Chapter 16
Congenital Neutropenia and Migration Defects

Thomas F. Michniacki, Saara Kaviany, and Kelly Walkovich

Abbreviations

AML	Acute myeloid leukemia
ANC	Absolute neutrophil count
CBCD	Complete blood count with differential
CGD	Chronic granulomatous disease
CHS	Chediak-Higashi syndrome
DADA2	Deficiency of adenosine deaminase 2
DEB	Diepoxybutane
DHR	Dihydrorhodamine
FISH	Fluorescence in situ hybridization
G-CSF	Granulocyte colony-stimulating factor
Hgb	Hemoglobin
HLH	Hemophagocytic lymphohistiocytosis
HSCT	Hematopoietic stem cell transplantation
MDS	Myelodysplastic syndrome
MMC	Mitomycin C (MMC)
MPO	Myeloperoxidase
SBDS	Shwachman-Bodian-Diamond syndrome
SCN	Severe congenital neutropenia
SDS	Shwachman-Diamond syndrome
WBC	White blood count

T. F. Michniacki (✉) · K. Walkovich
Division of Pediatric Hematology/Oncology, C.S. Mott Children's Hospital/University of Michigan, Ann Arbor, MI, USA
e-mail: tmich@med.umich.edu

S. Kaviany
Monroe Carell Jr. Children's Hospital at Vanderbilt, Nashville, TN, USA

J. A. Bernstein (ed.), *Primary and Secondary Immunodeficiency*,
https://doi.org/10.1007/978-3-030-57157-3_16

Introduction

Congenital conditions caused by an absolute or functional deficiency of neutrophils are associated with considerable morbidity and mortality in children and adults. Quantitative neutrophil disorders include severe congenital neutropenia, cyclic neutropenia, and bone marrow failure syndromes, notably Shwachman-Diamond syndrome [1]. Phagocyte functional deficits result from myeloperoxidase deficiency, chronic granulomatous disease, the leukocyte adhesion disorders, Chediak-Higashi syndrome, and neutrophil-specific granule deficiency [2]. The focus of this chapter is to touch upon many of these disorders and provide clinicians with the knowledge necessary to effectively diagnose and manage these clinically important immunodeficiency conditions (Table 16.1).

Case Presentation 1

A 5-year-old boy was being evaluated by a pediatric hematologist for worsening of neutropenia since the age of 1 year. A routine complete blood cell count with platelets and differential (CBCPD) obtained at 12 months of age showed a white blood cell count (WBC) 3.6×10^3/uL (absolute neutrophil count (ANC) 0.8×10^3/uL), hemoglobin (Hgb) 12.1 g/dL, and platelets 389×10^3/uL. The initial moderate neutropenia was attributed to a recent viral illness and the patient was monitored with annual CBCPDs by his pediatrician. At well-child visits his mother noted that his stools were often loose and voluminous. His growth overall was poor with height falling below the 5th percentile for age. He had no history of bleeding symptoms, persistent adenopathy, or infections requiring hospitalizations. At a recent dental visit, he was found to have gingivitis and notable oral ulcers. Physical examination was without limb or nail abnormalities, significant bruising/petechiae, leukoplakia, or pulmonary crackles. A repeat CBCPD was obtained by the hematologist noting a WBC of 1.3×10^3/uL (ANC 0.3×10^3/uL), Hgb 9.8 g/dL, and platelets 178×10^3/uL. Given CBCD abnormalities a bone marrow examination was performed showing overall marrow hypocellularity (average of 40%) with granulocyte and erythroid hypoplasia but no evidence of dysplastic changes, malignancy, or ringed sideroblasts. Additional laboratory studies revealed a reduced pancreatic isoamylase level, no evidence of abnormal chromosomal breakage upon cellular exposure to mitomycin C (MMC) or diepoxybutane (DEB), and normal telomere lengths within lymphocytes and granulocytes by fluorescence in situ hybridization (FISH) testing.

Table 16.1 Congenital neutropenia and migration defect disorders

Condition	Mutation	Inheritance	Clinical manifestations	Diagnostic testing	Management
Chediak-Higashi syndrome	*LYST*	AR	Ocular and cutaneous albinism, neurologic decline, large granules within granulocytes, recurrent infections, bleeding, risk of hemophagocytic lymphohistiocytosis (HLH)	Platelet aggregation studies, neurologic imaging, EMG/EEG, peripheral smear review, HLH evaluation studies, light microscopy of hair shafts, genetic testing	Antimicrobial prophylaxis, G-CSF, +/− interferon gamma, therapies targeting HLH, HSCT improves hematologic/immune dysfunction but fails to slow progression of neurologic abnormalities
Chronic granulomatous disease	*CYBB* *CYBA* *NCF1* *NCF2* *NCF4* *CYBC1*	XL AR AR AR AR AR	Catalase-positive organism infections, autoimmunity including granuloma formation/oral ulcers/urologic difficulties/colitis, hepatic dysfunction, risk of fungal disease and mulch pneumonitis	Neutrophil functional testing (DHR preferred), ESR/CRP trending, imaging to evaluate for infections with inflammatory marker elevations, genetic testing	Prophylaxis with TMP-SMX and itraconazole, +/− interferon-gamma, aggressive infection management with broad-spectrum antimicrobials, glucocorticoids for liver granulomas and mulch/nocardia pneumonitis, immunosuppression for significant auto-inflammation, HSCT
Cyclic neutropenia	*ELANE*	AD	Neutropenia every 14–35 days with associated monocytosis, severe bacterial and fungal infections (including *Clostridium*), osteopenia, oral ulcers and gingivitis, less risk of malignant conversion than severe congenital neutropenia	Bone marrow biopsy, frequent CBCDs to evaluate neutrophil count pattern, genetic testing	G-CSF, HSCT in severe refractory cases

(continued)

Table 16.1 (continued)

