

Two new thermophilic species of *Myceliophthora* from soil and compost piles

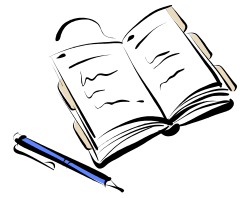
André Rodrigues, Márcia M.S. Moretti, Rafaella C. Bonugli-Santos,
Eleni Gomes, Lara D. Sette



Department of Biological Sciences
UESC – Santa Cruz State University
arodrigues@uesc.br



Summary



- The genus *Myceliophthora*
- Our study...
- Methods
- Results and discussion

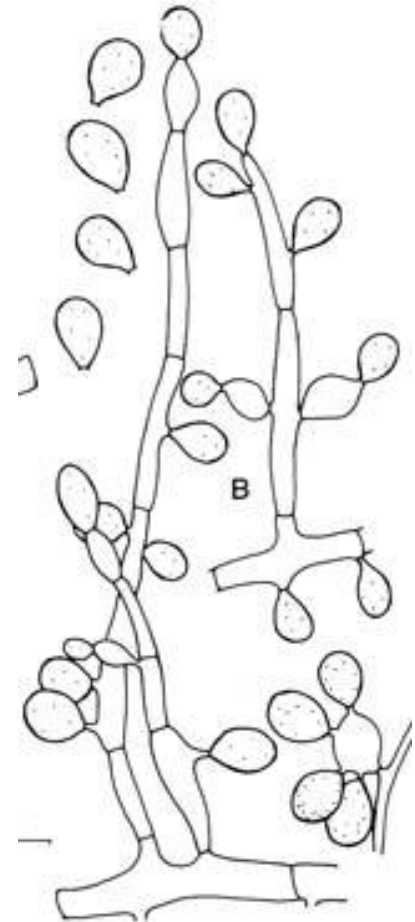
The genus *Myceliophthora*

- Type:

Costantin (1892): *Myceliophthora lutea*



Mat disease of mushrooms
(Vert-de-gris)

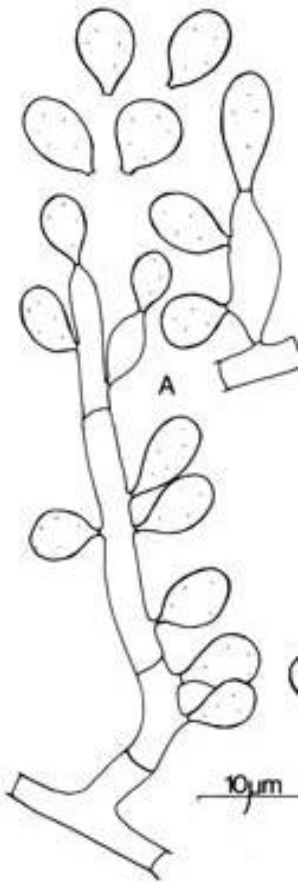


van Oorschot (1980)

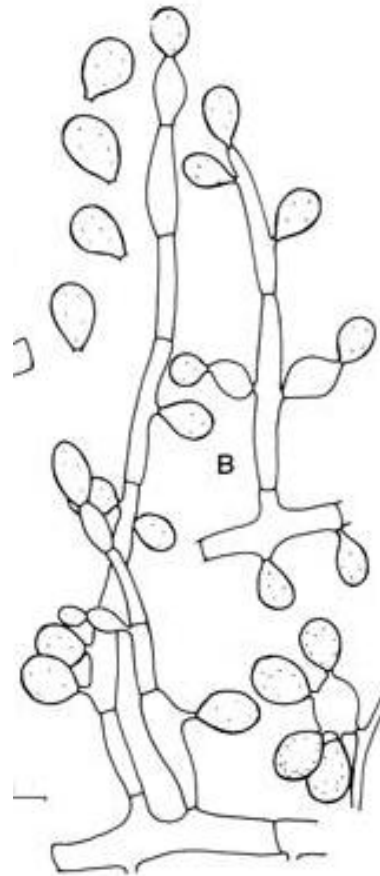
- Carmichael (1962): transferred to *Chrysosporium* sp.

