Disseminated *Mycobacterium avium* complex infection diagnosed from bone marrow biopsy and culture in a patient with human immunodeficiency virus – A case report

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SUMMARY

***Mycobacterium avium* complex (MAC) is one of the common non-tuberculous mycobacterium (NTM) organisms that typically affects patients with severe acquired-immunodeficiency syndrome with CD4 count of less than 50 cells/mm³ as an opportunistic infection. Here, we present a case of disseminated MAC diagnosed via bone marrow biopsy and culture in a severely immunocompromised patient with human immunodeficiency virus. An 18-year-old male, newly diagnosed human immunodeficiency virus who was poorly compliant with highly active antiretroviral therapy, presented with lethargy, prolonged fever, chronic diarrhoea, and significant weight loss. Biochemically, he had persistent spiking fever and bicytopenia and therefore a bone marrow trephine biopsy was obtained. Report of the bone marrow biopsy revealed granulomatous inflammation, Ziehl-Neelson stain was positive for acid-fast bacilli. Bone marrow aspirate culture detected MAC, but the sensitivity test was not available until few weeks later. He was empirically treated for disseminated NTM with Azithromycin, Ciprofloxacin, and Ethambutol while awaiting sensitivity report from the marrow blood culture. Unfortunately, the patient continued to deteriorate despite HAART and NTM treatment. Eventually, he succumbed to the disease after 3 weeks of treatment.**

INTRODUCTION

Mycobacterium avium complex (MAC) infection is an opportunistic infection commonly found in patients who are in an immunocompromised state. It can be caused by either *Mycobacterium avium* or *Mycobacterium intracellulare*. The advent of effective antiretroviral treatment (ART) in the modern era has resulted in a marked decline in MAC infection in human immunodeficiency virus (HIV) patients. In the United States, a large-scale observational retrospective cohort study from 1992 to 2015 reported a significant reduction in incidence from 65.3/1000 in 1992, which was prior to the beginning of highly active antiretroviral therapy (HAART) era in 1996, to 2.0/1000 in 2015.[1] However, the disease is prevalent among patients who presented late with advanced acquired-immunodeficiency syndrome (AIDS) disease and in those patients who were not on HAART. Till date, there have been no reported cases of MAC in bone marrow in Malaysia. Here, we report a case study of disseminated MAC (dMAC) infection in a HIV patient diagnosed by bone marrow biopsy and trephine.

CASE PRESENTATION

An 18-year-old male student was diagnosed with HIV in August 2021 through sexual transmission. He was warded from July 2021 to September 2021 for a splenic microabscess, which resolved after 31 days of intravenous (IV) Ampicillin-Sulbactam 3 g thrice a day. At that time, his CD4 count was 2 cells/mm³ and a CD8 count of 183 cells/mm³; serial blood cultures yielded no organisms. He was initiated with ART, which includes oral Emtricitabine-Tenofovir once daily and Efavirenz 600 mg once daily. Trimethoprim-sulfamethoxazole 2 tablets, once a day was prescribed for prophylaxis of *Pneumocystic carinii* infection.

He presented to us one month later, in October 2021, with progressive generalised body weakness, prolonged fever, and loose stool for the past month, which was 3 episodes per day and weight loss of 5 kg. He reported poor adherence to the ART medications. On examination, the patient was tachycardic with a pulse rate of 136 beats per minute and a body temperature of 38.6°C. He was cachexic, lethargic with pale conjunctiva, and clinically appeared dehydrated. Cardiovascular and lung examinations were unremarkable. Abdominal examination showed an enlarged liver measuring 16 cm, Traube's space was dull, and multiple shotty inguinal lymph nodes bilaterally.