Condition	Mutation	Inheritance	Clinical manifestations	Diagnostic testing	Management
Leukocyte adhesion deficiency	*ITGB2*	AR	Neutrophilia, umbilical cord separation delay, recurrent bacterial infections, inadequate wound healing, severe periodontitis/gingivitis, skin ulcerations, colitis, HPV infections	Absence of functional CD18 and CD11 on leukocytes via flow cytometry, genetic testing	Ustekinumab (IL-23/IL-12 antibody), quality oral hygiene, antimicrobial prophylaxis, GM-CSF not useful, HSCT in those with severe phenotypes
	SLC35C1	AR	Neutrophilia, recurrent infections, developmental delay, short stature with depressed nasal bridge, Bombay (hh) blood phenotype, no delay in umbilical cord separation, periodontal disease	Analysis of blood phenotype, absence of sialyl Lewis X (CD15a) on leukocytes via flow cytometry, genetic testing	Antimicrobial prophylaxis, quality oral hygiene, fucose supplementation (limited evidence)
	FERMT3	AR	Neutrophilia, recurrent bacterial infections, bleeding due to dysfunctional platelet aggregation, osteoporosis-like bone irregularities, delayed umbilical cord separation	Evaluation of integrin activation and expression on leukocytes, functional neutrophil and platelet assays, genetic testing	Antimicrobial prophylaxis, platelet transfusions in those with bleeding, HSCT
Myeloperoxidase deficiency	*MPO*	AR	Frequently asymptomatic, *Candida* or other fungal infections in the setting of diabetes or additional immunodeficiency	Histochemical staining for MPO, DHR	Antimicrobial prophylaxis in those with recurrent infections, excellent control of blood glucose levels in diabetics
Severe congenital neutropenia	*ELANE* *HAX1* *WAS* *G6PC3* *JAGN1*	AD AR XL AR AR	Severe neutropenia (often <200/uL), bone marrow biopsy with absence of myeloid precursors, recurrent bacterial/fungal infections, lack of dysmorphic features, risk of MDS/AML progression	Bone marrow biopsy, CBCDs, genetic testing	G-CSF daily, HSCT with severe disease

Shwachman-Diamond syndrome	*SDBS* *EFL1* *DNAJC21* *SRP54*	AR AR AR AD	Bone marrow hypocellularity with pancytopenia, pancreatic exocrine insufficiency, steatorrhea, skeletal abnormalities, hepatic disease, short stature, failure to thrive, elevated risk for MDS/AML	Bone marrow evaluation, ultrasound of pancreas, serum trypsinogen (< 3yo) or isoamylase (>3yo), genetic screening	G-CSF, annual bone marrow evaluations, pancreatic enzyme and fat-soluble vitamin replacement, HSCT in those with severe disease
Specific granule defects	*CEBPE* *SMARCD2*	AR AR	Recurrent bacterial/fungal infections, absence of specific granules within leukocytes, bleeding, frequent bi-lobed neutrophils (pseudo Pelger-Huet anomaly), risk of progression to MDS/AML	Light/electron microscopy evaluation of neutrophils, biochemical measurements of granule proteins, abnormal neutrophil chemotaxis, prolonged bleeding time, genetic testing	Aggressive management of infections, antimicrobial prophylaxis, GM-CSF with severe infections, HSCT in those with significant disease
Wiskott-Aldrich syndrome	*WAS*	XL	Immunodeficiency with recurrent infections, neutropenia, eczema, malignancy risk, bleeding, lymphadenopathy, hepatosplenomegaly, autoimmunity including immune-mediated thrombocytopenia	WAS protein analysis via flow cytometry on lymphocytes, reduced T lymphocytes and immunoglobulins, decreased mean platelet value, genetic testing	Platelet transfusions (irradiated), prophylactic antimicrobials, IVIG for hypogammaglobulinemia, low-dose IL-2 therapy, rituximab in those with autoimmune cytopenias, HSCT, gene therapy

AD autosomal dominant, *AML* acute myeloid leukemia, *AR* autosomal recessive, *CBCD* complete blood count with differential, *CRP* C-reactive protein, *DHR* dihydrorhodamine, *EEG* electroencephalogram, *EMG* electromyography, *ESR* erythrocyte sedimentation rate, *FISH* fluorescence in situ hybridization, *G-CSF* granulocyte colony-stimulating factor, *GM-CSF* granulocyte-macrophage colony-stimulating factor, *HLH* hemophagocytic lymphohistiocytosis, *HSCT* hematopoietic stem cell transplantation, *IL-2* interleukin-2, *IVIG* intravenous immunoglobulin, *MDS* myelodysplastic syndrome, *MPO* myeloperoxidase, *NBT* nitroblue tetrazolium, *TMP-SMX* trimethoprim-sulfamethoxazole, *XL* X-linked

Diagnosis/Assessment

Signs and Symptoms

Congenital bone marrow failure conditions can present at any age with cytopenias. The diagnosis of Shwachman-Diamond syndrome (SDS) most frequently manifests with intermittent and waxing/waning neutropenia during childhood [3, 4]. Anemia and thrombocytopenia may be present in patients with SDS but may be intermittent and asymptomatic. Bone marrow biopsy often reveals hypocellularity and hematopoietic cell line maturation delay/hypoplasia. Progression to aplastic anemia can occur with additional transformation to myelodysplastic syndrome and acute myeloid leukemia possible. Overall the risk in SDS for having malignant transformation is significant (18–36% at 30 years) but less so than patients with Fanconi anemia (40% by age 50) [3, 5]. SDS-associated neutropenia increases the risk of infections, including severe sepsis, in individuals diagnosed with the condition with additional neutropenic findings of oral ulcerations and gingivitis possible on physical examination. Quantitative and qualitative B- and T-cell defects may also be seen on laboratory analysis, further increasing infectious risk [5, 6].

