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Global collection of Mushroom Pathogens

Subtitle

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Productschap Tuinbouw

International Society for Mushroom Science

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1. Summary

The global mushroom industry is in need for effective diagnostic methods. These methods will help growers to take the right prevention measures and allow researchers to recognise new pathogens at an early stage. Effective diagnostic methods can only be developed if researchers have enough isolates of the pathogen at their disposal and if they are able to exchange methods. At the workshop of the Global Mushroom Disease Diagnostic Initiative, held in 2008 just before the ISMS conference in Cape Town, it was decided to try and build a collection of reference pathogen strains for the white button mushroom diseases present world wide. This collection can play a vital role in the development of diagnostic methods. In this project, researchers from South Africa (University of Pretoria), Ireland (Teagasc), New Zealand (CFR), US (Penn State University), Australia (University of Sydney) and the Netherlands (Wageningen University) combined relevant isolates into a fungal collection. Duplicates of this collection are located in Pretoria and Wageningen. This collectie contains isolates of fungal diseases collected from the main button mushroom producing countries (worldwide) en is accessible to scientific researchers that aim to develop diagnostic methods.

The Global Collection of Mushroom Pathogens currently contains 28 strains of *Cladobotryum*. Two of them have been identified as *Cladobotryum dendroides*, 5 of them have been identified as *Cladobotryum mycophilum* and the remaining 21 have been identified as being *Cladobotryum* strains (no identification to species level). Especially strains from China (the biggest mushroom producer) are lacking.

The Global Collection of Mushroom Pathogens currently contains 10 strains of *Mycogone perniciosa*. Four of them originate from Europe, one originates from China, three originate from South Africa and two originate from USA. Strains originating from Australia are lacking.

The Global Collection of Mushroom Pathogens currently contains 12 strains of *Trichoderma aggressivum*. Six of them originate from Europe, three originate from South Africa and three originate from USA. Strains originating from Australia and China are lacking. With respect to the other Trichoderma species, the Global Collection of Mushroom Pathogens currently contains 15 strains of *T. harzianum*, 11 strains of *T. atroviride* and 6 strains of *T. hamatum*. For the species *T. asperellum, T. aureoviride*, *T. citrinoviride, T koningii, T. longibrachiatum, T. saturnisporum* and *T. virens*, one single isolate is present. Next to this the collection contains 10 strains of uncharacterised *Trichoderma* species, all isolated from infected mushroom crops.

The Global Collection of Mushroom Pathogens currently contains 43 strains of *Verticillium* species (Figure 6). From Europe, 16 strains are present; 10 strains of *V. fungicola* var. *fungicola*, 3 strains of var. *aleophilum*, 1 strain of var. *flavidum*, 1 strain of *V. lamellicola* and 1 strain of *V. psalliotae*. From North America, 7 strains of *V. fungicola* var. *aleophilum*, 1 strain of *V. psalliotae* and 2 strains of *Verticillium fungicola* (without designation of variety) are present. From South Africa, 5 strains of *V. fungicola* var. *fungicola* and 2 strains of *Verticillium fungicola* (without designation of variety) are present. From Australia, 10 strains of *V. fungicola* var. *aleophilum*, are present. There are no strains available from China.

New strains can be added to the collection by offering them either to the South African National Collection of Fungi (PPRI) or the Department of Microbiology and Plant Pathology, University of Pretoria. Strains deposited in the South African collection will be shared with the mirror collection in Wageningen in the Netherlands. By doing so the full collection will always be available on two locations.

Interested parties can obtain strains from the Global Collection of Mushroom Pathogens. Strains will only be handed out by PPRI and/or the Department of Microbiology and Plant Pathology, University of Pretoria after under the terms of a material transfer agreement.

2. Introduction

In many places in the world, increasingly less chemical crop protection agents are available for use in mushroom cultivation. As a consequence, mushroom cultivation will loose the ability to use crop protection agents. As a consequence, good hygiene management, early detection and monitoring of pathogens (diagnostics), alternative crop protection agents and disease resistant mushroom varieties have to play an important role.
The number of researchers in several mushroom producing countries is decreasing already for a number of years. For an effective use of research funds, international collaboration on topics of mutual interest is important.

At the workshop of the Global Mushroom Disease Diagnostic Initiative, held in 2008 just before the ISMS conference in Cape Town, it was decided to try and build a collection of reference pathogen strains for the white button mushroom diseases present world wide. This collection will play a vital role in the development of diagnostic methods for mushroom diseases. This project was funded in part by the ISMS and in part by the Dutch Horticultural Board.

The Global Mushroom Disease Diagnostic Initiative is one of the results of a project funded by the Australian Mushroom Growers Association, titled Development of a disease monitoring system for the Australian mushroom industry". The goal of the Global Mushroom Disease Diagnostic Initiative, which is fostered by the ISMS, is to facilitate the development of diagnostic methods by:

- Facilitating exchange of knowledge
- Organise "face-to face" meeting whenever necessary
- Looking of opportunities for joint financing of projects
- Report to the industries of participating countries in a regular basis (thus showing the benefits of coöperation).

As mushroom industry in Australia is mainly focussed on cultivation of the button mushroom (*Agaricus bisporus*), the collection of mushroom pathogens is mainly focussed on fungal pathogens of *Agaricus bisporus*.

3. Method

In cooperation with the members of the Global Mushroom Disease Diagnostic Initiative (GMDDI) a list is made of pathogens (and numbers of isolates per pathogen) to be gathered in the collection. Next to this protocols will be developed for either sending out isolates from, or taking up isolates into the collection.
After this, samples for different countries were sent to the Mushroom Research Group, Plant Breeding Wageningen UR, checked for their status as a pure culture and stored in liquid nitrogen. After storage in liquid nitrogen a sample of each strain was revived from liquid nitrogen to check its survival. A duplicate of the collection is stored at the University of Pretoria.

4. Participating institutes and contact persons

The insitutes and contact persons participating in building and maintaining the Global Collection of Mushroom Pathogens are:

Plant Breeding
Postbus 386
6700AJ Wageningen, the Netherlands
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Lunnon Road, Hillcrest, 0083, South Africa
Contact personen Werner Rossouw (Werner.rossouw@up.ac.za)
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ARC Plant Protection Research Institute
Private Bag X134
Queenswood0001, South Africa
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5. Description of the collection.

Diseases in mushroom cultivation can be caused by viral, bacterial and fungal pathogens. The Global Collection of Mushroom Pathogens to date only contains fungal pathogens; 28 strains of *Cladobotryum* species, 10 strains of *Mycogone perniciosa*, 60 strains of *Trichoderma* species and 43 strains of *Lecanicillium/Verticillium* species. So currently the collection harbors 141 fungal pathogen strains. For details about the specific strains see Appendix I.

5.1 Geographical distribution of button mushroom production.

To estimate whether the collection of fungal pathogen strains is covering all relevant geographical regions, we first need to know what the main production area's of button mushrooms are. Sonnenberg et al. (2011) gives an overview of the main production areas. The main figures are summed in Table 1 and Figure 1. When looking at geographical area's, it can clearly be seen that the bulk of button mushroom production takes place in China

Table 1. Global production of button mushroom (*Agaricus bisporus*) in 2009.

Geographical area	X 1000 kg
China (mainly in provinces Jiangsu, Guangxi, Fujian, Shandong and Henan)	2.181.053
Europe (including countries; Poland, Netherlands, France, Spain, Italy, Germany, Ireland, UK, Belgium/Luxembourg, Hungary, Denmark, Austria, Romania, Serbia, Slovakia, Croatia, Bosnia, Macedonia, Bulgaria, Russia, Ukraine)	1.056.900
USA	356.936
South Korea	190.000
Australia	61.000
South Africa	20.000
New Zealand	8.500

Source: Sonnenberg et al. (2011)

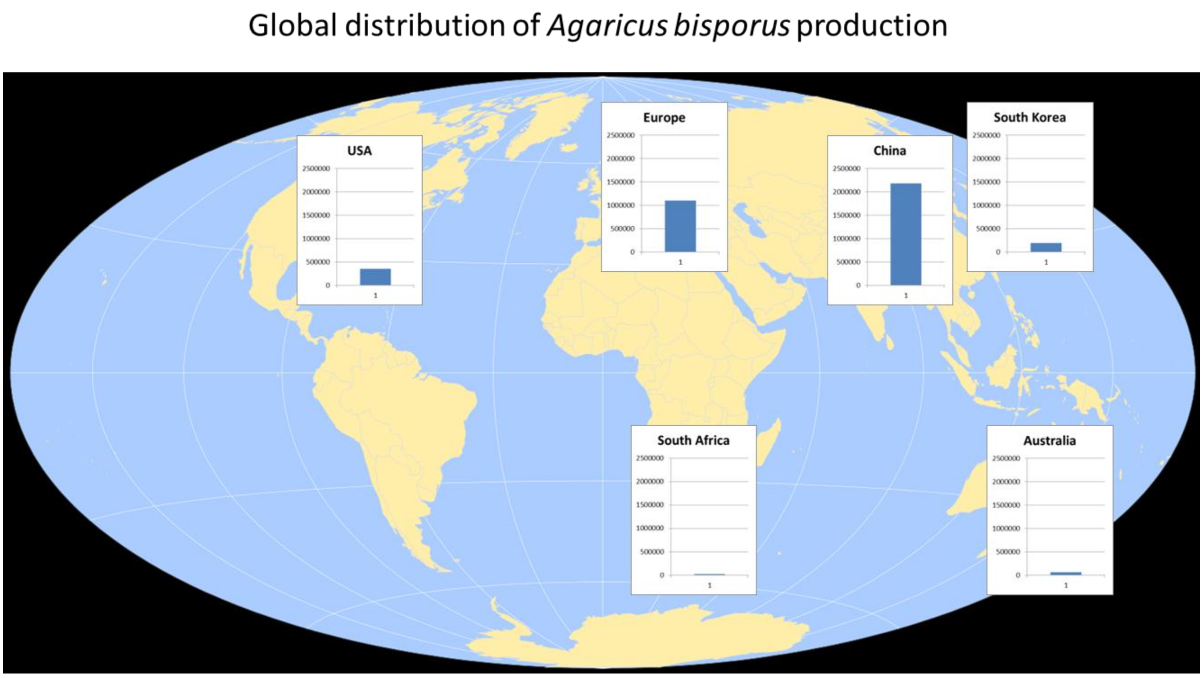

Figure 1. Global distribution of the main production area's of button mushrooms (*Agaricus bisporus*) in 2009. Adapted from Sonnenberg *et al.*, 2011)

and the various European countries. There is only limited information on the geographical spread of button mushroom production within China.
Within Europe, Poland, the Netherlands, France and Spain are major producers (Table 2). Other large sites of

Table 2. Mushroom production within Europe (Adapted from Sonnenberg et al., 2011).