The genus *Myceliophthora*

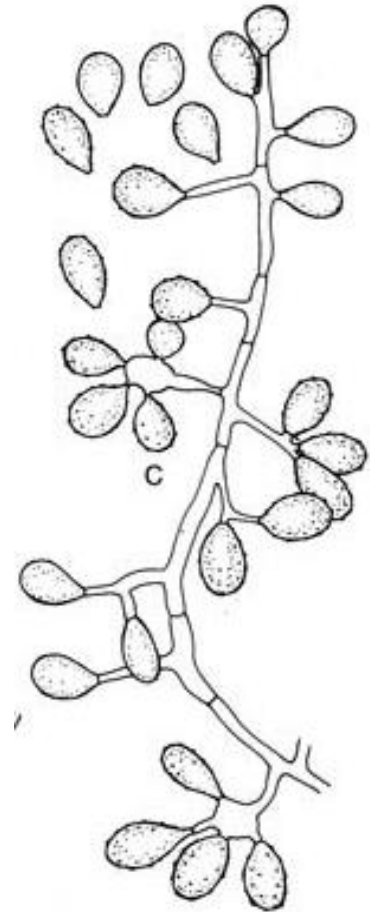
- van Oorschot (1977): reintroduced the genus



M. fergusii



M. lutea



M. thermophila

The genus *Myceliophthora*

van Oorschot (1980): revision

| <i>Myceliophthora</i> spp. | Teleomorph | Source |
|--------------------------------|-----------------------------------|----------------------|
| <i>M. thermophila</i> | <i>Corynascus heterothallicus</i> | soil |
| <i>M. fergusii</i> | <i>Corynascus thermophilus</i> | compost pile |
| <i>M. lutea</i> | unknown | cultivated mushrooms |
| <i>M. vellera</i> | unknown | soil |
| <i>Myceliophthora</i> anamorph | <i>Corynascus sepedonium</i> | soil, seeds |
| <i>Myceliophthora</i> anamorph | <i>Corynascus novoguineensis</i> | soil |
| <i>Myceliophthora</i> anamorph | <i>Arthroderma tuberculatum</i> | soil, feathers |
| <i>Myceliophthora</i> anamorph | <i>Ctenomyces serratus</i> | soil |

The genus *Myceliophthora*

McGinnis et al. (1992), Siegler et al. (1998),
Vidal et al. (2000)

1. anamorphs of Sordariales
2. Conidia with narrow basal attachments, borne directly on hyphae or pedicels or ampulliform swellings
3. Cellulolytic
4. Thermotolerant and thermophilic

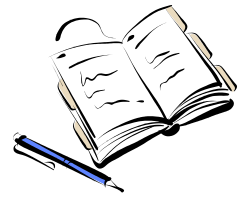
The genus *Myceliophthora*

Currently:

4 species and several unnamed anamorphs

| <i>Myceliophthora</i> spp. | Teleomorph | Source |
|--------------------------------|-----------------------------------|----------------------|
| <i>M. thermophila</i> | <i>Corynascus heterothallicus</i> | soil |
| <i>M. fergusii</i> | <i>Corynascus thermophilus</i> | compost pile |
| <i>M. lutea</i> | unknown | cultivated mushrooms |
| <i>M. hinnulea</i> | unknown | soil |
| <i>Myceliophthora</i> anamorph | <i>Corynascus sepedonium</i> | soil, seeds |
| <i>Myceliophthora</i> anamorph | <i>Corynascus novoguineensis</i> | soil |
| unknown | <i>Corynascus sexualis</i> | soil |
| <i>Myceliophthora</i> anamorph | <i>Corynascus similis</i> | soil |
| <i>Myceliophthora</i> anamorph | <i>Corynascus verrucosus</i> | soil |

Summary

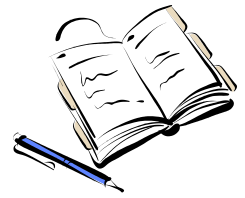


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Our study

- During a survey of thermophilic fungi for biotechnological applications, four isolates exhibited high **cellulolytic** (Moretti 2010) and proteolytic (Zanphorlin et al. 2010) activities
- Here, we propose two new species to accommodate these isolates

Summary



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Methods



Compost piles
Site: Olímpia, SP



Cultivated soil
Site: Ubarana, SP



Isolation



Characterization

Methods

Morphological

Macro and micromorphology: were observed of cultures grown on MA 2% and PDA at 45° C.

Radial growth: was determined on MEA, OA, PCA and PDA at 25, 36 and 45° C.

