Genetics of severe congenital neutropenia as a gateway to personalized therapy

Jean Donadieu[1] and Christine Bellanné-Chantelot[2]

[1]Centre de Référence des Neutropénies Chroniques, Registre National des Neutropénies Congénitales, Service d'Hémato-oncologie Pédiatrique, Hôpital Armand-Trousseau, Assistance Publique Hôpitaux de Paris, Paris, France; and [2]Sorbonne Université, Département de Génétique Médicale, Hôpital Pitié–Salpêtrière, Assistance Publique Hôpitaux de Paris, Paris, France

Severe congenital neutropenias (SCNs) are rare diseases, and to date about 30 subtypes have been described according to their genetic causes. Standard care aims to prevent infections and limit the risk of leukemic transformation; however, several subtypes may have additional organ dysfunction(s), requiring specialized care. Granulocyte colony-stimulating factor and hematopoietic stem cell transplantation are now the bedrock of standard care. Better understanding of SCN mechanisms now offers the possibility of adapted therapy for some entities. An inhibitor of sodium glucose cotransporter, an antidiabetic drug, may attenuate glycogen storage disease type Ib and glucose-6-phosphatase catalytic subunit 3 neutropenias by clearing 1,5-anhydroglucitol, the precursor of the phosphate ester responsible for these SCNs. Chemokine receptor CXCR4 inhibitors contribute to reversing the leukocyte defect in warts, hypoglobulinemia, infections, and myelokathexis syndrome. All these new approaches use oral drugs, which notably improve quality of life. Additionally, improved research into clonal evolution has highlighted some ways to potentially prevent leukemia, such as stimulating somatic genetic rescue, a physiological process that might limit the risk of leukemic transformation.

LEARNING OBJECTIVES
- Identify in the clinical presentation of a patient with neutropenia the key features suggesting a possible genetic cause
- Realize that in G6PC3 neutropenia and in GSDIB, gliflozine may be considered an alternative to standard care with GCSF and/or HSCT
- Learn that in CXCR4 WHIM syndrome, CXCR4 inhibitors offer a therapeutic approach

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Introduction

Interest in severe congenital neutropenias (SCNs) was heightened by the availability of a new therapeutic agent, granulocyte colony-stimulating factor (GCSF), at the beginning of the 1990s.[1] It is an understatement to say that GCSF dramatically transformed hematology medical practice. Even though GCSF is mainly indicated for chemotherapy-induced neutropenia, it has also focused light on the extremely rare group of diseases called SCNs. GCSF marketing was associated with a recommendation by health authorities to develop patient registries, primarily to assess its potential leukemic risk. Incidentally, registry studies have provided valuable evidence not only to demonstrate the efficacy of GCSF but also to show that high doses induce clonal hematopoiesis, leading to myelodysplasia or acute leukemia.[2,3] We now know that this risk can be limited by hematopoietic stem cell transplantation (HSCT) early

during SCN evolution.[4] But even if HSCT can control the risk of myelodysplastic syndrome and even if GCSF remains effective throughout a patient's lifetime, daily GCSF use engenders quality-of-life limitations, explaining poor compliance and thus, only partial efficacy, with sequelae, persistent symptoms, and sometimes life-threatening infections.[5] Such limitations are strong motivations to find new therapeutic approaches. And genetic analyses have opened the gate to potential novel agents.

SCN genetics: accurate and quick diagnoses for better classification and more therapeutic options

Since 1993, while registries were being progressively created, genetic technologies have advanced dramatically, and about 30 distinct SCN genetic entities have been described (Table 1).[6] Above all, genetics provides a reliable parameter to classify a patient's disease. Initially, determining

Table 1. List of genes and phenotypes associated with GNs (2022)