Country	X 1000 kg
Poland	250000 (1)
Netherlands	230000 (1)
France	102400 (1)
Spain	93500 (1)
Italy	64500 (1)
Germany	58000 (1)
Ireland	55000 (1)
UK	43000 (1)
Bulgaria	3000 (2)
Belgium/Luxembourg	34000 (1)
Hungary	20000 (1)
Denmark	2300 (1)
Austria	700 (1)
Russia	9751 (2)
Ukraine	30000 (2)
Others	24000 (1)

(1) Figures from Groupement Européen de Producteurs du Champignon (GEPC)

(2) Dedicated estimates

mushroom production are USA, South Korea, Australia and South Africa. In USA two main production areas can be distinghuished; Pennsylvania and California. Next to button mushroom, also other mushroom species are being cultivated. FAOStat gives an overview of mushroom production in different countries (regardless the species produced, see Appendix II).

In summary we can say that mushroom production is mainly focussed in China, the European countries, North America, Australia and South Africa. Ideally, the Global Collection of Mushroom Pathogens should contain isolates from each of these geographical regions.

5.2 *Cladobotryum* species.

Cobweb disease is caused by Cladobotryum species (Fletcher & Gaze, 2007; Geels et al., 1988). *Cladobotryum dendroides* (syn*. Dactylium dendroides*) which is the conidial state of *Hypomyces rosellus* has historically been considered to be the commonest cause. *Cladobotryum mycophilum*, the conidial state of *Hypomyces odoratus,* is also commonly found in Europe, North America and South Africa. Next to this also *Cladobotryum varium* (teleomorph, Hypomyces aurantius) has been described as inciting cobweb disease of mushrooms (McKay et al. 1999).
The Global Collection of Mushroom Pathogens currently contains 28 strains of *Cladobotryum*. Two of them have been identified as *Cladobotryum dendroides*, 5 of them have been identified as *Cladobotryum mycophilum* and the remaining 21 have been identified as being *Cladobotryum* strains (no identification to species level). Global distribution of the different *Cladobotryum* strains is shown in Figure 2. There are ample strains present from North America and South Africa. The number of strains originating from Europe and Australia is limited. From the biggest button mushroom producing country (China), no strains of *Cladobotryum* are present.

Geographical distribution of *Cladobotryum* isolates

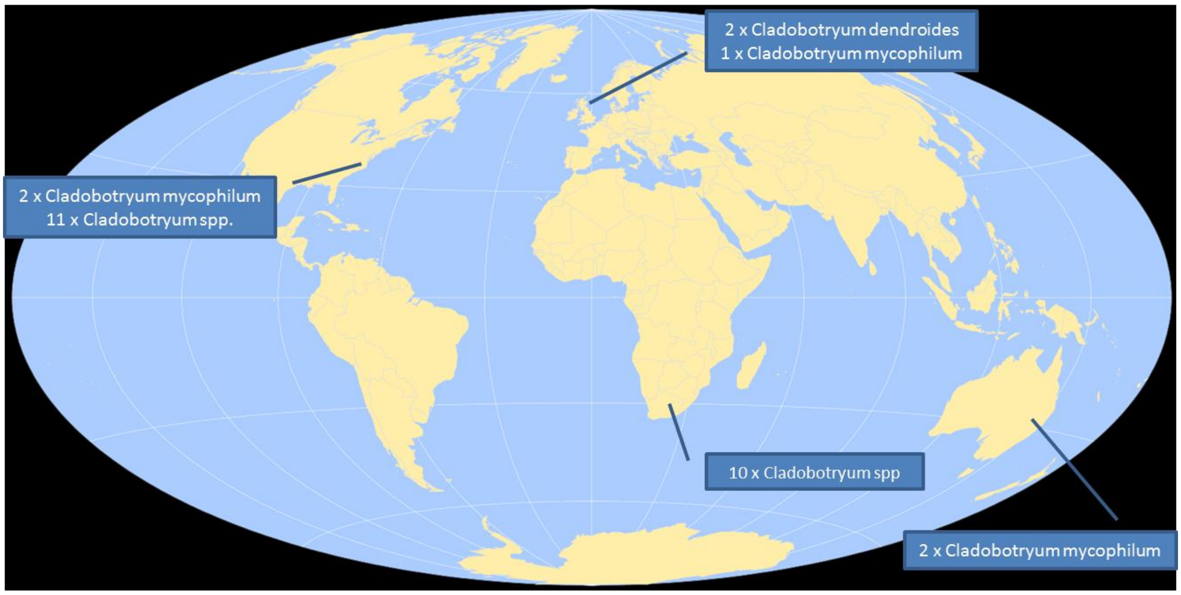

Figure 2. Global distribution of *Cladobotryum* strains that are present in the Global Collection of Mushroom Pathogens.

5.2.1 How well are these strains characterised?

The English isolates MES 13880 and 13881 (both *C. dendroides*) and isolate MES 13875 (*C. mycophilum*) have been determined at the International Mycological Istitute (IMI, Egham). The 2 Australian isolates of *C. mycophilum* (MES 13876 and 13877) have been identified by dr. Rodoni (Victoria DPI). All other isolates have no other identification than being *Cladobotryum* species isolated from diseased button mushroom cultures.
McKay et al. (1999) have compared morphological identification of 44 isolates of *Cladobotryum* species (many of which identified as *C. dendroides*) with a molecular identification using ITS sequences. Out of 29 isolates identified on morphological characteristics as *C. dendroides*, 27 were identified as *C. mycophilum* on basis of ITS sequences. From this it can be concluded that mis-identification is a real risk for *Cladobotryum dendroides*.
For molecular identification of *C. mycophilum*, 12 sequences of the ITS region are available at GenBank. *Cladobotryum dendroides* is the anamorph synonym of *Hypomyces rosellus* and can therefore be found under that name in GenBank. There are 28 sequences of *Hypomyces rosellus* : 10 for the partial RPB2 gene for RNA polymerase II subunit, 8 for the ITS region, 3 for the 28S rRNA and 5 for the partial tef gene for translation elongation factor 1-alpha.

5.2.2 Presence in other fungal collections?

Fungal strains can be found through the websites of the main fungal culture collections. An overview of presence of *Cladobotryum* strains in other fungal collections is given in Table 9. In some cases it is possible to link identical strains in different collections.

5.2.3 Scientific publications on selected strains within the collection

Strains MES 13880, 13881 and 13875 have been used in a study on fungicide resistance among *Cladobotryum* species (Grogan & Gaze, 2000). In this publication, strain 13880 is strain 187 (*C. dendroides* Type I, weakly resistant to thiabendazole). Strain 13881 is strain 164 (*C. dendroides* Type II, strongly resistant to thiabendazole). Strain 13875 is strain 222 (*C. mycophilum*, weakly resistant to thiabendazole).
None of the other *Cladobotryum* strains in the Global Collection of Mushroom Pathogens has ever been used in a publication.

Table 3. Presence of *Cladobotryum* strains pathogenic to *Agaricus bisporus* in other collections.

Species	This collection	Other collections	Region of origin	Remarks
C. dendroides	MES 13880	ATCC MYA-823[1]	UK	
C. dendroides	MES 13881	ATCC MYA 824[1]	UK	
C. dendroides		ATCC MYA 825[1]	UK	
C. dendroides		ATCC 204285[1]	USA	
C. dendroides		ATCC 32056[1]	Germany	
C. dendroides		ATCC 32057[1]	Germany	
C. dendroides		ATCC 62581[1]	India	
C. dendroides		IMI 359310[2]	UK	
C. dendroides		IMI 372795[2]	UK	
C. dendroides		MUCL 28202[4]	Belgium	EMBL: Y17092
C. dendroides		MUCL 1084[4], CBS 233.34[5]	USA	EMBL: Y17090
C. mycophilum	MES 13875	ATCC MYA 821[1]	UK	
C. mycophilum		ATCC MYA 822[1]	UK	
C. mycophilum		ATCC 204286[1]	Ireland	
C. mycophilum		IMI 267134	UK	EMBL: Y17095 & Y17101
C. mycophilum		IMI 372796[2]	UK	
C. mycophilum		IMI 368694[2]	UK	
C. mycophilum		DSM No.: 11940[3]	?	
C. mycophilum		MUCL 166	Belgium	
C. mycophilum		CBS 148.46	Canada	Y17100
C. mycophilum		CBS 818.69	Netherlands	Y17096 and Y17104
C. mycophilum		CBS 111.92	Germany	Y17098
C. varium		ATCC 36807[1]	?	
C. varium		ATCC 36827[1]	?	
C. varium		ATCC 36828[1]	?	
C. varium		ATCC MYA 466[1]	Germany	
C. varium		ATCC 248634[1]	India	
C. varium		MUCL 30376	?	Y17087
C. varium		MUCL 8223	Canada	Y17088

[1]ATCC: http://www.lgcstandards-atcc.org/?geo_country=nl
[2]CABI: http://www.cabi.org/default.aspx?site=170&page=3160
[3]DSMZ: http://www.dsmz.de/catalogues/catalogue-microorganisms.html
[4]BCCM (http://bccm.belspo.be/index.php)
[5]CBS (http://www.cbs.knaw.nl/databases/),
No strains of Cladobotryum were found in at the ICMP (http://scd.landcareresearch.co.nz/Search/Search/ICMP)

5.2.4 Conclusion

A relatively large number *Cladobotryum* strains are present in the collection. However, many of them have not been properly identified by molecular techniques. Next to this there appears to be an overrepresentation of strains from North America and South Africa. Strains from the European countries are present in the collections of ATCC, CABI and DSMZ. Strains of *Cladobotryum* species from China are not present in easily accessible culture collections. They may need to be sampled or obtained from Chinese scientists. Recently the presence of *Cladobotryum mycophilum* was reported in a crop of *Agaricus bisporus* in Korea (Back et al., 2010). The isolate (AB527074) was identified by amplification and sequencing of the ITS region. Perhaps this isolate may also be available.

5.3 *Mycogone* species.

Wet bubble disease is caused by the fungus *Mycogone perniciosa.* Its symptoms are well described by Geels et al. (1988) and Fletcher & Gaze (2007). A second species*, Mycogone rosae*, has also been associated with diseased mushrooms in the UK (Williams, 1939). *M. perniciosa* causes distortion of affected mushrooms and the characteristic undifferentiated lumps of tissue. *M rosae* produces spots or depressed areas on the cap surface, brown in colour, but with yellow discoloured tissue surrounding the spots (Williams. 1939). According to Fletcher & Gaze (2007) this pathogen is rarely seen in mushroom cultivation. There are no strains of *Mycogone rosae* present in the collection.
The Global Collection of Mushroom Pathogens currently contains 10 strains of *Mycogone perniciosa*. Four of them originate from Europe, one originates from China, three originate from South Africa and two originate from USA. Global distribution is shown in Figure 3. The *Mycogone perniciosa* strains are evenly divided over the main mushroom producing area's. However, strains originating from Australia are lacking.