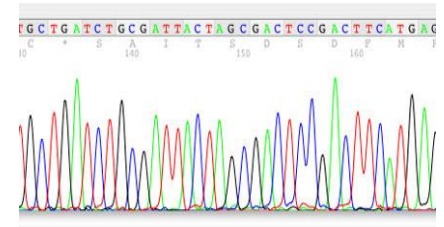
Methods

Molecular

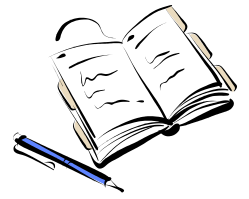


Amplification of ITS rDNA and D1/D2 26S rDNA

DNA sequencing (Sanger method)



Summary



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BLAST_N NCBI – GenBank results

| Isolate | D1/D2 26S rDNA | | | ITS rDNA | | |
|---------|----------------|-----|--|----------|-----|---|
| | length | % | GenBank closest relative | length | % | GenBank closest relative |
| M7.7 | 462 | 100 | <i>Corynascus sepedonium</i> FMR9123 (FJ666364) | 457 | 99 | <i>Myceliophthora</i> sp. T41 (FJ548836) |
| F.2.1.1 | 508 | 98 | <i>Corynascus sepedonium</i> FMR9123 (FJ666364) | 477 | 99 | <i>Myceliophthora</i> sp. T31 (FJ548837) |
| F.2.1.3 | 518 | 98 | <i>Corynascus sepedonium</i> FMR9123 (FJ666364) | 415 | 100 | <i>Myceliophthora</i> sp. T31 (FJ548837) |
| F.2.1.4 | 491 | 99 | <i>Corynascus sepedonium</i> FMR9123 (FJ666364) | 492 | 99 | <i>Myceliophthora</i> sp. T31 (FJ548837) |

 compost pile

 soil

Myceliophthora olimpiensis sp. nov.

Teleomorph: unknown

Type: **Brasil**: Olímpia-SP, ex compost pile, 04/24/09, Moretti (M.7.7.)

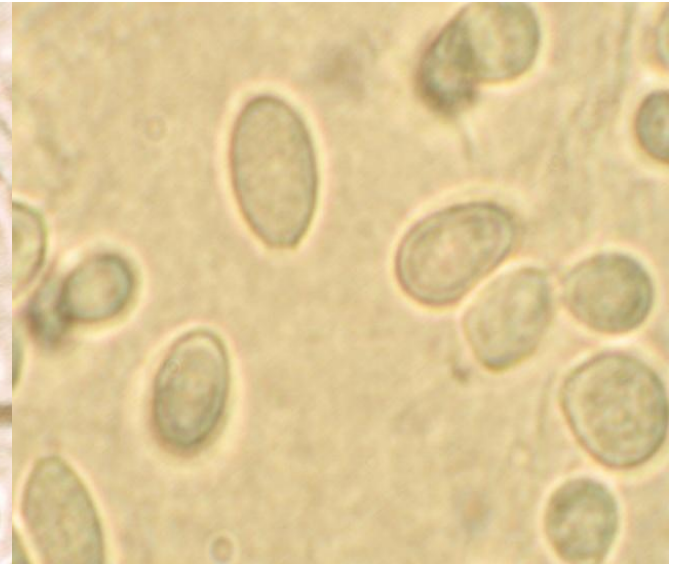
Remarks: colonies plane, pale brown. Conidia clavate or obovoid, with narrow truncate base, sessile or on ampulliform swellings (sometimes in chains), smooth walled, 5.65-7.91 x 3.39-4.52 μm .



culture on PDA



conidiophores



conidia

Myceliophthora olimpiensis sp. nov.

Radial growth (mm) after 3 days of incubation

| Media | Temperature (° C) | | |
|-------|-------------------|--------------|--------------|
| | 25 | 36 | 45 |
| MEA | 6,83 ± 0,40 | 34,81 ± 1,94 | 78,16 ± 2,22 |
| OA | 6,16 ± 0,75 | 26,83 ± 1,32 | 75,16 ± 1,94 |
| PCA | 5,33 ± 1,21 | 30,00 ± 0,63 | 90,00 ± 0,00 |
| PDA | 3,83 ± 0,75 | 30,66 ± 0,81 | 70,50 ± 5,08 |

Myceliophthora ubaranaensis sp. nov.

