The Differential Diagnosis and Bone Marrow Evaluation of New-Onset Pancytopenia

Elizabeth P. Weinzierl, MD, PhD, and Daniel A. Arber, MD

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Abstract

New-onset pancytopenia can be caused by a wide variety of etiologies, leading to a diagnostic dilemma. These etiologies range from congenital bone marrow failure to marrow space-occupying lesions, infection, and peripheral destruction, to name a few. Bone marrow examination, in addition to a detailed clinical history, is often required for an accurate diagnosis. The purpose of this review is to provide a brief overview of many of the causes of new-onset pancytopenia in adults and children, with emphasis on bone marrow findings and recommendations of additional testing and clinical evaluation when needed, with the overall aim of aiding the pathologist’s role as a consultant to the patient’s treating physician.

Pancytopenia is a common indication for bone marrow examination and can have numerous causes. Cytotoxic therapies, including myeloablative radiation therapy and chemotherapy, are common, but predictable, causes of pancytopenia in patients being treated systemically for neoplasia. New-onset pancytopenia outside this setting, in both children and adults, can prove to be a diagnostic dilemma, and causes include congenital and acquired bone marrow failure syndromes, marrow space-occupying lesions, peripheral destruction of hematopoietic cells, autoimmune disorders, infection, and ineffective marrow production. Often, the workup of new-onset pancytopenia is extensive and should include a detailed clinical, medication, recreational drug, and environmental exposure history. Although bone marrow examination often reveals an underlying condition causing pancytopenia, it is not always conclusive. Understanding the various disorders that may cause pancytopenia can aid in the recommendation of additional testing and clinical evaluation when the marrow studies are not specific for a single etiology. Here, we provide a systematic overview of many of the causes of new-onset pancytopenia.

Congenital Bone Marrow Failure Syndromes

Many diseases can cause bone marrow failure, resulting in greatly reduced to absent hematopoiesis with subsequent pancytopenia. Inherited causes of bone marrow failure encompass a small but important group of these diseases [Table 1]. Although rare, these diseases should always be considered as a potential cause of new-onset pancytopenia.
Disease | Defect | Supportive Clinical Findings | Supportive Laboratory Findings | Inheritance Pattern
--- | --- | --- | --- | ---
Fanconi anemia | Multiple genes involved (at least 16) | Skeletal abnormalities (radius, thumb); small stature; urogenital abnormalities; 40% with no physical findings | Increased chromosomal breakage in response to mitomycin C or diepoxylutein | AR (most) or XLR
Dyskeratosis congenita | Multiple genes involved in telomere maintenance | Leukoplakia; nail dystrophy; lacy skin pigmentation; pulmonary fibrosis | Genetic testing (a negative result does not rule out disease) | AR, XLR, or AD
Shwachman-Diamond syndrome | Most common mutation in Shwachman-Bodian-Diamond syndrome (SBDS) gene | Exocrine pancreatic insufficiency | Genetic testing (a negative result does not rule out disease); normal sweat chloride | AR
Congenital amegakaryocytic thrombocytopenia | Myeloproliferative leukemia virus oncogene (MPL) | Sequeleae of severe thrombocytopenia | Genetic testing (a negative result does not rule out disease); elevated thrombopoietin levels | AR
Hemophagocytic lymphohistiocytosis | Multiple genes involved, including perforin 1 (PRF1) and UNC13D | Fever; splenomegaly; hepatitis; neurologic symptoms; rash | Hemophagocytosis; hypertriglyceridemia; hypofibrinogenemia; low/absent NK cell activity; elevated serum ferritin; soluble CD25 >2,400 U/mL | AR or XLR

AD, autosomal dominant; AR, autosomal recessive; NK, natural killer; XLR, X-linked recessive.

Most importantly, if bone marrow findings are compatible and the clinical history is suggestive of FA, peripheral blood specimens should be sent for the definitive test, the chromosome breakage test, which demonstrates marked chromosome breakage after treatment with a cross-linking agent such as mitomycin C because of the underlying defect in DNA repair. In addition, bone marrow specimens should be sent for cytogenetic analysis because clonal cytogenetic abnormalities are found in approximately 65% of cases by age 30 years.

Dyskeratosis congenita (DC), initially described in 1910, is a rare inherited cause of pancytopenia and is characterized by the classic clinical triad of leukoplakia, nail dystrophy, and lacy skin pigmentation, but this classic triad is not necessary for diagnosis. The skin and nail abnormalities often present early in childhood, before 10 years, followed by bone marrow failure, which occurs by 20 years in 80% of patients. In addition, the disease course in 20% of patients is complicated by pulmonary manifestations of reduced diffusion capacity or restrictive pulmonary disease. Although bone marrow hypoplasia is the main pathologic abnormality seen in affected patients, a predisposition to malignancy is seen, with, according to one study, approximately 10% of patients developing malignancies, including MDS. The bone marrow findings in patients with DC range from normal to variable stages of aplasia, culminating in a hypoplasia indistinguishable from idiopathic AA. As with FA, this disease can prove a diagnostic challenge to the pathologist; however, there is no single test that definitively establishes the diagnosis. A first step is eliminating FA from the differential diagnosis because cells from patients with DC do not demonstrate increased chromosomal breakage with cross-linking agents. Clinical and family history is certainly of paramount importance and can often be the most
helpful clue to an accurate diagnosis. Further complicating the diagnosis is the genetic diversity of the disease, in that X-linked, autosomal recessive, and autosomal dominant forms of the disease exist. The implicated genes are involved in telomere maintenance, resulting in shortened telomeres in patients with DC mutations. Although several of these genes are available for clinical testing, a negative test result does not eliminate DC from the differential because the pathologic genetic mutations are uncharacteristic in approximately 50% of DC cases. 

