

*MicroBytes Case History No. 7*

*From Dr. Joe DiPersio...*

## **A Stubborn Finger Infection**

A 42 year old male with a previous history of diabetes mellitus and poor medication compliance presented with pain and drainage from a nodular non-erythematous distal left finger lesion, from which purulent material could be expressed (figure 1).



**Figure 1: Finger infection from a restaurant worker.**

He reportedly kept the lesion covered and noted that it would occasionally dry up only to reappear. The digit was not swollen and he had full range of motion.

The lesion persisted for two months without progression. No other lesions developed, and he was afebrile, without evidence of regional lymphadenopathy.

He was employed as a restaurant dishwasher and denied hunting, fishing, and exposure to soil or pets. Purulent material expressed from the lesion and tissue samples were sent for culture and histopathology.

Based on preliminary laboratory test results the patient received a single dose of Amphotericin B and was taken to surgery for extensive debridement of the lesion. The patient refused further therapy and his finger healed post-operatively.

However, a recurrence of the lesion at the same site three months later was treated successfully with a six-week course of Amphotericin B.

Cultures of the expressed material and debrided tissue yielded moderate growth of creamy white, yeast-like colonies on Sabouraud's dextrose agar incubated at 30°C for three days (figure 2).



**Figure 2: Creamy colonies seen on Sabdex Agar after three days.**

A [lactophenol cotton blue stain](#) of a colony suggested the diagnosis (figure 3).

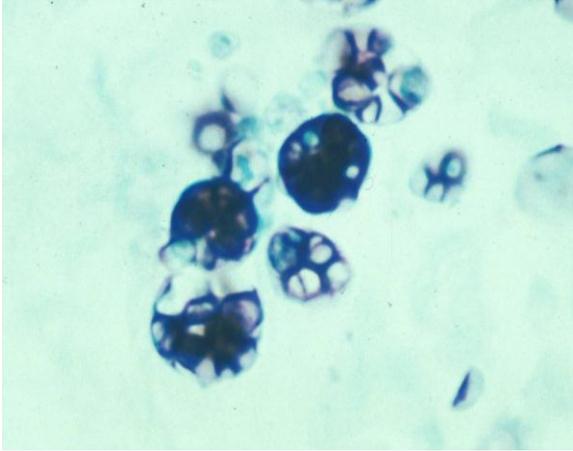


Figure 3: Structures seen on a Lactophenol Cotton Blue stain from the Sabdex colony.

Characteristic structures were also seen on H&E stains of tissue (figure 4).

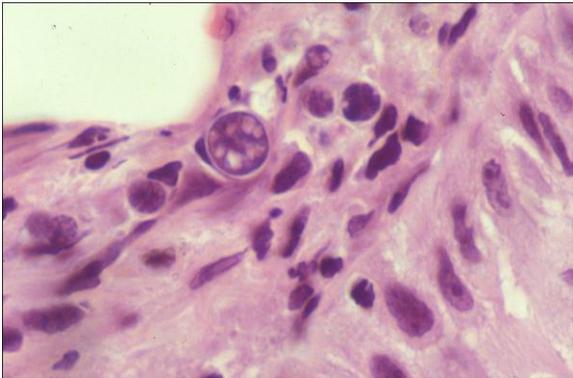


Figure 4: H&E Stain from infected tissue.

Organism identification was also confirmed using API-20C.

**What is the  
causative  
agent?**

# Discussion

**Diagnosis:** Cutaneous Protothecosis caused by the algae, *Prototheca wickerhamii*.

**Taxonomy:** Protothecosis is an uncommonly reported opportunistic infection caused by achlorophyllous algae within the family *Chlorellaceae* and genus *Prototheca*. The first reported human infection was from a farm worker in Sierra Leone in 1964.

**Habitat and Occurrence:** These organisms are ubiquitous in nature as saprophytes in soil, decaying plant matter and water. Infections occur in humans and occasionally in wild and domestic animals such as dogs, cats, and cows.