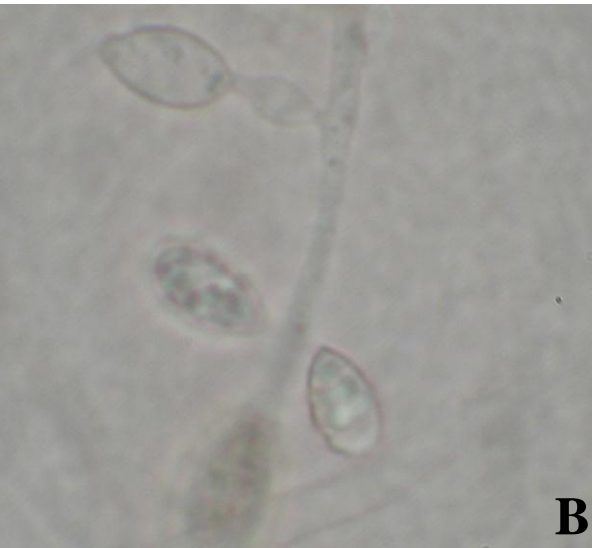
Teleomorph: unknown

Type: **Brasil:** Ubarana-SP, ex cultivated soil, 05/14/09, Moretti (F.2.1.4.)

Remarks: colonies floccose, pale brown, reverse uncolored. Conidia ellipsoid or obovoid, with truncate base, sessile or on ampulliform swellings (sometimes in chains), rough walled, 5.0-6.41 x 3.39-4.52 μm .



culture on PDA



conidiophores



conidia

Myceliophthora ubaranaensis sp. nov.

Radial growth (mm) after 3 days of incubation

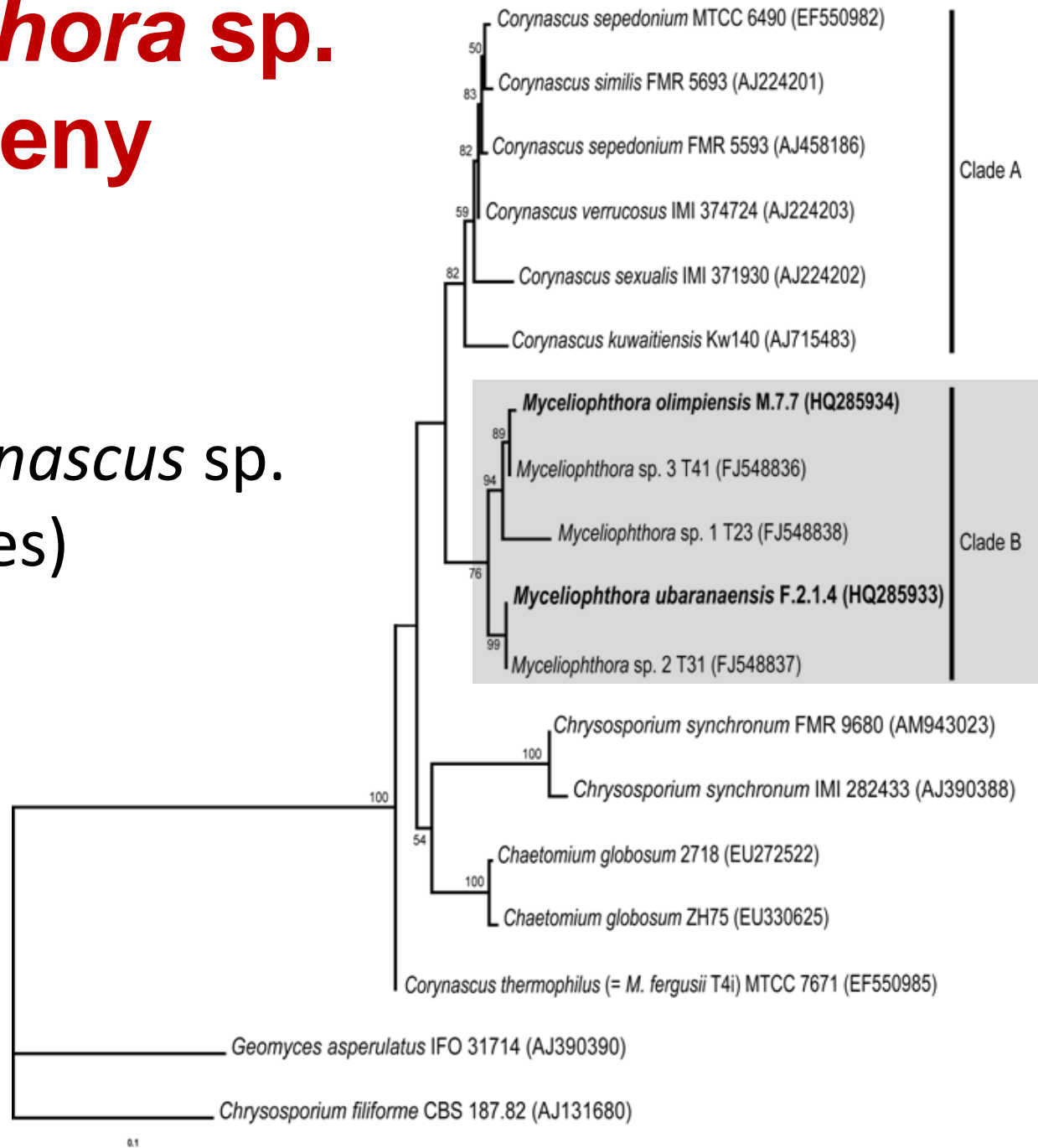
| Media | Temperature (° C) | | |
|-------|-------------------|--------------|--------------|
| | 25 | 36 | 45 |
| MEA | 5,16 ± 0,75 | 32,00 ± 1,89 | 90,00 ± 0,00 |
| OA | 5,00 ± 1,54 | 25,83 ± 1,60 | 75,00 ± 7,64 |
| PCA | 4,50 ± 0,54 | 21,33 ± 1,03 | 62,33 ± 2,06 |
| PDA | 3,16 ± 1,16 | 25,16 ± 0,75 | 71,83 ± 3,60 |