Shwachman-Diamond syndrome is another rare cause of pancytopenia, inherited in an autosomal recessive manner, and usually presenting in infancy with exocrine pancreatic insufficiency and bone marrow failure. Most cases are associated with mutations in chromosome 7 affecting the Shwachman-Bodian-Diamond syndrome (SBDS) gene. Neutropenia is the most common presentation of this bone marrow failure, and the marrow cellularity can be low, normal, or high, and mildly dysplastic features can be seen. The condition progresses to complete bone marrow failure in 20% to 25% of patients and to MDS/AML in 5% to 33% of patients. Again, bone marrow findings are nonspecific but should be evaluated to eliminate other causes of bone marrow failure. Cytogenetic abnormalities of chromosome 7 can be helpful clues to a diagnosis of Shwachman-Diamond syndrome, and genetic testing can also be helpful, but 10% of patients will not carry a mutation in the SBDS gene. In addition, other causes of pancreatic insufficiency should be entertained, with sweat chloride test results being normal in this disease, compared with cystic fibrosis.

Congenital amegakaryocytic thrombocytopenia (CAMT) is an additional cause of congenital bone marrow failure and is inherited in an autosomal recessive manner, often because of mutations in myeloproliferative leukemia virus oncogene (MPL, or the thrombopoietin receptor). Patients usually are seen as neonates with severe thrombocytopenia and its clinical sequelae, including petechiae, purpura, or bleeding of the skin, mucous membranes, and/or gastrointestinal tract. In approximately 25% of patients, the condition progresses to pancytopenia, usually between 3 and 4 years of age. On peripheral smear, platelets remain normal in size, and bone marrow biopsy, although initially normocellular and demonstrating reduced or absent megakaryocytes, can evolve quite early and quickly to aplasia. In its pancytopenic presentation, the differential diagnosis includes other bone marrow failure syndromes, and a complete workup, including chromosomal breakage studies, should be performed to eliminate such entities as FA. Genetic analysis for mutations in MPL can additionally be performed, but a negative result does not preclude the diagnosis of CAMT. Finally, patients with CAMT often have high circulating levels of thrombopoietin, which can also be tested.

Another rare and complex syndrome that can present with pancytopenia is hemophagocytic lymphohistiocytosis (HLH), which can be either primary or acquired. The clinical diagnosis of this syndrome requires the presence of 5 or more of the following: fever, splenomegaly, cytopenia involving 2 or more cell lines, hypertriglyceridemia or hypofibrinogenemia, hepatitis, low or absent natural killer cell activity, a serum ferritin level higher than 500 μg/L, soluble CD25 higher than 2,400 U/mL, or hemophagocytosis as demonstrated in bone marrow, spleen, or lymph node. First described in 1952, the primary familial form of this disorder occurs most often in infants younger than 1 year, and has been found to be the result of uncontrolled T-cell and histiocyte activation. Mutations in several proteins, including perforin 1, UNC13D, and syntaxin 11, have been implicated in this disease. Interestingly, the syndrome is most often triggered by a stimulus, such as infection, and numerous viruses, bacteria, and parasites have been implicated. Primary HLH can also occur in patients with immune deficiencies, including Chédiak-Higashi syndrome, Griscelli syndrome 2, and X-linked lymphoproliferative disease.

Because HLH is rapidly fatal, a high diagnostic suspicion should be entertained. Bone marrow biopsy can be helpful, and the finding of hemophagocytosis in the bone marrow, spleen, or lymph nodes is indeed one of the diagnostic criteria for HLH, as mentioned before. However, the sensitivity of bone marrow morphologic analysis for hemophagocytosis is surprisingly low; however, hemophagocytosis may be more easily identified on iron-stained marrow aspirate smears than typical Wright-stained smears. In a postmortem study, hemophagocytosis was seen in the bone marrow of only 9 of 23 children with HLH and was more commonly seen in sections of the spleen (17/24 patients) and lymph nodes (17/23 patients). Similarly, a Canadian study determined a sensitivity of only 60% in finding hemophagocytosis in the initial bone marrow biopsy of patients with HLH.

Aplastic Anemia

In addition to congenital causes of bone marrow failure, numerous acquired causes of bone marrow failure exist. Indeed, AA is relatively common, and is seen in both children and adults. Although the majority of cases are idiopathic, this disease can be caused by multiple etiologies, including drugs, chemicals, radiation, viruses, anorexia, and even pregnancy. Patients often present with an abrupt onset of pancytopenia and a remarkably reduced bone marrow cellularity.

Idiopathic AA and Paroxysmal Nocturnal Hemoglobinuria

The majority of cases of acquired AA are idiopathic in nature and can occur in both children and adults.
Although the exact mechanism is unknown, idiopathic AA is thought to result from an attack of effector T lymphocytes on hematopoietic stem cells, resulting in bone marrow failure and peripheral pancytopenia. Consistent with this theory, AA is remarkably responsive to immunosuppressive drugs, and long-term survival is estimated at approximately 75%. Severe AA is defined by the specific criteria shown in Table 2. The peripheral blood typically shows pancytopenia, with a relative lymphocytosis and without definite morphologic abnormalities of the cells that remain in the blood. Bone marrow findings typically demonstrate a markedly hypocellular marrow with a reduction of all cell lines, and admixed T lymphocytes may be relatively increased. The distinction in such cases from hypoplastic MDS can be challenging. Careful examination of aspirate smears and touch preparations for morphologic features of dysplasia, as well as correlation with cytogenetic and immunohistochemical studies and can be helpful for this differential diagnosis. Cytogenetic abnormalities more often associated with MDS are shown in Table 3. Immunohistochemical studies for CD34 may show an increase in immature cells in hypoplastic MDS. In addition, some patients with apparent AA evolve over time to have definite MDS.

