At the Tip of the Tongue: A Novel Human Infection by a

Microsporidium

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Microsporidia are unicellular obligate intracellular parasites that infect a wide range of invertebrates and vertebrates. At least 8 microsporidian genera have been identified as human pathogens: *Anncaliia* (previously *Brachiola*), *Encephalitozoon*, *Enterocytozoon*, *Vittaforma*, *Nosema*, *Pleistophora*, *Trachipleistophora*, and '*Microsporidium*' (a collective term for microsporidia for which the genus has not been determined) [1]. The majority of human microsporidium infections have been described as causing chronic diarrhea in patients with HIV/AIDS; however, ocular infections have also been documented in healthy individuals [2].

In 2009, a 67 year old woman presented with tongue nodules. At the time of presentation, the woman was immunosurppressed due to treatment for non-Hodgkin's lymphoma. Histopathology showed myositis characterized by clusters of small, ovoid, basophilic organisms within the myocytes. The microorganisms were also noted by Giemsa, Warthin-Starry silver, and Lillie-Twort Gram stains. An onslide embedding protocol [3] was modified to deparaffinize, resin-embed and thin section the tissue.

Abundant 2-3 µm spores in the myocytes were seen by light and electron microscopy (Fig.1-6). The polar filament coiled around the sporoplasm 10-14 times (Fig.5). Additionally, the polar filament was anisofilar with the last few coils being smaller in diameter and the nuclei were in a diplokaryotic arrangement (Fig.6). Other morphological features included lamellar polaroplast, manubroid, posterior vacuole, and ribosomes.

PCR of formalin-fixed paraffin-embedded tissue resulted in the identification of a microsporidium that is closely related to *Tubulinosema acridophagus*. *Tubulinosema acridophagus* is a parasite found in North American grasshoppers such as *Melanoplus* and *Schistocerca* [4] and probably in the fruit fly *Drosophila melanogaster* [5]. Prior to this case, the genus *Tubulinosema* had not been associated with human infection.

Electron microscopy is a powerful technique in conjunction with molecular and histopathological analyses. Electron microscopy aided in the diagnosis of this case and specific PCR testing which lead to identification of a new species of microsporidium associated with human infection.

References

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FIG. 1.H&E. FIG. 2.Warthin-Starry silver stain. Light microscopic images of spores in the tongue myocytes (100 x objective). FIG. 3. Electron micrograph (EM) of numerous spores. FIG. 4. EM of a spore showing a cross-section of the manubroid (**M**), which is the straight portion of the polar filament surrounded by the lamellar polaroplast (**LP**). Bar = 500 nm. FIG. 5. An electron dense exospore (**Ex**) and electron lucent endospore (**En**) compose the spore wall. A limited number of polar filaments (**PF**) are visible. Bar = 100 nm. FIG. 6. The contents of the spore (sporoplasm) are made up of a posterior vacuole (**PV**), diplokaryon (**N**), ribosomes, polaroplast and polar filaments (**PF**) which wraps around the area between the nuclei and the endospore layer. Bar = 100 nm.