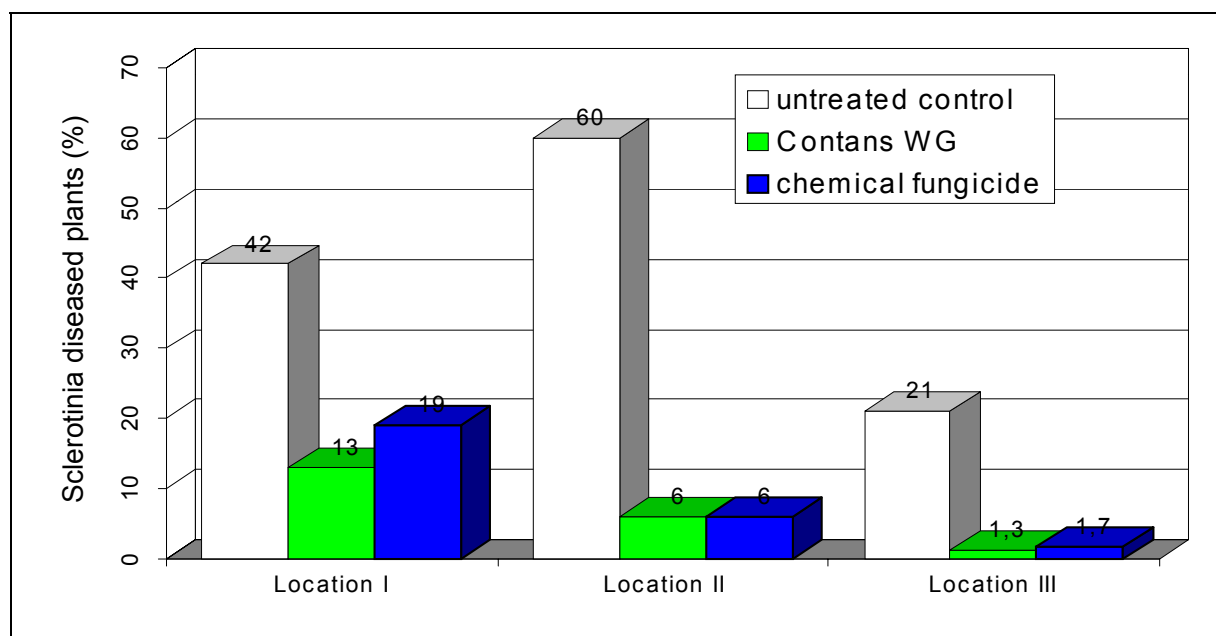


Fig. 2 Efficacy of Contans® WG against *Sclerotinia sclerotiorum* on oilseed rape in comparison with an untreated control and a treatment with a chemical fungicide

In 1997 there were carried out some further official tests to evaluate the efficacy of Contans® against *Sclerotinia sclerotiorum* on lettuce. The results (table) indicate a major reduction of vital sclerotia in the soil as well as a reduction of the amount of diseased plants.

Table: Results of official tests on lettuce

Locations	Percentage of remaining sclerotia after trial		Percentage of diseased plants	
	with Contans®	without Contans®	with Contans®	without Contans®
Rostock	1,3 %	57 %	8,6 %	34,3 %
Hannover	8 %	31 %	0 %	5 %
Magdeburg	not detected	not detected	2 %	6 %
Hamburg	1,8 %	38,2 %	no disease attack	no disease attack
Berlin	not detected	not detected	3 %	12 %

Shelf Life

The shelf life of Contans® depends directly on the viability of the spores of *C. minitans* in the formulation. In the freshly prepared preparation spore germinability is approximately 75 %. Therefore, to achieve 1×10^9 viable spores per gram of product, 1.33×10^9 spores per gram were incorporated into Contans®. A test has been carried out to investigate the shelf life. After 10 months at 23 °C and 4 °C respectively the viability of the formulated spores in the low temperature variation was more than 70 % (Fig. 3). That means the loss of viability was less than 20 %. So, storage for six months should provide an acceptable level of efficacy.

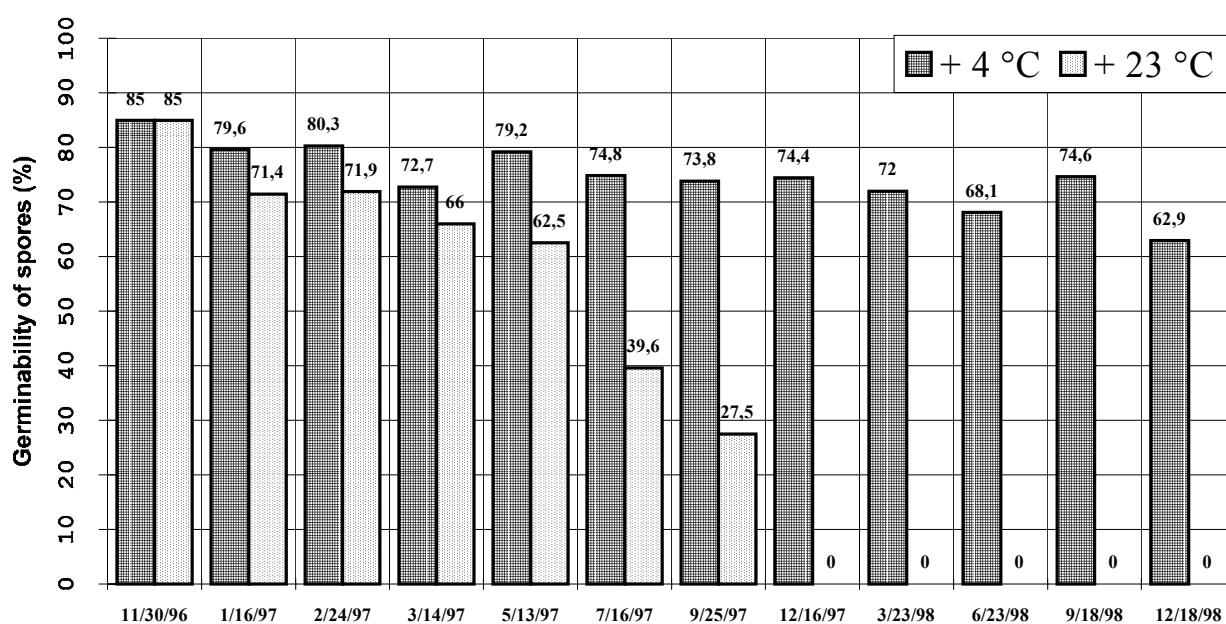


Fig. 3 Viability of the spores of *Coniothyrium minitans* in the Contans® formulation

Registration

Contans® is registered as a fungicide in Germany, Switzerland, Austria, France, Poland, Luxembourg, the Netherlands, Denmark, Italy, Spain, USA and Mexico. The European registration the application for the inclusion of the active substance into Annex 1 of Directive 914/414/EEC has been approved.

Patent Protection

German Patent (DE 19502065 A1, 7 December 1995) and United States Patent (5,766583, 16 June 1998) have been awarded for use of the strain of *Coniothyrium minitans*, CON/M/91-08, as a biological control agent against *Sclerotinia sclerotiorum* and *Sclerotinia minor*.

PCT application (DE95/00926) was filed covering Europe and Canada.

Another patent application to protect a new filter technique necessary for the separation of the pure spores from the fungus culture substrate was submitted in April 1997.

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