SKML 2007-1 MYCOLOGY EVALUATION

Sample A ***Absidia corymbifera***

Strain was part of SKML 01-2005. Score at that time was 48%, this time it is 62%.

Absidia spp. are filamentous fungi that are cosmopolitan and ubiquitous in nature as common environmental contaminants. They are found in plant debris and soil, as well as being isolated from foods and indoor air environment. They often cause food spoilage.

Pathogenicity:
Absidia corymbifera is a relatively rare cause of human zygomycosis. Zygomycosis is an opportunistic mycoses that manifests with pulmonary, rhinocerebral, cutaneous, gastrointestinal, renal or meningeal involvement. Disseminated zygomycosis may originate from these infections. Zygomycosis is very rarely observed in immunocompetent host.
Since *Absidia* spp. are cosmopolitan and ubiquitous in nature, they are also common laboratory contaminants. Thus, their isolation in culture requires cautious evaluation. Nevertheless, the growth of *Absidia*, particularly from clinical samples of patients with immunosuppression or diabetes mellitus, should be regarded as potentially significant. Also, the visualization of typical hyphae of zygomycetes group of fungi on direct microscopic examination, of particularly a sterile body site, should be considered significant even if the culture yields no growth (5).

Distribution:
Worldwide (1).

Lab diagnosis:
1. Macroscopic morphology:
   On Sabouraud agar at 30°C: colonies fast growing, expanding, white to grayish-brown (Fig. 1)
2. Microscopic morphology (Fig. 2):
   Wide nonseptate hyphae. (a few septa may occasionally be present). Rhizoids are sparingly produced. When present, sporangiophores arise on stolons from points between the rhizoids, but not opposite the rhizoids. The sporangiophores are branched and carry pyriform, relatively small (20-120 µm in diameter) sporangia. Sporangiophore widens to produce the funnel-shaped apophysis beneath the sporangium. The apophysis is very well-developed and typical. The columella, the tip of the sporangiophore that extends into the sporangium, is semicircular in shape and has a small projection on top. Sporangiospores are one-celled, hyaline to light black, round to oval in shape, smooth or rarely echinulate on surface and 3-4.5 µm in diameter. They are found in the sporangium and are released to the surrounding when the sporangium ruptures (Fig. 2).

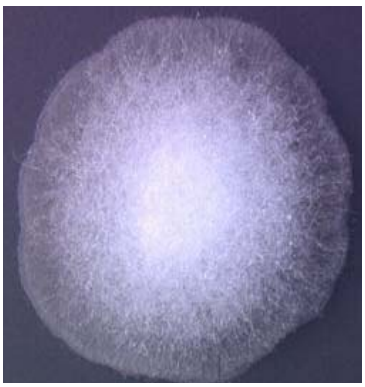

Fig. 1. Macroscopic morphology on Sabouraud agar (front)

Fig. 2. Microscopic morphology (rhizoids)

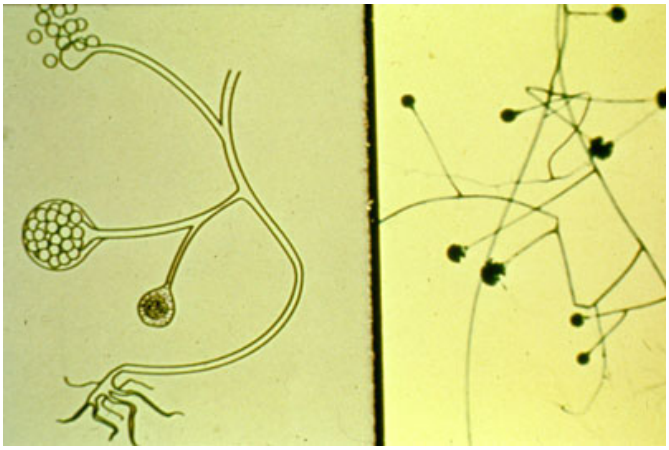

Fig. 3
Microscopic morphology

Fig. 4
Microscopic morphology
(sporangium with columella)

Difference between Absidia sp. and other zygomycetes are shown below.