Neutropenia subgroup	Gene, disease name	OMIM code	Main hematological features	Extrahematopoietic features	Inheritance, gene localization	Normal function of the gene-encoded protein
Congenital neutropenia usually without extrahematopoietic manifestation	*ELANE*, severe congenital or cyclic neutropenia	202700 162800	Severe and permanent neutropenia, maturation arrest, intermittent/cyclic with variable BM features	No	Dominant, 19q13.3	Protease activity Antagonism with α-1 antitrypsin
	CSF3R	202700	Permanent maturation arrest, unresponsive to GCSF	No	Recessive, 1p35-p34.3	Transmembrane GCSF receptor/ intracellular signaling
	WAS	301000	Severe permanent neutropenia, maturation arrest, monocytopenia	No	X-linked, Xp11.4-p11.21	Cytoskeleton homeostasis
	CXCR2		Severe permanent neutropenia, no maturation arrest	No	Recessive, 2q35	Chemokine receptor (CXCL12)
	SEC61A1		No maturation arrest	No	Dominant, 3q21.3	The translocon complex of proteins transports nascent polypeptides with a targeting signal sequence into the interior space of the ER
	SRP54		Maturation arrest	Some mutations are associated with exocrine pancreas deficiency, mental retardation	Dominant, 14q13.2	SRP is a protein-RNA complex that recognizes and targets proteins specific to the ER. SRP54 and SRP68 proteins are
	SRP68		Maturation arrest	No	Recessive, 17q25.1	components of that complex.
Congenital neutropenia with frequent extrahematopoietic manifestations, including innate-immunity deficiencies	*SBDS*, Shwachman–Diamond syndrome	260400	Mild neutropenia, dysgranulopoiesis, mild dysmegakaryopoiesis	Exocrine pancreas deficiency, metaphyseal dysplasia, mental retardation, cardiomyopathy	Recessive, 7q11.22	Ribosomal protein that regulates RNA expression
	EFL1, syndrome	260400	Mild neutropenia, dyserythropoiesis	Exocrine pancreas deficiency, metaphyseal dysplasia, mental retardation	Recessive, 15q25.2	Ribosomal protein that regulates RNA expression
	GATA2, syndrome	614038 614172	Mild neutropenia, monocytopenia, macrocytosis	Lymphedema, deafness, mycobacterial, HPV infections	Dominant, 3q21.3	Transcription factor
	G6PC3, severe congenital neutropenia	202700	Maturation arrest	Skin-prominent superficial venous network, atrial defect, uropathy	Recessive, 17q21	1,5AG6P hydrolysis
	SLC37A4, Glycogen Storage Disease Type Ib	232220	No maturation arrest	Hypoglycemia, fasting hyperlactacidemia, and glycogen overload in the liver	Recessive, 11q23.3	glucose-6-phosphate or 1,5AG6P transport to the ER
	TAFAZIN, Barth disease	302060	No maturation arrest	Hypertrophic cardiomyopathy, myopathic syndrome, 3-methylglutaconic aciduria	X-linked, Xq28	Tafazzin, phospholipid membrane homeostasis

Table 1. (continued)

Neutropenia subgroup	Gene, disease name	OMIM code	Main hematological features	Extrahematopoietic features	Inheritance, gene localization	Normal function of the gene-encoded protein
	CXCR4, WHIM syndrome	193670	No maturation arrest, myelokathexis	Lymphopenia, thrombocytopenia, cardiopathy type tetralogy of Fallot	Dominant, 2q21	Chemokine receptor (ligand CXCL12)
	JAGN1, severe congenital neutropenia	616022	Variable	Bone abnormalities, exocrine pancreatic deficiency	Recessive, 3p25.3	ER protein
	VPS13B, Cohen syndrome	216550	No maturation arrest	Psychomotor retardation, clumsiness, microcephaly, characteristic facial features, hypotonia, joint laxity, progressive retinochoroidal dystrophy, myopia	Recessive, 8q22-q23	Sorting and transporting proteins in the ER
	GFI1, severe congenital neutropenia	202700	Permanent/severe or mild neutropenia, sometimes maturation arrest	Internal ear (in mouse model), lymphopenia	Dominant, p22	Transcription factor regulating oncoproteins
	HAX1, Kostmann's disease	202700	Maturation arrest	Central nervous system: mental retardation or seizures	Recessive, 1q21.3	Antiapoptotic protein located in mitochondria and cytosol
	AP3B1, Hermansky-Pudlak syndrome type 2	608233	No maturation arrest	Albinism	Recessive, 5q14.1	Cargo protein or ER trafficking
	LAMTOR2, chronic neutropenia	610389	No maturation arrest	Albinism	Recessive, 1q21	Lysosome packaging
	USB1, poikiloderma with Clericuzio-type neutropenia	604173	No maturation arrest, minor dysgranulopoietic features	Poikiloderma	Recessive, 16q21	Unelucidated
	VPS45, severe congenital neutropenia	615285	Maturation arrest/myelofibrosis	Nephromegaly, hepatosplenomegaly, mental retardation	Recessive, 1q21.2	Role in segregation of intracellular molecules into distinct organelles
	TCIRG1, severe congenital neutropenia	202700	Variable	Skin angiomatosis	Dominant, 11q13.2	Unelucidated
	EIF2AK3, Wolcott-Rallison syndrome	604032	Maturation arrest	Insulin-dependent neonatal diabetes	Recessive, 2p11.2	ER stress
	CLPB syndrome	616254	Maturation arrest	Mental retardation, congenital cataracts, 3-methylglutaconic aciduria	Recessive/Dominant, 11q13.4	Unelucidated
	STK4 (MST1) syndrome	614868	Intermittent or autoimmune neutropenia	Atrial defect	Recessive, 20q13	Serine/threonine protein kinase
	SMARCD2		Dysplastic syndrome, no granules in neutrophils	Chronic diarrhea, bone abnormalities, low-set ears	Recessive, 17q23	Unelucidated

