

CASE REPORT

Brucellosis with Bone Marrow Granulomas- Two Case Reports

Majed Momin¹, Amitha G², Anamika Aluri³, Abhijeet Ingle⁴, Sandhya Rani C⁵

1- Consultant Pathologist, 2- Consultant Microbiologist, 3- Consultant Biochemist & Head Biochemistry lab, 4- Consultant Histopathologist. Yashoda Hospital, Malakpet, Nalgonda X-Roads, Hyderabad – 500036

Abstract

Brucellosis is zoonotic disease of worldwide distribution and still remains endemic in some developing countries. Clinical presentation of this systemic disease may be acute, insidious or chronic. The disease mimics many illnesses and presents diagnostic difficulties. Automated blood culture system and ELISA proved useful as new laboratory based diagnostic method. Although various regimens have been used in the treatment, a combination of doxycycline and rifampicin is the best treatment for human brucellosis. Here we present two cases of brucellosis who presented with history of fever, cough and weakness. On routine investigations anemia and hepatitis were present, bone marrow examination revealed non-caseating granuloma. In both these cases history and bone marrow findings mimicked chronic inflammatory condition like tuberculosis but final diagnosis of brucellosis was made on basis of bone marrow and blood culture results which were positive for Brucella.

Key Words: Anemia, Brucellosis, Hepatitis, Non-caseating bone marrow granulomas

Address for correspondence: Dr. Majed Momin, Consultant Pathologist, Department of Laboratory Medicine, Yashoda hospital, Malakpet, Nalgonda X-roads, Hyderabad – 500036. Mobile No. +919849076574, Email id: majedmomin878@yahoo.co.in

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Introduction

Brucellosis is a multisystem disease with non-specific symptoms that generally occur within 2 weeks but sometimes up to 3 months after inoculation. Human beings contract the disease as a result of consuming unpasteurized, contaminated goat's milk or soft cheese that had been infected with *Brucella melitensis* (*B. Melitensis*)^[1]. This Gram-negative, aerobic non spore forming coccobacillus is a free-living, soil-dwelling organism that usually infects goats and sheep. In infected host, the bacterium appears as intracellular localization, particularly within the reticuloendothelial system^[2]. There are six species of *Brucella* and four of which are known to infect humans^[3]. It is well documented that appropriate antimicrobial therapy improves the clinical and hematological status of patients with brucellosis.

Case - 1

A 63-year-old man referred to our center (Yashoda hospitals, Malakpet, Hyderabad) with

complaints of fever associated with chills, dry cough, generalized weakness, swelling over both feet for four weeks. Physical examination revealed an ill looking old man with mild pallor, body temperature 98.4 degree F, pulse rate 80/min, blood pressure 140/70 mm of Hg and respiratory rate 20/min. Systemic examination was unremarkable.

Results of laboratory tests made on admission were as follows: White blood cell count: 8900/mm³, Platelet count: 28500/mm³, Hemoglobin: 8.1g/dl with rouleux formation on peripheral blood smear. Erythrocyte sedimentation rate: 120 mm/hr, Creatine (Cr): 1.7 mg/dL, SGOT: 64 U/L, SGPT:83 U/L, Alkaline phosphatase: 343 U/L. Urinalysis revealed proteinuria. Plasma glucose 151mg/dl. Malarial antigen, Typhi-Ig M, Weil felix test and stool for ova and cyst came negative.

Total Iron: 24 mcg/dl, TIBC 224mcg/dl, Transferrin saturation: 10.7%. and serum protein electrophoresis absent M band and increase in alpha 1 and alpha 2 globulins suggestive of acute inflammatory disease. Reticulocyte count 3.0%. Direct coombs test came positive.

Chest X-ray was normal. HRCT Chest showed mild bilateral pleural effusion. Pleural fluid diagnostic tap was attempted, but unsuccessful due to minimal fluid.

Bone marrow aspiration and biopsy was done and bone marrow culture sent. Bone marrow aspiration cytology and trephine biopsy histology revealed non-caseating granulomas with surrounding lymphocytes. Stain for AFB (Z-N staining) was non-contributory. Bone marrow culture and Brucella Ig M were positive for brucella and TB-PCR negative. Mantoux test revealed skin induration 8mm after 48 hours.