Genus	Best growth	Sporangiophore	Apophysis	Columella	Sporangium	Rhizoid	Stylo-spore
Absidia	45°C	Branched, hyaline	Conical, not prominent	Dome-shaped	Pear-shaped	+, but usually indistinct	-
Apophysomyces	≥42°C	Usually unbranched, grayish-brown	Bell-shaped, not prominent	Usually dome-shaped, rarely elongated	Pear-shaped	+	-
Mortierella	40°C	Branched, hyaline	-	-	Spherical	+	+/-
Mucor	<37°C	Branched or unbranched, hyaline	-	+, in varying shapes	Spherical	-	-
Rhizomucor	54°C	Branched, brown	-	Spherical	Spherical	+	-
Rhizopus	45°C	Unbranched and brown mostly	Not prominent	Spherical or elongated	Spherical	+	-

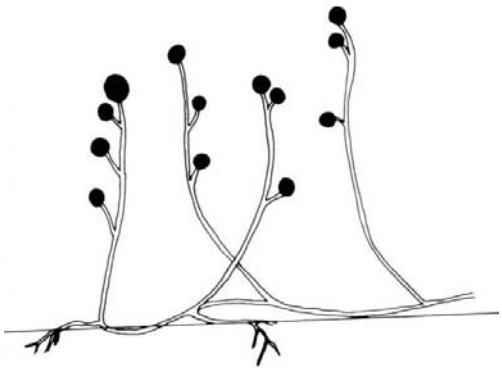

Fig. 5

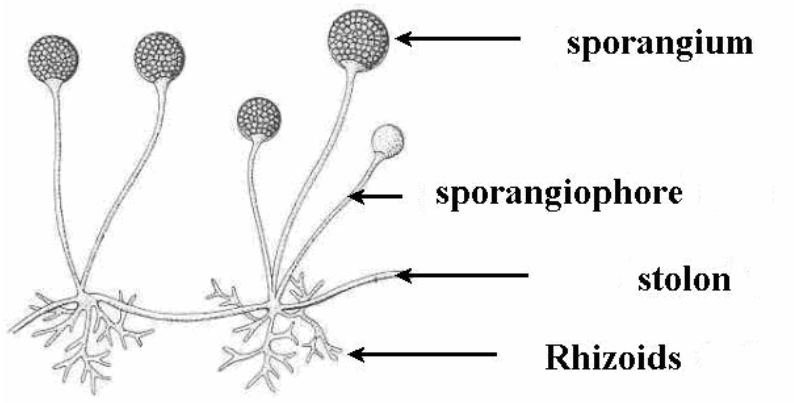

Fig. 6

Sample B ***Cryptococcus neoformans***

Cryptococcus is an encapsulated yeast. Following its first identification in nature from peach juice samples, the major environmental sources of *C. neoformans* have been shown to be either soil contaminated with pigeon droppings (*C. neoformans* var. *neoformans*) or eucalyptus trees and decaying wood forming hollows in living trees (*C. neoformans* var. *gattii*). *C. neoformans* var. *gattii* was also isolated from goats with pulmonary disease.

Pathogenicity:
C. neoformans is the causative agent of Cryptococcosis. Given the neurotropic nature of the fungus, the most common clinical form of cryptococcosis is meningoencephalitis. The course of the infection is usually subacute or chronic. Cryptococcosis may also involve the skin, lungs, prostate gland, urinary tract, eyes, myocardium, bones, and joints.
Until recently commonly encountered predisposing factor for development of cryptococcosis is AIDS. Since introduction of HAART, organ transplant recipients or cancer patients receiving chemotherapeutics or long-term corticosteroid treatment are major risk groups in the developed world. Serodiagnosis of this species breaks it out into one of 5 serotypes, A, B, C, D, and AD. *C. neoformans* var. *neoformans* (D), *C. neoformans* var. *gattii* (B, C), and *C. neoformans* var. *grubii* (A). The validity of *C. neoformans* var. *grubii* is still being argued and some authors place serotype A as *C. neoformans* var. *neoformans*. Serotype AD is still being characterized.

Distribution:
Worldwide.

Lab diagnosis:
1. Macroscopic morphology
   On Sabouraud agar at 30^{o}C: fast growing, soft, glistening to dull, smooth, usually mucoid, and cream to slightly pink or yellowish brown in color (Fig. 7).
2. Microscopic morphology
   globose yeast cells only (Fig. 8).