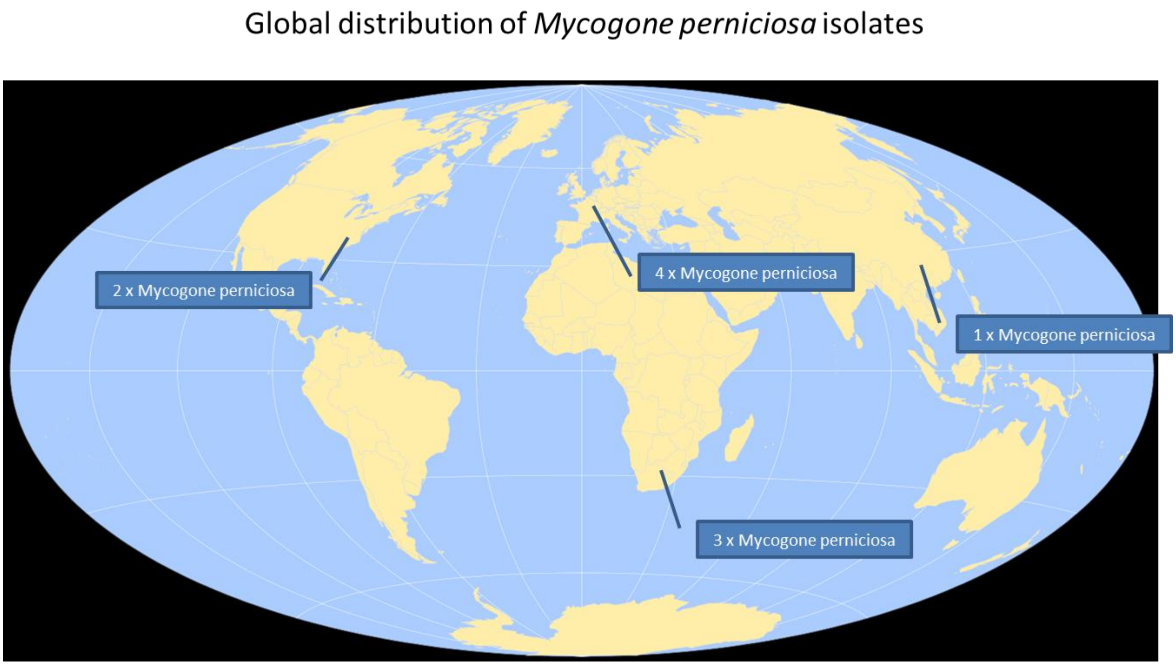

Figure 3 Global distribution of *Mycogone perniciosa* strains that are present in the Global Collection of Mushroom Pathogens

5.3.1 How well are these strains characterised?

All isolates of *Mycogone perniciosa* have been identified morphologically. *M. perniciosa* produces small thin-walled phialoconidia on Verticillium-like conidiophores together with much larger bicellular conidia (aleuriospores) that

5.3.2 Presence in other fungal collections?

Fungal strains can be found through the websites of the main fungal culture collections. An overview of presence of *Mycogone* strains in other fungal collections is given in Table 10. In some cases it is possible to link identical strains in different collections.

Table 4. Presence of Mycogone strains pathogenic to Agaricus bisporus in other collections.

Species	This collection	Other collections	Region of origin	Remarks
M. perniciosa	MES 11238	CBS 234.29[5]	Germany	
M. perniciosa	MES 11237	CBS 322.52[5]	Netherlands	
M. perniciosa	MES 11240	CBS 648.82[5]	Belgium	GenBank: FJ904634.1
M. perniciosa		MUCL 18464[4]	France	
M. perniciosa		MUCL 18993[4]	Belgium	
M. perniciosa		MUCL 28867[4]	Belgium	
M. perniciosa		DSM No.: 2339[3]	Germany	
M. perniciosa		ATCC 96432[1]	UK	
M. perniciosa		ATCC 96437[1]	Ontario, Canada	
M. perniciosa		ATCC 10934[1]	?	
M. perniciosa		ATCC 96436[1]	Ohio, USA	PSU 54
M. perniciosa		ATCC 96429[1]	Shanghai, China	
M. perniciosa		ATCC 42350[1]		GCRI 21,Gandy, Sci. Hortic. 8: 307-313, 1978
M. perniciosa		ATCC 60269[1]	Denmark	K Bech, Tidsskr. Planteavl. 86: 141-150, 1982
M. perniciosa		ATCC 96435[1]	PA, USA	PSU 53
M. perniciosa	MES 13944	ATCC 96439[1]	WA, USA	PSU 181
M. perniciosa	MES 13939	ATCC 96421[1]	Shanghai China	
M. perniciosa		ATCC 96425[1]	Shanghai China	
M. perniciosa		ATCC 96440[1]	Shanghai China	
M. perniciosa		ATCC 96426[1]	Fujian, China	
M. perniciosa		ATCC 22226[1]	PA, USA	
M. perniciosa		ATCC 96433[1]	Fujian, China	
M. perniciosa		ATCC 60268[1]	Denmark	K Bech, Tidsskr. Planteavl. 86: 141-150, 1982
M. perniciosa		ATCC 96434[1]	NY, USA	PSU 52
M. perniciosa		ATCC 96431[1]	Shanghai, China	
M. perniciosa		ATCC 96424[1]	Fujian, China	Strain 11
M. perniciosa		ATCC 96422[1]	Shanghai, China	Strain 3
M. perniciosa		ATCC 96438[1]	PA, USA	PSU 180
M. perniciosa		ATCC 96430[1]	Zhejiang, China	Strain 23
M. perniciosa		ATCC 96427[1]	UK	
M. perniciosa		IMI 342299[2]	UK	
M. perniciosa		IMI 359311[2]	UK	
M. rosae		MUCL 18465[4]	France	
M. rosae		CBS 132.97[5]	France	
M. rosae		CBS 527.80[5]	Germany	
M. rosae		CBS 563.78B[5]	Russia	
M. rosae		CBS 563.78A[5]	Russia	
M. rosae		CBS 488.67[5]	Poland	
M. rosae		IMI 147442[2]	?	
M. rosae		ICMP 5441[6]	New Zealand	

[1]ATCC: http://www.lgcstandards-atcc.org/?geo_country=nl
[2]CABI: http://www.cabi.org/default.aspx?site=170&page=3160
[3]DSMZ: http://www.dsmz.de/catalogues/catalogue-microorganisms.html
[4]BCCM (http://bccm.belspo.be/index.php)
[5]CBS (http://www.cbs.knaw.nl/databases/),
[6]ICMP (http://scd.landcareresearch.co.nz/Search/Search/ICMP)

develop on short, lateral hyphae and consist of a dark, spherical thick-walled, verrucose apical cell situated on a thin-walled basal cell (Glamoclija et al., 2008). These features allow a relatively easy identification. With respect to possibilities of molecular identification of *Mycogone perniciosa*, 12 sequences of the ITS region have been deposited in Genbank by Sharma,V.P. and Singh,S.K. These sequences are the result of an unpublished study on etiology and molecular characterization of wet bubble disease causing fungus *Mycogone perniciosa* in *Agaricus bisporus.*

5.3.3 Scientific publications on selected strains within the collection

Out of the 10 isolates of *Mycogone perniciosa*, only one appears to be used in published research. Meyer & Korsten (2008) published a nested PCR method for detection of *M. perniciosa* and used MES 11240 as a reference strain. Next to the reference strain they used 10 South African isolates of *M. perniciosa* but these are not in the Global Collection of Mushroom Pathogens.

5.3.4 Conclusion

Compared to the large numbers of *Cladobotryum* strains, *Lecanicillium* strains and *Trichoderma* strains, a total of 10 strains of *Mycogone perniciosa* appears quite low. For each geographical location, there are only 2 to 4 isolates. For the Chinese region there is only one isolate available. For the Australian continent, *Mycogone perniciosa* strains are even lacking. Chinese isolates of *Mycogone perniciosa* appear to be present in the ATCC collection. However, strains of Australian origin do not appear to be present in culture collection. It may be necessary to collect them.

5.4 *Trichoderma* species.

A number of different species and strains of *Trichoderma,* producing a range of symptoms are found in mushroom culture (Geels et al. 1988; Fletcher & Gaze (2007). The species most frequently associated with the mushroom crop are *Trichoderma atroviride*, *T. aureoviride*, *T. hamatum*, *T. harzianum*, *T. inhamatum*, *T. koningii*, *T. longibrachiatum*, *T. pseudokoningii*, *T. viride*, and *T. virens*. However, the most devastating *Trichoderma* species with respect to loss of yield is *Trichoderma aggressivum*.

Global distribution of *Trichoderma aggressivum* isolates

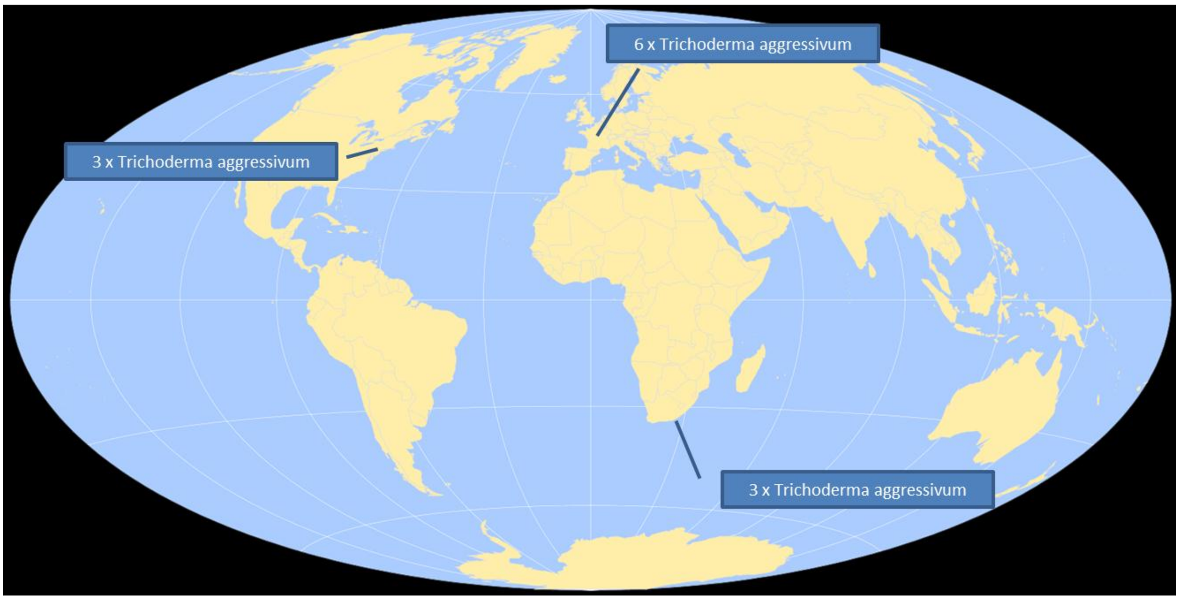

Figure 4. Global distribution of *Trichoderma aggressivum* strains that are present in the Global Collection of Mushroom Pathogens

