

Short Communication

First detection of intestinal microsporidia in Northern Nigeria

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Abstract:

Microsporidia are intracellular spore-forming protozoa that are increasingly being recognized as pathogens in humans.

Faecal samples were taken from 2250 HIV/AIDS and 1050 HIV-negative patients from Kano and Makurdi in Northern Nigeria, and were investigated for microsporidial infections by Giemsa staining technique (Light microscopy). In Kano, *Enterocytozoon bienuesi* was detected in 8 (14.17%) and *Encephalitozoon intestinalis* in 5 (2.60%) out of 192 HIV/AIDS patients screened. A mixed infection of both 0.52% was observed.

Results from Makurdi showed that *Enterocytozoon bienuesi* was detected in 13 (0.65%) and *Encephalitozoon intestinalis* in 96 (4.78%) out of 2008 HIV/AIDS patients examined. No mixed infection was observed. Microsporidial spores were not found in 1050 HIV-negative patients screened from both areas. There was a significant difference (χ^2 , $p < 0.05$) in infection rates between the HIV/AIDS and HIV-negative patients. This study aimed at detecting the prevalence of intestinal microsporidia to provide baseline data on the status of this disease in Nigeria.

Detection of Microsporidia in Immuno-compromised patients has not been described previously in this area.

Key Words: *Enterocytozoon bienuesi*, *Encephalitozoon intestinalis*, Microsporidial infections

Introduction

The term microsporidia is also used as a general nomenclature for the obligate intracellular protozoan parasites belonging to the Phylum Microsporidia. To date, more than 1,200 species belonging to 143 genera have been described as parasites infecting a wide range of vertebrate and invertebrate hosts.(1) The microsporidia spores of species associated with human infection measure from 1 to 4 μ m and that is a useful diagnostic feature. There are at least 14 microsporidian species that have been identified as human pathogens: *Enterocytozoon bienuesi*, *Encephalitozoon intestinalis*, *Encephalitozoon hellem*, *Encephalitozoon cuniculi*, *Pleistophora sp*, *Trachipleistophora hominis*, *T. anthropophthera* *Nosema ocularum*, *N. algerae*, *Vittaforma corneae*, *Microsporidium ceylonensis*, *M. africanum*, *Brachiola vesicularum*, *B. connori*. *Encephalitozoon intestinalis* was previously named *Septata intestinalis* but it was reclassified as based on its similarity at the morphologic, antigenic, and molecular levels to other species of this genus.(1)

Their role in human disease was not appreciated until the AIDS pandemic. Two microsporidia *Enterocytozoon bienuesi* and *Encephalitozoon intestinalis* have been identified as possible causes of diarrhoeal illness in HIV-infected patients.(2,3) Of the several species that infect man *Enterocytozoon bienuesi* were the first documented case and the most commonly recognized microsporidia that causes gastrointestinal disease in immuno-compromised patient particularly in HIV/AIDS. This parasite is commonly observed in HIV-infected patients with CD4 Lymphocytes counts of less than 50 cells/mm³ who complain of chronic diarrhoea, nausea, malabsorption and severe weight loss.(4,5) Whereas *Encephalitozoon intestinalis* causes both a disseminated and intestinal infections frequently associated with nephritis, sinusitis or bronchitis.(6) Cases of intestinal Microsporidiosis have been detected in HIV-seronegative asymptomatic individuals, and organ transplant recipients.(7,8) This study raises awareness on the presence of this disease which will help in the management of HIV/AIDS.

Methods

Study area are Makurdi on 9.5° N - 8.5° E located in Benue State, central Nigeria and Kano on 200 00' - 80 30' E located in Kano State, North East Nigeria.

Study populations were in-patients of Infectious Disease Hospital (IDH) Kano and General Hospital Makurdi. Each patient had a standardized clinical evaluation and provided a fresh stool specimen upon admission.

For microsporidial investigation, faecal specimen was homogenized in distilled water, filtered through a 300µm pore mesh sieve and centrifuged at 1500rpm, smears were prepared from sediments, fixed in methanol and stained with 10% Giemsa solution as described by Van-Gool et al., (9) and modified in this study, examined at x1000 magnification (oil emersion). Giemsa stained spores were broadly oval, with the cytoplasm staining light grey-blue with a dark stained nucleus Spores were classified as either small about 1.0 - 1.6 x 0.7 - 1.0µm (*Ent. bienuesi*) or large about 2.0 - 2.5 x 1.0 - 1.6µm (*E. intestinalis*).⁽⁶⁾

Results

Prevalence of microsporidia in stool samples of HIV/AIDS patients is shown in Tables 1 and 2. In Kano 14(7.29%) of the 192 patients examined had microsporidia, comprising; *Enterocytozoon bienuesi* 8/192 (4.17%), *Encephalitozoon intestinalis* 5/192 (2.60%) and mixed infection of both 1/192 (0.52%), while none of the 50 HIV-negative patients had microsporidia. In Makurdi, *Enterocytozoon bienuesi* was detected in 13/2008 (0.65%) and *Encephalitozoon intestinalis* in 96/2008 (4.78%) of the HIV/AIDS patients screened, none of the 1000 HIV-negative patients screened was positive. The infection rates between the HIV/AIDS and HIV-negative patients was significant (X², p<0.05).

