Isolated cervical cryptococcal lymphadenitis without meningitis in an immunocompetent human immunodeficiency virus-negative child: a rare case report

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Introduction: Cryptococcal infection is the second most common life-threatening, opportunistic infection in human immunodeficiency virus (HIV)-infected individuals after Mycobacterium tuberculosis. Most cryptococcal infections start in the respiratory tract and secondarily involve the central nervous system, skin and bone marrow but rarely disseminate to lymph nodes.

Case presentation: Cases of disseminated cryptococcal lymphadenitis with or without meningitis have been reported in HIV-infected as well as immunocompetent HIV-negative individuals. Here, we report a rare case of isolated cervical cryptococcal lymphadenitis without meningitis in an immunocompetent HIV-negative child diagnosed by fine-needle aspiration cytology (FNAC) of the involved lymph nodes.

Conclusion: Clinical presentations of extrapulmonary cryptococcosis in HIV-positive hosts are variable and vague. The present case scenario proves that the same might also be true for HIV-negative individuals. FNAC is a quick, feasible, economical and reliable method for the diagnosis of cryptococcal lymphadenitis when correlated with microbiological, biochemical and histopathological examinations.

Keywords: Cryptococcal lymphadenitis; Cryptococcus neoformans var. neoformans; fine-needle aspiration cytology; HIV negative; immunocompetent.

Introduction
Cryptococcosis is a chronic, subacute or (rarely) acute pulmonary, systemic or meningitic infection caused by the yeast Cryptococcus neoformans (Rippon, 1988). Cryptococcal infection is the second most common life-threatening, opportunistic infection in human immunodeficiency virus (HIV)-infected individuals after Mycobacterium tuberculosis (Das et al., 2002; Dinato et al., 2006; Suchitha et al., 2008; Srinivasan et al., 2010; Mardi & Kaushal, 2012; Deshmukh et al., 2013), and in immunocompromised hosts the yeast is disseminated to almost all organs of the body (Suchitha et al., 2008). Cryptococcal infections have a special predilection for the central nervous system, and meningitis is the commonest presentation (Rippon, 1988; Dinato et al., 2006). Lymph node involvement is rarely found in cryptococcal infection in either HIV-infected or HIV-negative individuals (Kumarguru et al., 2009; Srinivasan et al., 2010), although some cases of disseminated cryptococcal lymphadenitis with or without meningitis have been reported in both HIV-infected and immunocompetent HIV-negative individuals (Das et al., 2002; Agarwal et al., 2004; Taneja et al., 2008; Mitha et al., 2010; Oscar et al., 2010).

We present a rare case of isolated cervical cryptococcal lymphadenitis without meningitis in an immunocompetent HIV-negative child. To the best of our knowledge, no such presentation has been documented previously. Fine-needle aspiration cytology (FNAC) is an inexpensive, easily available, simple, reliable and rapid method of diagnosing cryptococcal lymphadenitis (Suchitha et al., 2008).

Case report
A male child aged 1 year and 10 months was admitted to the paediatric ward on 31 July 2013 with abdominal distension and fever of 1 month’s duration, yellowish discoloration of the skin and eyes, swelling of the legs for 1 week and occasional vomiting. There was no history of loss of consciousness or seizures, and his appetite was good. His mother provided a history of an uneventful pregnancy, term delivery at a hospital and no post-natal complications. The child had been treated for his ailments
with multiple combinations of antibiotics in a private hospital before attending our hospital. On examination, the child weighed 10 kg, was 85 cm in height, and was alert and conscious with a fever of 99 °F. Pallor and icterus were present along with swelling of the abdomen and legs. He had multiple, enlarged, non-tender, discrete post-auricular and posterior cervical lymph nodes, the largest of which measured 2.5 × 2.5 cm² (Fig. 1). Hepatomegaly (5 cm below the right mid-clavicular line) and splenomegaly (3 cm below the left mid-clavicular line) were present. No other abnormality was detected on examination of the respiratory, cardiovascular and central nervous systems. The patient was admitted with provisional diagnosis of fever with anaemia and hepatosplenomegaly under investigation. The differential diagnoses considered for this patient were: malaria, disseminated tuberculosis, leukaemia, lymphoma and HIV infection.