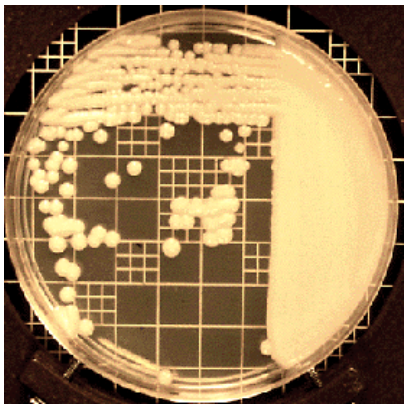

Fig. 7 Macroscopic morphology on Sabouraud agar (front)

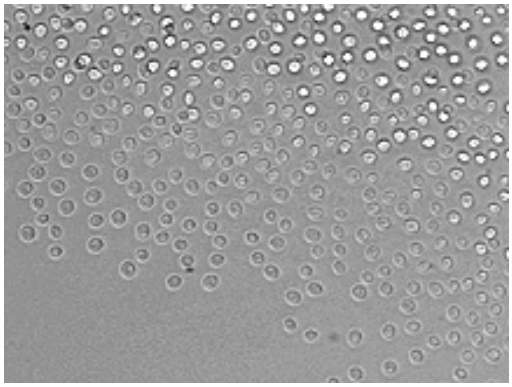

Fig. 8 Microscopic morphology

Difference between *C. neoformans* and other species are shown below.

Strain	Macroscopic morphology	Microscopic morphology	Supplementary test(s)
C. neoformans	Fast growing, soft, glistening to dull, smooth, usually mucoid, and cream to slightly pink or yellowish brown in color.	Globose yeast cells.	Urease: + Growth 37^{o}C: +
C. laurentii			Fails to grow at 37°C
Candida			Urease: -

Sample C ***Fusarium oxysporum***

Strain was part of SKML 01-2004. Score at that time was 31%, this time it is 40%.

Fusarium is a filamentous fungus widely distributed on plants and in the soil. It is found in normal mycoflora of commodities, such as rice, bean, soybean, and other crops. While most species are more common at tropical and subtropical areas, some inhabit in soil in cold climates. Some *Fusarium* species have a teleomorphic state.

Pathogenicity:
Fusarium is one of the emerging causes of opportunistic mycoses. Trauma is the major predisposing factor for development of cutaneous infections due to *Fusarium* strains. Disseminated opportunistic infections, on the other hand, develop in immunosuppressed hosts, particularly in neutropenic and transplant patients. *Fusarium* infections following solid organ transplantation tend to remain local and have a better outcome compared to those that develop in patients with hematological malignancies and bone marrow transplantation patients.

Distribution:
Worldwide

Lab diagnosis:

1. Macroscopic morphology
   On Sabouraud agar at 30^{o}C: Rapid growth. Colonies are initially white, becoming tinged with salmon and lavender at maturity (Fig. 9). Lavender to purple reverse. Salmon to orange sporodochia (mass of conidiophores) may be present.
2. Microscopic morphology
   Hyphae are septate and hyaline. Conidiophores are short and simple (usually not branched). Macroconidia usually produced abundantly, slightly sickle-shaped, thin-walled, with an attenuated apical cell and a foot-shaped basal cell. They are three to 5-septate (Fig. 10). Microconidia are abundant, mostly non-septate, ellipsoidal to cylindrical, slightly curved or straight occurring in false heads (a collection of conidia at the tip of the phialide) from short monophialides. Chlamydoconidia are present and often abundant, occurring singly and in pairs.

Fig. 9 Macroscopic morphology on Sabouraud agar (front)

Fig. 10 Microscopic morphology

Difference between *F. oxysporum* and other species are shown below.