Table 1. (continued)

Neutropenia subgroup	Gene, disease name	OMIM code	Main hematological features	Extrahematopoietic features	Inheritance, gene localization	Normal function of the gene-encoded protein
	SASH3		Maturation arrest	Autoimmune disease, viral infection(s)	X-linked, Xq26	Unelucidated
	CARD11		Maturation arrest, hyperlymphocytosis	Skin disorders	Dominant, 7p22.2	Unelucidated
	Mitochondrial DNA, Pearson syndrome	557000	Vacuolization of BM precursors, Perls staining reveals ring sideroblasts	Exocrine pancreatic insufficiency, sideroblastic anemia and thrombocytopenia usually later in life, delayed development with Kearns-Sayre syndrome; elevated lactate/pyruvate ratio	Complex inheritance	Deletion(s) in mitochondrial DNA

AP3B1, adaptor-related protein complex-3 subunit-β1; *CARD11*, caspase recruitment domain family member-11; *CLPB*, caseinolytic peptidase-B protein homolog; *EFL1*, elongation factor-like GTPase-1; *EIF2AK3*, eukaryotic translation initiation factor-2 alpha kinase-1; *GATA2*, transcription factor-2 binding to the DNA sequence "GATA"; *GFI*, gold furrows of hull 1, GATA1 transcription factor; *HAX1*, encodes hematopoietic cell-specific Lyn substrate-1 protein; *JAGN1*, Jagunal homolog-1; *LAMTOR2*, late endosomal/lysosomal adaptor, MAPK and MTOR activator-2; *SASH3*, sterile alpha motif (SAM) and Src homology-3 (SH3) domain containing-3; *SEC61A1*, SEC61 translocon-subunit alpha-1; *SLC3*, solute carrier family-3; *SMARCD2*, SWItch/sucrose nonfermentable (SWI/SNF)-related, matrix associated, actin dependent regulator of chromatin, subfamily D, member 2; *SRP*, signal-recognition particle; *STK4 (MST1)*, serine/threonine kinase-4 (macrophage stimulating-1; *TAZ*, tafazzin; *TCIRG1*, T-cell immune regulator 1, ATPase H+ transporting V0 subunit a3; *USB1*, U6 snRNA biogenesis phosphodiesterase-1; *VPS13B*, vacuolar protein sorting–associated protein 13B; *WAS*, Wiskott-Aldrich syndrome.

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a culprit gene derived from the laborious collection of biological material from phenotypically well-defined patients. Elastase, neutrophil expressed (*ELANE*) was the first gene identified,[7] followed by the Shwachman-Bodian-Diamond syndrome (*SBDS*) gene.[8] But once the medical community had access to more powerful genetic tools, such as targeted next generation sequencing and whole-exome sequencing, it became possible to study simultaneously the same panel of genes involved in this group of diseases regardless of the patient's phenotype. This powerful approach broke the lines separating classic phenotypes.[9] Some entities may share symptoms attributable to different genes, while very different clinical manifestations can be caused by the same genetic mutation(s). As a consequence, the term "SCN" is not completely accurate, and perhaps calling these entities genetic neutropenias would be more appropriate. Notably, genetic neutropenias (GNs) are not always severe, and the adjective "severe" is not justified by many patients' clinical status. Now, obviously, genetic expertise is the key to diagnosis. Although blood tests have become routine examinations, physicians must justify genetic testing, as it is widely accepted for only a restricted number of settings.

Indeed, population study results have shown that neutropenia is very common. Depending on geographic origin, neutropenia is detected in 1% (Caucasians) to 10% (those with African origins) of people, while GN prevalence is less than 1 in 10 000.[10,11] This situation explains why a clinical algorithm makes it possible to restrict genetic testing to suspected cases in an attempt to limit patient anxiety and avoid waste of medical resources. One algorithm, very easy to use, was reported for pediatric neutropenia, and its score can even be computed during a short phone call to specify the patient's characteristics. A rapid decision of whether or not to launch genetic research can then be made by calculating the score of the individual algorithm elements (Figure 1).[12]

From genetic diagnosis to therapeutic approaches: 3 situations

But once a genetic diagnosis is fully determined, how can this information change the natural history of a patient's disease? Here we develop 3 situations (4 diseases) in which genetic information, based on a large amount of basic scientific research, deciphers the molecular consequences of a genetic defect and leads to a specific, personalized therapeutic approach not yet at the same stage of development.

