Bone marrow evaluation of monocytosis

Cherie H Dunphy

Abstract

An absolute peripheral blood (PB) monocytosis is defined as greater than 1000 monocytes/μl. The differential diagnosis of an absolute peripheral monocytosis includes a reactive monocytosis and a neoplastic process that may be associated with various haematological neoplasms, that is chronic myelomonocytic leukaemia, myeloid and lymphoid neoplasms with eosinophilia and rearrangement of *PDGFRB*, juvenile myelomonocytic leukaemia, chronic myeloid leukaemia, as well as acute myeloid leukaemias with a prominent monocytic component. The diagnostic approach to a PB monocytosis, including appropriate evaluation of cytomorphological features and flow cytometric analysis of the PB, associated bone marrow findings, as well as the cytogenetic and molecular findings, will be discussed.

Keywords acute myeloid leukaemia; chronic myelomonocytic leukaemia; juvenile myelomonocytic leukaemia; monocytosis; reactive monocytosis

Differential diagnosis of peripheral blood monocytosis

Reactive monocytosis

An absolute peripheral monocytosis may occur secondary to chronic infectious processes, or may be associated with various immunological disorders (i.e. inflammatory gastrointestinal disorders, collagen vascular diseases and sarcoidosis), malignancies (including non-Hodgkin and Hodgkin lymphomas, plasma cell dyscrasias and, rarely, carcinomas) and benign haematological disorders (haemolytic anaemia, chronic neutropenia and immune thrombocytopenic purpura), as well as post-splenectomy.[1–3] In addition, peripheral monocytosis may occur secondary to administration of granulocyte-monocyte colony stimulating factor (GM-CSF), in the recovery phase after chemotherapy, after intravenous immunoglobulin therapy and after myocardial infarction.[4–6] Monocytosis may rarely be associated with carcinomas (particularly lung, colorectal and renal carcinomas); it has been demonstrated to be associated with the secretion of interleukin 6 by the neoplastic cells.[7,8] Such a case is illustrated in Figure 1.

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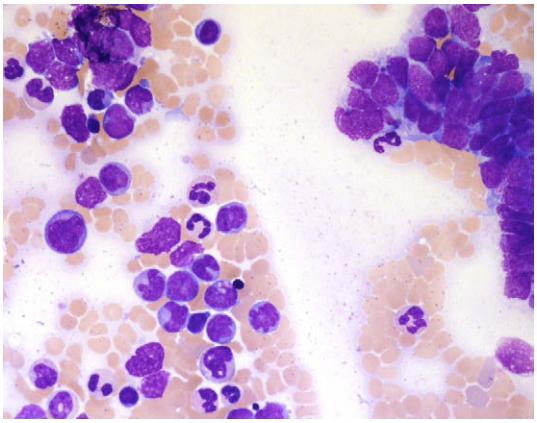

Figure 1 Patient with lung carcinoma which expressed interleukin 6 and was associated with an absolute peripheral blood monocytosis (8200 monocytes/μl) and an increase in monocytes in the bone marrow (BM) (18% monocytes). The BM aspirate demonstrates an increase in morphologically mature monocytes and adjacent metatastic carcinoma on the right side of the image.

Monocytosis associated with clonal haematopoietic neoplasms

A peripheral blood (PB) monocytosis may also occur in association with various clonal haematopoietic neoplasms. The descriptions and diagnostic criteria of these neoplasms will first be individually discussed,[9] followed by a description of the distinctive PB findings (cytomorphological and flow cytometric features), bone marrow (BM) findings (cytomorphological and immunophenotypic features), and cytogenetic and molecular features that may distinguish these entities from a reactive monocytosis and from one other. An absolute monocytosis in an adult requires close clinical follow-up if no apparent reactive cause can be clearly identified.

Chronic myelomonocytic leukaemia (CMML) is a clonal BM stem cell disorder in which monocytosis is a major defining feature. Diagnostic criteria for CMML are as follows:

1. persistent PB monocytosis (> 1000/μl)
2. no Philadelphia (Ph) chromosome or *BCR–ABL* fusion gene
3. fewer than 20% blasts (including myeloblasts or monoblasts) and/or blast equivalents (promonocytes) in the PB or BM
4. dysplasia in one or more myeloid cell lines.

If myelodysplasia is absent or minimal, the diagnosis of CMML may still be made if the other requirements are met: an acquired, clonal cytogenetic abnormality is detectable in the BM cells *or* the monocytosis has persisted for 3 months or longer, *and* all other causes of monocytosis have been excluded.

If greater than 20% blasts and/or promonocytes are present in the PB or BM, the World Health Organization (WHO) classification diagnosis is acute myeloid leukaemia.

CMML is further divided into CMML-1 and CMML-2, based on the percentage of blasts and/or promonocytes. In CMML-1, blasts and/or promonocytes represent fewer than 5% of peripheral white blood cells (WBCs) and fewer than 10% of BM nucleated cells. In CMML-2, blasts and/or promonocytes represent between 5% and 19% of peripheral WBCs or between 10% and 19% of BM cells. CMML-2 may also be diagnosed when there are fewer than 20% blasts in the PB or BM if Auer rods are identified. It

should always be kept in mind that a diagnosis of CMML should never be based on PB findings only and without BM examination, since there may be a higher percentage of immature cells (blasts and/or promonocytes) in the BM. It should also be noted that systemic mastocytosis may be associated with clonal haematological non-mast cell lineage diseases, particularly CMML. Thus, the possibility of concomitant systemic mastocytosis should be considered when diagnosing CMML, since mast cell resistance to most cytoreductive agents is of major importance for treatment planning.[10]