Strain	Macroscopic morphology	Microscopic morphology	Supplementary test(s)
F. oxysporum	White, becoming tinged with salmon and lavender at maturity. Reverse: lavender to purple.	Conidiophores short and simple. Macroconidia abundant, slightly sickle-shaped, thin-walled, foot-shaped basal cell, 3-5-septate. Microconidia abundant, mostly non-septate, ellipsoidal to cylindrical, slightly curved or straight occurring in false heads, short monophialides. Chlamydoconidia often abundant.	
F. proliferatum	White, becoming tinged with purple. Reverse: colorless to dark purple.	Hyphae septate and hyaline. Conidiophores are medium length, simple or branched. Conidiogenous cells are monophialides and polyphialides. Microconidia abundant, single-celled and clavate and borne from both monophialides and polyphiades in false heads and in chains. Macroconidia may be rare, and very similar to those see in *F. moniliforme*.	
F. solani	Woolly to cottony with cream to white. Reverse: cream.	Hyphae septate and hyaline. Conidiophores simple (non-branched) or branched monophialides (phialides with a single opening). Macroconidia are moderately curved, stout, thick-walled, usually 3-5-septate, are borne on short conidiophores that soon form sporodochia. Microconidia borne from long monophialides, are one to three-celled, occur in false heads only (in clusters of conidia at the tip of the phialide).	
F. subglutinans	Pink or vinaceous to violet.	Conodiophores erect and branched, macroconidia falcate to rather straight, 3-5-septate with foot-cell, microconidia unicellulair, ovoidal, ellipsoidal or allantoid.	
Acremonium sp.	Compact, flat or folded, occasionally raised center, glabrous, velvety, membrane-like, at the beginning, sometimes powdery texture. By aging, cottony, white, pale grey or pale pink. Reverse: uncolored or a pink to rose colored pigment production is observed.	Unbranched, solitary, erect phialides. At the apices of the phialides are the hyaline conidia, single or multicellular, fusiform with a slight curve or resemble a shallow crescent. These structural properties of conidia vary depending on the species.	

Sample D ***Trichophyton interdigitale***

Trichophyton is a dermatophyte which inhabits the soil, humans or animals. Related to its natural habitats, the genus includes anthropophilic, zoophilic, and geophilic species. Some species are cosmopolitan. Others have a restricted geographic distribution. *Trichophyton* is one of the leading causes of hair, skin, and nail infections in humans. Most of the *Trichophyton* species have teleomorphic forms and these teleomorphs are classified in the genus *Arthroderma*.

Pathogenicity:

T. interdigitale (formerly known as *T. mentagrophytes* var. *interdigitale*) is an anthropophilic fungus which is a common cause of tinea pedis, particularly the vesicular type, tinea corporis, and sometimes superficial nail plate invasion.

Distribution:

Worldwide

Lab diagnosis:

1. Macroscopic morphology
   On Sabouraud's dextrose agar, colonies are usally flat, white to cream in color with a powdery to suede-like surface and yellowish and pinkish brown reverse pigment, often becoming a darker red-brown with age (Fig. 11). *T. interdigitale* has a suede-like to downy surface whereas *T. mentagrophytes* has a characteristic granular appearance (Fig. 11 and 13).
2. Microscopic morphology
   Numerous subspherical to pyriform microconidia, occasional spiral hyphae and spherical chlamydospores are present, the latter being more abundant in older cultures. Occasional slender, clavate, smooth-walled, multiseptate macroconidia are also present in some cultures (Fig. 12).

