

Comparative Evaluation of Simultaneous Bone Marrow Aspiration and Bone Marrow Trephine Biopsy – A Tertiary Care Hospital Based Cross-Sectional Study

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Abstract: **Background:** A bone marrow evaluation encompasses bone marrow aspirate (BMA) and bone marrow biopsy (BMB) along with a review of peripheral smear examination. **Aim:** To compare the diagnostic utility of simultaneous BMA and BMB and to study the concordance rate between the two. **Material and methods:** All BMA and BMB that were simultaneously done between January 2012 and June 2014 in a Civil hospital, Ahmedabad were reviewed and analyzed in detail. **Results:** Seventy three aspiration and biopsy slides were reviewed. The commonest indication was anemia (40%) followed by pancytopenia (27%) pyrexia of unknown origin (19%); 4% aspirates biopsies were inadequate. The overall concordance rate between aspirates and biopsy was found to be 73.9%. Concordance was highest for acute and chronic leukemias, metastatic deposits, multiple myeloma, megaloblastic anemia, tuberculosis and least for lymphoproliferative disorders, myelofibrosis and marrow hypoplasia. Trephine biopsy was useful for patients' with acute leukemias, lymphoproliferative disorders, myelofibrosis, marrow hypoplasia, osteitis fibrosa cystica and metastasis. BMB and BMA could detect tuberculous granulomas in 85.17% and 57.14% cases, respectively. However, parasitic infections (malaria) could be diagnosed exclusively in BMA. **Conclusions:** BMA and BMB are important, useful complementary diagnostic tools giving a higher diagnostic yield when used in conjunction.

Keywords: Bone marrow aspiration, bone marrow biopsy, concordance

1. Introduction

Examination of bone marrow has wide applications in clinical medicine. It is a useful investigative tool for the diagnosis of many hematological and nonhematological disorders. It also forms an important prerequisite for follow-up of patients undergoing chemotherapy, bone marrow transplantation and other modalities of medical treatment. In addition, it is also a formidable weapon for noncontributory or inconclusive results during the clinical evaluation process. Ideally, a bone marrow examination encompasses examination of bone marrow histology, Imprint cytology, bone marrow aspirate (BMA) along with a review of peripheral smear. When these examinations are done simultaneously they complement each other. The aspirates are used to mainly study the morphology and maturation of cells and they can also be useful for additional flow cytometric, immunophenotyping, cytogenetic and molecular studies. The trephine biopsies on the other hand provide information on details of marrow architecture, cellularity and distribution of cells and is especially useful in diagnosing focal lesions like granulomas, lymphoid or carcinoma infiltrates.^(2,3)

2. Aim

To evaluate the usefulness of simultaneous BMA (Bone marrow aspiration) and BMB (Bone marrow biopsy) and study the concordance rate between two.

3. Material and Methods

The study was conducted in the Dept. of Hematology in a Civil Hospital, Ahmedabad between January 2012 and June 2014, a total of 73 patients underwent diagnostic Bone marrow aspiration and Bone marrow biopsy for diagnostic purposes.

Out of the above, 73 cases in which both BMA and BMB were carried out simultaneously were included in the study. For preparing the BMA, the standard technique of using Jamshidis needle was applied for collection. About 0.25-0.5 ml of aspirate was obtained from the posterior superior iliac spine into a syringe and delivered onto a clean glass slide. After the smears were made the slides were air-dried and were stained subsequently with May-Grunwald-Giemsa (MGG) stain. Trephine biopsies were performed using the Jamshidineedle with the length of biopsy ranging from 1 to 2 cm. The biopsies were fixed in Bouins fluid, decalcified for 48 hours and embedded in paraffin from which 4 μ thin sections were made and stained with hematoxylin and eosin (H&E) stain. Other special stains like reticulin, periodic acid-Schiff (PAS) and Perls were done, wherever indicated. BMA were considered inadequate when diluted with blood or doesn't contain sufficient particles. Trephine biopsies were considered inadequate when there were <3 intertrabecular spaces or when there was inadequate/total absence of hematopoietic elements.

4. Results

A total of 73 cases were reviewed from January 2012 to June 2014.

- 1) The age of the subjects ranged from three years to 83 years with a male predominance (2.4:1).
- 2) The commonest indication was refractory anemia (40%) followed by pancytopenia (27%) pyrexia of unknown origin (19%); 4% aspirates and biopsies were inadequate.
- 3) The overall concordance rate between aspirates and biopsy was found to be 73.9 %. The results of comparative evaluation was further subdivided into:
 - List of cases, which were concordant on both aspirates and biopsy (Table 1).
 - List of cases, where diagnosis was possible on BMB alone and aspirates were not contributory (Table 2).
 - List of cases, where diagnosis was possible on aspirates alone. BMB was not contributory
- 4) The highest positive concordance was seen in megaloblastic anemia and reactive marrow 37.03% and 20% respectively. Other cases with a good positive correlation, even though the number of cases was small, include Aplastic anemia, Acute and chronic leukemias, metastatic deposits, multiple myeloma and tuberculous granuloma.
- 5) Correlation was found to be least with myelofibrosis and marrow hypoplasia.