On admission, complete blood count revealed persistent bicytopenias: haemoglobin 5.9 g/dL, total white cell count $1.82 \times 10^3/\mu L$, and platelet $405 \times 10^3/\mu L$. The peripheral blood film showed hypochromic microcytic anaemia and leukopaenia, with lymphopenia and neutropenia likely secondary to underlying infection; no blast cells were seen. The liver enzymes were raised with aspartate transaminase (AST) -236 U/L, alanine transaminase 202 U/L, and alkaline phosphatase 829 U/L. Inflammatory markers were raised with C-reactive protein 168 mg/L and erythrocyte sedimentation rate 118 mm/h. Stool samples were sent for culture, ova and cyst, and *Clostridium Difficile* toxins, and the results were negative. The chest and abdominal radiographs were normal. Sputum samples sent for Acid Fast Bacilli smear and *Mycobacterium* Tuberculosis (MTB) cultures as well as

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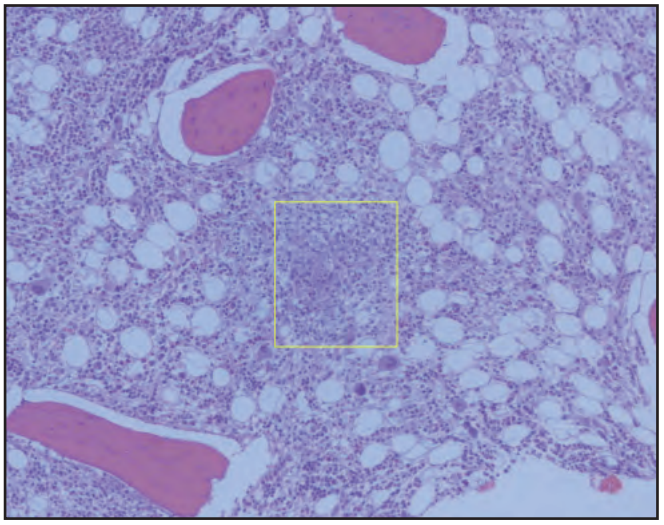

Fig. 1: A small foci of epithelioid granuloma surrounded by epitheloiod histiocytes (H&E stain, x20obj)

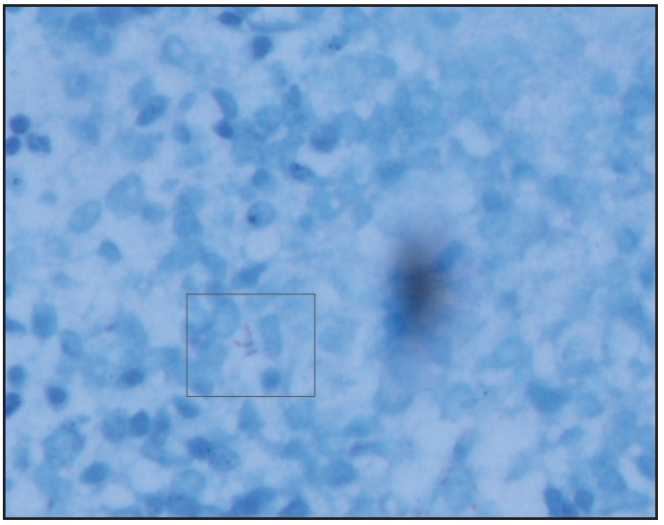

Fig. 2: Acid fast bacilli seen in the bone marrow (x60 obj)

Figure 1 and 2 are the slides of bone marrow trephine biopsy

GeneXpert MTB were negative. Hepatitis B and Hepatitis C serology as well as Rapid Plasma Reagin test were negative. With the evidence of hepatomegaly, persistent temperature spikes, severe bicytopenia, and persistent raised alkaline phosphatase, bone marrow trephine biopsy was obtained due to high clinical suspicion of haematological malignancies.

Findings of bone marrow biopsy revealed tuberculous granulomatous inflammation; Ziehl-Neelson stain was positive for acid-fast bacilli (Figures 1 and 2). Preliminary report yielded non-tuberculous mycobacterium (NTM) species. The case was discussed with the infectious disease consultant and the patient was given empirical treatment with Azithromycin, Ciprofloxacin, and Ethambutol while awaiting the susceptibility report. Rifampicin was not initiated in view of transaminitis.

