Triple protozoal enteropathy of the small intestine in an immunocompromised male: A rare histopathology report

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Abstract

Enteric protozoan parasites remain the most commonly encountered parasitic diseases in HIV infected patients. Opportunistic protozoal infections that infect GIT most commonly and cause diarrhea in HIV-infected patients are Cryptosporidium parvum, microsporidia and Isospora belli. Developing an infection with enteric protozoan parasites is dependent on absolute CD4+ cell counts, with lower counts associated with more severe, more atypical disease, and a greater risk of disseminated disease. We present histopathological features in a patient, where all three parasitic infections co-existed in HIV infected patient, who was under antitubercular therapy in addition to antiretroviral therapy and herpes zoster infection being treated by acyclovir.

Key words: Enteropathy, immunocompromised, protozoa

INTRODUCTION

Human immunodeficiency virus (HIV) infection is the most common cause for immunodeficiency state worldwide, with the hallmark of infection being depletion of CD4+ T lymphocytes, the essential components of the cell-mediated immunity. Enteric protozoan parasites remain the most commonly encountered parasitic diseases and continue to cause significant morbidity and mortality. Several secondary gastrointestinal (GIT) infections including bacterial, viral, parasitic, and fungal diseases besides primary HIV enteropathy is known. Opportunistic protozoal infections that infect GIT most commonly and cause diarrhea in HIV-infected patients are Cryptosporidium parvum, microsporidia, and Isospora belli. Immunosuppressed hosts are more likely to acquire infection after exposure and have more severe disease once the infection is established. They have disseminated infection rather than localized infection, and are unable to clear parasites, thereby, becoming chronic carriers.[1,2] Developing an infection with enteric protozoan parasites is dependent on absolute CD4+ cell counts, with lower counts associated with more severe, more atypical disease, and a greater risk of disseminated disease.[2] We present histopathological features in a patient, where all three parasitic infections coexisted in HIV-infected patient, who was under antitubercular therapy (ATT) in addition to antiretroviral therapy (ART) and Herpes Zoster infection being treated by acyclovir.

CASE REPORT

A 43-year-old male was diagnosed in November 2008 as HIV-positive and was on regular ART. He was admitted to hospital with complaint of passing loose stools 8-10 times per day since 1 month without associated pain abdomen, passage of blood or mucus in stool. He had an episode of diarrhea about
6 months back. It was diagnosed as cryptosporidial diarrhea on stool examination and he was treated with nitrazoxamide successfully.

The patient had several associated comorbidities. He was a documented case of disseminated tuberculosis since August 2010 and was on ATT. Presently, he is also a patient of Herpes Zoster (T6-T10 dermatome) and is on acyclovir therapy. On general physical examination, he was found to be a thinly built, well oriented, afebrile with a supine blood pressure of 90/66 mmHg and a regular pulse rate of 88/min. He had a significant left cervical and axillary lymphadenopathy with matted nodes. No organomegaly on abdominal examination was seen. Upper GI endoscopy revealed essentially normal esophagus and stomach and first part of duodenum. The second part of duodenum showed nodular folds. Ultrasonography of abdomen and pelvis showed calcified areas in segment VII of liver and multiple hypoechoic focal lesions (? granulomas).

The laboratory investigations revealed hemoglobin-10.9 g/dl, total leucocytic count -4,200/µl, differential leucocytic count was lymphocytic predominant (54%), and a platelet count of 1.5 lakh/µl. His absolute CD4+ T lymphocyte count was 219 cells/µl and viral load was less than 400 copies/ml. His liver and renal function tests and urine examination were essentially normal. Upon stool examination no ova, cyst, acid-fast bacilli, or any other organism was discerned. Stool was also negative for fat globules.

Histopathological examination of biopsy from second part of duodenum revealed mild atrophy in the form of blunting, shortening, and fusion of villi with crypt elongation at places. A focal disorganization of the epithelium, loss of nuclear polarity, cell crowding, and cell vacuolation was seen [Figure 1a]. Lamina propria showed an intense lymphoplasmacytic infiltrate along with few eosinophils and occasional macrophages. Numerous enterocytes showed intracytoplasmic loose parasitophorous vacuoles containing isosporidia schizonts and merozoites. These organisms were seen in the basal part of cytoplasm below and around the nucleus. The single organisms were the schizonts. Focally on luminal surface of occasional enterocytes, small bluish intracellular but extracytoplasmic round to oval amphophilic structures resembling cryptosporidia were seen. Also noted were foci of occasional enterocytes containing intracytoplasmic vacuoles with small, round to oval, bluish supranuclear refractile spores resembling microsporidia [Figure 1b]. No crypt destruction, dysplasia, or granulomas were seen. Lamina propria showed dilated vascular channels at places. Special histochemical stains to identify the parasites done were – Brown – Bren modification of Gram stain, PAS, Masson's trichrome, toluidine blue [Figure 1-d], Giemsa, modified acid fast, and Gomori's methanamine silver (GMS) stains. A histopathology opinion of enteritis due to triple infection by isosporidia, cryptosporidia, and microsporidia was offered.