5. Discussion

- 1) One of the objectives of our study was to examine the utility of simultaneous BMA and BMB in routine hematological practice. All the patients included in our study underwent simultaneous BMA and BMB evaluation. They were primarily newly presenting and undiagnosed cases. Generally, patients with hematological malignancies are referred to a GCRI.
- 2) The failure rate of BMA and BMB in our study was 4% and was mainly due to a diluted marrow or a dry tap. Inadequate biopsies mainly comprised of subcortical bony tissue with no marrow elements. Humphries et al reported a rate of 3.9% from 2,235 simultaneous bone marrow aspirations and biopsies.
- 3) The overall percentage of cases, which were concordant on both aspirates and biopsy was 73.9 %, which is in agreement with the study published by Ramji Rai et al⁶ who reported a positive concordance in 68.25% of the 160 patients' studied.
- 4) The highest correlation was seen with megaloblastic anemia and reactive marrow as 37.03% and 20% respectively. Most of our cases of megaloblastic anemia were diagnosed on BMA only with trephine biopsy being reported as hypercellular leading to the low concordance.
- 5) We had seven cases of acute leukemia (4 AML, 3 ALL) all of which were detected on trephine biopsy. Aspirates were diagnostic in only 71.4% of cases (4 AML, 1 ALL) as the BMA in the case of 2 ALL was diluted with peripheral blood, hence no opinion could be given.
- 6) Of the five cases of chronic leukemias (1 CLL, 4 CML), all the chronic lymphocytic leukemias were concordant on aspirate and biopsy. The myeloid leukemias showed

a 60% concordance. The aspirates were better able to classify the phases of CML (chronic phase n = 1, accelerated phase n = 2 and blastic phase n = 1) as compared to biopsy. One case of CML with myelofibrosis was inconclusive on aspirate.

- 7) Though, it was possible to diagnose both acute and chronic leukemias on peripheral smear examination, bone marrow evaluation was important as a baseline investigation for exact typing of leukemia and to be compared with subsequent aspirations during treatment.
- 8) There were two cases of multiple myeloma, all of which could be diagnosed with combined study of trephine biopsy and marrow aspiration. Sabharwal et al⁹ found that in cases where BMA was inconclusive for multiple myeloma, BMB complemented BMA as it helped to identify compact masses of plasma cells with no stroma. This was seen in 3 of our 7 cases and it was a significant histological feature for differentiation between myelomatous and nonmyelomatous plasmacytosis and the role of BMB proved invaluable for this.
- 9) All 3 cases of metastatic deposits (Adenocarcinoma) were detected on BMB. All the biopsies in these cases showed tumor associated desmoplastic reaction, demonstrated by reticulin stain. BMA and BMB showed a 66.6% concordance in the diagnosis of metastatic tumors. This is in agreement with other authors who have found a lower sensitivity (30-50%) on aspirates in the diagnosis of metastatic tumors as compared to biopsy.
- 10) The BMA and BMB is a useful tool in the diagnosis of unsuspected nonhematological malignancy. Though, the number of cases is small, it is in close agreement with the study by Ozkalemkas et al,¹¹ who studied 19 cases of unsuspected nonhematological malignancies in aspirates and biopsy
- 11) One case was diagnosed as osteitis fibrosa cystica on BMB. The patient
- 12) had osteolytic lesions and was being investigated for suspected multiple myeloma. BMB showed increased osteoclastic activity affecting subperiosteal, osteonal and endosteal surfaces. When this diagnosis was offered, follow-up investigations revealed the patient to have hyperparathyroidism due to a parathyroid adenoma. Though osteitis fibrosa cystica is uncommon, bone is a target tissue for parathyroid hormone (PTH) whose calcitropic effect is mediated largely via catabolic actions on it and can be useful in the diagnosis of metabolic bone disease. While the BMA done in a known case of chronic renal failure for anemia was inconclusive, BMB revealed features of high turnover renal osteodystrophy characterized by increased bone resorption predominating over bone formation.
- 13) Bone marrow hypoplasia (n = 2) and myelofibrosis (n = 1) could only be diagnosed on biopsy. The aspirates in all the above cases could not offer any definitive opinion. Bone marrow histology in myelofibrosis was classified according to the criteria by Bartl et al.¹⁴ Reticulin was graded from 1+ to 4+ as per the grading system proposed by Bauertneister et al.¹⁵ All the cases were found to be in the intermediate stage of myelofibrosis i.e. Grade II characterized by hyperplastic areas of marrow mixed with fibrotic areas consisting of

both reticulin and collagen fibers with reduction in erythrocytic and myelocytic elements.

