

## Epidemiology, Clinical, Immune, and Molecular Profiles of Microsporidiosis and Cryptosporidiosis among HIV/AIDS patients

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**Abstract :** The objective of this study was to determine the prevalence of intestinal parasites, with special emphasis on microsporidia and Cryptosporidium, as well as their association with human immunodeficiency virus (HIV) symptoms, risk factors, and other digestive parasites. We also wish to determine the molecular biology definitions of the species and genotypes of microsporidia and Cryptosporidium in HIV patients. In this cross-sectional study, carried out in Kinshasa, Democratic Republic of the Congo, stool samples were collected from 242 HIV patients (87 men and 155 women) with referred symptoms and risk factors for opportunistic intestinal parasites. The analysis of feces specimen were performed using Ziehl-Neelsen stainings, real-time polymerase chain reaction (PCR), immunofluorescence indirect monoclonal antibody, nested PCR-restriction fragment length polymorphism, and PCR amplification and sequencing. Odds ratio (OR) and 95% confidence intervals were used to quantify the risk. Of the 242 HIV patients, 7.8%, 0.4%, 5.4%, 0.4%, 2%, 10.6%, and 2.8% had Enterocytozoon bienewisi, Encephalitozoon intestinalis, Cryptosporidium spp., Isospora belli, pathogenic intestinal protozoa, nonpathogenic intestinal protozoa, and helminths, respectively. We found five genotypes of E. bienewisi: two older, NIA1 and D, and three new, KIN1, KIN2, and KIN3. Only 0.4% and 1.6% had Cryptosporidium parvum and Cryptosporidium hominis, respectively. Of the patients, 36.4%, 34.3%, 31%, and 39% had asthenia, diarrhea, a CD4 count of  $<100$  cells/mm<sup>3</sup>, and no antiretroviral therapy (ART), respectively. The majority of those with opportunistic intestinal parasites and C. hominis, and all with C. parvum and new E. bienewisi genotypes, had diarrhea, low CD4+ counts of  $<100$  cells/mm<sup>3</sup>, and no ART. There was a significant association between Entamoeba coli, Kaposi sarcoma, herpes zoster, chronic diarrhea, and asthenia, and the presence of 28 cases with opportunistic intestinal parasites. Rural areas, public toilets, and exposure to farm pigs were the univariate risk factors present in the 28 cases with opportunistic intestinal parasites. In logistic regression analysis, a CD4 count of  $<100$  cells/mm<sup>3</sup> (OR = 4.60; 95% CI 1.70-12.20; P = 0.002), no ART (OR = 5.00; 95% CI 1.90-13.20; P = 0.001), and exposure to surface water (OR = 2.90; 95% CI 1.01-8.40; P = 0.048) were identified as the significant and independent determinants for the presence of opportunistic intestinal parasites. E. bienewisi and Cryptosporidium are becoming more prevalent in Kinshasa, Congo. Based on the findings, we recommend epidemiology surveillance and prevention by means of hygiene, the emphasis of sensitive PCR methods, and treating opportunistic intestinal parasites that may be acquired through fecal-oral transmission, surface water, normal immunity, rural area-based person-person and animal-human infection, and transmission of HIV. Therapy, including ART and treatment with fumagillin, is needed.

**Keywords :** diarrhea, enterocytozoon bienewisi, cryptosporidium hominis, cryptosporidium parvum, risk factors, africans

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