Classically SDS is associated with exocrine pancreatic dysfunction causing poor growth, insufficient nutrient absorption, steatorrhea, and voluminous stool output, although these findings might not always be present [3, 6]. Asymptomatic patients may still have fatty infiltration within the pancreas on ultrasound imaging and show abnormal pancreatic enzyme testing (reduced fecal elastase, serum trypsinogen, or serum pancreatic isoamylase levels) [3]. The absence of ringed sideroblasts on marrow assessment assists in differing the condition from Pearson syndrome, which additionally has pancreatic dysfunction [7]. Interestingly, pancreatic abnormalities may spontaneously improve as patients age. Hepatic manifestations have also been reported, including hepatomegaly with transaminitis and hyperbilirubinemia findings consistent with cholestasis [3, 5].

Physical examination or imaging may reveal skeletal abnormalities, including metaphyseal dysplasia and thoracic/pelvic dystrophies [3, 6]. Malformed upper extremities and thumbs may be seen but less frequently than in other congenital bone marrow failure syndromes, such as Fanconi anemia, Diamond-Blackfan anemia, or congenital amegakaryocytic thrombocytopenia [8, 9]. Finally, behavioral and neurocognitive manifestations can be present in those with SDS, with some individuals having developmental delay or neuropsychological disorders [5, 6].

Diagnostic Testing

Bone marrow biopsy assists in verifying suspected marrow hypocellularity and evaluating for promyelocytic arrest (a phenomenon associated with severe congenital neutropenia) as well as ruling out additional causes of cytopenias, including malignancy and autoimmune processes. Marrow analysis should include diagnostic pathology, flow cytometry, karyotyping, and FISH screening for dysplastic genetic rearrangements. Telomere length testing is usually normal, unlike in dyskeratosis congenita, and

chromosomal breakage upon exposure to DEB and MMC is unremarkable, which differs from Fanconi anemia [8, 10]. Idiopathic or acquired causes of aplastic anemia may manifest similarly with bone marrow hypocellularity but typically present with more pronounced acute anemia and thrombocytopenia and a lack of significant gastrointestinal concerns [11, 12]. Testing for deficiency of adenosine deaminase 2 (DADA2) through quantitative enzyme analysis should be considered as such patients may present with bone marrow failure but typically manifest with early-onset stroke and rheumatologic complaints [13]. In children with pancreatic dysfunction and pulmonary symptoms, cystic fibrosis, often through sweat chloride testing, should be ruled out [14]. Serum trypsinogen testing is most accurate in those <3 years of age, while pancreatic isoamylase analysis is superior if 3 years of age or greater [3, 6].

Genetic testing may verify a suspected diagnosis of SDS. The condition is autosomal recessive in nature with most patients (around 90%) having mutations involving the Shwachman-Bodian-Diamond syndrome (SBDS) gene on chromosome 7q.11. However, up to 10% of patients may fail to have abnormal genetic testing [3, 5]. The SBDS protein appears to have important functions in ribosomal biogenesis, mitotic spindle function. and maintenance of the bone marrow stromal environment. It is still fully unclear how deficits in the abovementioned functions of SBDS lead to the varied clinical manifestations of SDS [5]. Recent studies have shown that additional genes involved in ribosomal biogenesis, DNAJC21, ELF1, and SRP54, may also be associated with a SDS phenotype [6, 15–17].

Management/Outcome

Given the significant clinical variability between patients and the rarity of the disorder, the natural history of SDS is overall poorly defined [3, 6]. CBCPDs should be monitored frequently (at least every 3 months) given risk of progression to significant bone marrow failure and possibility of transformation to dysplasia or myeloid neoplasm. Annual bone marrow evaluations should be strongly considered. Patients with clinically significant neutropenia and a history of recurrent infections may benefit from daily or intermittent dosing of granulocyte colony-stimulating factor (G-CSF) [3, 5]. The need for blood product transfusions is rare with SDS unless considerable marrow hypocellularity is present. Pancreatic enzyme replacement and supplementation of fat-soluble vitamins is necessary in those with significant pancreatic dysfunction but may be discontinued in patients who have resolution of their pancreatic phenotype as they age [3, 5].

Hematopoietic stem cell transplantation should be considered in those with SDS-associated severe aplastic anemia and is often necessary in patients who have progressed to myelodysplastic syndrome (MDS) or acute myeloid leukemia (AML) [5]. Outcomes posttransplantation were previously poor in this population but have improved recently with optimization of supportive care, usage of reduced intensity conditioning regimens, and enhanced prevention of graft-versus-host disease measures, although those with AML continue to have a poor prognosis [6, 18, 19].

Case Presentation 2

An 18-year-old female is hospitalized for her second bout of *Candida* sepsis. She additionally had a history of frequent mucocutaneous fungal infections and polyarthritis and was recently diagnosed with type 1 diabetes mellitus. CBCPDs obtained during admission showed neutrophilia and normal absolute lymphocyte counts. Quantitative T- and B-cell subsets showed normal absolute values with proliferation of T cells to antigens and mitogens additionally unremarkable. Immunoglobulin levels, including IgE, were normal. Dihydrorhodamine oxidation (DHR) testing was abnormal but genetic testing for mutated genes associated with chronic granulomatous disease was negative.

Diagnosis/Assessment

Signs and Symptoms

Myeloperoxidase (MPO) is abundantly found within neutrophils and enzymatically reacts with hydrogen peroxide and chloride to produce hypochlorous acid as part of the respiratory burst pathway that facilitates destruction of phagocytosed microbes [20]. MPO deficiency is one of the most common primary phagocytic disorders but very frequently manifests without any clinical symptoms [21]. Those with complete MPO deficiency may present with an elevated risk of disseminated or invasive candidiasis infections. A concurrent diabetes mellitus diagnosis further increases this risk. Human MPO-deficient neutrophils appear to have a reduced ability to kill *Aspergillus fumigatus*, but patients with MPO deficiency seem to have a less significant risk of *Aspergillus* infection than those with chronic granulomatous disease (CGD) [22, 23]. MPO deficiency is further associated with autoimmune clinical symptoms, revealing the enzyme's importance in regulation of inflammation [24, 25].