- 14) We found that 75% of granulomatous lesions in bone marrow were diagnosed by BMB alone. Aspirates were diagnostic in only 25% cases. All these cases were tuberculous granulomas. Though AFB stain was positive in only one case, the clinical history, presentation, laboratory and radiological findings helped in reaching the diagnosis of tuberculous granuloma in the others. BMB had a higher yield in detecting granulomas as compared to BMA, which is consistent with other studies^(16,17)

6. Conclusion

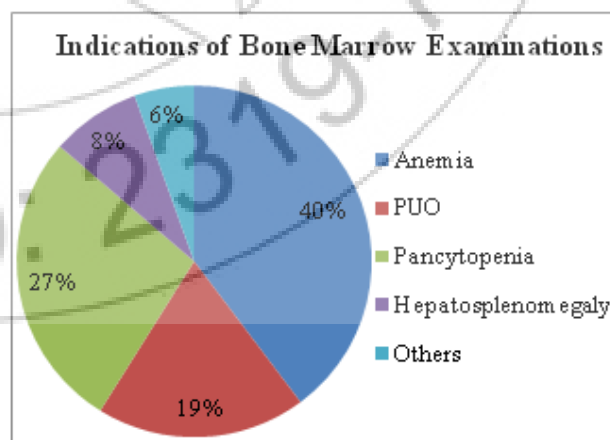
The utility and efficacy of BMB as compared to BMA have been discussed and debated. The answer, though complicated remains essentially the same. Both procedures complement each other with aspiration smears being primarily used for a cytological diagnosis and trephine biopsies helpful for a histological diagnosis as cellularity, fibrosis and architectural patterns are better visualized. Metastatic deposits and focal lesions also have a better diagnostic yield on BMB. This view has been supported by other authors. Bone marrow examination was a very useful tool in the evaluation of unsuspected conditions such as metabolic bone disorders and metastasis as observed in our study.

Despite the growing complexity and dependence on newer methodologies and ancillary assays including immunochemistry, cytogenetic analysis, flow cytometry and molecular assays, which may have augmented and refined the diagnostic criteria formerly obtained by light microscopy, the traditional role of examination of BMA and histopathological evaluation of BMB remains as important as it has been in the past.

References

- [1] Riley RS, Hogan TF, Pavot DR, Forysthe R, Massey D, Smith E, et al. A pathologist's perspective on bone marrow aspiration and biopsy: I. Performing a bone marrow examination. *J Clin Lab Anal* 2004;18(2):70-90.
- [2] Bain BJ. Bone marrow aspiration. *J Clin Pathol* 2001;54(9):657-63.
- [3] Bain BJ. Bone marrow trephine biopsy. *J Clin Pathol* 2001;54(10):737-42.
- [4] Lee SH, Erber WN, Porwit A, Tomonaga M, Peterson LC; International Council for Standardization In Hematology. ICSH guidelines for the standardization of bone marrow specimens and reports. *Int J Lab Hematol* 2008;30(5):349-64.
- [5] Humphries JE. Dry tap bone marrow aspiration: clinical significance. *Am J Hematol* 1990;35(4):247-50.
- [6] Toi PCh, Varghese RG, Rai R. Comparative evaluation of simultaneous bone marrow aspiration and bone marrow biopsy: an institutional experience. *Indian J Hematol Blood Transfus* 2010;26(2):41-4.
- [7] Bain BJ, Clark DM, Lampert IA, Wilkins BS (Eds.). *Bone marrow pathology*. 3rd edition, Blackwell Science: Italy 2001:p.372-3.