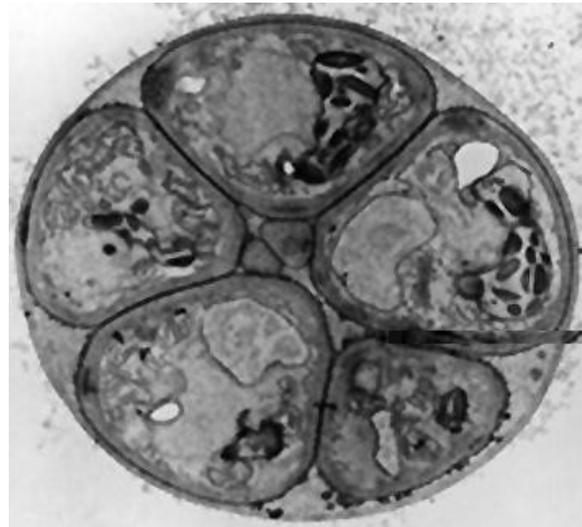
Five species have been described, but only two have been reported as human pathogens. *P. wickerhamii* is more common in human infections and *P. zopfii* in animals. Human Protothecosis generally presents in three clinical forms.

**Pathogenesis:** Cutaneous and/or subcutaneous infection is the most common form and occurs in patients with liver or renal illness, cancer, AIDS, or other immunosuppressive disorders. Olecranon (outside bony portion of the elbow) bursitis is seen more commonly in non-immunosuppressed patients who have repeated trauma to the affected elbow. Disseminated Protothecosis is rare and occurs mainly in severely immunosuppressed patients.

**Morphology:** *Prototheca* are unicellular organisms measuring 3 to 30  $\mu\text{m}$  in diameter, spherical to oval in shape that reproduce asexually by internal septation

and irregular cleavage, with subsequent rupture and release of endospores.

A sporangium (also called a “theca”) may contain up to 16 or more daughter cells or sporangiospores (figure 5).



**Figure 5:** Thin section of *P. wickerhamii* containing five sporangiospores (electron micrograph, x16,250)

The enlarging sporangiospores eventually rupture the parent cell, releasing spores that go on to develop mature endosporulating cells. Colonies of *Prototheca* appear yeast-like on common laboratory media.

Unlike fungi, they lack glucosamine in their cell walls and unlike bacteria they do not contain muramic acid. They are differentiated from bacteria and yeast by their cell size, shape, and mode of reproduction.

**Culturing:** *P. wickerhamii* and *P. zopfii* grow aerobically on standard laboratory media including [Sabouraud's Dextrose Agar](#). Like *Cryptococcus neoformans* they are inhibited by cyclohexamide, but not

chloramphenicol. Yeast-like colonies appear 24-72hrs after incubation at 25 to 35°C.

**Staining:** A wet prep from a colony unstained or stained with [lactophenol cotton blue](#) or [calcofluor](#) generally reveals the characteristic morphology of sporangiospores within a sporangium. Many commercial biochemical yeast identification systems can identify *Prototheca* to the genus level. [Trehalose assimilation](#) is a key test to differentiate *P. wickerhamii* from *P. zopfii*. Morphologically, the sporangia of *P. zopfii* are larger and more oval than those of *P. wickerhamii*, but there may be overlapping of these traits.

*Prototheca* are best visualized in tissue specimens stained with Gomori Methenamine Silver (GMS) that show the distinctive internal cleavage of the sporangia which is described as a “morula” or daisy-like appearance (figure 6A).