Diagnostic Testing

Given that oxidation of DHR to rhodamine 123 requires the presence of hydrogen peroxide, a diagnosis of MPO deficiency can yield abnormal DHR results. Such a finding could lead the clinician to assume a diagnosis of chronic granulomatous disease in this patient. However, abnormal DHR testing may also be present with significant glucose-6-phosphate dehydrogenase deficiency [26]. Histochemical staining for MPO within neutrophils and a normal nitroblue tetrazolium assay result may aid in making the correct diagnosis [22, 26]. Additional disorders of neutrophil function, such as the leukocyte adhesion defect disorders, Chediak-Higashi syndrome, and neutrophil-specific granule deficiency, should also be considered within the differential diagnosis [2].

Human immunodeficiency virus (HIV) should be ruled out in those with severe disseminated fungal infections and a history of chronic mucocutaneous *Candida*

infection [27]. Further causes of such infections, including CARD9 and STAT3 deficiencies, should be considered [28, 29]. Even in those found to have a deficiency of MPO, additional causes of the patient's symptomology should be ruled out given the overwhelming majority of MPO-deficient patients are asymptomatic, as incorrectly attributing a patient's clinical manifestations to MPO deficiency may result in overlooking the true cause of their immunodeficiency.

Management/Outcome

Most individuals with MPO deficiency do well and are asymptomatic. Therefore, treatment interventions are often unnecessary. Aggressive management of acute infections in those with a history of invasive organisms should be undertaken. In such individuals, prophylactic antifungal therapy may be helpful. Specific treatments targeting the pathologic cause of the disorder, MPO deficiency, are lacking. Supportive care options include optimal control of serum glucose values in diabetic patients and avoidance when possible, of therapies such as corticosteroids that further exacerbate the risk of fungal infections [22].

Case Presentation 3

An 8-month-old female was hospitalized for her second episode of suppurative lymphadenitis. She also had a history of numerous bouts of pneumonia, as well as persistent stomatitis. During these episodes CBCPDs obtained demonstrated significant neutropenia (absolute neutrophil counts ranging from 0.1×10^9/L to 0.3×10^9/L). Both episodes of lymphadenitis were confirmed to be caused by *Staphylococcus aureus*. She had a normal evaluation of circulating B, T, and NK cells. Her immunoglobulin (IgA, IgM, and IgG) serum concentrations were normal.

Diagnosis/Assessment

Signs and Symptoms

Severe congenital neutropenia (SCN) is characterized by severe neutropenia, with absolute neutrophil counts (ANC) less than 0.2×10^9/L on at least three separate occasions in a one-month time period [1, 30]. Clinically, these patients present with severe bacterial infections early in life. Genetic evaluations have detected both inherited (autosomal dominant, X-linked, or recessive in situations frequently with consanguinity) and spontaneous mutations. Dominant forms are caused by neutrophil elastase (*ELANE*) mutations, while patients with X-linked inheritance will manifest mutations in the Wiskott-Aldrich syndrome (*WAS*) gene. Kostmann

syndrome is the autosomal recessive form of the disorder due to pathogenic variants in *HAX1*, characterized by early stage maturation arrest of myeloid differentiation [31]. The incidence of SCN is approximately one in one million [1]. Phenotypically, these patients may present with mouth sores, gingivitis, otitis media, respiratory infections, cellulitis, and skin abscesses. They also may have mild hepatosplenomegaly on exam [1, 30].

Diagnostic Testing

Frequent CBCPD testing over a month period is required for the diagnosis of SCN and to rule out cyclic patterns to the neutropenia. Cyclic neutropenia is an autosomal dominant congenital granulopoietic disorder caused by mutations in the *ELANE* gene as well [1, 32, 33]. This disorder is characterized by periods of normal neutrophil counts oscillating with severe neutropenia classically within a 21-day cycle with 5–7 days of profound neutropenia. Cyclic neutropenia is estimated to effect approximately 0.6 per 1 million people. At times of the neutropenic nadir, reciprocal monocytosis is observed. Clinically, these patients may experience oral ulcers, stomatitis, or cutaneous infections during periods of neutropenia. Diagnosis is made by obtaining CBCPD approximately two to three times a week, for 2 months [1, 33]. Genetic testing confirming pathogenic variants in *ELANE* can also support the diagnosis. Treatment is focused on utilizing G-CSF to minimize the length of neutropenia per cycle such that cycles are typically reduced to 9–11 day cycles with 1–2 days of profound neutropenia [1].

Severe congenital neutropenia patients may demonstrate peripheral blood eosinophilia and monocytosis on the CBCPDs obtained at the time of diagnosis and also during times of profound neutropenia. They may also demonstrate mild thrombocytosis and anemia of chronic disease [1, 30]. Their bone marrow evaluation will demonstrate arrest of myeloid cell maturation at the promyelocytic stage. This maturation arrest in the bone marrow is not seen in immune or idiopathic neutropenia making this assessment a key component of evaluation. Antineutrophil antibody testing may additionally be positive on testing in those suspected of an immune-mediated neutropenia, although a negative result does not rule out a diagnosis of the condition and a positive result does not rule out a diagnosis of congenital conditions, such as SCN [34].

Management/Outcome

The incidence of fatal disease in SCN has decreased tremendously due to the use of G-CSF. The vast majority (95%) of patients respond well to G-CSF, although dosing can be quite variable between patients [1, 30]. In SCN patients requiring extremely high G-CSF dosing (greater than 8–10 mcg/kg/day) or those with severe infections despite apparently adequate G-CSF dosing, hematopoietic stem cell transplant should be considered [35, 36].

Approximately 10–20% of patients with the initial diagnosis of SCN will go on to develop MDS/AML [1, 37]. The development of MDS or AML does not appear to be related to the use of G-CSF. The development of MDS may present with subtle changes in platelet count or hemoglobin or pathologically flawed bone marrow failing to respond appropriately to increases in the dosing of G-CSF. Due to the risk of MDS/AML, annual bone marrow evaluations and every three-month CBCPD are recommended in patients with SCN [37]. In patients who demonstrate cytogenetic abnormalities or MDS on bone marrow evaluation, stem cell transplantation should be pursued, as traditional chemotherapy agents are not effective in these patients [1, 35, 36].

