

Bone marrow aspirates and trephine biopsy sections in various haematological disorders.

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Abstract:

BACKGROUND: Bone marrow examination is crucial diagnostic modality for evaluation of various hematological and non hematological disorders. However, marrow aspirate smears and biopsy sections, even though performed simultaneously, are often assessed at different points of time due to different processing methods. And the assessment of bone marrow and trephine biopsy sections is done to reach the appropriate diagnosis .

AIM: The aim of this study is to compare the diagnostic accuracy and the rate of concordance between the two modalities of bone marrow examination.

MATERIALS AND METHODS: About 150 simultaneously performed bone marrow aspirates and BM trephine biopsies were done and the results are retrospectively analyzed over a period of 1 year. The concordance between the two was analysed. The concordance rates for different hematological disorders were calculated and recorded as high for >75%, moderate for 45%–75%, and low for <45% .

RESULTS: A high concordance was found in cases of megaloblastic anemia, leukemias, non-Hodgkin's lymphoma, and multiple myeloma; moderate concordance was found in hypoplastic marrow and concordance was low in Hodgkin's lymphoma, chronic myeloid leukemia (CML) in blast phase, metastatic, and granulomatous involvement of bone marrow.

CONCLUSION: Bone marrow aspiration alone is sufficient for the diagnosis of megaloblastic anemia and most of the hematological malignancies. Bone marrow biopsy is more appropriate for detection of disorders with focal marrow involvement such as lymphoproliferative disorders, metastatic cancer, focal blast crisis in CML, granulomatous lesions, and hypoplastic marrow. However, it is strongly recommended that both should be reviewed simultaneously to ensure maximum diagnostic accuracy.

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I. Introduction

Bone marrow is involved in variety of hematological and non-hematological disorders. The hematological disorders include acute leukemia, myeloproliferative neoplasm (MPN), hemato-lymphoid neoplasm, nutritional deficiency diseases. On the other hand non-hematological disorders include infectious diseases infiltrating the bone marrow such as tuberculosis, parasitic infections and metastatic deposits [1].

Although, diseases of bone marrow present with various clinical symptoms and also involve the blood but peripheral blood picture alone does not reflect the nature of disease process. Depending upon diagnosis suspected from the clinical features and peripheral blood examination, indications for bone marrow examination can be summarized. Therefore, complete hematological evaluation of cases where bone marrow examination was indicated includes BMA smear and bone marrow trephine biopsy as they are complementary to each other [2]. Hence, we attempted to correlate both these parameters to arrive at a more conclusive final diagnosis.

II. Materials And Methods

The study has been conducted for bone marrow examination on 150 cases presenting with anaemia, fever and organomegaly. The patients had a male to female sex ratio of 1.6:1 and a wide range of age from 3 years to 80 years . It includes both indoor and outdoor patients, who were suspected of having their bone marrow involvement by any hematological or non-hematological disorders. The relevant history and bio-data of patients was recorded and informed consent was taken. Patients were investigated for complete blood count, coagulation profile, reticulocyte count and peripheral blood film (PBF) examination. BMA and BMB were done Section simultaneously for these patients. In patients of thrombocytopenia, five minutes of firm pressure was applied at the end of the procedure. However, as a precaution, the patients were kept in lying down position on his/her back for a further 10–15 minutes to apply more prolonged pressure.

BMA was performed by Salah's marrow puncture needle; smears prepared were stained with leishman stain. Prussian blue for iron demonstration was done in selected cases and iron grading was done [3]. BMB was taken by Jamshidi biopsy needle and specimens were fixed in 10% formalin fixative, decalcified in 10% formic acid-5% formaldehyde and processed with paraffin-wax embedding. Sections, 1µm-thick, were cut and were stained by Hemotoxylin and Eosin (H & E) stain. The staining for reticulin fibers with Gomori's Silver impregnation method was done in selected cases and grading was done [4]. Bone marrow biopsy and aspiration findings were analysed in context of clinical signs, symptoms, other laboratory investigations and diagnosis reached.

III. Results

Total cases studied were categorized further on the basis of peripheral blood features and accepted indications were summarized [Table-1]. The results were analysed as per the extent of correlation between BMA and BMB.

Out of the 150 cases studied, in 69 cases both the procedures were comparable [Table2] . BMA was reported as suggestive of aplastic anaemia in 27 cases but BMB revealed hypo cellular marrow only in 9 cases where as aplastic anaemia in rest of the 18 cases [Table 3] .

There were 3 cases which was diagnosed as MPN by BMA and the BMB revealed reactive marrow.

There were 21 cases, where diagnosis was made only on BMB, BMA was not contributory [Table 4] .