The bone marrow aspirate culture detected MAC and this microorganism was identified through the Line Probe Assay. Susceptibility testing using the broth microdilution method showed a minimum inhibitory concentration (MIC) of <0.12 for rifampicin, < 0.5 for ethambutol, 2.0 for ciprofloxacin, and 0.25 for clarithryomycin. Despite undergoing treatment for disseminated MAC infection for 3 weeks, there was no clinical improvement and was complicated with septic shock due to hospital-acquired infection. Despite adequate resuscitation and timely administration of broad-spectrum antibiotics, in his case, he was given parenteral Piperacillin-Tazobactam. Unfortunately, his condition continued to deteriorate, eventually succumbed to the disease.

DISCUSSION

MAC is one of the common NTM organisms that are ubiquitous in the environment. Its niche includes water, soil, and dust. The mode of transmission is usually via ingestion, inhalation, or dermal contact with the environment.[2] It typically affects patients who are immunocompromised, such as those with haematological malignancies, patients receiving immunosuppressants after solid organ transplantation, as well as severe AIDS with CD4 count <50 cells/mm^3 as an opportunistic infection.

The disease has a long incubation period with an insidious onset. It presents as either a disseminated or localised syndrome, i.e. lymphadenitis, osteomyelitis, pericarditis, cutaneous or soft tissue abscesses, or central nervous system involvement. Disseminated MAC with multiorgan involvement can be found in as high as 20–40% of patients with advanced immunosuppressed states who were not on effective ART or undiagnosed HIV patients.

Clinical presentation of MAC is vague and includes prolonged fever, night sweats, significant weight loss, fatigue, diarrhoea, and abdominal pain. Lack of specific symptoms often make the clinical suspicion of NTM infection difficult. Disseminated MAC may present as pancytopenia or leukopaenia due to bone marrow infiltration as well as elevated alkaline phosphatase. In patients who are on HAART, MAC disease can present as immune reconstitution syndrome (IRIS), which is a systemic inflammatory response that is indistinguishable from active MAC disease, often with absent bacteremia. MAC-associated IRIS often presents as a localized abscess and lymphadenitis. The immune response is related to a temporary increase in CD4 counts and a rapid reduction in HIV ribonucleic acid counts.

The diagnosis of dMAC is based on clinical features and is confirmed by the isolation of MAC organisms from sterile cultures such as blood, lymph nodes, bone marrow biopsy, or other sterile tissue or body fluid samples. Although blood culture remains the gold standard in detecting dMAC as it is a less invasive procedure and has almost 100% sensitivity, the downside of blood culture is its long turnaround time, which takes up to 6 weeks. On the other hand, bone marrow trephine biopsy is able to detect acid-fast bacillus (AFB) -stain

positive and granulomatous formation early in dMAC, enabling early initiation of treatment for its rapid diagnostic strategy. However, further staining is required to identify the type of acid-fast microorganism. In our case, we were able to detect AFB from the bone marrow biopsy within a few days of the procedure and the marrow aspirate culture later identified MAC using Line Probe Assay. This enabled early initiation of empirical treatment.

The infiltration of MAC organisms into the bone marrow indicates rapid disease progression and reflects severe immunocompromise.[3] A study by Hussong et al. concluded that bone marrow biopsy staining has an important value as it allows rapid recognition of AFB microorganisms with a mean time of 1.1 days as compared to bone marrow aspirate culture and MTB blood culture, which took 19 days and 16 days, respectively.[4] The study concluded that routine staining of core biopsy from bone marrow allows prompt identification and early initiation of antimicrobial treatment, which may improve the survival outcome for these patients. Prior to the availability of ART, MAC infection was the most common opportunistic infection and contributed substantially to the mortality and morbidity in severely compromised HIV patients with CD4 counts 50 cells/μL.[5] However, since the advent of ART, the incidence of MAC infection has decreased exponentially, although patients with low CD4 counts are still at risk.[6]