Case Presentation 4

A 2-year-old boy was admitted to the local hospital for evaluation of fever and abdominal pain. His previous history included frequent respiratory infections, recurrent ear infections, and occasional episodes of high fevers. He was noted to have significant lymphadenopathy and hepatosplenomegaly on exam, in addition to pallor and areas of skin hypopigmentation. His laboratory evaluation demonstrated pancytopenia, coagulopathy, and elevated transaminases. In particular, his lymphocytes were noted to have rod-shaped cytoplasmic organelles with a central linear density. His bone marrow evaluation also demonstrated giant intracytoplasmic inclusions in the myeloid cells, as well as evidence of hemophagocytosis.

Diagnosis/Assessment

Signs and Symptoms

Chediak-Higashi syndrome (CHS) is an autosomal recessive disorder caused by mutations in the lysosomal trafficking regulator (*LYST)*, leading to formation of giant lysosomes or lysosome-related organelles [38]. Phenotypically, CHS is defined by immunodeficiency, oculocutaneous albinism, and hemophagocytic lymphohistiocytosis (HLH). Patients with CHS also have bleeding phenomena as a result of deficient platelet dense bodies [39, 40]. The immunodeficiency is characterized by incomplete degranulation and chemotaxis defects of neutrophils [41].

To aid in clinical diagnosis, these patients typically have partial albinism with hair colors ranging from gray to white, as well as eye pigmentation changes that can result in photosensitivity. The infections associated with CHS are typically pyogenic, especially involving the respiratory tract and skin. Due to the abovementioned platelet defects, they may have easy bruising and mucosal bleeding. Their neurologic manifestations can include weakness, ataxia, sensory defects, and neurodegeneration [40].

Diagnostic Testing

The hallmark of a CHS diagnosis is the presence of large cytoplasmic granules in granulocytes. Platelet aggregation studies reflect on the deficient platelet granules. Evaluation of hair shafts demonstrates large, speckled pigment clumps [39]. The classic bone marrow evaluation demonstrates abnormal granules in all stages of myeloid cell maturation, as well as evidence of hemophagocytic lymphohistiocytosis. Ultimately genetic testing of the *CHS1/LYST* gene can confirm diagnosis [40].

Other similar disorders include Griscelli syndrome, which also results in partial albinism, respiratory tract infections, hypogammaglobulinemia, and variable cellular immunodeficiency [42]. Griscelli syndrome type 2 patients also demonstrate immunodeficiency phenotypes and hemophagocytic lymphohistiocytosis but these patients do not have the giant granules as seen in CHS [43]. Hermansky-Pudlak also results in oculocutaneous albinism and bleeding phenotypes despite a normal platelet count due to the absent platelet dense bodies. This syndrome, however, does not involve neutrophils or natural killer cell dysfunction or result in the accelerated phase seen in CHS [44].

Other conditions with chemotactic defects of neutrophils include hyper-immunoglobulin (hyper-Ig) E syndrome. Patients present with elevated IgE levels (typically >200 IU-mL), as well as recurrent *Staphylococcal* infection of the respiratory tract and lung [45]. Skeletal and dental abnormalities, in addition to dermatitis, are also seen in patients with hyper-IgE [46].

Management/Outcome

Diagnostic workup is important to identify CHS patients prior to development of the "accelerated phase" if possible. Approximately 85% of patients with CHS will enter this phase, which can occur anytime between birth and early childhood. In the accelerated phase, patients demonstrate fever, pancytopenia, hepatosplenomegaly, lymphadenopathy, coagulopathy, and jaundice. This accelerated phase is a nonmalignant infiltrate of lymphohistiocytes across multiple organs [39, 40]. It is typically precipitated by infection, in particular Epstein-Barr virus infections [40, 47]. Unfortunately, development of the accelerated phase can be fatal as result of severe anemia, bleeding complications, or infectious complications. In patients who do survive past the first decade of life (<20%), progressive neurological dysfunction typically leads to further complications [39, 40].

Supportive care including prophylactic antibiotics given immune dysfunction is key in patients with CHS. Treatment of the hematologic and immune defects of CHS is bone marrow transplant; however the neurological outcomes are not improved after transplantation. Higher mortality is associated with patients who enter transplant at the time of the accelerated phase [48, 49].

Case Presentation 5

A 3-year-old girl was referred to pediatric hematology for evaluation of a persistent mild-moderate leukocytosis in the setting of recurrent *Staphylococcal aureus* skin infections. In review of her history, she was hospitalized for the first 2 weeks of life due to omphalitis and a presumed leukemoid reaction. With intravenous antibiotics, her omphalitis resolved, and her leukocytosis improved, although her parents note she had persistent neutrophilia even at discharge. In addition, she has received antibiotics three additional times for extensive skin abscesses. Incision and drainage of each abscess was attempted and yielded positive growth of *Staphylococcal aureus* but no pus. The complete blood count obtained at her 1-year well-child visit showed her white blood cell count (WBC) was 21.8×10^3/uL (absolute neutrophil count (ANC) 17.2×10^3/uL), hemoglobin (Hgb) 12.1 g/dL, and platelets 482×10^3/uL. A dihydrorhodamine flow assay previously demonstrated a normal oxidative burst. Physical exam was notable for poor dentition with diffuse gingival hyperemia and two ulcerative skin lesions on her left leg. Flow cytometric analysis for CD18 expression on neutrophils was decreased and CD11b expression was nearly absent.

Diagnosis/Assessment

Signs and Symptoms

Patients with leukocyte adhesion deficiency 1 (LAD1), particularly those with severe disease, routinely present in the newborn period often with omphalitis, delayed umbilical cord separation and recurrent infections, especially with *Staphylococcus aureus* and gram-negative bacilli. Of note, delayed umbilical cord separation is not reported in other forms of LAD but is frequently found in patients with urachal anomalies [50]. Moreover, delayed umbilical cord separation is not uniformly seen in patients with LAD1. Laboratory evaluation of LAD1 patients demonstrates a neutrophil predominant leukocytosis that may be subtle during periods of wellness but exuberant with infections. The physical exam is notable for the lack of pus formation and poor wound healing. Older patients have near universal periodontal disease secondary to dysregulation of the IL17/23 axis [51] with many patients suffering complete loss of their teeth by late adolescence. Additionally, LAD1 patients are prone to inflammatory skin lesions that are pyoderma gangrenosum-like in appearance although notably on biopsy do not have a neutrophilic infiltrate [52], inflammatory bowel disease [53], and HPV-related warts [54].