There were seen 30 cases in which diagnosis were possible to make with or without complementary findings of BMA. But pattern of involvement of marrow was detected only on BMB [Table 5] .

IV. Conclusion

The bone marrow examination is valuable investigation in hematology practice. BMA and BMB both are important procedures for the diagnosis of hematological and non-hematological conditions. These procedures are also useful for follow up of the patients undergoing chemotherapy [5,6].

In the present study 69 out of total 150 cases showed comparable results between BMA and BMB. Out of these 69 cases, indication for bone marrow examination was anaemia in 48 cases and both BMA and BMB examination revealed erythroid hyperplasia with either micro normoblastic or megaloblastic proliferation .These observations were nearly similar to the findings seen in a study conducted by Ch Toi P et al., [7] . But iron stained sections of BMB showed differences in iron content from that of BMA smears. Stuart-Smith SE et al., have also shown in a study that aspirate smears reflect bone marrow iron stores more reliably than formic acid decalcified trephine biopsy sections [8] . Rest of the 21 out of these 69 cases were diagnosed as acute leukemia by peripheral blood film examination in all, except 6 cases, where it was confirmed on bone marrow examination. Younus U and associates emphasized that although BMA confirms the diagnosis of acute leukemia, bone marrow biopsy specimen complements the peripheral blood and aspirate findings in providing additional information for the diagnosis and especially prognosis of acute leukemia [9]. Also various investigators have studied the diagnostic value of antibodies suitable for use on paraffin wax embedded sections in the diagnosis of acute leukemia in sections from bone marrow biopsy specimens [10].

In 27 out of total 150 cases, BMA smear interpretation was suggestive of aplastic anaemia, where as BMB revealed hypocellular marrow with focal hyper cellular areas in 9 cases. Rest of the 18 cases were ultimately confirmed as aplastic anaemia. As BMB gives the qualitative and quantitative assessment of cellularity, therefore confirms the diagnosis and overcomes the limitation of BMA .

An important limitation of bone marrow obtained by aspirate is the admixing of marrow and sinusoidal blood, which may not allow for reliable estimates of marrow cellularity. Also It is necessary that finding of a 'dry tap' should never be dismissed as being due to faulty technique and always needs a bone marrow biopsy for further evaluation [13]. In the present study, out of total 150 cases, 15 cases were reported as bloody tap/dry tap on BMA. Which were turned out to be hypocellular/ hypercellular marrow on BMB. Also, 3 cases of leukoerythroblastic blood picture, on repeated BMA attempts was found to be of dry tap. BMB showed myelofibrosis along with additional finding of few epithelioid cells. On correlation with clinical details possibility of Koch's pathology was kept. Reticulin stained sections revealed grade-3 fibrosis. Ch Toi P et al., have mentioned that 80% cases of granulomatous lesions were diagnosed by BMB alone [7] . It was observed in a single treated case of multiple myeloma that marrow was normocellular on aspiration. Whereas, BMB examination revealed the focal collections of plasma cells. Babarovic E and fellows have mentioned the role of BMB for detection of minimal residual disease after treatment in case of multiple myeloma [14].

We have encountered 18 cases of MPN's out of which diagnosis was possible to be made from PBF and BMA in 6 cases only. Role of trephine biopsy is not only in differentiation of MPN but also to assess the overall marrow cellularity, histotopography, morphology of megakaryocytes as well as blasts, and degree of Myelofibrosis. Rest of the four non diagnostic aspirates in patients who had grade-3 marrow fibrosis highlights the importance of trephine biopsy. BMA does not have much role in diagnosis of primary myelofibrosis because

diffuse osteomyelosclerosis, intrasinusoidal hematopoiesis and vascular proliferation as seen in present study, which is characteristic of primary myelofibrosis, can be confirmed and graded on BMB only. Also, reticulin stain which gives accurate platform for grading of fibrosis to get done is possible on BMB only.

There were 6 cases of NHL in present study where BM biopsy renders information which cannot be determined from aspiration such as spatial distribution and extent of infiltrates, overall cellularity and fibrosis. This also implies that trephine biopsy may be more useful in post chemotherapy patients to assess the residual tumour cell burden and degree of chemotherapy response [17].

There were 3 cases of Chronic Lymphocytic Leukemia (CLL) with diffuse involvement of marrow, which was seen on biopsy section while aspiration showed only that marrow is involved. Bone marrow examination in case of CLL should always include a trephine biopsy because bone marrow aspirate gives very little information beyond that already available from examination of blood. Pattern of marrow involvement by leukemic cells could be only analysed by trephine biopsy. Also, trephine biopsy permits an accurate assessment of extent of infiltration and gives information of prognostic importance.