Inhibitor of sodium glucose cotransporter to treat G6PC3 and GSDIB SCNs

Glucose-6-phosphatase catalytic subunit 3 (G6PC3) and glycogen storage disease type Ib (GSDIB) neutropenias are caused by a defect in the glucose-6-phosphatase (G6Pase) enzyme.[13,14] For years, the mechanism of neutropenia in those entities remained unclear. The main enigma was the lack of a direct role of G6Pase, which is composed of several units (ie, the endoplasmic reticulum (ER)/cytosol transporter in GSDIB and the catalytic subunit 3 in G6PC3 GNs) in the neutrophils' metabolic machinery. Even if a genetic defect was clearly documented, a key was missing to understand these 2 diseases because G6Pase did not appear to be responsible for a direct defect in the neutrophil-energy pathway. This mystery was solved when the role of 1,5-anhydroglucitol phosphate was identified, and the 2 defect proteins involved in GSDIB and in G6PC3 GN were shown to play an important

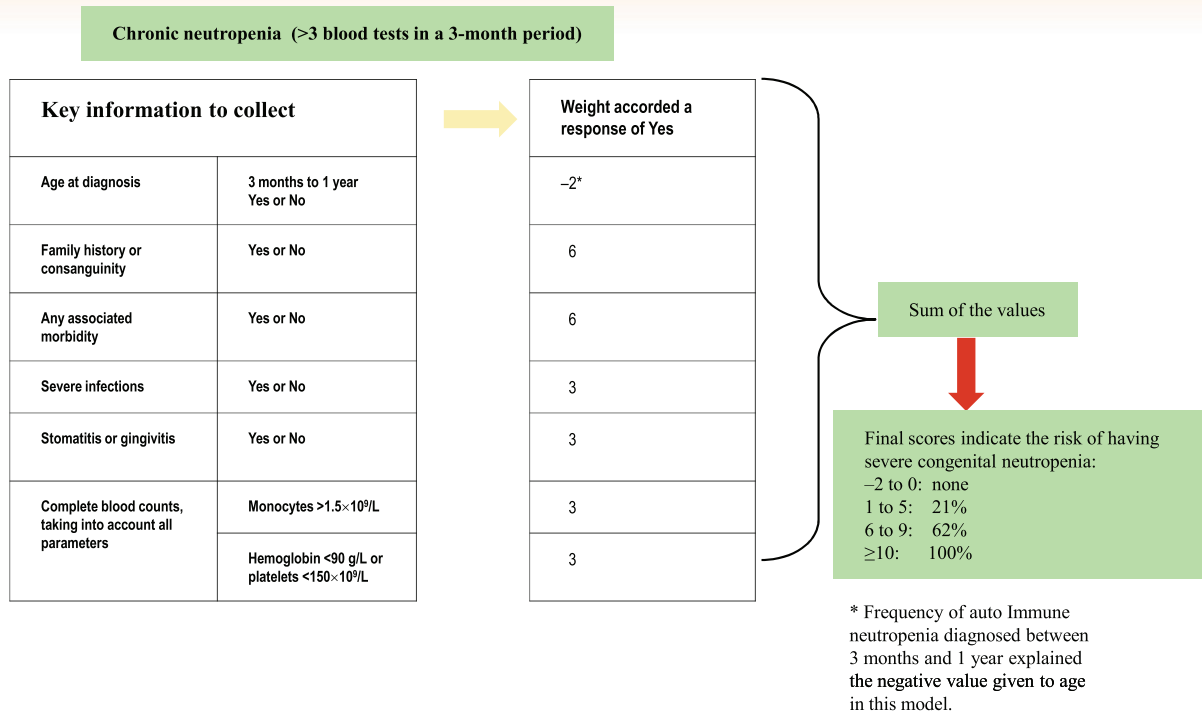## When genetic testing is important to evaluate chronic neutropenia: a quick algorithm

Chronic neutropenia (>3 blood tests in a 3-month period)

Key information to collect		Weight accorded a response of Yes
Age at diagnosis	3 months to 1 year Yes or No	−2*
Family history or consanguinity	Yes or No	6
Any associated morbidity	Yes or No	6
Severe infections	Yes or No	3
Stomatitis or gingivitis	Yes or No	3
Complete blood counts, taking into account all parameters	Monocytes >1.5×10^9/L	3
	Hemoglobin <90 g/L or platelets <150×10^9/L	3

Sum of the values

Final scores indicate the risk of having severe congenital neutropenia:
−2 to 0: none
1 to 5: 21%
6 to 9: 62%
≥10: 100%

* Frequency of auto Immune neutropenia diagnosed between 3 months and 1 year explained the negative value given to age in this model.