- [8] Charles KS, Winfield DA, Angel C, Goepel J. Audit of bone marrow aspirates and trephine biopsies in multiple myeloma - a single centre study. *Clin Lab Haematol* 2004;26(6):403-6.
- [9] Sabharwal BD, Malhotra V, Aruna S, Grewal R. Comparative evaluation of bone marrow aspirate particle smears, imprints and biopsy sections. *J Postgrad Med* 1990;36(4):194-8.
- [10] Grann V, Pool JL, Mayer K. Comparative study of bone marrow aspiration and biopsy in patients with neoplastic disease. *Cancer* 1966;19(12):1898-900.
- [11] Ozkalemkas F, Ali R, Ozkocaman V, Ozcelik T, Ozan U, Ozturk H, et al.
- [12] The bone marrow aspirate and biopsy in the diagnosis of unsuspected nonhematologic malignancy: a clinical study of 19 cases. *BMC Cancer* 2005;5:144.
- [13] Malluche HH, Koszewski N, Monier-Faugere MC, Williams JP, Mawad H. Influence of the parathyroid glands on bone metabolism. *Eur J Clin Invest* 2006;36(Suppl 2):23-33.
- [14] Schwarz C, Sulzbacher I, Oberbauer R. Diagnosis of renal osteodystrophy. *Eur J Clin Invest* 2006;36(Suppl 2):13-22.
- [15] Gupta R, Setia N, Arora P, Singh S, Singh T. Hematological profile in pyrexia of unknown origin; role of bone marrow trephine biopsy vis-à-vis aspiration. *Hematology* 2008;13(5):307-12.
- [16] Basu D, Saravana R, Purushotham B, Ghotekar LH. Granulomas in bone marrow - a study of fourteen cases. *Indian J Pathol Microbiol* 2005;48(1):13-6.
- [17] Frisch B, Bartl R, Burkhardt R, Jäger K. Histologic criteria for classification and differential diagnosis of chronic myeloproliferative disorders. *Haematologia (Budap)* 1984;17(2):209-26.
- [18] Bauermeister DE. Quantitation of bone marrow reticulin - a normal range. *Am J Clin Pathol* 1971;56(1):24-31.
- [19] da Silva MR, Stewart JM, Costa CH. Sensitivity of bone marrow aspirates in the diagnosis of visceral leishmaniasis. *Am J Trop Med Hyg* 2005;72(6):811-4.
- [20] Westerman MP. Bone marrow needle biopsy: an evaluation and critique. *Semin Hematol* 1981;18(4):293-300.



Distribution of cases with positive correlation between biopsy and aspirates(n=54)

Diagnoses	Number of cases
Acute leukemia	5
Chronic leukemia	4
Metastatic deposits	3
Multiple myeloma	2
Tuberculosis	4
Megaloblastic	20
Reactive	10
Aplastic anemia	6

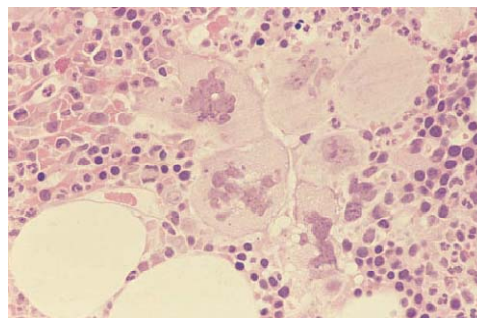


Figure 4: Myelofibrosis cellular phase clusters of megakaryocyte with hyperlobulated nuclei

Diagnoses made on Bone Marrow Biopsy alone; Aspirate was non contributory(n=13)

Diagnoses	Number of cases
Tuberculous granuloma	2
Marrow hypoplasia	2
Lymphoproliferative disorders	1
Metastasis	2
Myelofibrosis	2
CML	1
ALL	2
Osteitisfibrocystica	1

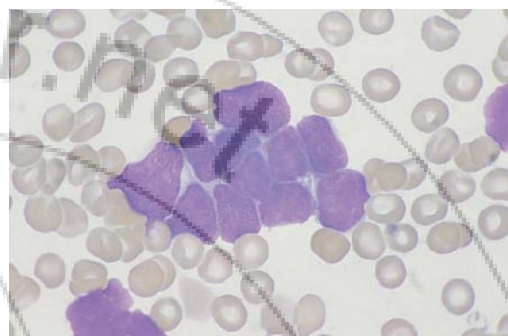


Figure 5: Metastatic Adenocarcinoma

Diagnoses made on Aspirates alone; Biopsy was non contributory(n=3)

Diagnoses	Number of cases
Malaria	1
Granuloma	1
MegaloblasticAnemia	1

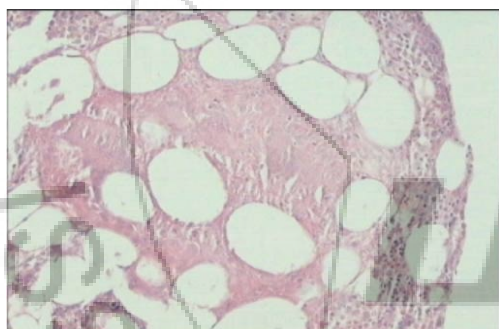


Figure 1: BMB: Granuloma formation

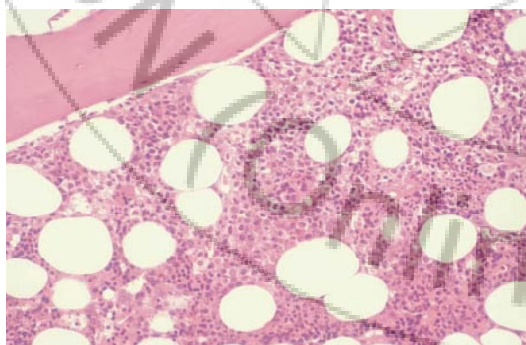


Figure 2: Leukemoidreaction