In one case of thrombocytopenia in present study BMA and BMB findings were consistent to each other. But an additional finding on BMB was normal arrangement of megakaryocytes which were seen increased in number on BMA [19].

Hence, it was observed from the above discussion that bone marrow evaluation is an important and effective tool in diagnosing and evaluating hematological and non-hematological disorders. Complete evaluation of bone marrow samples includes a brief patient history, hematological profile, BMA smears and biopsy sections [20]. A correlation of bone marrow aspiration and biopsy showed that both the procedure were complementary to each other. The BMA generally provides an excellent cytomorphological details which enables hematopathologist in recognising the abnormal hematopoietic cells or the non-native cells in case of non-hematological disorders. Whereas, a bone marrow trephine biopsy demonstrates the topographic arrangement of hematopoietic cells within the marrow framework and hence gives a more representative view of the cellularity of the marrow and allows infiltration to be recognized clearly. BMB examination has definite edge over BMA in the detection of minimal residual diseases, staging of lymphoma and for the diagnosis of acute leukemia in relapse cases which are otherwise clinically silent.

[Table 1]:

Accepted indications	No. of cases	Percentage
Pancytopenia	60	40
Anaemia	57	38
Acute leukemia	15	10
Chronic leukemia	12	08
Leucoerythroblastic blood picture	03	02
Thrombocytopenia	03	02

Table showing accepted indications of bone marrow examination

[Table2]:

PBF	BMA	BMB	NO. OF CASES	PERCENTAGE
AML	AML	AML	09	6
ALL	ALL	ALL	06	4
PANCYTOPENIA	AML	AML	03	2
PANCYTOPENIA	ALL	ALL	03	2
ANEMIA	ANEMIA	ANEMIA	48	32
TOTAL			69	46

showing cases with similar findings on bone marrow aspiration and bone marrow biopsy

[Table3]:

PBF	BMA	BMB	NO. OF CASES	PERCENTAGE
PANCYTOPENIA	HYPOCELLULAR	APLASTIC ANEMIA	18	12
PANCYTOPENIA	HYPOCELLULAR	HYPOCELLULAR	09	06
TOTAL			27	18

Showing cases where findings on bone marrow aspiration were suggestive only.

[Table 4]:

PBF	BMA	BMB	NO. OF CASES	PERCENTAGE
PANCYTOPENIA	Bloody tap	Hypocellular	9	6
PANCYTOPENIA	DRY TAB	Hypocellular	6	4
Leucocyte-throblastic	DRY TAB	Epithelioid cell	3	2
Anaemia	Normocellular marrow	Focal collection of plasma cells	3	2
Total			21	14

showing cases where diagnosis was possible only on bone marrow biopsy

TABLE 5

PBF	BMA	BMB	NO. OF CASES	PERCENTAGE
CML	Bloody tap	Prefibrotic phase of MPN	6	4
ANEMIA	Dry tap	Myelofibrosis with myelosclerosis	3	2
PANCYTOPENIA	MPN	MPN with MF (prefibrotic phase)	3	2
PANCYTOPENIA	Bloody tap	MPN with MF	3	2
PANCYTOPENIA	Dry tap	MPN with MF	3	2
CLL	CLL	DIFFUSE INVOLVEMENT	3	2
ANEMIA	NHL	NODULAR INVOLVEMENT	3	2
Pancytopenia	NHL	Diffuse involment	3	2
Thrombocytopenia	Megakaryocytosis	Megakaryocytosis	3	2
Total			30	20

Showing cases where patterns of involvement were seen on biopsy

V. Conclusion

The present study showed that BMA and BMB are easy, rapid, costeffective and more or less are of equal value in various hematological and non-hematological disorders of bone marrow. Although, methods are complementary on correlation but in some cases one or other of these methods is more conclusive and also have important diagnostic value even by using basic standard fixation and embedding procedures, with the aspiration smears being primarily useful for cytological diagnosis and biopsy sections mainly helpful to identify histological features like architectural patterns, grading of fibrosis, pattern of infiltration with lymphomas and granulomatous conditions. Both of the procedures should be done simultaously as they play important role in providing findings, which are mandatory for making final diagnosis.