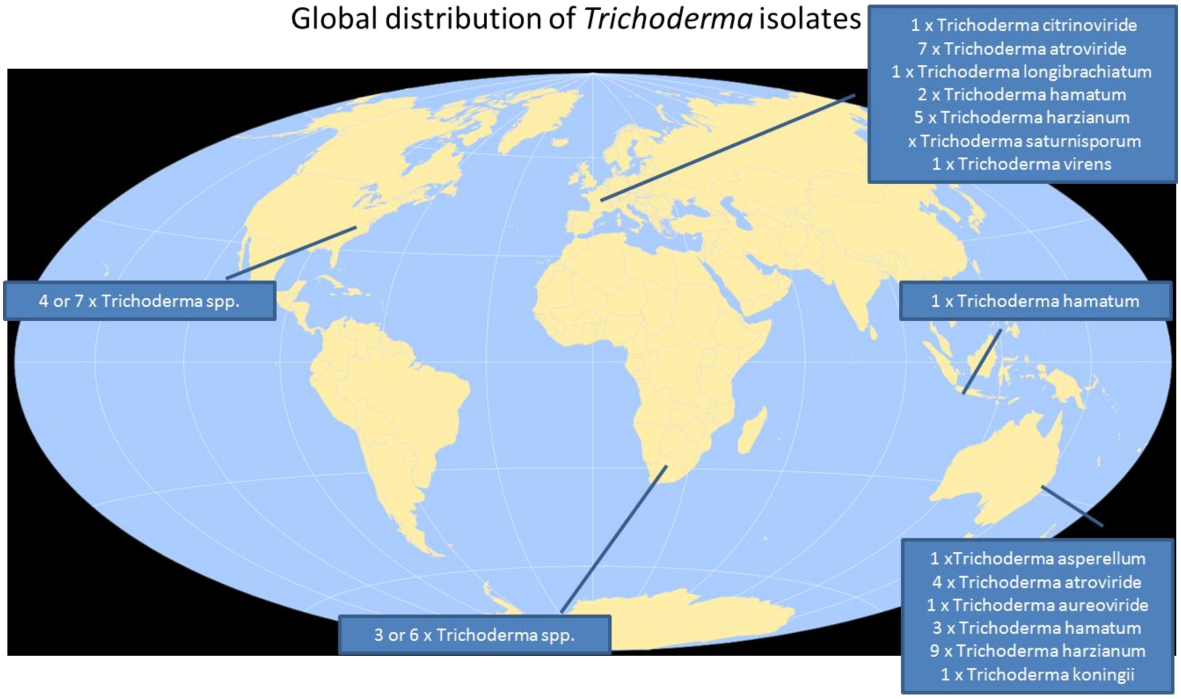

Figure 5 Global distribution of *Trichoderma* strains that are present in the Global Collection of Mushroom Pathogens

The Global Collection of Mushroom Pathogens currently contains 12 strains of *Trichoderma aggressivum*. Six of them originate from Europe, three originate from South Africa and three originate from USA. Global distribution is shown in Figure 4. The *T. aggressivum* strains are evenly divided over the main mushroom producing area's. However, strains originating from Australia and China are lacking. However, to this date there are no reports of *Trichoderma aggressivum* causing problems in Australian or Chinese crops.

With respect to the other Trichoderma species, the Global Collection of Mushroom Pathogens currently contains 15 strains of *T. harzianum*, 11 strains of *T. atroviride* and 6 strains of *T. hamatum*. For the species *T. asperellum, T. aureoviride*, *T. citrinoviride, T koningii, T. longibrachiatum, T. saturnisporum* and *T. virens*, one single isolate is present. Next to this the collection contains 10 strains of uncharacterised *Trichoderma* species, all isolated from infected mushroom crops. Global distribution of these other *Trichoderma* species is shown in Figure 5.

The *T. harzianum* strains originate mainly from Europe (Netherlands, 4 strains; Belgium, 1 strain; Italy, 1 strain) and Australia (9 strains). Also the *T. atroviride* strains originate mainly from Europe (Netherlands, 4 strains, Germany, 2 strains, Belgium, 1 strain) and Australia (4 strains). The *T. hamatum* strains originate from Europe, Indonesia and Australia. The other species of Trichoderma weed molds also originate from Europe or Australia.

The uncharacterised *Trichoderma* isolates, on the other hand, predominantly originate from South African and USA.

5.4.1 How well are these strains characterised?

Trichoderma strains are often difficult to identified by morphological characteristics. Next to this, in an ongoing project the taxonomy is being revised. In summary it is difficult to discriminate between species and the names of the species are changing. With respect to *Trichoderma* species that are causing problems in button mushroom cultivation a major study was performed by Samuels et al. (2002), which resulted in a taxonomic key to *Trichoderma* species commonly found associated with commercially grown *A. bisporus*. In this study its was shown that *T. aggressivum* was strongly separated from their closest relative, the morphologically similar *T. harzianum*, by molecular sequences of the ITS-1 region of nuclear rDNA and a fragment of translation elongation factor gene (EF-lα). Based on morphological characters, *T. aggressivum* can be distinguished from *T. harzianum* and *T. atroviride* most readily by rate of growth. Of these, only *T. harzianum* grows well and sporulates at 35°C, while *T. atroviride* is the slowest growing. *Trichoderma aggressivum* f. *aggressivum* and f. *europaeum* are effectively indistinguishable morphologically although they have subtly different growth rates at 25°C on SNA and statistically significant

micromorphological differences. In summary this means that identifying strains of *Trichoderma* species on basis of morphological characteristics is difficult. Molecular data appear to be more helpful.
The taxonomy of Trichoderma species is under revision by the International Subcommission on Trichoderma and Hypocrea Taxonomy (http://isth.info/). Their website presents a collection of easy tools for quick molecular identification of Hypocrea/Trichoderma species.
Out of the 11 *T. aggressivum* strains in the Global Collection of Mushroom Pathogens, 6 have been identified by using DNA sequences. The remaining 5 strains have been identified morphologically.
Out of the 15 strains of *T. harzianum*, the 6 European isolates have been identified by using DNA sequences. The remaining 9 Australian isolates have been identified morphologically.
Out of the 11 strains of *T. atroviride*, 6 European isolates have been identified using DNA sequences. A seventh European isolate has been identified using molecular techniques (ITS-RFLP). The remaining 4 Australian isolates have been identified using morphological techniques.
Next to this, 3 strains of *T. hamatum* and the isolates of *T. citrinoviride, T. longibrachiatum, T. saturnisporum* and *T. virens* have been identified using DNA sequences.

5.4.2 Presence in other fungal collections?

Fungal strains can be found through the websites of the main fungal culture collections (see http://www.cabri.org/ for a convenient aggregate site). As *Trichoderma* species are quite ubiquitous, you will find them in many culture collections. For instance, *T. harzianum* and *T atroviride* have been isolated not only from diseased mushroom crops but also from many other substrata. Because of the large number of *Trichoderma* strains in other collections, only *Trichoderma aggressivum* strains are listed in Table 11. *T. aggressivum* can be considered more or less as specific for mushroom cultivation, although Table 11 shows that it was also isolated from a water treatment system. In some cases it is possible to link identical strains in different collections. Remarkably, no *T. aggressivum* strains were found in the collections of MUCL, DSMZ, ICMP and ATCC.

Table 5. Presence of Trichoderma aggressivum strains in other collections.

Species	This collection	Other collections	Region of origin	Remarks
T. aggressivum	MES 11410	CBS 450.95[1]	Canada	Genbank: AF359260, AF400265
T. aggressivum	MES 11431	CBS 100528[1]	PA, USA	
T. aggressivum	MES 11433	CBS 100526[1]	Ireland	GenBank: AF400265, FJ442434, AF348096
T. aggressivum		CBS 433.95[1]	UK	GenBank: AF348097
T. aggressivum		CBS 100525[1]	UK	rDNA sequences ITS : DQ328879, DQ328903 Actin : AF442826, FJ442433 RPB2 : AF545541 Elongation Factor 1 : AF348095, AF534614
T. aggressivum		CBS 689.94[1] = IMI359824	UK	GenBank: AF348089
T. aggressivum		CBS 100527[1]	PA, USA	rDNA sequences ITS : DQ328880, DQ328904
T. aggressivum		CBS 123943[1]	USA	
T. aggressivum		CBS 115901[1]	Israel	Isolated from water treatment system, textile industry
T. aggressivum		CBS 123782[1]	PA, USA	
T. aggressivum		IMI 393969[2]	Ireland	AF348098 AF456924
T. aggressivum		IMI 393970[2]	USA	Genbank: AF348094 AF345950
T. aggressivum		IMI 393971[2]	Canada	

[1]CBS (http://www.cbs.knaw.nl/databases/),
[2]CABI: http://www.cabi.org/default.aspx?site=170&page=3160

5.4.3 Scientific publications on selected strains within the collection

The *Trichoderma aggressivum* isolates shown in Table 12, have been used in a number of studies.

Collection nr	Other collections	References
MES 11410	CBS 450.95	Samuels et al., 2002; Muthumeenakshi et al., 1994; Kullnig-Gradinger et al., 2002, Hatvani et al., 2007, Szczech et al., 2008, Kredics et al (2009); Antal et al., 2005
MES 11431	CBS 100528	Samuels et al., 2002; Hatvani et al., 2007; Antal et al., 2005
MES 11433	CBS 100526	Samuels et al., 2002; Hatvani et al., 2007, Sobieralski et al., 2009, 2010; Krause et al., 2006; Kredics et al., 2009; Antal et al., 2005

5.4.4 Conclusion

Trichoderma species that cause problems can be divided into two groups. The species most frequently associated with the mushroom crop are *Trichoderma atroviride*, *T. aureoviride*, *T. hamatum*, *T. harzianum*, *T. inhamatum*, *T. koningii*, *T. longibrachiatum*, *T. pseudokoningii*, *T. viride*, and *T. virens*. They can be considered as weed moulds. *T. aggressivum* stands out as the most devastating *Trichoderma* species with respect to loss of yield.
The Global Collection of Mushroom Pathogens contains quite some *T. aggressivum* isolates from Europe, North America and South Africa. From China and Australia there are no reports of problems with *T. aggressivum*. Based on the dispersed distribution of the strains over geographical regions, there is a chance that an adequate amount of genetic diversity is present in the collection.
For the “weed mould” Trichoderma species, it is less likely that enough genetic diversity is present in the collection. Except for of *T. harzianum*, *T. atroviride* and *T. hamatum* only one strain is present for most species. On the other hand, these species may be less interesting because they cause less damage.