Table 2
Criteria for Severe Aplastic Anemia

At least 2 of the following peripheral blood findings:
- Reticulocytes <1%, corrected for hematocrit
- Absolute neutrophil count <500/μL (0.5 × 10⁹/L)
- Platelets <20,000/μL (20 × 10⁹/L)

AND
- Bone marrow biopsy with <25% normal cellularity
- Bone marrow biopsy with <50% normal cellularity in which less than 30% of the cells are hematopoietic

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Paroxysmal nocturnal hemoglobinuria (PNH) demonstrates a peculiar relationship with AA. Because of a defect in the phosphatidylinositol glycan complementation class A (PIGA) gene, which leads to a defect in GPI-linked proteins, PNH cells demonstrate an increased sensitivity to complement activation, leading to hemolytic anemia. PNH can arise de novo; in this setting, the classic presentation includes hemolysis, pancytopenia, and/or venous thrombosis with a normocellular to hypercellular bone marrow. Interestingly, patients who present with PNH clones can eventually progress to AA, and patients with AA can develop PNH clones. Indeed, up to two thirds of patients with pancytopenic AA concomitantly have small clones of PNH cells, and about 30% of patients with PNH have preceding AA. However, despite the frequency of small PNH clones in patients with AA, progression to clinical PNH is infrequent; one recent study found a 2.1% incidence of PNH at 5 years.

**Table 3**

Cytogenetic Abnormalities Associated With MDS

<table>
<thead>
<tr>
<th>Abnormality</th>
<th>Note</th>
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<tbody>
<tr>
<td>−7 or del(7q)</td>
<td></td>
</tr>
<tr>
<td>−5 or del(5q)</td>
<td></td>
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<tr>
<td>i(17q) or t(17p)</td>
<td></td>
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<tr>
<td>−13 or del(13q)</td>
<td></td>
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<tr>
<td>del(11q)</td>
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<tr>
<td>del(12p) or t(12p)</td>
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<tr>
<td>del(9q)</td>
<td></td>
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<tr>
<td>idic(X)(q13)</td>
<td></td>
</tr>
<tr>
<td>t(11;16)(q23;p13.3)</td>
<td></td>
</tr>
<tr>
<td>t(3;21)(q26.2;q22.1)</td>
<td></td>
</tr>
<tr>
<td>t(1;3)(p36.3;q21.2)</td>
<td></td>
</tr>
<tr>
<td>t(2;11)(p21;q23)</td>
<td></td>
</tr>
<tr>
<td>inv(3)(q21q26.2)</td>
<td></td>
</tr>
<tr>
<td>t(6;9)(p23;q34)</td>
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MDS, myelodysplastic syndrome.

*In the setting of persistent cytopenias of unknown etiology, these abnormalities are considered presumptive evidence of MDS. Adapted from Brunning et al.*

**Image 2** A 55-year-old man with pancytopenia with no significant medical history. **A**, A peripheral smear demonstrated occasional ovalocytes. Although most neutrophils appeared morphologically normal, a subset of dysplastic forms was seen (Wright, ×60). The aspirate smears were aspiculate and hemodilute and are not shown. **B**, A core biopsy specimen showed 10% to 20% hypopcellularity. No obvious clusters of immature cells were noted (H&E, ×40). **C**, A CD34 immunohistochemical stain, however, demonstrated 5% to 10% increased blasts (×60). No ringed sideroblasts were seen by iron stain (not shown). Cytogenetics reportedly demonstrated t(3;21)(q26;q22) and del(7), consistent with a diagnosis of myelodysplastic syndrome (refractory anemia with excess blasts-1).
Drugs

Drugs are among the most common causes of acquired AA, chief among them being chloramphenicol. Because of a known but as-of-then unquantified relationship of chloramphenicol with AA, the 1960s California State Senate requested a study to further assess this risk. By investigating every fatality in the state of California during a 1.5-year period, it assessed a 13-fold increase in risk for the development of AA in patients taking chloramphenicol. Moreover, this risk usually occurred after the second or third course of chloramphenicol treatment and was not related to dosage. Since then, additional drugs have been shown to be associated with the development of AA, including nonsteroidal anti-inflammatory drugs, antithyroid drugs, corticosteroids, penicillamine, allopurinol, and gold. The etiology of such aplasia is thought to be a direct cytotoxic effect or immune-related idiosyncratic response. A more extensive list of drugs reportedly associated with marrow aplasia is provided in Table 4.