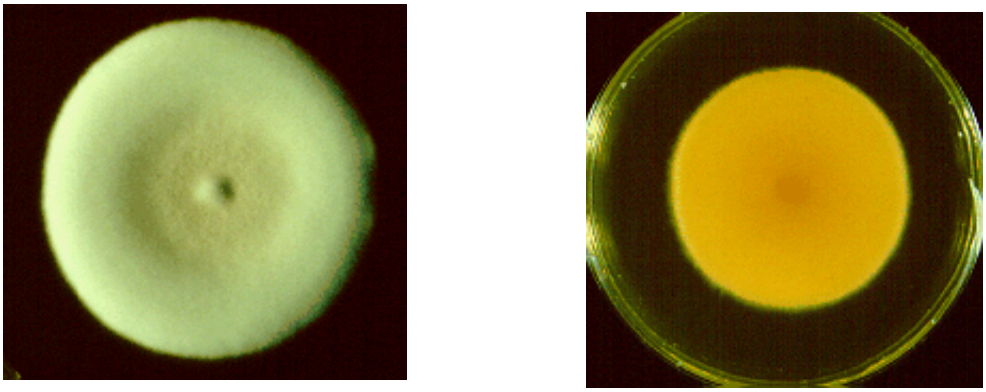

Fig. 11 Macroscopic morphology front (left) and back (right) on Sabouraud agar

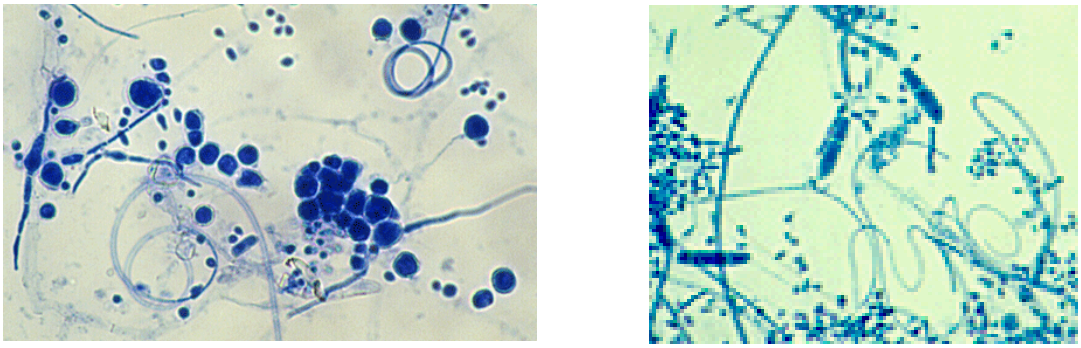

Fig. 12 Microscopic morphology

Difference between *T. interdigitale* and other species are shown below.

Strain	Macroscopic morphology	Microscopic morphology	Supplementary test(s)
T. interdigitale	Flat, white to cream with a powdery to suede-like surface and yellowish and pinkish brown reverse pigment, often becoming a darker red-brown with age.	Numerous microconidia, occasional spiral hyphae, spherical chlamydospores in older cultures. Occasional slender, clavate, smooth-walled, multiseptate macroconidia.	Hair perforation test: + Urease: +,-
T. mentagrophytes	Flat, white to cream, with a powdery to granular surface (Fig. 13). Sometimes central folding or raised central tufts or pleomorphic suede-like to downy areas. Reverse: yellow-brown to reddish-brown.	Numerous single-celled microconidia, hyaline, smooth-walled, predominantly spherical to subspherical in shape, occasional clavate to pyriform forms; spiral hyphae and smooth, thin-walled, clavate shaped, multicelled macroconidia may also be present.	Hair perforation test: +,- Urease: +
T. raubitschenkii / T. rubrum	Flat to slightly raised, white to cream, suede-like to downy. Reverse: yellow-brown to wine-red.	Downy type: slender clavate microconidia and no macroconidia. Granular type: clavate to pyriform microconidia and thin-walled, cigar-shaped macroconidia.	Hair perforation test: -
T. tonsurans	Suede-like to powdery, flat with a raised centre or folded, often with radial grooves, pale-buff to yellow to dark-brown. Reverse: yellow-brown to reddish-brown to deep mahogany.	Hyphae relatively broad, irregular, branched with numerous septa. Numerous microconidia long clavate to broad pyriform, borne at right angles to the hyphae. Very occasional smooth, thin-walled, irregular, clavate macroconidia. No spiral hyphae	Hair perforation test: -
M. persicolor	Flat, white to pinkish, with a suee-like to granular texture and a peripheral fringe. Reverse: orange to red.	Abundant, spherical to pyriform microconidia, macroconidia rarely produced.	Hair perforation test: +

Fig. 13 Macroscopic morphology of *T. mentagrophytes* on Sabouraud agar

Literature

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The macroscopic descriptions is according to the literature above. This may differ from your results that can depend on the composition of the Sabouraud agar plates.

Brun S, Bouchara JP, Bocquel A, Basile AM, Contet-Audonneau N, Chabasse D. Evaluation of five commercial Sabouraud gentamicin-chloramphenicol agar media. Eur. J. Clin. Microbiol. Dis. 2001; 20:718-723