5.5 *Lecanicillium / Verticillium* species.

Lecanicillium fungicola (Preuss) Zare and Gams [synonyms: *Verticillium fungicola* (Preuss) Hassebrauk, *Verticillium malthousei* (Preuss) Ware] is the causal agent of dry bubble disease (Geels et al., 1988; Fletcher & Gaze, 2007; Berendsen et al., 2010).
The Global Collection of Mushroom Pathogens currently contains 43 strains of *Verticillium* species (Figure 6). From Europe, 16 strains are present; 10 strains of *V. fungicola* var. *fungicola*, 3 strains of var. *aleophilum*, 1 strain of var. *flavidum*, 1 strain of *V. lamellicola* and 1 strain of *V. psalliotae*. From North America, 7 strains of *V. fungicola* var. *aleophilum*, 1 strain of *V. psalliotae* and 2 strains of *Verticillium fungicola* (without designation of variety) are present. From South Africa, 5 strains of *V. fungicola* var. *fungicola* and 2 strains of *Verticillium fungicola* (without designation of variety) are present. From Australia, 10 strains of *V. fungicola* var. *aleophilum*, are present. There are no strains available from China.

5.5.1 How well are these strains characterised?

Gams and Van Zaayen (1982) distinguished three varieties of *Verticillium fungicola*: var. *fungicola*, var. *aleophilum* and var. *flavidum*. Recently, it has been concluded that, on the basis of the internal transcribed spacer (ITS) region and small subunit rDNA sequences, *V. fungicola* is more closely related to the often insect-pathogenic species of the genus *Lecanicillium* than to the plant-pathogenic species of the genus *Verticillium* (Zare and Gams, 2008). *Verticillium fungicola* and its varieties *fungicola* and *aleophilum* were therefore renamed *Lecanicillium fungicola*. *Verticillium fungicola* var. *flavidum* was redefined as a separate species: *Lecanicillium flavidum*. The latter species differs from *L. fungicola* in ITS sequence, its optimum and maximum temperatures for growth, and, morphologically, in the repeated branching of its conidiophores (Zare and Gams, 2008). In general, it is var. *aleophilum* that affects crops in Canada and the USA, whereas, in Europe, var. *fungicola* is the main causal agent of the disease (Collopy et al., 2001; Largeteau et al., 2004).
The majority of the *Verticillium* strains in the Global Collection of Mushroom Pathogens has been identified morphologically. Collopy et al (2001) performed molecular phylogenetic analyses on a large number of isolates of

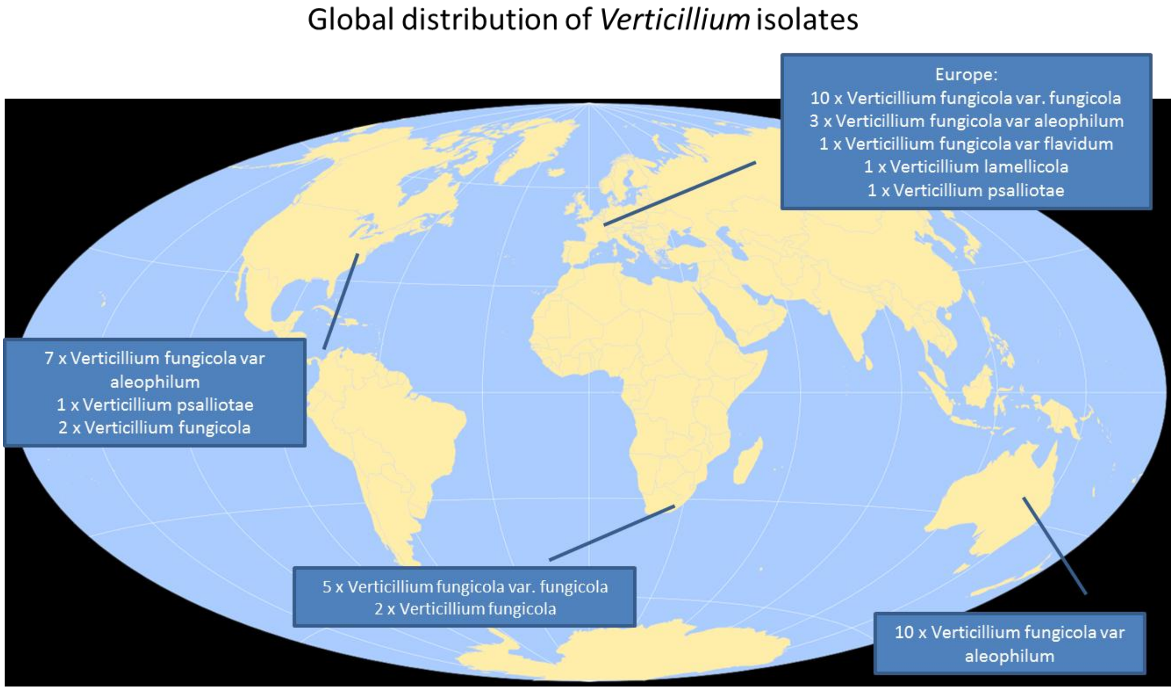

Figure 6 Global distribution of *Verticillium* strains that are present in the Global Collection of Mushroom Pathogens

Verticillium fungicola collected from various Pennsylvania mushroom farms and a large number of isolates of *Verticillium* spp. collected during the last 50 years from various geographic locations. Sequence analysis of internal transcribed spacers 1 and 2 (ITS1 and ITS2) and 5.8S regions of the nuclear ribosomal DNA (rDNA) transcriptional unit showed identical rDNA sequences were obtained for all Pennsylvania isolates studied and the ex-type strain of *V. fungicola* var. *aleophilum*. Sequence analysis of European isolates revealed a close relationship to the ex-type strain *V. fungicola* var. *fungicola*. No European-like isolates of *V. fungicola* var. *fungicola* were detected in the collection of North American isolates examined. Results from RAPD primers strongly indicate the presence of a clonal population of V. fungicola among Pennsylvania isolates.
Largeteau et al., (2006) analysed eighteen isolates of *V. fungicola* var. *fungicola* and four var. *aleophilum* isolates for DNA polymorphism. RAPD and AFLP markers delineated three French isolates from a homogeneous group containing the other var. *fungicola* isolates. This work emphasized that, like the American var. *aleophilum*, the var. *fungicola* in Europe is genetically homogeneous.
MES11451, MES11449, MES11525 and MES13869 were molecularly studied by Collopy et al. (2001). MES 11459, MES11449, MES 11454 and MES 11501 were molecularly studied by Largeteau et al. (2006).

5.5.2 Presence in other fungal collections?

Fungal strains can be found through the websites of the main fungal culture collections (see http://www.cabri.org/ for a convenient aggregate site). *Verticillium* species that are pathogens for *Agaricus bisporus*, can be found in many culture collections Table 12).

5.5.3 Scientific publications on selected strains within the collection

As mentioned above, MES11451, MES11449, MES11525 and MES13869 were molecularly studied by Collopy et al. (2001), while MES 11459, MES11449, MES 11454 and MES 11501 were studied by Largeteau et al. (2006).
MES11451, MES11449 and MES13869 have also been studied by Romaine et al. (2002) for development of a Verticillium fungicola specific PCR method.
MES13869 and MES11451 have been used in a phylogenetic study by Farrag et al. (2009).
MES 11451 was studied by Yokoyama et al (2004) for the development of a mating specific PCR method.

Table 6. Presence of Verticillium strains pathogenic to Agaricus bisporus in other collections.

Species	This collection	Other collections	Region of origin	Remarks
Verticillium fungicola var. aleophilum	MES 11451[5]	CBS 357.80	Netherlands	Gams & Van Zaayen, (1982), GENBANK AF324876
Verticillium fungicola var. fungicola	MES 11449	CBS 440.34 = ATCC 22607 = MUCL 9781	UK	Gams & Van Zaayen, (1982), Ware (1933), Collopy et al., 2001, Yokoyama et al., 2006, GENBANK AF324874, AB124635, AB107135.
Verticillium fungicola var. fungicola	MES 13868	CBS 194.79	UK	
Verticillium fungicola var. flavidum	MES 13869	CBS 342.80	France	AF324877, EF641878
Verticillium fungicola		ATCC 32843	USA (PA)	Wuest & Forer (1975), strain 01.
Verticillium fungicola		ATCC 32837	USA (PA)	Wuest & Forer (1975), strain B0.
Verticillium fungicola		ATCC 62492	?	
Verticillium fungicola		ATCC 26869	USA (PA)	Wuest et al. (1974), strain BC69
Verticillium fungicola		ATCC 32845	Switzerland	Wuest & Forer (1975), strain S1.
Verticillium fungicola		ATCC 18163	Tundra, Alaska	(Strain IFO 8578 [A-1-5])
Verticillium fungicola		ATCC 32838	USA (PA)	Wuest & Forer (1975), strain C7
Verticillium fungicola		ATCC 32844	USA (PA)	Wuest & Forer (1975), strain P5
Verticillium fungicola		ATCC 62493	UK	Strain 8A8
Verticillium fungicola		ATCC 32848	USA (PA)	Wuest & Forer (1975), strain 87
Verticillium fungicola		ATCC 32839	UK	Wuest & Forer (1975), strain E1
Verticillium fungicola		ATCC 32842	Korea	Wuest & Forer (1975), strain K2
Verticillium fungicola		ATCC 32847	USA (CA)	Wuest & Forer (1975), strain T5
Verticillium fungicola		ATCC 32841	USA (PA)	Wuest & Forer (1975), strain GB-LF-7014
Verticillium fungicola		ATCC 32849	USA (PA)	Wuest & Forer (1975), strain 90
Verticillium fungicola		ATCC 60267	Denmark	K Bech (1982) Tidsskr. Planteavl. 86: 141-150, strain 1682.
Verticillium fungicola		ATCC 26867	USA (CA)	Wuest et al. (1974), strain ML2
Verticillium fungicola		ATCC 32840	USA (PA)	Wuest & Forer (1975), strain G1
Verticillium fungicola		ATCC 22227	USA (PA)	
Verticillium fungicola		ATCC 32846	USA (PA	Wuest & Forer (1975), strain T1
Verticillium fungicola		ATCC 26868	USA (CA)	Wuest et al. (1974), strain ML4
Verticillium lamellicola		ATCC 58906		Kuter, (1984)
Verticillium lamellicola		ATCC 22614 = CBS 342.37	Germany	
Verticillium lamellicola		ATCC 22613 = CBS 343.37	?	
Verticillium lamellicola		ATCC 22602 = CBS 138.37	Germany	
Verticillium psalliotae		ATCC 18912	Canada	
Verticillium psalliotae		ATCC 16541 = CBS 650.85		HACC 188, Thirumalachar MJ. Antiamoebin, an anthelmintic and antiprotozoal antibiotic, and a method for producing the same. US Patent 3,657,419 dated Apr 18 1972