Table 4

Drugs and Chemicals Associated With Acquired Aplastic Anemia

<table>
<thead>
<tr>
<th>Drugs and Chemicals</th>
<th>Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Allopurinol</td>
<td>Chloramphenicol, streptomycin, tetracycline, methicillin, mebendazole, sulfonamides, trimethoprim/sulfamethoxazole, flucytosine</td>
</tr>
<tr>
<td>Antibiotics</td>
<td></td>
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<tr>
<td>Anticonvulsants</td>
<td>Hydantoins, carbamazepine, phenacemide</td>
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<tr>
<td>Antidiabetes drugs</td>
<td>Tolbutamide, chlorpropamide</td>
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<tr>
<td>Antithiamine drugs</td>
<td>Cimetidine, ranitidine, chlorpheniramine</td>
</tr>
<tr>
<td>Antiprotozoals</td>
<td>Quinacrine, chloroquine</td>
</tr>
<tr>
<td>Antithyroid drugs</td>
<td>Methimazole, methylthiouracil, propylthiouracil</td>
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<tr>
<td>Benzene</td>
<td></td>
</tr>
<tr>
<td>Carbimazole</td>
<td></td>
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<tr>
<td>Carbonic anhydrase inhibitors</td>
<td>Acetazolamide, methazolamide</td>
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<tr>
<td>Cytotoxic drugs used in cancer chemotherapy</td>
<td></td>
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<tr>
<td>Estrogens</td>
<td></td>
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<tr>
<td>Gold</td>
<td></td>
</tr>
<tr>
<td>Insecticides</td>
<td></td>
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<tr>
<td>Lithium</td>
<td></td>
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<tr>
<td>Methyldopa</td>
<td></td>
</tr>
<tr>
<td>Nonsteroidal anti-inflammatory drugs</td>
<td>Phenytoin, indomethacin, ibuprofen, sulindac, aspirin</td>
</tr>
<tr>
<td>d-penicillamine</td>
<td></td>
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<tr>
<td>Potassium perchlorate</td>
<td></td>
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<tr>
<td>Quinidine</td>
<td></td>
</tr>
<tr>
<td>Sedatives</td>
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</table>

* Adapted from Shimamura and Guinan.

Image 3 – A 61-year-old man, after heart transplantation, with pancytopenia. A, A peripheral blood smear demonstrated occasional hypersegmented neutrophils, rare nucleated RBCs, and occasional left-shifted myeloid cells (Wright, ×60). B, A core biopsy specimen demonstrated a hypocellular marrow with trilineage hematopoiesis (H&E, ×20). Flow cytometry was noncontributory. Overall, the findings were nonspecific, and the clinical team attributed the patient’s pancytopenia to his immunsuppressive regimen.

In addition to drugs, simple chemicals are known to be associated with the development of AA. Benzene, an industrial solvent, has been implicated, perhaps more than any other chemical, in the development of AA and subsequent leukemia. In addition, insecticides such as dichlorodiphenyltrichloroethane (DDT), γ-hexachlorocyclohexane (lindane), and hydrocarbon-based glue vapors have been associated with increased rates of AA.

Not surprisingly, radiation toxicity is also associated with AA. Although the earliest manifestation of acute radiation sickness is the “hematopoietic syndrome,” characterized by a decline in hematopoietic stem cells with resulting cytopenias, the nadir in these cytopenias usually occurs 1 to 4 weeks after exposure. In addition, iatrogenic radiation therapy for malignancy has been shown to cause or exacerbate pancytopenia.

**Infection**

Infection is an additional cause of pancytopenia in both children and adults. Several infections are known to induce bone marrow failure, and these are often treatable and reversible. Infection can also lead to acquired HLH, which can be rapidly fatal without aggressive intervention.

Parvovirus B19 is a virus traditionally associated with bone marrow failure. Although parvovirus B19 is commonly associated with megaloblastic anemia, in one study, 9 of 167 children with a clinical syndrome suggestive of parvovirus had an “AA picture,” and of these, 4 were shown to be acutely infected with parvovirus. In another study, serologic tests showed that 40.7% of 27 patients with AA demonstrated parvovirus anti-IgM antibodies compared with 5% of 20 control patients. Bone marrow biopsy in patients with acute parvovirus infection generally demonstrates a decrease in erythroid precursors with only rare giant pronormoblasts and large eosinophilic nuclear inclusion bodies. Patients with suppressed immune systems, however, have more chronic infection and often show numerous infected large cells with inclusions on marrow examination.

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**Image 4**

A 45-year-old man after kidney and pancreas transplantation 8 months earlier, presenting with cytopenias. **A**, An aspirate smear demonstrated numerous giant proerythroblasts with pale cytoplasmic inclusions (arrows). **B**, A core biopsy specimen demonstrated a normocellular marrow with numerous large erythroid precursors with viral inclusions. No erythroid maturation was seen. **C**, An immunohistochemical stain for parvovirus highlighted many of these large cells. A diagnosis of cytopenias associated with parvovirus infection was made.
HIV has been shown to cause bone marrow failure and subsequent pancytopenia. The degree of hematologic findings in the course of HIV infection varies widely. Initial infection often leads to a lymphopenia followed by an atypical lymphocytosis, which may or may not be associated with a transient pancytopenia. However, after a period of clinical latency, in which the bone marrow may be initially hypercellular, the bone marrow becomes hypocellular with a resulting pancytopenia. Interestingly, all lineages can appear dysplastic in HIV, thus bringing up a differential diagnosis that includes MDS, as such, a diagnosis of MDS in the setting of HIV infection should be made with caution.

Hepatitis-associated AA is a well-recognized disease that preferentially affects young men approximately 2 to 3 months after an episode of acute hepatitis. It is usually fatal if left untreated, and no association with drugs, toxins, or hepatitis A, B, or C viruses has been elucidated. Interestingly, this disease is probably not uncommon; indeed, hepatitis has been documented in 2% to 5% of cases of AA in the West and 4% to 10% of cases in Asia.