Based on histopathology report, patient was put on tablets niazoxanide, Septran, Cifran, and Fluconazole. He has since improved and is under regular follow-up in the outpatient department and ART center of the hospital.

**DISCUSSION**

Opportunistic infections are generally restricted to severely immunosuppressed individuals.
and are considered AIDS-defining illnesses in HIV-infected patients, as they almost always occur when the CD4+ T-cell count falls below 200 cells/µl. Opportunistic parasitic infections can cause severe morbidity and mortality. Since many of these infections are treatable, an early and accurate diagnosis is important. This can be accomplished by a variety of methods such as direct demonstration of the parasite, spores and by serological tests to detect antigen and/or specific antibodies. Adherence to conventional procedure may not be appropriate in patients with AIDS. For example, antibody response may be poor in these patients and therefore immunodiagnostic tests have to be interpreted with caution.

Cryptosporidial disease commonly caused by *C. parvum*, has a predilection for the proximal small bowel, resulting in nonspecific thickening of the duodenum, jejunum, and proximal ileum. Cryptosporidiosis causes severe chronic and even fatal diarrhea with malabsorption and dehydration. Cryptosporidium species other than *C. parvum* and *C. hominis* were identified in 9.4% of the patients analyzed in a European study in HIV-infected. In untreated HIV infected patients with CD4+ T cell counts of < 300/l, the incidence of cryptosporidiosis is approximately 1% per year. Laboratory diagnosis is made by demonstrating oocysts in the specimens using modified acid-fast stain. Cryptosporidial infection is confined to luminal surface of enterocytes from base of crypts to the tips of the atrophied villi. Villus atrophy and crypt hyperplasia are nonspecific and can be present in varying degrees as in the case of microsporidiosis and isosporidiasis. Cryptosporidium characteristically is intracellular but extracytoplasmic and bulges from the apical surface of the epithelium. Heavy burdens usually correlate with severe mucosal damage and even infiltration of neutrophils. Therapy is predominantly supportive, and marked improvements have been reported in the setting of effective ART.

Microsporidia on the other hand are small, unicellular, obligate intracellular parasites that reside in the cytoplasm of enteric cells. The main species causing disease in human is *Enterocytozoon bieneusi* and *Septata intestinalis*. The mode of transmission for the various microsporidia remains obscure. It is accepted that infection occurs via the orofecal route. The clinical manifestations are similar to those for cryptosporidia and include abdominal pain and diarrhea. Sometimes a coinfection with cryptosporidiosis may be seen. The small size of the organism may make it difficult to detect; however, diagnosis is made by demonstrating spores in stool specimens by modified Trichrome stain or in the tissue biopsy specimen [Figure 1c]. Microsporidia-infected bowel displays villous atrophy and pleomorphism, crypt hyperplasia, and increased number of intraepithelial lymphocytes. The tip of infected villus appears as progressively more disorganized aggregates of crowded cells (piling up) that are undergoing degeneration and necrosis. Many enterocytes contain bluish supranuclear plasmodia, some of which mold the nucleus. Individual sloughing enterocytes are tear shaped and invariably contain refractile spores. Definitive diagnosis may require electron-microscopic examination of a stool specimen, intestinal aspirate, or intestinal biopsy specimen.

Isosporiasis caused by *I. belli* oocysts ingestion of which produces protracted and sometimes a profuse diarrhea. The oocysts are demonstrated in fecal smears by modified acid-fast stain. Upper
GI endoscopy with biopsy is a diagnostic method of choice, if oocysts are not visualized in stool. The morphology reveals a nonspecific villous atrophy and crypt hyperplasia like cryptosporidium and microsporidium. Eosinophilia of lamina propria may be a striking feature. Histopathological changes are more extensive than due to mere presence in the infected cells, suggesting that the parasite releases toxic agents, which cause hypersensitivity reaction. Parasite is intracellular in location, relatively large in size, and is surrounded by parasitophorous vacuole making its identification easy. The clinical syndromes of isospora infection are identical to those caused by cryptosporidia. The important distinction is that infection with isospora is relatively easy to treat with trimethoprim-sulfamethoxazole.

To the best of our knowledge, it is the first case being reported in India with triple infection diagnosed on tissue biopsy, when the stool examination did not reveal the parasites. Parasite stool examination continues to be recommended for immunocompromised with persistent and/or recurrent diarrhea. Simple histochemical stains help in identification of the parasites. Identification of parasites causing intestinal infections helped in specific therapy limiting the morbidity and providing symptomatic motor relief. The take-home message from this presentation to our young colleagues and clinicians is that in a known immunosurveillance case, one must look out for additional tissue pathogens for timely and correct therapy.

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