Figure 1. A quick algorithm to identify potential genetic neutropenia.

role in the removal of a toxic metabolite, 1,5-anhydroglucitol-6-phosphate (1,5AG6P), in neutrophils.[15] 1,5AG6P is produced by phosphorylation of 1,5-anhydroglucitol (1,5AG), a glucose analogue derived from food and normally present in the plasma. As 1,5AG6P is an inhibitor of hexokinase, the enzyme that allows glucose metabolism by glycolysis, it has to be destroyed as soon as it is formed. This is the function of G6PC3, an enzyme related to the classical G6Pase but that has only little G6Pase activity. The transport of 1,5AG6P from the cytosol to the lumen of the ER is carried out by the glucose-6-phosphate transporter encoded by *SLC37A4*, the gene mutated in GSDIB. The cytosolic accumulation of 1,5AG6P due to mutations in G6PC3 or in SLC37A4 leads to the inhibition of glycolysis. This mechanism explains neutrophil dysfunction and apoptosis in GSDIB and G6PC3-deficiency GNs.[15] That finding had potential clinical impact, as it opened the way to reverse the clinical manifestations of neutropenia and neutrophil dysfunction in GSDIB and G6PC3-deficient patients. Indeed, prescribed off-label, inhibitors of sodium glucose cotransporter (eg, empagliflozin or dapagliflozin) that are routinely used clinically as antidiabetic drugs to treat type 2 diabetes inhibit renal glucose uptake, cause glycosuria, block renal 1,5AG reabsorption, and lower plasma glucose (to the renal glucose threshold) and 1,5AG concentrations. Empagliflozin was also shown to decrease intracellular 1,5AG6P in G6PC3-deficient mice and normalize their absolute neutrophil counts.[15] That finding has now been confirmed in humans[16]: the clinical use of inhibitors of sodium-glucose linked transporter type 2 did not interfere with patients' glucose levels (which, in this context, are low or normal) but increased

1,5AG clearance and allowed myeloid maturation and function. For the first 4 reported cases,[16] all with GSDIB, empagliflozin, during short-term follow-up of less than 1 year, demonstrated both diminished 1,5AG and that GCSF support could be dramatically lowered or even withdrawn while infectious events were controlled.[17] Additional cases with longer follow-up have confirmed those observations and offer a cheaper approach and much more efficient management of both GSDIB and G6PC3 GNs.[18,19]

CXCR4 inhibitors and WHIM syndrome

Warts, hypoglobulinemia, infections, and myelokathexis (WHIM) syndrome is a very rare GN caused by a mutation of the CXCR4 chemokine receptor. In 1964, Zuelzer and Krill described an exceptional congenital neutropenia associated with bone marrow (BM) hyperplasia of mature neutrophils: myelokathexis.[20] In 1990, Wetzler proposed the acronym WHIM, reflecting the manifestations of human papillomavirus (HPV)-induced warts, hypogammaglobulinemia, and bacterial infections together with myelokathexis. WHIM syndrome is characterized by heterogeneous disease manifestations that include severe infectious episodes, HPV-associated warts, panleukopenia, and hypogammaglobulinemia.[21,22] Its clinical onset and complications are more variable than originally suspected. Neutropenia is associated with lymphopenia and monocytopenia, which are almost always observed in patients suffering from this disorder; HPV infections are extremely common. WHIM syndrome is a potentially fatal disease mostly because of immune-system alterations and HPV infections.[21,22]

Genetic analyses of WHIM syndrome patients identified inherited heterozygous autosomal dominant mutations in the *CXCR4* gene encoding the receptor of the CXC α-chemokine (CXCL12),[23] which regulates hematopoiesis and the peripheral trafficking of neutrophil and lymphocyte subsets. CXCR4 was first studied for its role as a human immunodeficiency virus coreceptor. The functional consequences of the variants are dramatic, as they confer a CXCR4 gain of function responsible for the WHIM syndrome–associated panleukopenia. Current WHIM syndrome therapies are intravenous immunoglobulin and GCSF injections and antibiotic prophylaxis.[24] Those treatments may limit the severe infections affecting patients, but they fail to control potentially lethal HPV or mycobacterial infections. Since the genetic findings showed CXCR4 gain of function, agents inhibiting CXCR4 function appear promising. Those treatments, initially developed to control HIV infection, were obviously abandoned but were later shown to possibly act as a stem cell mobilizer.