Verticillium psalliotae	ATCC 22608 = CBS 345.37	Germany	
Verticillium psalliotae	ATCC 22098 = CBS 463.70	Israel	
Verticillium psalliotae	ATCC 60266	Denmark	K Bech (1982) Tidsskr. Planteavl. 86: 141-150, strain 265
Verticillium psalliotae	ATCC 22609 = CBS 344.37	Germany	
Verticillium fungicola	IMI 188936	UK	
Verticillium fungicola	IMI 246427	UK	
Verticillium fungicola	IMI 246428	UK	
Verticillium fungicola	IMI 268001	Australia	
Verticillium fungicola	IMI 289231	UK	
Verticillium fungicola var. aleophilum	IMI 289232	UK	
Verticillium fungicola var. flavidum	IMI 289233	UK	
Verticillium fungicola	IMI 290546	UK	
Verticillium fungicola var. aleophilum	IMI 290953a	UK	
Verticillium fungicola	DSM No.: 2352	Germany	
Verticillium fungicola	MUCL 8126	France	
Verticillium fungicola	MUCL 8219	UK	
Verticillium fungicola	MUCL 21766	Belgium	
Verticillium fungicola	MUCL 34289	Germany	
Verticillium lamellicola	MUCL 594	Belgium	
Verticillium lamellicola	MUCL 11470	Sweden	
Verticillium lamellicola	MUCL 11536	Germany	
Verticillium lamellicola	MUCL 44211	Belgium	
Verticillium lamellicola	MUCL 45415	Belgium	
Verticillium psalliotae	MUCL 9800 = CBS 505.48 = IMI 92016	UK	
Verticillium psalliotae	MUCL 18310	Belgium	
Verticillium psalliotae	MUCL 31517	Argentina	
Verticillium psalliotae	MUCL 39924	?	
Verticillium psalliotae	MUCL 41064	Brazil	
Verticillium fungicola var. flavidum	CBS 530.81	Belgium	
Verticillium fungicola var. flavidum	CBS 649.82B	Belgium	
Verticillium fungicola var. flavidum	CBS 290.80	Germany	
Verticillium fungicola var. flavidum	CBS 238.80	USA (NY)	
Verticillium fungicola var. flavidum	CBS 879.73	Netherlands	
Verticillium fungicola var. flavidum	CBS 300.70D	Austria	
Verticillium fungicola var. aleophilum	CBS 501.89	Netherlands	

Verticillium fungicola var. aleophilum	CBS 507.81C	Netherlands	
Verticillium fungicola var. aleophilum	CBS 507.81B	Netherlands	
Verticillium fungicola var. aleophilum	CBS 507.81A	Netherlands	
Verticillium fungicola var. aleophilum	CBS 270.79B	?	yellow mutant of CBS 300.70A
Verticillium fungicola var. aleophilum	CBS 270.79A	?	albino mutant of CBS 300.70A
Verticillium fungicola var. aleophilum	CBS 300.70A	Australia	
Verticillium fungicola var. fungicola	CBS 648.80	Netherlands	
Verticillium fungicola var. fungicola	CBS 134154	Netherlands	
Verticillium fungicola	CBS 122449	Spain	
Verticillium fungicola	CBS 114332	Estonia	
Verticillium fungicola	CBS 112974	Finland	
Verticillium fungicola	ICMP 3343	New Zealand	
Verticillium fungicola	ICMP 3802	New Zealand	
Verticillium fungicola	ICMP 3848	New Zealand	
Verticillium fungicola	ICMP 3856	New Zealand	
Verticillium fungicola	ICMP 5508	New Zealand	
Verticillium fungicola	ICMP 5510		
Verticillium fungicola	ICMP 5511 = IFO 6624		
Verticillium fungicola	ICMP 5513 = IFO 6962		
Verticillium fungicola	ICMP 10403	New Zealand	
Verticillium fungicola	ICMP 10404	New Zealand	
Verticillium fungicola	ICMP 10405	New Zealand	
Verticillium fungicola	ICMP 10833	New Zealand	
Verticillium fungicola	ICMP 10834	New Zealand	
Verticillium fungicola	ICMP 16518	?	
Verticillium psalliotae	ICMP 2492	New Zealand	
Verticillium psalliotae	ICMP 5509	New Zealand	
Verticillium psalliotae	ICMP 5512	New Zealand	

[1]ATCC: http://www.lgcstandards-atcc.org/?geo_country=nl
[2]CABI: http://www.cabi.org/default.aspx?site=170&page=3160
[3]DSMZ: http://www.dsmz.de/catalogues/catalogue-microorganisms.html
[4]BCCM (http://bccm.belspo.be/index.php)
[5]CBS (http://www.cbs.knaw.nl/databases/),
[6]ICMP (http://scd.landcareresearch.co.nz/Search/Search/ICMP)

5.5.4 Conclusion

It can be concluded that the Global Collection of Mushroom Pathogens contains isolates from 4 major mushroom producing geographical areas. Also other collections can provide a wealth of isolates from these regions. However, nor the global Collection of Mushroom Pathogens, nor the other collections contain isolates from China.

6. Expanding the collection

As becomes evident from comparing the main mushroom producing geographical regions with the origin of the isolates, additional isolates are needed to obtain full coverage. For this two sources can be found. First of all, isolates can be obtained from other collections. Next to this, new isolates can be isolates from diseased mushroom crops.

6.1 Exchange with other collections

An overview of different fungal collections can be found on websites such as http://www.biorepositories.org/. Below a list is provided of fungal culture collections, worldwide (Table 13).

Table 7. List of fungal culture collections.

Acronym	WDCM Number	Collection	Region
ACM	WDCM 13	University of Queensland Microbial Culture Collection	Oceania
DAR	WDCM 365	Plant Pathology Herbarium	Oceania
DFP	WDCM 102	DFP Culture Collection	Oceania
DMPMC	WDCM 454	Department of Microbiology	Oceania
FRR	WDCM 18	Food Science Australia, Ryde	Oceania
VPRI	WDCM 851	Victorian Plant Disease Herbarium	Oceania
WAC	WDCM 77	Department of Agriculture Western Australia Plant Pathogen Collection	Oceania
CCFC	WDCM 150	Canadian Collection of Fungal Cultures	America
DFF	WDCM 50	Forest Pathology Culture Collection, Pacific Forest Research Centre	America
NoF	WDCM 744	The Fungus Culture Collection of the Northern Forestry Centre	America
SCCM	WDCM 920	Sporometrics Culture Collection of Microorganisms	America
UAMH	WDCM 73	University of Alberta Microfungus Collection and Herbarium	America
ATCC	WDCM 1	American Type Culture Collection	America
FGSC	WDCM 115	Fungal Genetics Stock Center	America
NRRL	WDCM 97	Agricultural Research Service Culture Collection	America
SRRC	WDCM 751	United States Department of Agriculture, Agricultural Research Service	America
ACCC	WDCM 572	Agricultural Culture Collection of China	Asia
CFCC	WDCM 995	China Forestry Culture Collection Center	Asia
CGMCC	WDCM 550	China General Microbiological Culture Collection Center	Asia
CICIM	WDCM 897	The Culture and Information Centre of Industrial Microoganisms of China Universities	Asia
HKUCC	WDCM 798	The University of Hong Kong Culture Collection	Asia
YM	WDCM 832	Strains Collection of Yunnan Institute of Microbiology, Yunnan University, China	Asia
JSCC		http://www.nbrc.nite.go.jp/jscc/aboutjsccc.html	Asia
PPRI	WDCM 351	National Collections of Fungi: Culture Collection	Africa

6.2 New isolates from diseased mushroom crops

Only pure, mite free cultures of fungal mushroom pathogen can be added to the collection. New strains will preferably be deposited to either the South African National Collection of Fungi (PPRI) or the Department of Microbiology and Plant Pathology, University of Pretoria.
Contact details are:

BIOSYSTEMATCS DIVISION: Mycology Unit
Plant Protection Research Institute
P/Bag X134
QUEENSWOOD
0121
Tel number 012 304 9570/1/3/60
Fax 012 325 6998
e-mail: KwindaG@arc.agric.za
JacobsR@arc.agric.za

Dept of Microbiology and Plant Pathology
University of Pretoria
Hillcrest
Pretoria
0002
Tel number (012) 420-3850
Fax (012) 420-4588

New isolates can only be accepted if the following minimum of data can be provided;

- Isolate number (accession Code (e.g. ATCC) or code assigned by researcher)
- Fungus name
- Depositor/Collector
- Host/Substrate (Genus & Species of the host)
- Locality (Geographic origin – specify country and area/region if relevant)
- Date collected
- Isolation method
- Person who made the identification

Strains deposited in the South African collection will be shared with the mirror collection in Wageningen in the Netherlands. By doing so the full collection will always be available on two locations.

7. Publicity on the availability of the collection

The availability of the collection has been anounced in Mushroom Business, a magazine that is read by a large number of people in the mushroom industry. Next to this, an excel sheet listing all strains is published on the ISMS website.

Hi Prof. Korsten. Do you have additional ideas for making the industry/research institutes/diagnostic labs aware of the possibility to obtain strains from the collection.

8. Delivering isolates from the collection

Interested parties can obtain strains from the Global Collection of Mushroom Pathogens. Strains will only be handed out by PPRI and/or the Department of Microbiology and Plant Pathology, University of Pretoria after under the terms of a material transfer agreement. A minimal handling fee of Rand 100 (≈ $ 10,- / ≈€ 8,-) per culture is payable to the organization that will provide you with the isolate.

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Appendix I.
Strains in the Global Collection of Mushroom Pathogens

For normal Appendices.

Important: Make a separate section for each Appendix by copying an existing Appendix with a section break at the end. Each Appendix should have a separate numbering, I-1, I-2, I-3 for the pages of the first Appendix, II-1, II-2, II-3 for the pages of the second Appendix etc.. The page numbers after the Roman numerals in the header are automatically generated by Word. The Appendix numbering and the Roman numerals are not automatically generated and should be inserted by hand in the Appendix title and in the Odd and Even Header of that section (this is best done in the 'normal view' mode).