The differential diagnosis for patients with suspected LAD1 includes LADII (impaired fucosylation of macromolecules, especially selections) and LADIII (defective beta integrin activation) as well as other related defects that share a common link of a defect in adhesion protein or flawed regulation of adhesion proteins that do not allow phagocytes to migrate from the peripheral blood across the

endothelium and into tissue. Importantly, LADII patients can be clinically distinguished by the high frequency of neurologic manifestations, craniofacial anomalies, and the presence of the rare Bombay erythrocyte phenotype, while LADIII is accompanied by bleeding diatheses.

Diagnostic Testing

Criteria for the diagnosis of LAD1 was published in 1999 and relies heavily on the decreased expression of CD18 on leukocytes (<5% expression) in the appropriate clinical setting of recurrent or deep-seated infections, delayed umbilical cord separation, poor wound healing, and leukocytosis [55]. Updated flow cytometric analysis for LAD1 suggests that both the assessment of CD18 and CD11 (through either CD11a, CD11b, or CD11c) expression on leukocytes be routinely performed as some patients with LAD1 may have expression or residual function of CD18; however, all LAD1 patients have near absent expression of CD11 subunits. Utilizing both CD18 and CD11a measurements increases the sensitivity of the assay and minimizes the risk of delayed or missed diagnoses [56]. Beyond functional testing, genetic sequencing is being increasingly utilized to identify biallelic pathogenic variants in the common beta chain of the beta-2 integrin family (*ITGB2).* Identification of familial pathogenic variants is being increasingly used in prenatal counseling.

LADII patients can be diagnosed through flow cytometric analysis of peripheral blood leukocytes to assess for the absence of properly fucosylated macromolecules, particularly of sialyl Lewis X expression, i.e., CD15a. Confirmation of LADII is obtained through sequencing the gene encoding the GDP-fucose transporter. LADIII patients primarily rely on identification of pathogenic variants in the kindlin-3 gene, although functional assessment of integrin function is also useful.

Management/Outcomes

Optimal management of LAD1 is dependent on the severity of disease. Historically, severity of clinical features and the magnitude of functional defects have been directly related to the degree of CD18 expression [57], although the correlation is imperfect as several cases of LAD1 are now recognized to have normal expression of CD18, albeit nonfunctional.

Patients are generally classified into severe and moderate subgroups with a relative assignment of less than 2% CD18 expression of the β2 integrins considered severe disease. Patients with severe disease have historically been assigned a very poor prognosis with most patients dying in the first decade of life without hematopoietic stem cell transplant (HSCT) [58]. HSCT is the only current curative option for LAD1, although gene therapy has been explored [59]. Patients with mild/moderate disease respond to conservative measures including optimizing oral hygiene, aggressive antimicrobial therapy for infections, and prevention of infection through prophylactic antibiotics as needed and vaccination, particularly immunization against HPV.

For the management of periodontal and/or skin disease related to LAD1, ustekinumab, a monoclonal antibody of the p40 subunit common to IL-12 and IL-23, has been used with favorable results [51]. LADII management relies on the use of antibiotics and fucose supplementation, while LADIII management is focused on management of bleeding complications and consideration of HSCT [60].

Conclusion

Congenital neutropenia and migration defect disorders may manifest with various phenotypes, but all can cause an elevated risk of serious bacterial, fungal, and even viral infections. Early diagnosis is essential to decreasing the morbidity and mortality of these conditions with crucial diagnostic laboratory analyses including functional neutrophil studies, flow cytometry, and direct pathologic evaluation of the bone marrow and peripheral blood. Supportive care often entails usage of antimicrobial prophylaxis, aggressive management of infectious complications, and regular administration of G-CSF. Frequently hematopoietic stem cell transplantation can be curative but is not without risks.

Clinical Pearls/Pitfalls

- Congenital bone marrow failure syndromes may present with a reduction in only one cell line and may not diagnostically become apparent until adulthood.
- Abnormal physical exam findings may be crucial to assisting in the diagnosis of congenital neutropenia and migration defect disorders.
- Significant enzymatic deficiencies in MPO and glucose-6-phosphate dehydrogenase may lead to abnormal DHR testing results, leading to an incorrect presumed diagnosis of chronic granulomatous disease.
- Clinically asymptomatic and well-appearing African-Americans may present with ANC values less than 500/microL which is likely consistent with a diagnosis of Duffy null benign ethnic neutropenia, rather than severe congenital neutropenia.
- Griscelli syndrome may present similarly to Chediak-Higashi syndrome with albinism, immunodeficiency, and hemophagocytosis but Chediak-Higashi syndrome may be differentiated by microscopic examination of patient hair shaft and peripheral blood granulocytes.
- Delayed umbilical cord detachment should raise suspicion for LAD1, but evaluation for urachal anomalies should also be considered.
- HSCT remains the only curative therapy for leukocyte adhesion defects, but IL-23 directed therapy with ustekinumab has shown promise in improving oral and skin manifestations of LAD1.
- Patients with LADII can be easily differentiated from LAD1 patients by the presence of the rare Bombay blood group in patients with LADII.