Case reports have described other organisms that can cause bone marrow suppression, including leptospirosis and dengue fever. A summary list of such entities is provided in Table 5.

Numerous viruses, bacteria, protozoa, and fungi are also known to cause pancytopenia via an acquired HLH syndrome. Patients with this disease present similarly to those with primary HLH, as described before, and are diagnosed using the same criteria. This disease affects all age groups, and most patients have no underlying immune defect. Although an exhaustive list of etiologic infectious agents has been implicated, leading causes include Epstein-Barr virus, cytomegalovirus, herpes simplex virus, adenovirus, and leishmania.

Pregnancy

Pregnancy has also been reported to be associated with bone marrow failure; in fact, AA was first described in a pregnant woman. Since then, multiple cases have been reported of women developing pancytopenia during pregnancy. Interestingly, although in several of those patients the pancytopenia improved after abortion or delivery, many consider the association between pregnancy and bone marrow failure to be purely coincidental.

Autoimmune Disease

Autoimmune diseases can also present with new-onset pancytopenia, which can have multiple etiologic factors. Indeed, hematologic abnormalities are often part of the diagnostic criteria for such diseases, including systemic lupus erythematosus (SLE). Approximately 57% to 78% of patients with SLE are anemic, possibly because of a combination of anemia of chronic disease, renal insufficiency, autoimmune hemolytic anemia, and microangiopathic hemolytic anemia. Similarly, approximately 47% of patients with SLE have neutropenia, which may be because of autoimmune destruction, bone marrow suppression, hypersplenism, and/or drugs. In addition, 10% to 25% of patients with SLE have thrombocytopenia, which may be immune-mediated in etiology. Multiple studies have investigated bone marrow findings in patients with SLE and peripheral cytopenias. Although these findings are nonspecific, bone marrow biopsies often demonstrate hypoplasia, dyserythropoiesis, and increased reticulin fibrosis, and one study also reported megakaryocytic atypia.

A more concerning pancytopenic presentation of SLE is secondary HLH, also called macrophage activation syndrome (MAS), which is histologically identical to primary HLH and can similarly be rapidly fatal. Interestingly, MAS can occur in many autoimmune diseases, and has been described in systemic juvenile idiopathic arthritis, Kawasaki disease, and adult-onset Still disease, among others. As in primary HLH, MAS is often thought to be initiated by a precipitating event such as infection or drugs. Although many currently use the primary HLH criteria for diagnosing MAS, this practice remains suboptimal. Systemic juvenile idiopathic arthritis, for instance, often presents with a leukocytosis, so neutropenia, one of the criteria for primary HLH, often does not appear until late in the course of disease. Moreover, as in primary HLH, hemophagocytosis is often not found on bone marrow biopsy.

Although drug-induced aplasia was previously discussed, drugs can also cause immune-mediated cytopenias through the formation of antibodies with cross-reactivity to the drug and to hematopoietic cells or through drug antigen-antibody complexes that passively bind hematopoietic cells and fix complement. This phenomenon has been most frequently associated with quinine, sulfonamides, and rifampin; however, unicytopenias or bicytopenias are much more common and well described than pancytopenia, which is extremely rare.
Sequestration

Splenomegaly occurs with many diseases and is known to lead to hypersplenism with resulting pancytopenia. The mechanism of such pancytopenia has long been thought to be a combination of hemolysis, sequestration, and premature destruction of blood cells. Indeed, up to 90% of the peripheral platelet mass, 30% of the red cell mass, and 65% of granulocytes can be sequestered in a massive spleen. The often proliferative bone marrow findings in such cases may be unhelpful unless they help elucidate the underlying cause of splenomegaly, such as lymphoma. Not unexpectedly, in patients whose cytopenias are caused by splenic sequestration, splenectomy can be essentially curative.

Nutritional Deficiencies

New-onset pancytopenia can sometimes result from nutritional deficiency. Copper deficiency, which can occur because of long-term total parenteral nutrition, gastrointestinal surgery, weight reduction surgery, excessive zinc intake, and even renal failure, can lead to hematologic abnormalities, including pancytopenia. Bone marrow findings often demonstrate vacuolization of erythroid and myeloid precursors, increased stainable iron in plasma cells, and ringed sideroblasts. Although these bone marrow findings, as well as the peripheral pancytopenia are reversible with copper replacement, patients with such copper deficiency have been initially misdiagnosed with MDS, and have even been referred for allogeneic bone marrow transplantation. If copper deficiency is suspected, additional laboratory studies such as serum iron and transferrin saturation, which are usually normal, and serum copper and ceruloplasmin levels, which are uniformly low, can be helpful.