Coll. name WUR	Coll. name PPRI	Old name of strain	Other collections	Year	Genotyping	country of origin	Deposit by	origin details
Cladobotryum dendroides								
MES 13880	MUG-Fc-00054	MYA-823	ATCC MYA-823		no	UK		Grogan & Gaze (2000)
MES 13881	MUG-Fc-00055	MYA-824	ATCC MYA-824		no	UK		Grogan & Gaze (2000)
Cladobotryum mycophilium								
MES 13875	MUG-Fc-00056	MYA-821	ATCC MYA-821		no	UK		Grogan & Gaze (2000)
MES 13876	MUG-Fc-00221	AMC00107		2007	no	Australia	Rodoni, DPI Victoria	
MES 13877	MUG-Fc-00222	AMC00307		2007	no	Australia	Rodoni, DPI Victoria	
MES 13878	MUG-Fc-00243	Type II #315		2003	no	USA		Kaolin Mushrooms
MES 13879	MUG-Fc-00238	Type II #316		2003	no	USA		Gaspari Mushrooms Farm
***Cladobotryum* species**								
MES 13882	MUG-Fc-00240	PS#294		2000	no	USA		Yeatman, Chester County
MES 13883	MUG-Fc-00236	PS#299		2000	no	USA		Sunset Mushrooms, Chester County
MES 13884	MUG-Fc-00235	PS#300		2000	no	USA		Kaolin Mushrooms, Chester County
MES 13885	MUG-Fc-00234	PS#301		2000	no	USA		Monterey Mushrooms, Temple Farm
MES 13886	MUG-Fc-00239	PS#302		2000	no	USA		Creekside Mushrooms Farm
MES 13887	MUG-Fc-00237	PS#304		2003	no	USA		Pietro Mushrooms Farm
MES 13889	MUG-Fc-00223	ACC28		2008	no	USA	Romaine	Penn State Uni, USA
MES 13890	MUG-Fc-00224	ACC177		2008	no	USA	Romaine	Penn State Uni, USA
MES 13891	MUG-Fc-00225	ACC305		2008	no	USA	Romaine	Penn State Uni, USA
MES 13892	MUG-Fc-00226	ACC308		2008	no	USA	Romaine	Penn State Uni, USA
MES 13893	MUG-Fc-00227	ACC314		2008	no	USA	Romaine	Penn State Uni, USA
MES 13894	MUG-Fc-00228	CS			no	RSA	Meyer	Country Mushrooms
MES 13895	MUG-Fc-00229	N22-6			no	RSA	Meyer	Denny's Shongweni
MES 13896	MUG-Fc-00230	Phes1			no	RSA	Meyer	Denny's Phesantekraal

MES 13897	MUG-Fc-00231	G22-18			no	RSA	Meyer	
MES 13898	MUG-Fc-00232	CS51-21			no	RSA	Meyer	
MES 13899	MUG-Fc-00233	CS51-23			no	RSA	Meyer	
MES 13900	MUG-Fc-00241	G51-29			no	RSA	Meyer	
MES 13901	MUG-Fc-00051	G21-64		2009	no	RSA	Meyer	Casing soil, Country Mushrooms
MES 13902	MUG-Fc-00052	CS21-12		2010	no	RSA		Meyer, Denny's Deodar
MES 13903	MUG-Fc-00053	CS21-3		2010	no	RSA		Meyer, Denny's Deodar
Mycogone perniciosa								
MES 11238		M020704	CBS 234.29	1929	no	Germany	Wollenweb er	Grower
MES 11237		M020703	CBS 322.52	1948	no	NL	Bels-Koning	Mushroom bed
MES 11240		M020706	CBS 648.82	1982	FJ904634	Belgium	Brand & Gams	host Agaricus silvicola
MES 11792		M050201		2005	no	NL		Grower
MES 13939	MUG-Fc-00063	ATCC96421	ATCC96421		no	China (Shanghai)	Romaine	Fruitbody Shanghai
MES 13940	MUG-Fc-00050	CS11		2009	no	RSA	Meyer	Boland Mushrooms
MES 13941	MUG-Fc-00247	DW1-4		2011	no	RSA	Meyer	DocWim's
MES 13942	MUG-Fc-00064	MYC Z F9		2006	no	RSA	Meyer	
MES 13943	MUG-Fc-00220	ACC176		2008	no	USA	Romaine	Penn State Uni, USA
MES 13944	MUG-Fc-00226	PS#181		1982	no	USA		Ostrum MR Farm WA
Trichoderma aggressivum* f. *aggressivum								
MES 11410		CBS 45095	CBS 450.95	1992	AF359260, AF400265	Canada	Seaby	CBS; Th.4/ Britisch Columbia
MES 11431		T041102	CBS 100528	1998		USA	D. Royse	Pennsylvania Mushroom compost
Trichoderma aggressivum* f. *europeanum								
MES 11433		T041104	CBS 100526	1998	AF400265, FJ442434, AF348096	Ireland	D. Royse	mushroom compost
MES 12999		Walkro/ 08-07		2007	SEQ	NL		Compost yard
MES 13004		ProefcenterB/02-10		2007	SEQ	Belgium		Research Station Belgium
MES 13916	MUG-Fc-00062	CBS 101525	CBS 101525	1998	Genbank AF359256	Italy	Geels	
MES 13905	MUG-Fc-00242	PS#326		2050	no	USA		Th4, Chester Co Farm,
MES 13063		Champworld		2008	SEQ	NL		Grower
MES 13067		Wijers		2008	SEQ	NL		Grower
Trichoderma aggressivum								
MES 13906	MUG-Fc-00246	Royal2		2011	no	RSA	Van Greuning	Royal Mushrooms
MES 13907	MUG-Fc-00047	CS21-5		2009	no	RSA	Meyer	Casing soil, Deodar Denny's
MES 13908	MUG-Fc-00045	CS11-66		2009	no	RSA	Meyer	Phase 2 compost, Meadow mushrooms
Trichoderma asperellum								
MES 13926	MUG-Fc-00209	AMT01807		2007	no	Australia	Clift	Sidney Uni
Trichoderma atroviride								
MES 11409		CBS 15910D		1989	ITS-RFLP	NL		tested in experiment; identification confirmed by

								CBS
MES 11279		930405		1993	SEQ	Netherlands		Grower
MES 11284		940323		1994	SEQ	Germany		Grower
MES 11322		951124		1995	SEQ	Germany		Grower
MES 11382		980825		1998	SEQ			Grower
MES 13002		Proefcenter B/21-08-07		2007	SEQ	Belgium		Research Station Belgium
MES 13083		PM-50/machine		2008	SEQ	Netherlands		-
MES 13909	MUG-Fc-00194	AMT00107		2007	no	Australia	Rodoni	DPI Victoria
MES 13910	MUG-Fc-00207	AMT01607		2007	no	Australia	Clift	Sidney Uni
MES 13911	MUG-Fc-00210	AMT01907		2007	no	Australia	Clift	Sidney Uni
MES 13912	MUG-Fc-00213	AMT02207		2007	no	Australia	Clift	
Trichoderma aureoviride								
MES 13927	MUG-Fc-00212	AMT02107		2007	no	Australia	Clift	Sidney Uni
Trichoderma citrinoviride								
MES 11313		951115		1995	SEQ	NL		Grower
Trichoderma hamatum								
MES 11397		9543023		1995	SEQ	NL		Grower; casing soil
MES 11346		960531		1996	SEQ	DK		Compost yard
MES 00005		Jamur ceguk		?????	SEQ	Indonesia		-
MES 13913	MUG-Fc-00196	AMT00307		2007	no	Australia	Rodoni	DPI Victoria
MES 13914	MUG-Fc-00201	AMT01007		2007	no	Australia	Rodoni	DPI Victoria
MES 13915	MUG-Fc-00244	AMT00707		2007	no	Australia	Rodoni	DPI Victoria
Trichoderma harzianum								
MES 11293		949906		1994	SEQ			-
MES 12998		Dongenzicht/07-2006		2006	SEQ	NL		Grower
MES 12991		Jacobs 2007		2007	SEQ	NL		Grower
MES 13006		Peffer/03-2008		2008	SEQ	NL		Grower
MES 13003		Proefcenter B/1-27-09		2009	SEQ	Belgium		Research Station Belgium
MES 13917	MUG-Fc-00197	AMT00407		2007	no	Australia	Rodoni	DPI Victoria
MES 13918	MUG-Fc-00198	AMT00507		2007	no	Australia	Rodoni	DPI Victoria
MES 13919	MUG-Fc-00199	AMT00807		2007	no	Australia	Rodoni	DPI Victoria
MES 13920	MUG-Fc-00202	AMT01107		2007	no	Australia	Rodoni	DPI Victoria
MES 13921	MUG-Fc-00206	AMT01507		2007	no	Australia	Clift	Sidney Uni
MES 13922	MUG-Fc-00208	AMT01707		2007	no	Australia	Clift	Sidney Uni
MES 13923	MUG-Fc-00211	AMT02007		2007	no	Australia	Clift	Sidney Uni
MES 13924	MUG-Fc-00214	AMT02307		2007	no	Australia	Clift	Sidney Uni
MES 13925	MUG-Fc-00215	AMT02407		2007	no	Australia	Clift	Sidney Uni
Trichoderma koningii								
MES 13928	MUG-Fc-00195	AMT00207		2007	no	Australia	Rodoni	DPI Victoria
Trichoderma longibrachiatum								
MES 13084		Walkro Nr.5		2008	SEQ	NL		-
Trichoderma saturnisporum								
MES 11270		930115		1993	SEQ	NL		Experimental Station Horst
Trichoderma species								
MES 13929	MUG-Fc-00046	CS21-2		2009	no	RSA	Meyer	Deodar Denny's