References

1. Walkovich K, Boxer LA. How to approach neutropenia in childhood. Pediatr Rev. 2013;34:173–84.
2. Amulic B, Cazalet C, Hayes GL, Metzler KD, Zychlinsky A. Neutrophil function: from mechanisms to disease. Annu Rev Immunol. 2012;30:459–89.
3. Myers KC, Bolyard AA, Otto B, Wong TE, Jones AT, Harris RE, et al. Variable clinical presentation of Shwachman-Diamond syndrome: update from the North American Shwachman-Diamond Syndrome Registry. J Pediatr. 2014;164:866–70.
4. Lachowiez C, Meyers G. Identifying unexpected inherited marrow failure syndromes in adults presenting with Aplastic Anemia, MDS or solid tumors. Blood. 2016;128:5071–1.
5. Myers KC, Davies SM, Shimamura A. Clinical and molecular pathophysiology of Shwachman-Diamond syndrome: an update. Hematol Oncol Clin North Am. 2013;27:117–128, ix.
6. Nelson AS, Myers KC. Diagnosis, treatment, and molecular pathology of Shwachman-Diamond Syndrome. Hematol Oncol Clin North Am. 2018;32:687–700.
7. Farruggia P, Di Marco F, Dufour C. Pearson syndrome. Expert Rev Hematol. 2018;11:239–46.
8. Triemstra J, Pham A, Rhodes L, Waggoner DJ, Onel K. A review of Fanconi anemia for the practicing pediatrician. Pediatr Ann. 2015;44:444–5, 448, 450 passim.
9. Dokal I, Vulliamy T. Inherited aplastic anaemias/bone marrow failure syndromes. Blood Rev. 2008;22:141–53.
10. Savage SA. Human telomeres and telomere biology disorders. Prog Mol Biol Transl Sci. 2014;125:41–66.
11. Young NS. Aplastic Anemia. N Engl J Med. 2018;379:1643–56.
12. Kallen ME, Dulau-Florea A, Wang W, Calvo KR. Acquired and germline predisposition to bone marrow failure: diagnostic features and clinical implications. Semin Hematol. 2019;56:69–82.
13. Michniacki TF, Hannibal M, Ross CW, Frame DG, DuVall AS, Khoriaty R, et al. Hematologic manifestations of deficiency of adenosine deaminase 2 (DADA2) and response to tumor necrosis factor inhibition in DADA2-associated bone marrow failure. J Clin Immunol. 2018;38:166–73.
14. VanDevanter DR, Kahle JS, O'Sullivan AK, Sikirica S, Hodgkins PS. Cystic fibrosis in young children: a review of disease manifestation, progression, and response to early treatment. J Cyst Fibros. 2016;15:147–57.
15. Dhanraj S, Matveev A, Li H, Lauhasurayotin S, Jardine L, Cada M, et al. Biallelic mutations in DNAJC21 cause Shwachman-Diamond syndrome. Blood. 2017;129:1557–62.
16. Carapito R, Konantz M, Paillard C, Miao Z, Pichot A, Leduc MS, et al. Mutations in signal recognition particle SRP54 cause syndromic neutropenia with Shwachman-Diamond-like features. J Clin Invest. 2017;127:4090–103.
17. Bezzerri V, Cipolli M. Shwachman-diamond syndrome: molecular mechanisms and current perspectives. Mol Diagn Ther. Epub ahead of print 9 November 2018. https://doi.org/10.1007/s40291-018-0368-2.
18. Cesaro S, Oneto R, Messina C, Gibson BE, Buzyn A, Steward C, et al. Haematopoietic stem cell transplantation for Shwachman-Diamond disease: a study from the European group for blood and marrow transplantation. Br J Haematol. 2005;131:231–6.
19. Dalle J-H, Peffault de Latour R. Allogeneic hematopoietic stem cell transplantation for inherited bone marrow failure syndromes. Int J Hematol. 2016;103:373–9.
20. Aratani Y. Myeloperoxidase: its role for host defense, inflammation, and neutrophil function. Arch Biochem Biophys. 2018;640:47–52.
21. Parry MF, Root RK, Metcalf JA, Delaney KK, Kaplow LS, Richar WJ. Myeloperoxidase deficiency: prevalence and clinical significance. Ann Intern Med. 1981;95:293–301.
22. Ren R, Fedoriw Y. The molecular pathophysiology, differential diagnosis, and treatment of MPO deficiency. J Clin Exp Pathol;2. Epub ahead of print 2012. https://doi.org/10.4172/2161-0681.1000109.