Morphological comparisons

| <i>Myceliophthora</i> | Optimum growth (° C) | Colony aspect | Conidia | Teleomorph |
|------------------------|----------------------|-------------------------|--|---------------------------|
| <i>M. olimpiensis</i> | 45 | plane, pale brown | obovoid-ellipsoid, smooth-walled, 5.65-7.91 x 3.39-4.52 µm | unknown |
| <i>M. ubaranaensis</i> | 45 | floccose, pale brown | obovoid-ellipsoid, rough-walled, 5-6.4 x 3.39-4.52 µm | unknown |
| <i>M. fergusii</i> | 45 | floccose, pinkish-cream | pyriform-clavate, smooth, 4.8-12 x 2.8-5.2 µm | <i>C. thermophilus</i> |
| <i>M. hinnulea</i> | 40 | Plane, red | pyriform-ovate, verrucose, 8-9 x 6-7.5 µm | unknown |
| <i>M. lutea</i> | 33 | Felty, pale yellow | pyriform-globose, smooth, 3.8-9 X 3-6 µm | unknown |
| <i>M. thermophila</i> | 33 - 36 | Woolly, pale brown | pyriform-obovoid, smooth, 4.5-11 x 3-4.5 µm | <i>C. heterothallicus</i> |

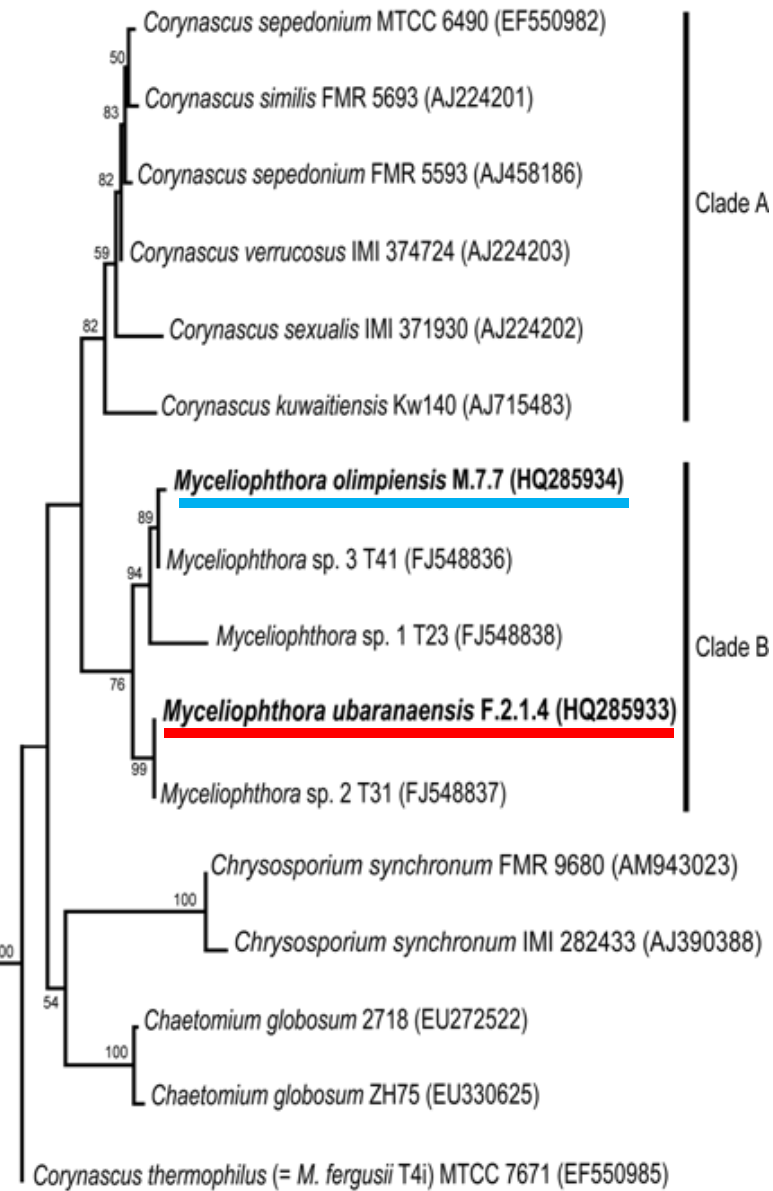
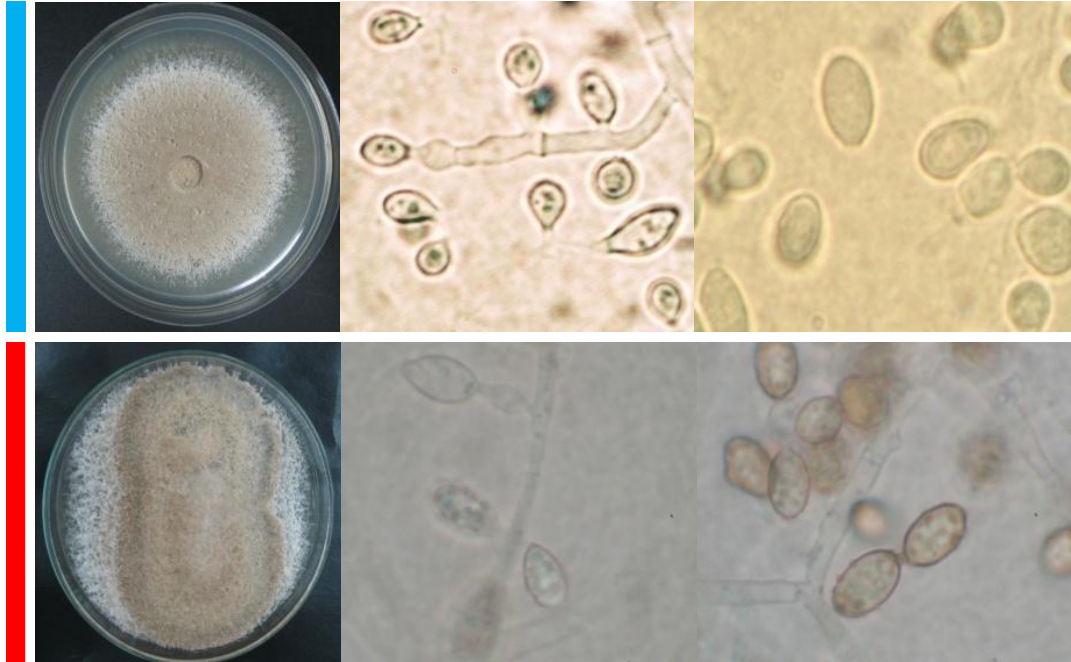
Myceliophthora sp. phylogeny

Affinity with *Corynascus* sp.
(Sordariales)



Neighbor-joining tree of *Myceliophthora* isolates inferred from a 476-base-pair fragment of ITS-rDNA. Numbers on branches are bootstrap support values derived from 1000 pseudoreplicates.

Myceliophthora sp. phylogeny



Neighbor-joining tree of *Myceliophthora* isolates inferred from a 476-base-pair fragment of ITS-rDNA. Numbers on branches are bootstrap support values derived from 1000 pseudoreplicates.

Summary

- *M. olimpiensis* sp. nov. – compost piles
- *M. ubaranaensis* sp. nov. – soil samples
- Thermophilic with distinct morphological and molecular markers
- Both are cellulolytic as well as proteolytic strains (data not shown).

Acknowledgments



Thank you!