Enterocytozoon bienuesi is more prevalent in Kano, while *Encephalitozoon intestinalis* in Makurdi. Figure 1 shows microsporidial spores found in study.

Table 1: Prevalence of intestinal microsporidiosis in Stool Samples of Patients with HIV/AIDS in Kano

Sources of Stool samples	Overall		Microsporidial Species Found		
			<i>Enterocytozoon bienuesi</i>	<i>Encephalitozoon intestinalis</i>	Mixed (<i>Ent. bienuesi</i> & <i>E. intestinalis</i>)
	No. Exam.	No. +ve*(%)	No. +ve(%)	No. +ve(%)	No. +ve(%)
HIV/AIDS patients	192	14 (7.29)	8 (4.17)	5 (2.60)	1(0.52)
HIV-ve patients	50	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Total	142	14	8	5	1

*+ve - Positive

Table 2: Prevalence of intestinal microsporidiosis in Stool Samples of Patients with HIV/AIDS in Makurdi

Sources of Stool samples	Overall		Microsporidial Species Found		
			<i>Enterocytozoon bienuesi</i>	<i>Encephalitozoon intestinalis</i>	Mixed (<i>Ent. bienuesi</i> & <i>E. intestinalis</i>)
	No. Exam.	No. +ve*(%)	No. +ve(%)	No. +ve(%)	No. +ve(%)
HIV/AIDS patients	2008	109 (5.43)	13 (0.65)	96 (4.78)	0(0.00)
HIV-ve patients	1000	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)
Total	3008	109	13	96	0

*+ve - Positive

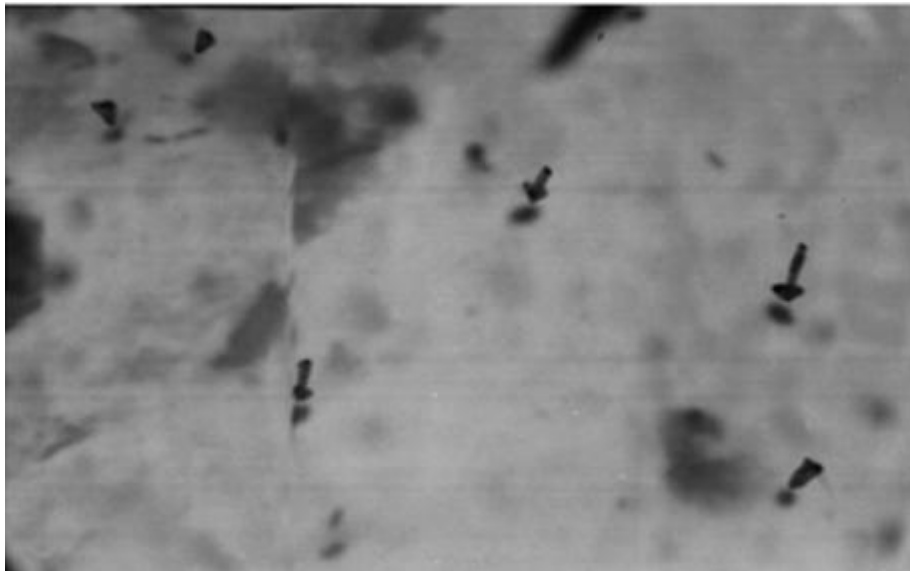


Figure 1: Oval Spores with dark stained nucleus of *Enterocytozoon bienuesi* (Arrow head) and *Encephalitozoon* species (Arrow) in faecal concentrate. (Magnification X1000)

Discussion

This report described an emerging gastrointestinal protozoan in North central, Nigeria, where they have not been studied. Microsporidia generally *Enterocytozoon bienuesi* caused up to 70% of otherwise unexplained cases of chronic diarrhoea involving patients with HIV/AIDS and low CD4 lymphocytes counts (10), the modified Giemsa staining technique for stool provides a useful means of screening clinical specimen.(9)

In the present study, *Enterocytozoon bienuesi* and *Encephalitozoon intestinalis* were detected areas studied with *Enterocytozoon bienuesi* being more prevalent in Kano and *Encephalitozoon intestinalis* in Makurdi. The occurrence of microsporidia in HIV/AIDS patients and not in HIV-negative patients conforms to earlier reports that microsporidia occurs in immuno-compromised patients particularly in HIV/AIDS.(5,11)

A final deduction from this study is the appreciation of the increasing prevalence of microsporidia and that most patients might have antigens and antibody levels, indicative of sub clinical infections, suggesting that this parasite could be a serious hazard to AIDS and other immuno-deficient patients due to causes other than AIDS, or probably due to most tropical diseases like malaria, schistosomiasis etc., and that infection are not at present being diagnosed. Since presently there is no satisfactory treatment for microsporidial infections, there is need for making chemotherapy for microsporidial disease a priority area of research.

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