Indeed, the AMD3100 compound was patented under the name Plerixafor to mobilize autograft stem cells. Successfully tested on a very limited number of WHIM syndrome patients, that drug efficiently mobilized granulocytes, lymphocytes, and monocytes,[25,26] and even controlled HPV infections, in 3 patients.[27] Plerixafor development was not pursued, but another compound, mavorixafor, was developed by another company. After phase 1 and phase 2 studies,[28] that latter molecule is now being evaluated in a phase 3 double-blind randomized trial; its results are expected by the end of 2022. The findings of the phase 2 study on mavorixafor showed that neutrophil and, in general, all leukocyte defects were corrected by this oral drug, with the added advantage of limiting HPV infections.[28] Notably, because CXCR4/CXCL12 interacts strongly with the GCSF-3 receptor (CSF3R) axis early during myelopoiesis, inhibiting CXCR4 enables neutrophil mobilization via a pathway other than GCSF. And now an oral compound may become available to avoid neutropenia in other chronic subtypes as well as post chemotherapy.

Somatic genetic rescue: clonal hematopoiesis is not always bad news

In addition to a high risk of infections, almost all GNs carry an elevated risk of leukemic transformation, for which the percentage depends on the gene involved. The trajectory from the naive germ line at birth to leukemia is a multistep mutational process. The high frequency of clonal evolution can be viewed as premature aging of clonal hematopoiesis, but clonal architecture is strongly dependent on the germline variant on which it develops: clonal hematopoiesis differs markedly in *ELANE* neutropenia, Shwachman-Diamond syndrome (SDS), and GATA2 syndrome.[29-31]

SDS, a recessive multisystem disorder characterized by exocrine pancreas deficiency, mild neutropenia, and various other organ dysfunctions, is caused by compound heterozygous mutations of the *SBDS* gene and offers an initial insight into the possible role of somatic genetic rescue to prevent leukemic transformation.[32] The SBDS protein is an essential cofactor for elongation factor-1. Biallelic *SBDS* mutations impair the release of antiassociation, eukaryotic translation-initiation factor-6 (EIF6) from the 60S ribosomal unit. The cell tries to escape that blockage in different ways. One quite deleterious method that directly involves the leukemic outcome is the overstimulation of the TP53 pathway,[33] induced by ribosomal stress.[34] The involvement of

TP53 mutations in SDS has been documented several times during SDS patients' lifetimes. During the "chronic phase," the TP53 clone frequently has a low variant-allele frequency, and at myelodysplastic syndrome onset, the *TP53* variant-allele frequency is high, typically with biallelic variants.[30,35] But it is not the sole mutational event observed during the course of SDS. The second frequent molecular event concerns the *EIF6* variant.[30] The occurrence of such a molecular event was suggested by a common cytogenetic finding in SDS patients' BM: the del20q clone, which harbors *EIF6*. But other molecular events (eg, point mutations, reciprocal chromosomal translocation) limit EIF6 concentration or EIF6 binding to the 60S subunit and reverse the ribosome-assembly and protein-synthesis defects.[36] Such mechanisms suggest that EIF6 inhibition might constitute a valuable strategy to compensate for the *SBDS* defect and represents a promising therapeutic strategy for SDS.[36,37]

CLINICAL CASE

The patient is an Algerian-born male, issued from a consanguineous family, who has previously been reported in a case series (as patient 5643).[38] He was first seen in our unit at the age of 7 years, when he had already experienced about 12 distinct severe infectious episodes, including bacterial meningitis and 3 types of pneumonitis since birth in addition to recurrent oral infections. He failed to thrive and had chronic diarrhea. Morphologically, he had a narrow thorax and thin skin with a prominent superficial venous network on the limbs and the abdomen. Echocardiography detected aortic insufficiency, without hemodynamic consequences. Initial blood tests showed profound neutropenia (absolute neutrophil count, 240/mm³), with mild anemia (hemoglobin, 9.8g/dL) and a normal platelet count. His first BM cytology did not reveal any significant abnormalities, with a myeloid to nucleated erythroid cell ratio of 5:1 and no maturation arrest. At that time, the sole diagnostic genetic test available was for *ELANE*, and no such variants were identified. His chronic diarrhea was difficult to analyze because of the presence of steatorrhea and a fat-soluble-vitamin deficiency but showed no documented exocrine pancreas deficiency. GCSF (5-7μg/kg/d) was prescribed, along with pancreatic enzyme and nutritional support. The patient returned to Algeria and continued to be treated between there and France. Repeated blood counts showed recurrent thrombocytopenia. Endoscopy of the digestive tract at 10 years old found inflammation; inflammatory bowel disease was diagnosed and treated with steroids. This situation, with recurrent infections and difficulty obtaining GCSF, resulted in several infections, including a colonic abscess and a colonic fistula necessitating a hemicolectomy that incurred major surgical complications. Although a molecular diagnosis of *SBDS* mutation was sought when he was 10 years old, it excluded a pathogenic variant; G6CP3-deficient neutropenia was finally diagnosed when he was 15 years old as the disease had just been identified by Sanger sequencing.[13]