23. Falcone EL, Holland SM. Invasive fungal infection in chronic granulomatous disease: insights into pathogenesis and management. Curr Opin Infect Dis. 2012;25:658–69.
24. Neutrophil-mediated regulation of innate and adaptive immunity: the role of myeloperoxidase. https://www.hindawi.com/journals/jir/2016/2349817/. Accessed 15 Oct 2019.
25. Kutter D, Devaquet P, Vanderstocken G, Paulus JM, Marchal V, Gothot A. Consequences of total and subtotal myeloperoxidase deficiency: risk or benefit ? Acta Haematol. 2000;104:10–5.
26. Milligan KL, Mann D, Rump A, Anderson VL, Hsu AP, Kuhns DB, et al. Complete myeloperoxidase deficiency: beware the 'false-positive' dihydrorhodamine oxidation. J Pediatr. 2016;176:204–6.
27. Blood GA. Human immunodeficiency virus (HIV). Transfus Med Hemother. 2016;43:203–22.
28. Corvilain E, Casanova J-L, Puel A. Inherited CARD9 deficiency: invasive disease caused by Ascomycete Fungi in previously healthy children and adults. J Clin Immunol. 2018;38:656–93.
29. Farmand S, Kremer B, Häffner M, et al. Eosinophilia and reduced STAT3 signalling affect neutrophil cell death in autosomal-dominant Hyper-IgE syndrome. Eur J Immunol. Epub ahead of print 13 October 2018. https://doi.org/10.1002/eji.201847650.
30. Welte K, Zeidler C, Dale DC. Severe congenital neutropenia. Semin Hematol. 2006;43:189–95.
31. Boztug K, Klein C. Genetics and pathophysiology of severe congenital neutropenia syndromes unrelated to neutrophil elastase. Hematol Oncol Clin North Am. 2013;27:43–60, vii.
32. Horwitz MS, Corey SJ, Grimes HL, Tidwell T. ELANE mutations in cyclic and severe congenital neutropenia: genetics and pathophysiology. Hematol Oncol Clin North Am. 2013;27:19–41, vii.
33. Lange RD, Jones JB. Cyclic neutropenia. Review of clinical manifestations and management. Am J Pediatr Hematol Oncol. 1981;3:363–7.
34. Newburger PE, Dale DC. Evaluation and management of patients with isolated neutropenia. Semin Hematol. 2013;50:198–206.
35. Carlsson G, Winiarski J, Ljungman P, Ringdén O, Mattsson J, Nordenskjöld M, et al. Hematopoietic stem cell transplantation in severe congenital neutropenia. Pediatr Blood Cancer. 2011;56:444–51.
36. Fioredda F, Iacobelli S, van Biezen A, Gaspar B, Ancliff P, Donadieu J, et al. Stem cell transplantation in severe congenital neutropenia: an analysis from the European Society for Blood and Marrow Transplantation. Blood. 2015;126:1885–92; quiz 1970.
37. Skokowa J, Dale DC, Touw IP, Zeidler C, Welte K. Severe congenital neutropenias. Nat Rev Dis Primers. 2017;3:17032.
38. Gil-Krzewska A, Saeed MB, Oszmiana A, Fischer ER, Lagrue K, Gahl WA, et al. An actin cytoskeletal barrier inhibits lytic granule release from natural killer cells in patients with Chediak-Higashi syndrome. J Allergy Clin Immunol. 2018;142:914–927.e6.
39. Introne W, Boissy RE, Gahl WA. Clinical, molecular, and cell biological aspects of Chediak-Higashi syndrome. Mol Genet Metab. 1999;68:283–303.
40. Kaplan J, De Domenico I, Ward DM. Chediak-Higashi syndrome. Curr Opin Hematol. 2008;15:22–9.
41. Clark RA, Kimball HR. Defective granulocyte chemotaxis in the Chediak-Higashi syndrome. J Clin Invest. 1971;50:2645–52.
42. Mancini AJ, Chan LS, Paller AS. Partial albinism with immunodeficiency: Griscelli syndrome: report of a case and review of the literature. J Am Acad Dermatol. 1998;38:295–300.
43. Minocha P, Choudhary R, Agrawal A, Sitaraman S. Griscelli syndrome subtype 2 with hemophagocytic lympho-histiocytosis: a case report and review of literature. Intractable Rare Dis Res. 2017;6:76–9.
44. El-Chemaly S, Young LR. Hermansky-Pudlak Syndrome. Clin Chest Med. 2016;37:505–11.
45. Dinauer MC. Disorders of neutrophil function: an overview. Methods Mol Biol. 2007;412:489–504.
46. Al-Shaikhly T, Ochs HD. Hyper IgE syndromes: clinical and molecular characteristics. Immunol Cell Biol. 2019;97:368–79.

47. Merino F, Henle W, Ramírez-Duque P. Chronic active Epstein-Barr virus infection in patients with Chediak-Higashi syndrome. J Clin Immunol. 1986;6:299–305.
48. Thakor A, Geng B, Liebhaber M, Moore T, Wang K, Roberts RL. Successful stem cell transplantation in Chediak-Higashi syndrome. J Allergy Clin Immunol Pract. 2015;3:271–2.
49. Eapen M, DeLaat CA, Baker KS, Cairo MS, Cowan MJ, Kurtzberg J, et al. Hematopoietic cell transplantation for Chediak-Higashi syndrome. Bone Marrow Transplant. 2007;39:411–5.
50. Razvi S, Murphy R, Shlasko E, Cunningham-Rundles C. Delayed separation of the umbilical cord attributable to urachal anomalies. Pediatrics. 2001;108:493–4.
51. Moutsopoulos NM, Zerbe CS, Wild T, Dutzan N, Brenchley L, DiPasquale G, et al. Interleukin-12 and interleukin-23 blockade in leukocyte adhesion deficiency type 1. N Engl J Med. 2017;376:1141–6.
52. Simpson AM, Chen K, Bohnsack JF, Lamont MN, Siddiqi FA, Gociman B. Pyoderma gangrenosum-like wounds in leukocyte adhesion deficiency: case report and review of literature. Plast Reconstr Surg Glob Open. 2018;6:e1886.
53. Uzel G, Kleiner DE, Kuhns DB, Holland SM. Dysfunctional LAD-1 neutrophils and colitis. Gastroenterology. 2001;121:958–64.
54. Leiding JW, Holland SM. Warts and all: human papillomavirus in primary immunodeficiencies. J Allergy Clin Immunol. 2012;130:1030–48.
55. Conley ME, Notarangelo LD, Etzioni A. Diagnostic criteria for primary immunodeficiencies. Representing PAGID (Pan-American Group for Immunodeficiency) and ESID (European Society for Immunodeficiencies). Clin Immunol. 1999;93:190–7.
56. Levy-Mendelovich S, Rechavi E, Abuzaitoun O, Vernitsky H, Simon AJ, Lev A, et al. Highlighting the problematic reliance on CD18 for diagnosing leukocyte adhesion deficiency type 1. Immunol Res. 2016;64:476–82.
57. Anderson DC, Schmalsteig FC, Finegold MJ, Hughes BJ, Rothlein R, Miller LJ, et al. The severe and moderate phenotypes of heritable Mac-1, LFA-1 deficiency: their quantitative definition and relation to leukocyte dysfunction and clinical features. J Infect Dis. 1985;152:668–89.
58. Etzioni A. Genetic etiologies of leukocyte adhesion defects. Curr Opin Immunol. 2009;21:481–6.
59. Qasim W, Cavazzana-Calvo M, Davies EG, Davis J, Duval M, Eames G, et al. Allogeneic hematopoietic stem-cell transplantation for leukocyte adhesion deficiency. Pediatrics. 2009;123:836–40.
60. Stepensky PY, Wolach B, Gavrieli R, Rousso S, Ben Ami T, Goldman V, et al. Leukocyte adhesion deficiency type III: clinical features and treatment with stem cell transplantation. J Pediatr Hematol Oncol. 2015;37:264–8.