Figure- 1: Bone marrow aspiration smear- Granuloma with epithelioid cells & lymphocytes

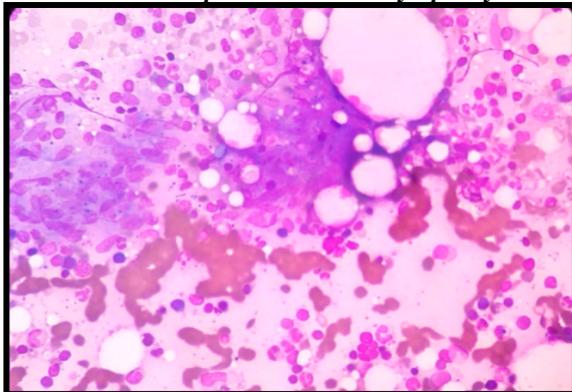
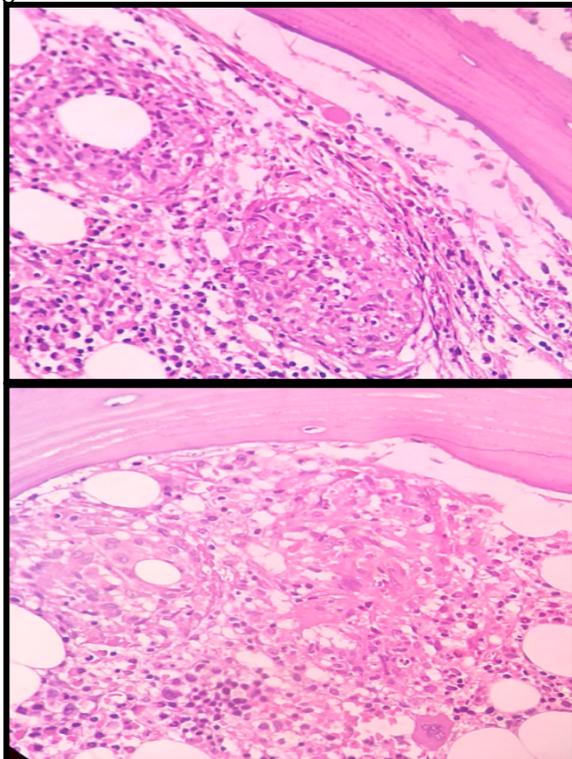


Figure- 2: Bone marrow biopsy- Non-caseating granuloma



Patient treated as a case of Brucellosis and Doxycycline (2×100 mg/day) and Rifampicin (1×600 mg/day) were started. The patient received this treatment for six weeks. One week after the initiation of the treatment, our patient's symptoms subsided and he became afebrile. Also he had normal liver function tests and serum creatinine.

Case – 2

A 51-year-old female patient was admitted with history of fever associated with chills and dry cough for 15 days. She also had history of loose stools three episodes. Vital signs were normal except for a temperature of 38° C. Her physical examination was otherwise normal except for scar of hysterectomy which was done 10 years back.

The results of laboratory tests are as follows: hemoglobin: 10.0 g/dl, erythrocyte: 4.18x10¹²/L, leukocyte: 7300/cumm (neutrophils 50%, lymphocyte 45%, monocyte 03%, eosinophils 02%), thrombocytes: 3.5x10³ / L, sedimentation rate: 40 mm/h.

Liver function tests were as follows: aspartate aminotransferase: 224U/L (14-60); alanine amino transferase: 143 U/L (9-69), Alkaline phosphatase: 110U/L and total bilirubin, 0.6mg/dL, (0.1 mg/dLdirect).

Haemostatic tests were in normal range. Serological tests for EBV, CMV, hepatitis B, hepatitis C, Typhi Ig M, rubella, toxoplasma Ig M were all negative. Widal Test, smear for malarial parasite and direct antiglobulin test were negative. Urinalysis revealed proteinuria. Brucella Ig M was positive.

Chest X ray show normal study. Abdominal ultrasonography was normal with post-hysterectomy status.

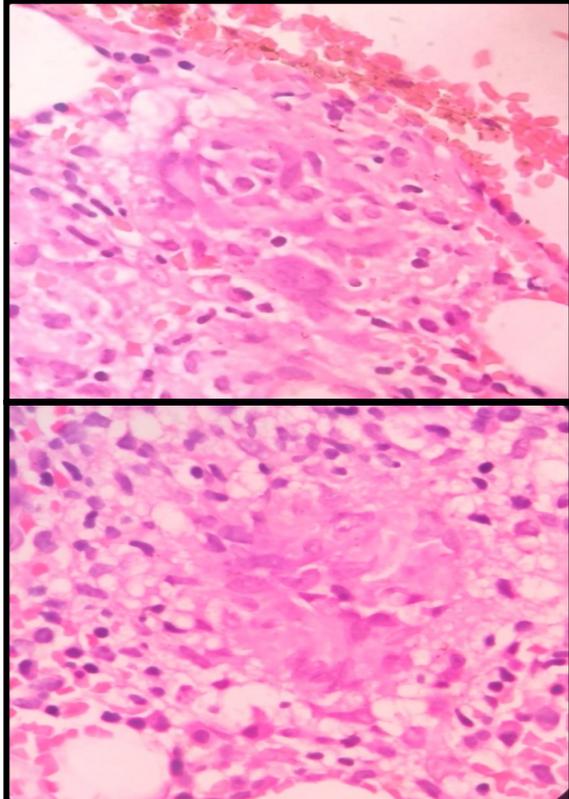
As a part of PUO, bone marrow aspiration and biopsy and bone marrow culture planned.