MES 13930	MUG-Fc-00048	G11-63		2009	no	RSA	Meyer	casing soil, Highveld Mushrooms
MES 13931	MUG-Fc-00049	K21-53		2009	no	RSA	Meyer	casing soil, Denny's Phesantekraal,
MES 13932	MUG-Fc-00203	AMT01207		2007	no			Morley
MES 13933	MUG-Fc-00204	AMT01307		2007	no			Morley
MES 13934	MUG-Fc-00205	AMT01407		2007	no			Morley
MES 13935	MUG-Fc-00216	ACC179		2008	no	USA	Romaine	Penn State Uni, USA
MES 13936	MUG-Fc-00217	ACC209		2008	no	USA	Romaine	Penn State Uni, USA
MES 13937	MUG-Fc-00218	ACC149		2008	no	USA	Romaine	Penn State Uni, USA
MES 13938	MUG-Fc-00219	ACC207		2008	no	USA	Romaine	Penn State Uni, USA
Trichoderma virens								
MES 11307		950906		1995	SEQ			Grower
Verticillium fungicola var. aleophilum								
MES 11451		357.80	CBS 357.80	1979	GENBANK AF324876	NL		Gams & Van Zaayen, (1982)
MES 11539		V020610		2002	no	NL		Dry bubble incidence Experimental Station Horst
MES 11525		V011207 (=V06)		????	no	USA		Dr.Royce Penn State Univer.Chester country PA Claude Fordyce
MES 11459		V X02		????	no	Germany		ex Les Miz 60, Germany
MES 13847	MUG-Fc-00179	AMV01707		2007	no	Australia		Rodoni, DPI Victoria
MES 13848	MUG-Fc-00180	DAR36011		2007	no	Australia	Clift	Sidney Uni
MES 13849	MUG-Fc-00181	DAR43412		2007	no	Australia	Clift	Sidney Uni
MES 13850	MUG-Fc-00182	DAR34255		2007	no	Australia	Clift	Sidney Uni
MES 13851	MUG-Fc-00183	DAR43407		2007	no	Australia	Clift	Sidney Uni
MES 13852	MUG-Fc-00184	DAR43408		2007	no	Australia	Clift	Sidney Uni
MES 13853	MUG-Fc-00185	DAR42124		2007	no	Australia	Clift	Sidney Uni
MES 13854	MUG-Fc-00186	DAR43117a		2007	no	Australia	Clift	Sidney Uni
MES 13855	MUG-Fc-00187	DAR36024		2007	no	Australia	Clift	Sidney Uni
MES 13856	MUG-Fc-00222	DAR31481a		2007	no	Australia	Clift	Sidney Uni
MES 13857	MUG-Fc-00188	ACC117		2008	no	USA	Romaine	Penn State Uni, USA
MES 13858	MUG-Fc-00189	ACC175		2008	no	USA	Romaine	Penn State Uni, USA
MES 13859	MUG-Fc-00190	ACC171		2008	no	USA	Romaine	Penn State Uni, USA
MES 13860	MUG-Fc-00191	ACC1185		2008	no	USA	Romaine	Penn State Uni, USA
MES 13861	MUG-Fc-00192	ACC123		2008	no	USA	Romaine	Penn State Uni, USA
MES 13862	MUG-Fc-00193	ACC121		2008	no	USA	Romaine	Penn State Uni, USA
Verticillium fungicola var. fungicola								
MES 11449		440.34	CBS 440.34 = ATCC 22607	1934	GENBANK AF324874	UK		Gams & Van Zaayen, (1982); Ware (1933).
MES 11454		R1		1974	no	NL		Grower
MES 11463		V 7902		1979	no	NL		tested at Experimental Station Horst
MES 11467		V 9401 (94980)		1994	no	NL		Identified by CBS; very sensitive to Sporgon
MES 11477		V 9601		1996	no	NL		Steenbekkers, Alem. 28-02-1996
MES 11495		V970325		1997	no	NL		Bovist-achtige mol. Ook clusters/ Bovee Ysselsteyn
MES 11500		V981222		1998	no	France		From casing soil; Identified

								by CBS; not very sensitive to Sporgon
MES 11501		V9909		1999	no	NL		Grower
MES 11542		V021105		2002	no	NL		Isolated from experiment at Experimental Station Horst
MES 13863	MUG-Fc-00168	AMV00107		2007	no	RSA	Clift	Sidney Uni
MES 13864	MUG-Fc-00169	AMV00207		2007	no	RSA	Clift	Sidney Uni
MES 13865	MUG-Fc-00171	AMV00607		2007	no	RSA	Clift	Sidney Uni. (Not a pure culture, difficult to obtain one)
MES 13866	MUG-Fc-00174	AMV01007		2007	no	RSA	Rodoni	DPI Victoria
MES 13867	MUG-Fc-00178	AMV01507		2007	no	RSA	Rodoni	DPI Victoria
MES 13868	MUG-Fc-00057	CBS 194.79	CBS 194.79	1979	no	UK	Fletcher	CBS
Verticillium lamellicola								
MES 11474		V951109B		1995	no	Poland		Grower; Identified by CBS
Verticillium psalliotae								
MES 11502		V990615G		1999	no	NL		Isolated from composting garden waste
Verticillium fungicola var. flavidum								
MES 13869	MUG-Fc-00059	CBS 342.80	CBS 342.80	1980	AF324877, EF641878	France	Gourbière	CBS
Verticillium fungicola								
MES 13870	MUG-Fc-00223	PS#262		1988	no	Canada		DR; V16; Markham Mushroom
MES 13871	MUG-Fc-00225	PS#182		1986	no	USA		PA; LN-4, Chester Co
MES 13872	MUG-Fc-00224	PS#263		1998	no	USA		Spawn
Verticillium species								
MES 13873	MUG-Fc-00043	G21-54		2009	no	RSA	Meyer	casing soil, Country Mushrooms
MES 13874	MUG-Fc-00245	MM1-1		2011	no	RSA	Meyer	Monandi

Appendix II.
FAOStat data on mushroom production

FAOStat (http://faostat.fao.org/) also collects data on mushroom production. These data are based either on official data, or they may be aggregated data (may include official, semi-official or estimated data). In other cases FAO makes an estimate. And in some cases data are just not available. Regardless the quality of the data, they do not discriminate between different mushroom species. The data apply both to mushrooms that are grown on (semi-) industrial scale and to mushrooms that are collected in nature (like truffles and boletes). Figures therefore differ from those mentioned in paragraph. Nevertheless these data can be used to find out in which regions of the world almost no mushroom production takes place. An overview of world wide mushroom production is given in Table 3.

Table 8. Mushroom production world wide (not only button mushroom).

Continent	Mushroom production in 2010 (tonnes)	Production as % of world production
World (Total)	7,443,133	
Asia (Total)	5,122,059	68.8 %
Europe (Total)	1,821,728	35.6 %
Americas (Total)	432,399	8.4 %
Oceania (Total)	49,508	0.9 %
Africa (Total)	17,439	0.3 %

Mushroom production in Asia

According to FAOStat, Asia produced 5,122,059 tonnes of mushrooms in 2010 (Table 4). Main mushroom producing countries in Asia are China (4,833,725 tonnes), Indonesia (61,376 tonnes), Japan (59,550 tonnes), India (41,000 tonnes), Iran (29,009 tonnes), South Korea (26,250 tonnes), Vietnam (21,213 tonnes), Thailand (6,925 tonnes) and North Korea (6,540 tonnes). In Asia, many species of edible mushrooms are being cultivated with button mushrooms ranking third (. estimated production of button mushrooms in China in 2009 was 2.181.053 tonnes).

Table 9. Mushroom production in Asia.

Geographical area	Mushroom production in 2010 (tonnes)	Production as % of Asian production
Asia + (Total)	5,122,059	
Eastern Asia China (4,833,725 tonnes), Japan (59,550 tonnes), South Korea (26,250 tonnes), and North Korea (6,540 tonnes)	4,926,300	96.2 %
South-Eastern Asia Indonesia (61,376 tonnes), Vietnam (21,213 tonnes) and Thailand (6,925 tonnes)	90,171	1.8 %
Southern Asia India (41,000 tonnes) and Iran (29,009 tonnes)	70,009	1.4 %
Western Asia Turkey (21,559 tonnes), Israel (9,500 tonnes) and Jordan (1,030 tonnes)	34,279	0.7 %
Central Asia + (Total)	1,300	0.03 %

Mushroom production in Europe

According to FAOStat, Europe produced 1,821,728 tonnes of mushrooms in 2010 (Table 5). Main mushroom producing countries in Europe are Italy, Spain, Netherlands, Poland and France. According to paragraph 5.1.1., production of button mushroom in Europe is 1.056.900 tonnes. This would mean that more than half of the mushrooms produced in Europe is *Agaricus bisporus*. A comparison with paragraph 5.1.1. also learns that although Italy appears to be a large producer of mushrooms, the production of button mushrooms is only about 1/8 of its total production.

Table 10. Mushroom production in Europe (not only button mushroom).

Geographical area	Mushroom production in 2010 (tonnes)	Production as % of European production
Europe + (Total)	1,821,728	
Southern Europe Italy (800,000 tonnes) and Spain (126,700 tonnes)	942,192	51.7 %
Western Europe Netherlands (266,000), France (119,346 tonnes), UK (69,300 tonnes), Ireland (65,000 tonnes), Germany (60,000 tonnes)	496,844	27.3 %
Eastern Europe Poland (173,448 tonnes), Hungary (14,026 tonnes), Ukraine (11,000 tonnes), Romania (9,973 tonnes) and Serbia (5,000 tonnes).	225,625	12.4 %
Northern Europe Denmark (9,541 tonnes), Finland (1,645 tonnes), Lithuania (10,434 tonnes)	157,067	8.6 %

Mushroom production in the America's

For the American continent, FAOStat reports a total mushroom production of 432.399 tonnes in 2010 (Table 6). This production is located entirely in Northern America. There is virtually no industrial mushroom production in South- or Central America or in the Caribbean.
The two producing countries in Northern America are USA (83% of total production in 2010) and Canada (17% of total production in 2010). Mexico does not appear to produce much mushrooms. Comparison with production data for USA for button mushroom (356.936 tonnes in 2009) shows that Agaricus bisporus is the main mushroom species produced.

Table 11. Mushroom production in the Americas (not only button mushroom).

Geographical area	Mushroom production in 2010 (tonnes)	Production as % of American production
Americas + (Total)	432,399	
Northern America USA (359,469 tonnes), Canada (72,930 tonnes)	432,399	100 %
Central America	0	
Caribbean	0	
South America	0	

Mushroom production in Oceania (including Australia and New Zealand).

According to FAOStat, Oceania produced 49,508 tonnes of mushrooms in 2010 (Table 7). This production was based entirely in Australia and New Zealand. Australia produced 41,295 tonnes of mushrooms in 2010, while New Zealand produced 8,213 tonnes. Based on the figures mentioned above, it can be concluded that these two countries mainly produce button mushrooms. Melanesia, Micronesia and Polynesia produced virtually no mushrooms on industrial scale.

Table 12. Mushroom production in the Oceania (not only button mushroom).

Geographical area	Mushroom production in 2010 (tonnes)	Production as % of Oceanian production
Oceania + (Total)	49,508	
Australia and New Zealand	49,508	100%
Australia (41,295 tonnes), New Zealand (8,213 tonnes)		
Melanesia	0	
Micronesia	0	
Polynesia	0	

Mushroom production in Africa

For the African continent, FAOStat reports a total mushroom production of 17439 tonnes in 2010 (Table 8). The bulk of this production is based in Southern Africa. Minor production of mushrooms takes place in Eastern Africa and Northern Africa. There is virtually no industrial mushroom production in Westerm and Middle Africa.

Table 13. Mushroom production in Africa (not only button mushroom).

Geographical area	Mushroom production in 2010 (tonnes)	Production as % of African production
Africa + (Total)	17,439	
Southern Africa	12,217	70.0 %
Eastern Africa	2,933	16.8 %
Northern Africa	2,289	13.1 %
Morroco (1,943 tonnes), Algeria (220 tonnes), Tunesia (126 tonnes)		
Middle Africa		
Western Africa		

If we compare these data with the production of button mushroom reported in paragraph 5.1.1. for South Africa (20.000 tonnes in 2009), we can conclude that mainly button mushroom is produced in South Africa.