Folate and B₁₂ deficiency are classic causes of macrocytic anemia, and although these deficiencies commonly present with anemia and thrombocytopenia, they can...
occasionally present with pancytopenia. In the West, folate deficiency is rare but can occur because of increased demand, decreased absorption, or nutritional deficiencies, as seen with chronic alcoholism. Pregnant women in the West occasionally have folate-deficient pancytopenia, which is correctable with folate administration. B₁₂ deficiency is also exceedingly rare in the West and is much more commonly associated with pernicious anemia and chronic atrophic gastritis than with nutritional deficiency. In other areas of the world, however, such as India, nutritional etiologies of folate and B₁₂ deficiency are quite common and can often lead to pancytopenia. A 1989 study based in India found that of 139 patients with megaloblastic anemia, 76% had B₁₂ deficiency, 6.8% had folate deficiency, and 8.8% had a combination of both. Of this entire group, 43.8% had pancytopenia. Additional studies from India found B₁₂ and folate deficiency to be significant causes of pancytopenia. In one study, 72% of cases of pancytopenia were attributed to megaloblastic anemia caused by folate and/or B₁₂ deficiency, and in another study, 22% of cases were attributed to the same etiology. Regardless of the etiology, bone marrow aspiration and biopsy are quite characteristic with folate/B₁₂ deficiency and demonstrate a hypercellular marrow with erythroid hyperplasia and megaloblastic maturation. Additional laboratory findings can be diagnostically helpful, such as peripheral macrocytosis, hypersegmented neutrophils, and decreased serum cobalamin and red cell folate levels.

Anorexia nervosa has also been reported to lead to bone marrow failure, most likely through severe vitamin and mineral deficiency. In a study of the bone marrow of 44 patients with a diagnosis of anorexia nervosa, the bone marrow was hypoplastic or aplastic in 39%, with a significant fraction of these cases demonstrating gelatinous degeneration or serous atrophy. Interestingly, only 1 of these patients demonstrated frank pancytopenia, with the majority...
Hematopathology / Review Article

Leukemias and MDSs

Hematopoietic neoplasms cause pancytopenia in both children and adults, and acute leukemias are among the most common of these neoplasms. Acute lymphoblastic leukemia accounts for approximately 80% of all childhood leukemias and is common in adults. The clinical presentation is demonstrating anemia and/or leukopenia. Notably, these patients demonstrate complete bone marrow recovery with dietary and therapeutic intervention.

Marrow Space Infiltrating Lesions

Hematopoietic cells are generally produced in the bone marrow; therefore, it stands to reason that entities that occupy marrow space can lead to pancytopenia through direct replacement, interference with ongoing hematopoiesis, or concomitant fibrosis. Common etiologies include hematopoietic neoplasms and metastases. A general overview to the most common of these entities is provided here.

Leukemias and MDSs

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variable, but is often a manifestation of the patient’s underlying pancytopenia resulting from the replacement of bone marrow by lymphoblasts. Common symptoms and signs include fatigue, easy bruising, and infection, as well as lymphadenopathy, hepatosplenomegaly, and bone pain. Although B-lymphoblastic leukemia generally presents with cytopenias, the peripheral blood is often involved by B lymphoblasts, and therefore the leukocyte count may be decreased, normal, or markedly elevated. Bone marrow examination is usually diagnostic and generally demonstrates replacement of marrow space by sheets of B lymphoblasts. Of course, flow cytometric and cytogenetic studies should be performed concomitantly with bone marrow aspirate and biopsy for proper classification. Prognosis is generally favorable in children, with a 10-year 63% event-free survival seen in children; however, this number drops to 25% to 35% in adults.\(^93\)

Acute lymphoblastic leukemia is generally a disease of childhood, whereas AML is usually a disease of adults, accounting for 80% of the acute leukemic cases in adults.\(^94\)

Currently, the World Health Organization classifies such leukemias into several categories, including AML with myelodysplasia-related changes, therapy-related myeloid...
neoplasms, AML with recurrent genetic abnormalities, and AML, not otherwise specified. As with acute lymphoblastic leukemia, patients with AML often have complications of cytopenias, including weakness, fatigability, bleeding, or infection. Blasts may not be present on the peripheral smear, but usually can be found. Other initial clues may be helpful, such as dysgranulopoiesis in the case of AML with myelodysplasia-related changes and a clinical history of cytotoxic therapy in the case of therapy-related myeloid neoplasms. Bone marrow aspirate and biopsy, concurrent immunophenotyping, cytogenetics, and molecular studies are diagnostic and allow for correct classification.

Similarly, other hematopoietic neoplasms, such as the spectrum of MDSs, can present with pancytopenia, and many of these syndromes are defined, at least in part, by their cytopenias. Primarily a disease of adults, MDS is commonly characterized by progressive bone marrow failure, with several of the subtypes often progressing to AML. Not unexpectedly, the more “high-grade” MDS categories that demonstrate extensive bone marrow failure, such as refractory cytopenia with multilineage dysplasia and refractory anemia with excess blasts, more commonly present with pancytopenia. Less often, compared with the MDSs, the combined myelodysplastic/myeloproliferative diseases, including chronic myelomonocytic leukemia, can also occasionally present with pancytopenia, but most cases will show an elevated WBC count. Bone marrow aspiration and biopsy with concurrent cytogenetics studies is crucial in patients suspected of having MDS or MDS/myeloproliferative disease to exclude acute leukemia, and the aspirate smears typically demonstrate varying levels of dysplasia in 1 or more cell lines.