His blood investigations revealed the following: haemoglobin level of 6.0 g dL⁻¹, total leucocyte count 15 600 cells mm⁻³, neutrophils 40 %, lymphocytes 51 %, monocytes 2 %, eosinophils 7 %, erythrocyte sedimentation rate of 67 mm at the end of 1 h, platelets 400 × 10⁹ cells mm⁻³, urea 19 mg dL⁻¹, creatinine 0.6 mg dL⁻¹, total bilirubin 7.9 mg dL⁻¹, direct bilirubin 4.4 mg dL⁻¹, indirect bilirubin 3.5 mg dL⁻¹, aspartate aminotransferase 160 U l⁻¹, alanine transaminase 54 U l⁻¹, total protein 6.6 g dL⁻¹, albumin 2.8 g dL⁻¹, globulin 3.8 g dL⁻¹, albumin:globulin ratio 0.7, potassium 4.0 mmol l⁻¹, sodium 136 mmol l⁻¹ and chloride 102 mmol l⁻¹. A peripheral blood smear showed dimorphic anaemia with eosinophilia (7 %) and was negative for malarial parasites. A test for HIV status was non-reactive as per the National AIDS Control Organisation (NACO) guidelines (NACO, 2007). Haematological, histological, immunological and microbiological tests for hepatitis B and C, lymphoma and immunoglobulin subclasses and quantities were all within normal limits (Kim et al., 2005). CD4 and CD8 counts were 689.23 and 515.34, respectively, giving a CD4:CD8 ratio of 1.34 and 8.66 % CD4 (Cytomics FC 500 MPL, software version MXP-2.2; Beckman Coulter). High-performance liquid chromatography for haemoglobinopathy showed normal results. A chest X-ray showed no radiological abnormality (Fig. 2). Ultrasonography of the whole abdomen reported hepatosplenomegaly with diffuse hepatic steatosis and minimal ascites. No mediastinal or retroperitoneal lymphadenitis was observed (Fig. 3).

A cerebrospinal fluid (CSF) direct mount showed two lymphocytes per high-power field and no pus cells or microorganisms. A Gram stain (centrifuged sample) of the CSF did not show any pus cells or microorganisms. Acid-fast staining from gastric lavage and CSF were negative. CSF inoculated on blood agar, chocolate agar, Sabouraud dextrose agar (SDA), Sabouraud cycloheximide chloramphenicol agar (SCCA) and brain–heart infusion broth was sterile. Urine and blood cultures were sterile. A stool culture did not show any growth of enteric pathogens.

Gram staining, Giemsa staining, a wet (normal saline) mount and an India ink preparation of the enlarged lymph node specimen obtained by FNAC showed round and oval yeast cells 6–20 μm in diameter with some showing narrow-based budding surrounded by halos suggestive of capsules (Fig. 4). Haematoxylin and eosin, Grocott–Gomori methenamine, periodic acid–Schiff and mucicarmine stains also showed budding yeast cells with capsules (Wong et al., 2011) (Fig. S1, available in the online Supplementary Material). No malignancy was detected on histopathological examination. An acid-fast stain of the lymph node sample did not reveal any acid-fast bacilli. Cultures on SDA and SCCA showed creamy-white, round, moist, convex, pasty colonies (Das et al., 2002; Dharmshale et al., 2006) (Fig. S2). A urease test carried out on the culture along with a negative control (Candida albicans ATCC 90028) and positive control (Cryptococcus neoformans var. neoformans ATCC 32608) showed urease production by the test organism (Agarwal et al., 2004).

**Fig. 1.** Multiple, firm, enlarged, non-tender, discrete post-auricular and posterior cervical lymph nodes, the largest of which measured 2.5 × 2.5 cm².

**Fig. 2.** Chest X-ray showing no radiological abnormality.
Nitrate was reduced to nitrite. A sugar assimilation test showed that the yeast assimilated sucrose, trehalose, glucose, cellobiose, galactose, xylose, raffinose, dulcitol, inositol and maltose (Taneja et al., 2008). Melibiose and lactose were not assimilated (Fig. S4). Thus, mycologically the yeast was confirmed as Cryptococcus neoformans var. neoformans (Talerman et al., 1970; Rippon, 1988; Suchitha et al., 2008; Chander, 2009; Anvikar et al., 2011). A Vitek 2 system identified the yeast as Cryptococcus neoformans (YST ID; bioMérieux). An antifungal susceptibility test (AST YST; bioMérieux) showed that fluconazole, flucytosine, voriconazole and amphotericin B were susceptible with MICs of $\leq1$, $\leq1$, $\leq0.12$ and $0.5\ \text{mg}\ \text{ml}^{-1}$, respectively. A diagnosis of cryptococcal lymphadenitis was made and treatment started with fluconazole 100 mg once daily (Dharmshale et al., 2006; Dinato et al., 2006; Mitha et al., 2010; Tsai et al., 2010; Bao et al., 2013). Within a week of treatment, there was a marked clinical improvement along with a reduction in lymph node size. The largest node that was originally $2.5 \times 2.5\ \text{cm}^2$ had reduced to $0.5 \times 0.5\ \text{cm}^2$ in size. A plan for follow-up for every 15 days or earlier if necessary was made and the patient was discharged on 12 August 2013 with advice to continue the drug.

