Reviewer’s report

Title: Histopathological Evaluation OF Ocular Microsporidiosis by different stains

Version: 1 Date: 3 February 2006

Reviewer: Jan Orenstein

Reviewer’s report:

Ocular Microsporidiosis

General: This is an important manuscript for those confronted with opportunistic infections such as microsporidia, especially in the age of HIV/AIDS. The question of how best to make the diagnosis of ocular microsporidiosis is new and the approach is well defined. It is interesting that the authors use two stains that are not generally used in rendering the diagnosis, i.e., modified acid fast and acridine orange. Why were they chosen? They apparently omit the commonly used modified Brown-Brenn or Brown-Hopps Gram stains, others have gotten excellent results with these stains. Although, they address it in their Discussion section, it is not clear why the authors didn’t present the results of two common successful diagnostic preparations, i.e., scrapings and touch imprints. Otherwise the methods are generally appropriate and well defined.

The biggest problem with this manuscript and the one that requires major compulsory revision is in the photomicrographs. It is difficult to correlate the descriptions of the spore appearance in the text to that in the figures. Perhaps the problem is that they are of too low magnification. Since the viewing was at 1,000x, why are most of the photographs at 500x? It is impossible to identify the central band or the polar dot in any figures. Incidentally, it has not been proven that the band, seen by LM, corresponds to the coiled polar filament. Because the figures are problematic, the Results and Discussion sections, and the conclusions are also a problem. Figure 1 is much too low magnification to confirm the description. There is a discrepancy between the description of the spore color in GMS in the text and in Table 2, brown versus black (the correct color). The acid fast stain is noted in the text to stain the spores bright red on a bluish background, however in the figure (4a), they are all dark blue, and the Masson trichrome (3d), which is almost black as opposed to the red mentioned in the text. The Cacofluor white (3a) barely stains the spores, which is not so if done correctly. The same is true for the other fluorochrome stains. The picture of the acridine orange stain (3b) does not correspond to the text at all.

It is interesting that the ease of standardizing the Gram chromotrope technique is alternatively described as easy versus not being standardized in their country. It would be very important to have speciated these microsporidia, which could have been done at the CDC. The writing is generally clear and there are only a few typos.

What next?: Unable to decide on acceptance or rejection until the authors have responded to the major compulsory revisions

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No
Declaration of competing interests:

I declare that I have no competing interests.