After developing a gut fistula at 18 years of age, he returned to France. A prescription of anti–tumor necrosis factor α (TNF-α) for about 5 years, combined with long-term GCSF, maintained a limited quality of life, as his disease affected his ability to go to school and later work. At the age of 23 years, he was
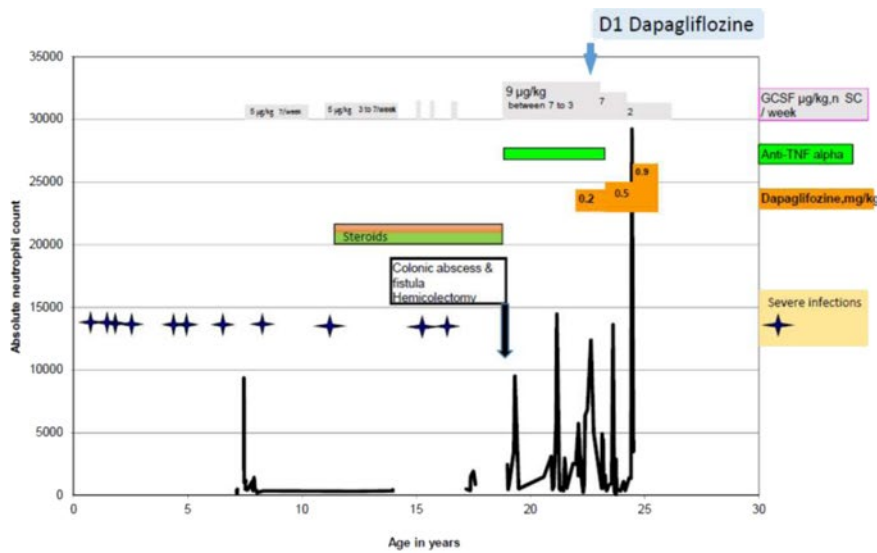
These 2 graphs summarize the medical history of the patient UPN 5643, who is presenting a G6PC3 neutropenia. The left graph depicts all his medical history, while the lower graph develop his recent period and the change related to Dapagliflozine onset.
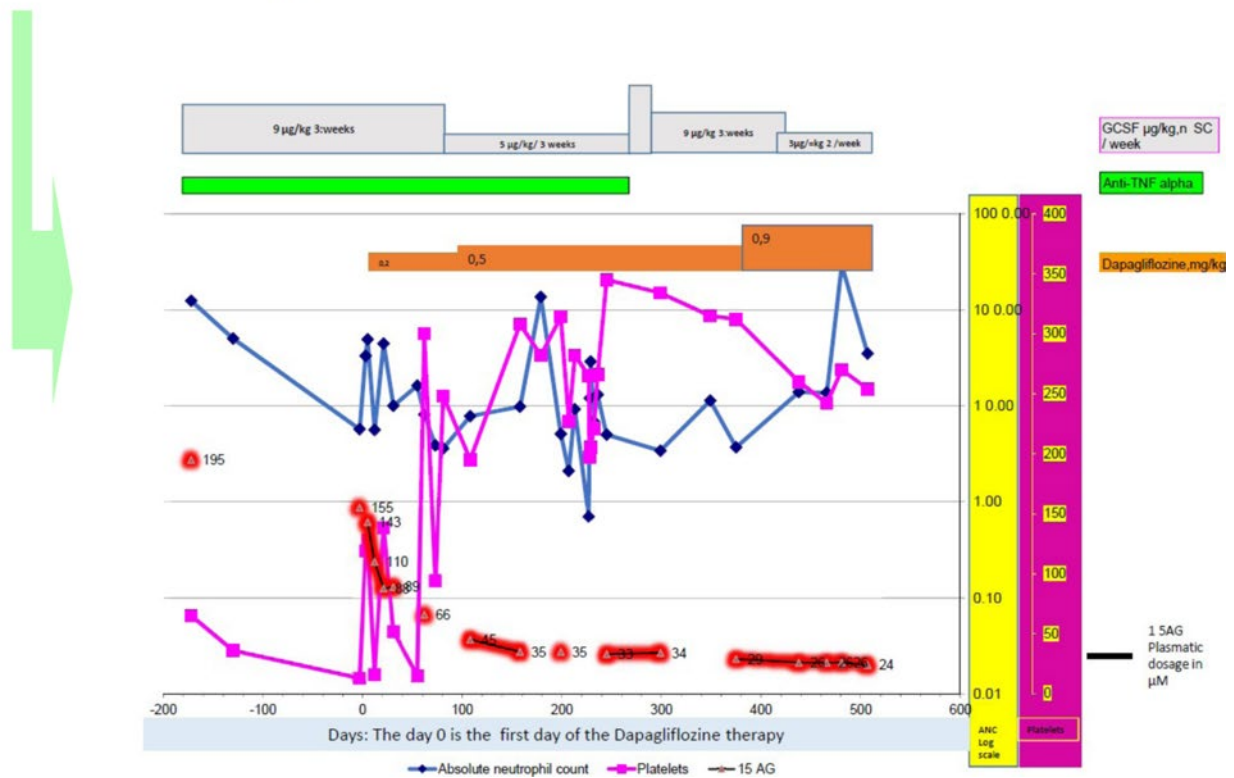
Figure 2. A visual summary of the medical history of a patient with G6PC3 neutropenia treated by gliflozine.