References

- [1]. S Neal, Young Aplastic anaemia, myelodysplasia and related bone marrow syndromes. In: Kasper, Braunwald, Fauci, Hauser, Longo, Jameson, editors. Harrison's Principles of Internal medicine 2005 1516th Edition New Delhi McGraw Hill:617 [Google Scholar]
- [2]. JR Krause, An appraisal of the value of the bone marrow biopsy in the assessment of proliferative lesions of the bone marrow. *Histopathology* 1983 7(5):627-44. [Google Scholar]
- [3]. E Gale, J Torrance, T Bothwell, EH Kass, The quantitative estimation of total iron stores in human bone marrow. *J Clin invest.* 1963 42(7):1076-82. [Google Scholar]
- [4]. DE Bauermeister, Quantitation of bone marrow reticul- a normal range. *Am J Clin Pathol.* 1971 56:24-31. [Google Scholar]
- [5]. RS Riley, TF Hogan, DR Pavot, R Forysthe, D Massey, E Smith, A pathologist's perspective on bone marrow aspiration and biopsy: Performing a bone marrow examination. *J Clin Lab Anal.* 2004 18(2):70-9. [Google Scholar]
- [6]. A Islam, Bone marrow aspiration prior to bone marrow core biopsy using the same bone marrow biopsy needle. A good or bad practice. *J Clin Pathol.* 2007 60:212-15. [Google Scholar]
- [7]. P Ch Toi, R Varghese G'Boy, R Rai, Comparative evaluation of simultaneous bone marrow aspiration and bone marrow biopsy. An institutional experience. *Indian J Hematol Blood Transfus.* 2010 26(2):41-44. [Google Scholar]
- [8]. SE Stuart Smith, DA Hughes, BJ Bain, Are routine iron stains on bone marrow trephine biopsy specimens necessary? *J Clin Pathol.* 2005 58(3):269-72. [Google Scholar]
- [9]. U Younus, K Saba, J Aijaz, MH Bukhari, S Naeem, Significance of Bone Marrow Histology in the Diagnosis of Acute Myeloid Leukemia. *ANNALS.* 2011 17(1):5-8. [Google Scholar]
- [10]. SM Krober, A Greschniok, Acute lymphoblastic leukemia: correlation between morphological/immunohistochemical and molecular biological findings in bone marrow biopsy specimens. *Mol pathol.* 2000 53(2):83-87. [Google Scholar]
- [11]. R Milosevic, G Jankovic, N Antonijevic, V Jovanovic, D Babic, M Colovic, Histopathologic characteristics of bone marrow in patients with aplastic anaemia. *Srp Arh Celok Lek.* 2000 128(5-6):200-04. [Google Scholar]
- [12]. BJ Bain, Clark DM, Wilkins BS, editors. *Bone marrow pathology. Histopathology* 2010 4th Edition UK: Wiley Blackwell Publishing:11 [Google Scholar]
- [13]. JE Humphries, Dry tap bone marrow aspiration: clinical significance. *Am J Hematol.* 1990 35(4):247-50. [Google Scholar]
- [14]. E Babarovic, T Valkovic, S Stifter, I Budisavljevic, I Seili-Bekafigo, A Duletic-Nacinovic, Assessment of Bone Marrow Fibrosis and Angiogenesis in Monitoring Patients With Multiple Myeloma. *Am J Clin Pathol.* 2012 137:870-78. [Google Scholar]
- [15]. KH Yookarin, D Basu, DK Dutta, Bone marrow trephine biopsy findings in chronic myeloid leukemia. *Malaysian Journal of pathology.* 2002 24:37-43. [Google Scholar]

- [16]. V Mahajan, V Kaushal, S Thakur, R Kaushik, A comparative study of bone marrow aspiration and bone marrow biopsy in haematological and non haematological disorders –An institutional experience. *JACM*. 2013 14(2):133-35. [Google Scholar]
- [17]. S Goyal, UR Singh, U Rusia, Comparative Evaluation of Bone Marrow Aspirate with Trephine Biopsy in Hematological Disorders and Determination of Optimum Trephine Length in Lymphoma Infiltration. *Mediterr J Hematol Infect Dis*. 2014 6(1)e2014002. doi: 10.4084/MJHID.2014.002. eCollection 2014 [Google Scholar]
- [18]. YS Kim, RJ Ford, JA Faber, B-cell chronic lymphocytic leukemia/small lymphocytic lymphoma involving bone marrow with an interfollicular pattern. *Am J Clin Pathol*. 2000 114(1):41-46. [Google Scholar]
- [19]. J Steven, Jubelirer The role of the bone marrow examination in the diagnosis of immune thrombocytopenic purpura: case Series and Literature Review. *Clinical and Applied Thrombosis/Hemostasis*. 2002 8:173-76. [Google Scholar]
- [20]. Bain B J, Bone marrow aspiration. *J Clin Pathol* 2001 54:657-63. [Google Scholar]