An interesting diagnostic dilemma in the setting of new-onset pancytopenia can be differentiating hypoplastic MDS from AA; this differentiation was discussed briefly before. To
reiterate, both can present with profound hypocellularity, but certain clues can help in the differentiation. MDS often presents with dyserythropoietic RBCs, dysplastic granulocytes, and hypogranular platelets, which are not often seen in the setting of AA. Bone marrow biopsies can be variably patchy in both diseases, but islands of immature cells as well as fibrosis are much more common in MDS, and immunohistochemical studies for CD34 may show an increase in immature cells in hypoplastic MDS. Certain chromosomal rearrangements are also a feature of MDS and not AA and can help in this distinction (Table 3).96

Non-Hodgkin Lymphomas and Chronic Leukemias

Non-Hodgkin lymphomas and chronic leukemias can also lead to pancytopenia, but such presentations are rare unless there is significant bone marrow replacement, autoimmune cytopenias, or splenomegaly. One of the chronic leukemias that most often presents with pancytopenia is hairy cell leukemia, which was first described in 1958 and comprises approximately 2% of all leukemias.97 In addition to generalized pancytopenia, patients often have a concurrent monocytopenia. According to a 1994 study, patients generally have a hemoglobin concentration less than 8.5 g/dL (85 g/L), neutrophil count less than 500/µL (0.5 × 10^9/L), and platelet count less than 50 × 10^9/L (50 × 10^9/L). Circulating hairy cells are usually present; however, the findings can be subtle.98 Bone marrow core biopsies are helpful—hairy cells demonstrate diffuse, interstitial bone marrow infiltration in more than 99% of cases.99 However, the appearance of hairy cells can be deceiving because of their resemblance to marginal zone lymphoma, prolymphocytic leukemia, monocytoid B-cell lymphoma, large granular lymphocytic leukemia (LGL), and mast cell disease on aspirate smears, and their interstitial pattern on bone marrow biopsy may mimic erythroid precursors. Immunophenotyping is thus of great importance in this diagnosis. Interestingly, the mechanism of pancytopenia in hairy cell leukemia is most likely multifactorial, resulting from leukemic replacement of marrow, reticulin fibrosis, and splenomegaly with hypersplenism.

LGL can also present with a clinical picture similar to that of acquired AA. In one institution’s series of patients with T-LGL, 4% of patients had AA.100 It is postulated that the etiology of this pancytopenia is the expansion of cytotoxic T cells, which then directly suppress hematopoiesis through direct cellular cytotoxicity or the production of cytokines. In these patients, peripheral blood and bone marrow findings are similar to those of acquired AA; however, there may be a relative or absolute increase in large granular lymphocytes in the blood and/or an increased interstitial infiltrate of CD3+, CD8+, granzyme B+ lymphocytes in clusters in the bone marrow biopsy specimen.101 In addition, there may be a distinct population of CD8 dim T cells with CD57 expression seen on flow cytometry.102,103

Plasma cell myeloma can occasionally present with pancytopenia, albeit rarely. Anemia by itself is a presenting hallmark of the disease, along with bone pain, fatigue, and weight loss, and is thought to be because of numerous factors, including bone marrow replacement, erythropoietin deficiency, and renal failure.104 Concomitant thrombocytopenia and granulocytopenia can be seen in up to 10% of patients at the time of diagnosis.105,106 Bone marrow aspiration and biopsy are generally diagnostic, and one large study demonstrated 10% or more clonal plasma cells in 96% of patients.105

Fibrotic Diseases

Diseases that cause fibrosis of the bone marrow can also lead to pancytopenia, most likely because of both ineffective hematopoiesis and subsequent splenomegaly with hypersplenism. The differential diagnosis of bone marrow fibrosis is extensive and includes primary myeloid neoplasms, malignant lymphomas, metastatic carcinomas, inflammatory reactions, granulomatous reactions, and osteopaties.

Many myeloid neoplasms can lead to myelofibrosis, but perhaps the myeloid neoplasms most commonly associated with bone marrow fibrosis are the myeloproliferative neoplasms. Among these neoplasms, primary myelofibrosis most often presents with bone marrow fibrosis and cytopenias.107,108 In the fibrotic stage of this disease, patients generally have anemia, and often thrombocytopenia, because of splenomegaly. However, the WBC count can be variable.109 As fibrosis becomes more severe, the patient’s cytopenias worsen.110 At diagnosis, the blood smear is often leukoerythroblastic with marked poikilocytosis, including teardrop-shaped RBCs, nucleated RBCs, giant platelets, and left-shifted myeloid cells. The bone marrow biopsy in this fibrotic phase demonstrates decreased cellularity, megakaryocytic atypia and clustering, and reticulin/collagen fibrosis.111,112 Additional diagnostic criteria that are helpful for a diagnosis include demonstration of a JAK2 V617F mutation (seen in approximately 50% of patients) and an increase in serum lactate dehydrogenase levels.

Of the malignant lymphomas, Hodgkin lymphoma involving the marrow is usually associated with fibrosis. In the modern era, bone marrow involvement is uncommon in Hodgkin lymphoma; in a study of 174 patients with Hodgkin disease, only 19 had bone marrow involvement at the time of presentation.111 However, when the bone marrow is involved, patients can have marrow fibrosis and pancytopenia, and, in the absence of Reed-Sternberg cells, diagnosis can be challenging.112 Interestingly, the degree of cytopenias does not appear correlated with the extent of marrow fibrosis, suggesting a possible additional mechanism of bone marrow suppression in patients with Hodgkin lymphoma.113 Moreover, patients with an acute clinical form of Hodgkin lymphoma often present with fibrosis and pancytopenia. In a study of 9...
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is most common in patients with lung, breast, or prostate carcinoma. As in primary myelofibrosis, the mechanism of bone marrow failure is believed to occur because of replacement of hematopoietic tissue by abnormal tissue, with hypersplenism resulting from extramedullary hematopoiesis. Diagnosis is usually straightforward with appropriate immunohistochemical studies and sufficient clinical history.