Bone marrow aspirate was diluted and scanty. Peripheral blood for culture sent since bone marrow was scanty. Bone marrow histology examination reveals granulomas consisting epithelioid cells and lymphocytes. Caseation necrosis was not seen. Stain for AFB (Z-N Staining) not contributory. Bone marrow biopsy TB-PCR was sent for analysis (Figure-3).

In view of non caseating granuloma and positive brucella Ig M, patient was treated as a case of brucellosis with Doxycycline (2×100 mg/day)

and Rifampicin (1×600 mg/day). Patient responded very well for treatment regime and liver function test subsided within one week and became afebrile. Patient discharged with advice to continue the same treatment for next six weeks and follow up.

Figure- 3: Bone marrow granulomas in biopsy



Discussion

Brucellosis is one of the leading infections causing pyrexia of unknown origin (PUO) in developing countries [4,5]. In spite of high prevalence of brucellosis in animals and favorable factors for transmission to human beings, the diagnosis of brucellosis can be missed easily due to lack of clinical suspicion and appropriate diagnostic facilities. This delay in diagnosis can result in increased incidence of sequelae. Brucellosis is a multi-systemic disease [6] which can mimic various diseases as in our both cases patient's clinical history mimicked tuberculosis, while lab finding showed evidence of hepatic and hematological involvement. Hepatic involvement in brucellosis is not rare and hepatomegaly has been documented in 15–20% of cases [7], however, liver function tests are usually mildly elevated [8].

In patients with brucellosis anemia results most commonly from bone marrow suppression, hypersplenism, autoimmune hemolysis or alteration of iron metabolism secondary to infection. The possible mechanism for thrombocytopenia and pancytopenia have been suggested to include hypersplenism or bone marrow aplasia. Bone marrow granulomata have no significant role in the hematological changes. The erythrocyte sedimentation rate is normal or mildly elevated and it is of Prognostic rather than diagnostic significance if it is during the early stage of the disease [9].

Isolation and identification of infectious agent is still considered gold standard for the definitive diagnosis. *Brucella* sps. can be isolated from blood, bone marrow, tissues (in focal complications) and body fluids, however, the isolation is dependent on stage of the disease, type of samples taken and culture methods used. Isolation from bone marrow is considered to be more sensitive at any stage of disease than blood but should be restricted to specific cases only because of painful procedure of sample collection [10,11].

ELISA are indirect primary binding assays and detect the antibodies present in test sample by using antiglobins/bacterial cell receptors labeled with isotopes, fluorochromes or enzymes as detecting molecules. ELISA has been found to be an acceptable alternative to blood culture for the diagnosis of brucellosis [12].

Different antibiotic regimens have been employed in the treatment of brucellosis including the following in various combinations: TMP/SMZ, rifampicin, doxycycline, ciprofloxacin, gentamicin and streptomycin. The mean duration of treatment is usually 6 weeks, but in case of complications like infective endocarditis or spinal involvement, therapy may be prolonged for up to 3 months [13-17].

Conclusion

Brucellosis is an important medical problem in developing countries. These cases emphasize that *Brucella* can affect any organ and system of the human body. The prevalence of involvement of the bone marrow in brucellosis may be underestimated as there is no significant role of granulomata in peripheral hematological change and painful procedure of marrow itself. However both our patients' marrow showed

evidence of non-caseating granuloma, use of bone marrow procedure and bone marrow aspirate culture essential in assessment of febrile patient.

Since the different treatment regime available, by proper diagnosis we can avoid long term medication as brucellosis mimicking other chronic granulomatous diseases like tuberculosis. Nevertheless, we believe that brucellosis should be considered in the differential diagnosis of hepatitis and bone marrow non-caseating granulomas for those who live in or have visited endemic areas.

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