**Discussion**

Cryptococcal infection mostly starts in the respiratory tract, resolves spontaneously and is usually asymptomatic (Kim et al., 2005). Immunocompromised states, most commonly HIV infection, facilitate the survival and dissemination of Cryptococcus to various parts of the body, although dissemination has been well documented even in
immunocompetent HIV-negative individuals (Suchitha et al., 2008). Dissemination of infection to various body organs is common in spite of resolution of the primary lung lesions. The central nervous system is the commonest site of secondary involvement resulting in meningoencephalitis (Kim et al., 2005; Dharmshale et al., 2006; Mitha et al., 2010). Organs such as the skin, respiratory tract, gastrointestinal tract and bone marrow are seen to be involved, in order of decreasing frequency, but lymph node involvement has been reported to be rare (Talerman et al., 1970; Suchitha et al., 2008). Cryptococcal lymphadenitis with meningitis with or without HIV infection, cryptococcal mesenteric lymphadenitis with meningitis with or without HIV infection and disseminated cryptococcosis in HIV-negative individuals have been well documented (Talerman et al., 1970; Shravanakumar et al., 2003; Agarwal et al., 2004; Suchitha et al., 2008; Taneya et al., 2008; Mitha et al., 2010; Bao et al., 2013). Here, we have reported a unique case of isolated cervical cryptococcal lymphadenitis in an immunocompetent HIV-negative child without meningitis, lung involvement or mediastinal lymphadenitis. Lymphonodular cryptococcosis, a rare entity seen in paediatric patients with selective lymph node involvement, is mostly described with multiple-site lymph node dissemination, which was not found in this case (Supparatpinyo, 1991).

Cryptococcal lymphadenitis is an uncommon form of cryptococcosis and is considered to be an AIDS-defining criteria according to the Centers for Disease Control and Prevention guidelines (Suchitha et al., 2008). However, the fact that HIV-non-infected individuals may also present with cryptococcal lymphadenitis has been proven by this study as well as by previous studies (Agarwal et al., 2004; Mitha et al., 2010). In developing countries like India, tubercular lymphadenitis is more common than cryptococcal lymphadenitis or may be coexistent (Anvikar et al., 2011; Deshmukh et al., 2013). Hence, it would be a logical choice to rule out tuberculosis by performing an acid-fast stain from the lymph node samples.

Gram staining, Giemsa staining and histopathological examination of lymph node samples revealed minimal granulomatous and inflammatory cellular reactions in this case, which is a typical feature of cryptococcal lymphadenitis, especially in children. Similar findings were shown by Shravanakumar et al. (2003) and Srinivasan et al. (2010).

Cryptococcus neoformans var. neoformans was identified as the fungal agent in this case. This finding is in contrast to most studies where Cryptococcus neoformans var. neoformans was found to be associated with immunocompromised HIV-infected individuals, whilst Cryptococcus gattii was the fungal agent implicated in HIV-negative immunocompetent patients (Rippon, 1988; Agarwal et al., 2004; Chander, 2009; Mitha et al., 2010).

Early diagnosis of disseminated cryptococcosis is difficult as it includes a large number of differential diagnoses such as tuberculosis, malignancies, Molluscum contagiosum, Blastomyces dermatitidis, Histoplasma capsulatum and Kaposi’s sarcoma, which can easily mimic the lesions (Anvikar et al., 2011). Clinical acumen and a high index of suspicion are the key factors for early diagnosis and initiation of treatment, which leads to a favourable outcome by preventing dissemination of infection. Mortality in untreated cases of disseminated cryptococcosis can be as high as 80 % (Dharmshale et al., 2006).

Fluconazole is the drug of choice for localized cryptococcal infection without central nervous system involvement (Dharmshale et al., 2006; Dinato et al., 2006; Jadhav et al., 2010; Mitha et al., 2010; Tsai et al., 2010; Bao et al., 2013). In our case, antifungal susceptibility tests showed that the fungus was sensitive to fluconazole. Thus, fluconazole was started in this patient as soon as the diagnosis was confirmed.

This case has given us new insight, showing that isolated cryptococcal lymphadenitis can present without meningitis in an immunocompetent, HIV-negative child. Hence, an immunocompetent host with enlarged lymph nodes showing no improvement with antibiotics, especially anti-tubercular, should undergo FNAC followed by fungal culture, a direct mount, India ink preparation and special stains like haematoxylin and eosin, periodic acid–Schiff and mucicarmine to rule out cryptococcal lymphadenitis, even though no meningitis or involvement of other sites may be present. Clinical presentations of extrapulmonary cryptococcosis in HIV-positive hosts are variable and vague. The present case scenario indicates that the same might also be true for HIV-negative individuals.

FNAC is a quick, feasible, economical and reliable method for the diagnosis of cryptococcal lymphadenitis when correlated with microbiological, biochemical and histopathological examinations. It provides samples for culture and is repeatable when patients return for follow-up during treatment.

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References