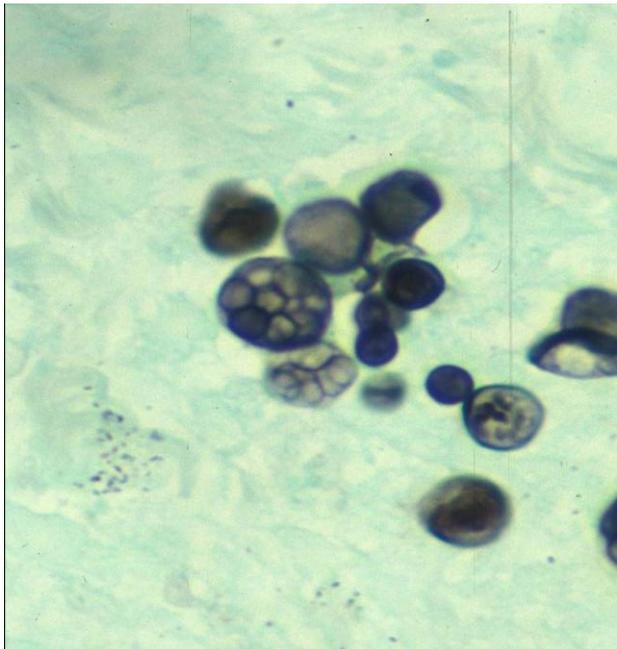


Figure 6a: A GMS stain showing the intact sporangia.

If sporangia have been broken open (figure 6B), the smaller sporangiospores (2 to 10 µm in diameter) may be confused with other potential fungal pathogens, but their size and the absence of budding will help in the differentiation.

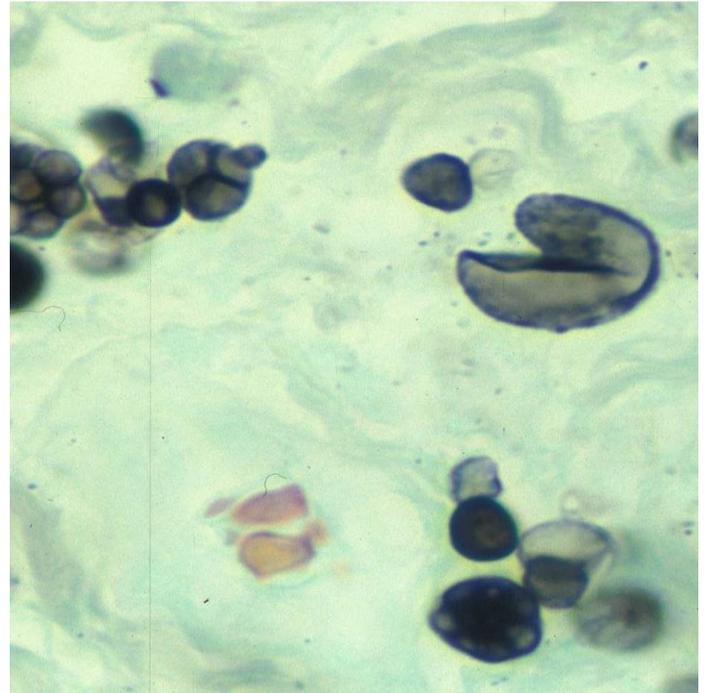


Figure 7: A GMS stain showing a ruptured sporangium releasing endospores.

**Treatment and Prognosis:** Although *Prototheca* are of low virulence, treatment can be difficult especially in severely immunocompromised patients. Infections may persist for many months and tend to recur and even spread. Surgical excision alone may cure some localized cases, but antimicrobial therapy is also necessary for persistent or deeper infections.

Amphotericin B with and without surgery has been the most common form of treatment especially in disseminated cases. Other anti-fungal agents have been used in cutaneous forms of the disease.

Although Protothecosis is still a rarely reported human infection, its worldwide incidence may be underestimated, due to misdiagnosis.



**Joseph DiPersio, PhD, DABMM**

*Dr. DiPersio is currently the Director of Microbiology Research at Summa Health System in Akron, Ohio and is an Associate Professor of Clinical Microbiology in Pathology at Northeastern Ohio University's College of Medicine.*

*He is a Diplomate of the American Board of Medical Microbiology and has directed clinical microbiology laboratories for over 30 years.*

*Dr. DiPersio has also published over one hundred scientific journal articles and national meeting abstracts.*



[Sales@HardyDiagnostics.com](mailto:Sales@HardyDiagnostics.com)  
[www.HardyDiagnostics.com](http://www.HardyDiagnostics.com)  
(800) 266-2222

1430 West McCoy Lane  
Santa Maria, CA 93455