Chronic myelononocytic leukaemia associated with eosinophilia (CMML-eos) represents a subset of CMML (< 1–2% of patients with CMML), in which there is an associated eosinophilia (i.e. a PB eosinophil count > 1500/μl). A small subset of these cases is also associated with a major manifestation of BM eosinophilia. The updated 2008 WHO classification created a new category of myeloid neoplasms characterized by eosinophilia and expression of fusion genes involving specific tyrosine kinases and apparently originating from a pluripotent (lymphoid and myeloid) stem cell. Therefore, cases of CMML with eosinophilia should show absence of *PDGFRA*, *PDGFRB* or *FGFRI* rearrangement. Myeloid neoplasms associated with *PDGFRB* rearrangement usually present with prominent eosinophilia, which is often accompanied by peripheral neutrophilia and monocytosis and a hypercellular BM with increased eosinophils and myeloid hyperplasia. The defining genetic feature of this disease is fusion of the *PDGFRB* gene at 5q31–33 with another partner gene. Most cases have *ETV6–PDGFRB* fusion due to a t(5;12) (q31–33;p12). It is important to recognize this entity since myeloid neoplasms with *PDGFRB* rearrangement are sensitive to imatinib therapy.

Juvenile myelomonocytic leukaemia (JMML) is a clonal haematopoietic disorder of childhood (> 95% of patients are younger than 4 years of age at diagnosis) and, more rarely, in the neonatal period. JMML is characterized by a principal proliferation of granulocytic and monocytic lineages. Essentially all patients show leucocytosis (but usually < 100,000/μl) and monocytosis. Unlike chronic myeloid leukaemia, basophilia is not a characteristic feature of JMML. The majority of patients are anaemic and thrombocytopenic. Levels of fetal haemoglobin (HbF) are consistently elevated for age, typically in the 20–80% range, in approximately 60% of cases. Diagnostic criteria for JMML include the following:

1. peripheral blood monocytosis greater than 1000/μl
2. blasts (including promonocytes) are fewer than 20% of the peripheral WBCs and BM cells
3. no Ph chromosome or *BCR–ABL* fusion gene
4. plus two or more of the following: HbF increased for age; immature granulocytes in PB; WBC count greater than 10,000/μl; clonal chromosomal abnormality (especially monosomy 7); in vitro hypersensitivity of myeloid progenitors to GM-CSF.

Antibodies directed against GM-CSF have been shown to selectively inhibit growth of JMML colonies, whereas antibodies against a variety of other growth factors do not: cultured non-adherent PB cells are approximately ten times more sensitive to exogenous GM-CSF than controls, but show normal sensitivity to interleukin-3 and G-CSF.[6,7]

Chronic myeloid leukaemia, BCR–ABL+ (CML): an absolute monocytosis may also occur in association with Ph+ CML; however, not generally as the prominent feature, as seen in CMML. When high WBC counts are observed, an absolute monocytosis (> 1000/μl) is common in patients with chronic myeloproliferative neoplasms, such as CML. However, in such cases, there is usually no relative monocytosis, that is monocytes are less than 8% of all PB leucocytes.[11]

Most cases of CML are associated with *BCR–ABL* fusion genes that encode a $p210^{BCR-ABL}$ fusion protein. In these cases, monocytosis is not a prominent feature, although the circulating monocytes are generally part of the clonal process (see the section below regarding flow cytometric immunophenotyping). In rare cases of Ph+ CML, there is expression of $p190^{BCR-ABL}$ instead of $p210^{BCR-ABL}$; and approximately half of these patients exhibit a prominent monocytosis, with haematological features intermediate between classical CML and CMML.[12] In such cases, fluorescent in-situ hybridization (FISH) analysis for *BCR–ABL* fusion is crucial for an accurate diagnosis of CML and to avoid misdiagnosis as CMML.

Acute myeloid leukaemias (AMLs) with a monocytic component (acute myelomonocytic leukaemia (AMML) and acute monoblastic or monocytic leukaemia (AMoL)): AMLs with a monocytic component (AMML and AMoL) may be associated with a peripheral monocytosis. As stated above in the discussion of CMML, if greater than 20% blasts and/or promonocytes are observed in the PB, AML may be diagnosed instead of CMML. Even if there are fewer than 20% blasts and/or promonocytes in the PB, a BM examination is necessary to exclude an AML. AMML and AMoL are defined and may be distinguished by findings in the BM, which will be discussed below.

Evaluation of PB monocytosis

Morphological evaluation

As can be surmised from the discussion above, cytomorphological evaluation of the PB smear is often critical in distinguishing disease entities that are associated with a PB monocytosis. In reactive monocytoses, the monocytic cells appear mature, with folded nuclear membranes, lacking prominent nucleoli and with abundant cytoplasm that may demonstrate cytoplasmic vacuolization. In CMML-1, blasts and/or promonocytes represent fewer than 5% of the peripheral WBCs, and the remaining monocytic cells appear mature. In CMML-2, blasts and/or promonocytes represent 5–19% of the peripheral WBCs and may contain Auer rods. Promonocytes are considered 'blast equivalents' and are characterized by their single prominent nucleolus. Blasts are characterized by a high nuclear:cytoplasmic ratio, very fine chromatin and varying numbers of nucleoli. In CMML, mild basophilia is sometimes present, and eosinophils are usually normal or slightly increased in number. As described above, in cases of CMML associated with eosinophilia (the eosinophil count in the PB is > 1500/μl) the possibility of a myeloid/lymphoid neoplasm associated with *PDGFRB* rearrangement should be considered. The cytomorphology of the monocytic cells in JMML are similar to those described in CMML. In addition, eosinophilia and basophilia may be observed in a minority of JMML patients.