

## Chapter 2

# Bone Marrow Sampling and Processing

### ABSTRACT

*Bone marrow aspirate and biopsy are complementary for proper assessment. Adequate sampling and processing are critical for accurate examination. Technical artifacts may compromise marrow interpretation. The multistep processing of the bone marrow involves various fixation, decalcification, and embedding protocols. Formalin fixation, EDTA decalcification, and paraffin embedding are the most common and the least injurious applied routine methods, allowing the application of many ancillary tests, such as immunohistochemistry (IHC), molecular tests, and in situ hybridization (ISH). Hematologic stains used for routine morphologic examination include the wright-Giemsa stain for bone marrow aspirate smears, hematoxylin and eosin, May Grunwald-Giemsa, and reticulin stain for bone marrow biopsy sections. Quality assurance measures are essential throughout bone marrow preparation to avoid diagnostic errors. Improper specimen collection, inadequate processing, and staining are possible sources of errors.*

### INTRODUCTION

An accurate bone marrow diagnosis builds on adequately managed specimens from sampling to reporting using standardized processes and technical procedures.

This chapter discusses the bone marrow evaluation stages with their standards and best practices. The primary learning objectives are to acquire the necessary technical skills and background knowledge for the best practices in managing a bone marrow sample, including:

1. Rationale and clinical indications for bone marrow examination
2. The types and procedures for bone marrow sampling with their advantages and limitations
3. The critical technical points in bone marrow processing
4. The common stains for morphologic evaluation of aspirate smears and biopsy sections
5. The quality assurance measures applied to bone marrow processing
6. The primary sources of technical errors that could affect marrow preparation and compromise diagnostic decision

## **The Diagnostic Bone Marrow Evaluation Passes Through Three Major Practical Stages**

1. The pre-examination stage includes bone marrow sampling, processing, and staining
2. Examination, including:
  - a. Cytomorphology and Histopathologic examination
  - b. Selection and application of ancillary tests guided by the preliminary findings and suggested clinical diagnosis and differential diagnosis
3. Interpretation and integrated reporting of all examined facets finalized into a diagnostic entity or spectrum of likely possibilities and recommended confirmation methods

## **PRE-EXAMINATION**

### **Bone Marrow Sampling**

The bone marrow examination is an essential part of most hematologic diagnoses. A biopsy is a gold standard in this respect; in some cases, it is indispensable for diagnosis, and in others, it provides an additional advantage. However, bone marrow biopsy and aspiration are complementary.

Managing the bone marrow specimens starts with the requisition, usually ordered by the patient's physician. Retrieving all relevant information is the first thing before commencing the procedure. Pertinent information includes the indications and clinical suspicion, the patient's demographics, clinical history, clinical and imaging findings, pathology reports, previous treatment, and biochemical and hematologic tests performed. Request for further data will depend on the patient's condition, e.g., type and duration of chemotherapy.

- **The common indications for bone marrow evaluation in clinical practice include:**
  1. Abnormal peripheral blood findings not reconciled by clinical and laboratory investigations, e.g., in distinguishing primary production defect from increased destruction of circulating cells
  2. Direct diagnostic evaluation of hematolymphoid neoplasms, e.g., acute leukemia, chronic myeloproliferative disorders, myelodysplasia, and myeloma.
  3. Pathologic staging of neoplasms: e.g., non-Hodgkin and Hodgkin lymphoma and metastatic carcinoma
  4. Evaluation of suspected congenital hematologic disorders such as constitutional anemia and their differentiation from clonal marrow failure disorders
  5. Infectious disease investigation such as fever of unknown origin (FUO), especially if blood or other tissue sampling is non-informative
  6. Post-therapy evaluation and monitoring of minimal residual disease status in neoplastic disorders
- **In some conditions, bone marrow biopsy provides a unique diagnostic advantage, such as when:**
  1. The diagnosis requires a whole tissue architecture, e.g., myeloproliferative disorders, granulomatous diseases, and significant hypocellularity.
  2. Expected focal marrow involvement, e.g., focal infiltrates with lymphoma, metastasis, and focal inflammatory lesions.

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