The current standard treatment for MAC infection is multidrug therapy, which consists of Clarithromycin or Azithromycin, Ethambutol, and Rifabutin in combination. Alternative drugs include fluoroquinolones or Amikacin. Primary prophylaxis for MAC is no longer recommended in today's practice, regardless of CD4 count.[7] Once a diagnosis is made, the treatment duration for MAC is a minimum of 12 months. After the 12-month therapy, secondary prophylaxis, either using Clarithromycin or Azithromycin, should be continued until the CD4 count reaches at least 100/mm^3. In our case report, our patient was treated empirically with Azithromycin, Ciprofloxacin, and Ethambutol while waiting for the culture sensitivity report.

Disseminated MAC carries a high mortality rate in patients with AIDS and the prognosis is poor. Kobayashi et al reported a mortality rate of 29% ($p = 0.022$) from a single-center study on HIV patients with dMAC who were on ART, particularly those with MAC bacteremia and low CD4 count.[8] Our patient succumbed to the illness despite 3 weeks of combination treatment consisting of empirical antibiotics with ART. This is likely due to rapid disease progression and severe sepsis from the infection.

CONCLUSIONS

The diagnosis of MAC is challenging as the clinical features are generally nonspecific. Patients with AIDS who presented with fever, persistent anemia, and high alkaline phosphatase should raise a high clinical suspicion for dMAC infection. Isolation of MAC from blood cultures has a long waiting period for susceptibility as mycobacterium is a slow-growing microorganism. More studies need to be done to evaluate the role of routine trephine biopsy for prompt diagnosis of dMAC in severely immunocompromised patients.

REFERENCES

1. Collins LF, Clement ME, Stout JE. Incidence, long-term outcomes, and healthcare utilization of patients with human immunodeficiency virus/acquired immune deficiency syndrome and disseminated Mycobacterium avium complex from 1992–2015. Open Forum Infect Dis. 2017; 4(3).
2. Nishiuchi Y, Iwamoto T, Maruyama F. Infection sources of a common non-tuberculous mycobacterial pathogen, Mycobacterium avium complex. Front Med 2017; 4(27): 2-10.
3. Corti M, Palmero D. Mycobacterium avium complex infection in HIV/AIDS patients. Expert Rev Anti Infect Ther 2008;6(3):351–63.
4. Hussong J, Peterson LR, Warren JR, Peterson LC. Detecting disseminated Mycobacterium avium complex infections in HIV-positive patients: the usefulness of bone marrow trephine biopsy specimens, aspirate cultures, and blood cultures. Am J Clin Pathol 1998; 110(6): 806-9.
5. Kobayashi T, Nishijima T, Teruya K. High mortality of disseminated non-tuberculous mycobacterial infection in HIV-infected patients in the antiretroviral therapy era. PLoS One. 2016; 11(3): e0151682.
6. Yangco BG, Buchacz K, Baker R, Palella FJ, Armon C, Brooks JT. Is primary Mycobacterium avium complex prophylaxis necessary in patients with CD4 <50 cells/μL who are virologically suppressed on cART?. AIDS Patient Care STDS. 2014; 28(6): 280-83.
7. Brooks JT, Song R, Hanson DL, Wolfe M, Swerdlow DL. Adult and adolescent spectrum of disease working group, discontinuation of primary prophylaxis against Mycobacterium avium complex infection in HIV-infected persons receiving antiretroviral therapy: observations from a Large National Cohort in the United States, 1992–2002. Clin Infect Dis. 2005; 41(1): 549-53.
8. Kobayashi T, Nishijima T, Teruya K, Aoki T, Kikuchi Y, Oka S. High mortality of disseminated non-tuberculous mycobacterial infection in HIV-infected patients in the antiretroviral therapy era. PloS One. 2016; 11(3): e0151682.