Autoimmune fibrosis is an additional cause of fibrosis-mediated cytopenias, including pancytopenia. SLE can certainly present with pancytopenia and bone marrow fibrosis, but noncharacterized autoimmune diseases with positive autoimmune serology can also lead to modest reticulin fibrosis.

patients with such a clinical presentation, 7 had pancytopenia, and the majority of cases involved the spleen, bone marrow, and liver. Marrow fibrosis was present in 5 patients, and the noninvolved areas of the bone marrow biopsy were either hypocellular or hypercellular. Similarly, in an additional study of patients with Hodgkin lymphoma and a rapid clinical course, patients presented with fever, pancytopenia, liver dysfunction, and extensive infradiaphragmatic disease and bone marrow involvement.

Metastatic carcinoma can also lead to bone marrow involvement and subsequent marrow fibrosis, but this occurs in fewer than 10% of patients with metastatic carcinoma and is most common in patients with lung, breast, or prostate carcinoma. As in primary myelofibrosis, the mechanism of bone marrow failure is believed to occur because of replacement of hematopoietic tissue by abnormal tissue, with hypersplenism resulting from extramedullary hematopoiesis. Diagnosis is usually straightforward with appropriate immunohistochemical studies and sufficient clinical history.

Autoimmune fibrosis is an additional cause of fibrosis-mediated cytopenias, including pancytopenia. SLE can certainly present with pancytopenia and bone marrow fibrosis, but noncharacterized autoimmune diseases with positive autoimmune serology can also lead to modest reticulin fibrosis.

A 75-year-old man with splenomegaly and pancytopenia. A, A peripheral smear revealed lymphoid cells with moderate amounts of cytoplasm, some with cytoplasmic villi. B, A core biopsy specimen demonstrated a slightly hypercellular marrow with an interstitial infiltrate of monocytoid cells with a “fried egg” appearance. C, Reticulin fibrosis was increased. D, A CD20 immunostain highlighted the interstitial pattern of infiltrating lymphocytes. Concomitant flow cytometry demonstrated a population of \( \kappa \)-restricted B cells expressing CD25, CD103, and CD11c, diagnostic of hairy cell leukemia.
Autoimmune fibrosis can be difficult to distinguish from primary myelofibrosis, but peripheral blood smears often lack teardrop poikilocytosis, and bone marrow biopsy generally demonstrates reactive lymphoid infiltrates in the absence of megakaryocytic clustering. Splenomegaly is also usually absent.\(^{118,119}\)

Granulomatous disease can also lead to pancytopenia. In a study of 38 adults with disseminated miliary tuberculosis, 3 patients presented with pancytopenia, and all 3 demonstrated granulomata in their bone marrow.\(^{120}\) The mechanism of pancytopenia in such patients is unclear, but could be multifactorial, including hypersplenism, increased histiocytic phagocytosis, and bone marrow infiltration by tubercular granulomas.\(^{121}\)

Osteopathies can also lead to bone marrow fibrosis and subsequent pancytopenia. Vitamin D–dependent rickets in breastfed infants has been reported to lead to marrow fibrosis and pancytopenia, most likely because of secondary hyperparathyroidism, which can cause osteosclerosis.\(^{122}\) Patients with primary hyperparathyroidism and patients with renal osteodystrophy receiving long-term dialysis have also been known to develop bone marrow fibrosis, osteomalacia, increased osteoclastic activity, and splenomegaly, with resulting pancytopenia.\(^{123}\) Additional laboratory
A 68-year-old man with pancytopenia. A, A peripheral blood smear demonstrated pancytopenia in the setting of leukoerythroblastosis. Dacrocytes and ovalocytes were also identified (Wright, ×60). B, and C, A core biopsy specimen demonstrated a hypercellular marrow with clusters of atypical megakaryocytes and dilated sinusoids (B, H&E, ×20; C, H&E, ×60). Reticulin fibrosis was reportedly increased (not shown). Concurrent molecular studies demonstrated a V617F mutation in JAK2. Overall, the findings were most consistent with a diagnosis of primary myelofibrosis.

A granzyme stain (×60) and F, a T-cell intracytoplasmic antigen stain (×60) both demonstrated increased cytoplasmic staining in the lymphocyte population. Upon further workup, the patient was found to harbor a T-cell receptor γ gene rearrangement, and a diagnosis of large granular lymphocytic leukemia was made.
studies, such as serum calcium and parathyroid hormone levels, can be helpful in this setting.

To summarize, the causes of new-onset pancytopenia are vast and extremely varied. Here we have provided a general overview of some of the more common causes of pancytopenia in both children and adults; however, this list is by no means complete, and patients with complex conditions may have multifactorial causes of pancytopenia. The diagnosis, especially in the absence of obvious neoplasm, can be extraordinarily challenging even in the presence of sharp clinical acumen; therefore, numerous studies, including peripheral smear examination and bone marrow biopsy, are often required to make an accurate diagnosis.

From the Department of Pathology, Stanford University, Stanford, CA.

Address reprint requests to Dr Weinzierl: Dept of Pathology, Stanford University, 300 Pasteur Dr, Lane 235, Stanford, CA 94305-5324; eweinzie@stanford.edu.

References


Image 13: A 34-year-old woman with pancytopenia with lupus and end-stage renal disease. A, A peripheral smear demonstrated numerous teardrop cells and elliptocytes (Wright, ×60). An aspirate smear was aspiculate and is not shown. B, A bone marrow core biopsy demonstrated extensive fibrosis, bone scalloping, and numerous osteoclasts (H&E, ×20). The findings were consistent with a diagnosis of renal osteodystrophy.


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