Chronic microsporidian infection of the nasal mucosae, sinuses and conjunctivae in HIV disease

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Abstract
A case of chronic infection of the nasal mucosae, sinuses and conjunctivae with a microsporidian parasite in association with HIV infection and immune deficiency is reported. This microsporidian resembles both Encephalitozoon cuniculi and the newly described Encephalitozoon hellem by electron microscopy. This occurred in an adult male resident in the UK with no history of foreign travel. Although there are previous descriptions of conjunctival infections from the USA, this is the first description of infection of the nasal epithelium. Further studies are underway to classify this protozoan.

Introduction
Microsporidia are obligate, intracellular, spore-forming protozoal parasites. They are widespread in the natural world and have long been recognized as a cause of disease in non-human hosts. However, only six microsporidial infections in patients without HIV infection have been documented. Reports of microsporidial infection in patients with AIDS first appeared in 1985, describing a new genus and species Enterocytozoon biennis, predominantly infecting the small intestine and usually presenting with chronic diarrhoea and weight loss. Studies have suggested that 30% of patients with HIV disease and pathogen negative chronic diarrhoea have Enterocytozoon biennis infection. Recent work utilizing duodenal biopsy specimens from HIV infected individuals with and without diarrhoea, and presumptively HIV negative individuals, has provided evidence of the pathogenicity of Encephalitozoon biennis, without evidence of a state of commensal carriage.

Encephalitozoon cuniculi usually infects non-human mammals. Infection usually begins in intestinal epithelial cells, and in animals first liver and then other extra-intestinal sites are infected, presumably via blood, lymph, or infected macrophages. In late infection central nervous system vasculitis and interstitial nephritis predominate. Two non-HIV infected children with encephalitozoon infection and CNS involvement have been described. Infection with protozoa morphologically identical to Encephalitozoon cuniculi was first described in HIV infection in association with a hepatic lesion, and then in a case of peritonitis.

Reports of microsporidian ocular infection in patients with AIDS first appeared early in 1990. Ocular infection presents with symptoms of foreign-body sensation, blurred vision or photophobia. Ophthalmological examination discloses conjunctivitis, decreased visual acuity, and a diffuse punctate keratopathy. Corneal or conjunctival scrapings or biopsies stained with Giemsa reveal dark-staining spores. Confirmation of the identity of the infecting microsporidia in these cases of hepatitis, peritonitis, and kerato-conjunctivitis has been by demonstrating their morphological similarity to Encephalitozoon cuniculi by electron microscopy. However, Didier et al recently isolated such microsporidia in cell culture from AIDS patients with kerato-conjunctivitis. Three such isolates were shown to be a new species, Encephalitozoon hellem, morphologically similar to Encephalitozoon cuniculi by electron microscopy, but distinct by SDS-PAGE analysis.

Case report
A 26 year old married bisexual man was first shown to be HIV antibody positive at routine screening in 1986. In January 1988 he remained well with CD4 0·40 x 10^9/l, HIV p24 Ag +ve, β2-microglobulin 4·1 mg/l. He first developed bilateral conjunctivitis in October 1988. Nasal obstruction and discharge became prominent in February 1989. By this stage he was unwell with weight loss and relapsing fevers and investigations showed CD4 0·19 x 10^9/l, HIV p24 Ag < 100 μg/ml, β2-microglobulin 4·6 mg/l. Zidovudine was commenced at 1 g/day with a good initial response. However, he continued to have episodic conjunctivitis, chronic nasal obstruction and discharge, as well as clinical and radiological evidence of sinusitis. This condition failed to respond to multiple courses of antibiotics and nasal decongestants. In June 1990 a diffuse punctate keratopathy was noted in both eyes, ENT examination showed multiple nasal polyps, and CT showed extensive opacities in the maxillary antra, and ethmoid and sphenoid sinuses, as well as minor cerebral atrophy. A full description of the ophthalmological findings and subsequent ocular response to treatment is being published elsewhere.

He was admitted to hospital in October 1990 with Pneumocystis carinii pneumonia and treated with intravenous pentamidine. Following recovery a formal nasal polypectomy was performed under general anaesthesia. After formalin fixation nasal polypectomy specimens...
were embedded in paraffin wax (Ral wax 1, BDH) and 4 μm sections were stained with haematoxylin and eosin, PAS and Grocott stains. A specimen was also embedded in methacrylate derived plastic (Immunobed, Park Scientific Ltd, Northampton) and 2 μm sections were stained by a two-stage May-Grünwald-Giemsa method. These preparations were examined by light microscopy. They revealed a polypoid nasal mucosa with a neutrophil infiltrate within the epithelium and a neutrophil, lymphocyte and plasma cell infiltrate in the adjacent submucosa. Many superficial epithelial cells contained numerous round...
opacification of the maxillary antra and ethmoid sinuses.

Figure 3 Oblique coronal CT post-second polypectomy pre-albendazole showing opacification of the maxillary antra and ethmoid sinuses.

puted tomographic evidence of bony destruction of the maxillary antra. Administration of albendazole produced symptomatic improvement and radiological regression of sinus infection. The manifestations of his ocular pathology are typical of previously described cases but this is the first case in an HIV infected subject described outside the USA. This subject has frequent contact with cats and dogs in his home environment and has never travelled outside the UK. We know that Encephalitozoon cuniculi can spread horizontally among mammals via contaminated excreta and one could postulate domestic animals as a source of this patient's infection.

This subject's symptoms of ocular and nasal infection were first noted up to 2 years prior to demonstration of his microsporidal infection. This symptom complex developed relatively early in the course of his immune deficiency, in parallel with symptoms of systemic HIV disease at a time when his CD4 count was of the order of 0-200 x 10^6/L. We found encephalitozoon easy to demonstrate by Giemsa stains or electron microscopy once suitable tissue specimens were obtained.

This case report extends the known tissue tropism of these organisms. This case appears to be a rarity, but precise definition of the frequency of microsporidal upper respiratory tract infection may depend on the development of serological assays. This in turn may depend upon propagation of such organisms and currently protozoa derived from the patient's polypectomy specimens are growing in tissue culture (Canning E U, personal communication). This man's infection appeared to respond well to albendazole. Encephalitozoon may be overlooked using light microscopy with certain stains and electron microscopy is at present vital in identification.

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