prescribed dapagliflozin at the initial dose of 0.2mg/kg in addition to his long-term therapy. Figure 2 shows his neutrophil, platelet, and blood 1,5AG evolutions over time. He initially received GCSF (9µg/kg) 3 times per week in conjunction with anti-TNF-α. Because of a partial neutrophil count and 1.5AG-clearance responses, the dapagliflozin dose was progressively increased up to 0.9mg/kg/d. Oral dapagliflozin has been continued since then (he is now 25 years old) for a total of 16 months. Notably, after 6 months the anti-TNF-α was withdrawn, and the inflammatory bowel disease did not recur. An attempt to stop GCSF was complicated by an oral

infection; however, it has now been tapered down to 3µg/kg twice weekly.

Conclusion

The standard care of GNs is based on GCSF and HSCT. Driven by the extensive development of genetic background analyses of such entities, better understanding of the mechanisms at work now offers some possibilities of adapted therapy. ISGTL2, an antidiabetic drug, may partially reverse GSDIB and G6PC3 GNs by clearing 1,5AG, which is responsible for the associated

neutropenia. CXCR4 inhibitors contribute to reversing the leukocyte defect in WHIM syndrome. Concerning the leukemic transformation risk, better understanding of the clonal evolution raises the possibility of preventing leukemia by stimulating somatic genetic rescue, a physiological process that might limit the risk of such progression.

Conflict-of-interest disclosure

Jean Donadieu: no competing financial interests to declare.
Christine Bellanné-Chantelot: no competing financial interests to declare.

Off-label drug use

Jean Donadieu: off-label drugs discussed are empagliflozin (Jardiance) and dapagliflozin (Forxiga).
Christine Bellanné-Chantelot: off-label drugs discussed are empagliflozin (Jardiance) and dapagliflozin (Forxiga).

Correspondence

Jean Donadieu, Hôpital Armand-Trousseau, 26 Ave du Dr Arnold Netter, 75012 Paris, France; e-mail: jean.donadieu@aphp.fr.

References

1. Bonilla MA, Gillio AP, Ruggeiro M, et al. Effects of recombinant human granulocyte colony-stimulating factor on neutropenia in patients with congenital agranulocytosis. *N Engl J Med*. 1989;320(24):1574-1580.
2. Donadieu J, Leblanc T, Bader Meunier B, et al; French Severe Chronic Neutropenia Study Group. Analysis of risk factors for myelodysplasias, leukemias and death from infection among patients with congenital neutropenia. Experience of the French Severe Chronic Neutropenia Study Group. *Haematologica*. 2005;90(1):45-53. https://www.researchgate.net/publication/8088087.
3. Rosenberg PS, Alter BP, Bolyard AA, et al; Severe Chronic Neutropenia International Registry. The incidence of leukemia and mortality from sepsis in patients with severe congenital neutropenia receiving long-term G-CSF therapy. *Blood*. 2006;107(12):4628-4635.
4. Rotulo GA, Beaupain B, Rialland F, et al. HSCT may lower leukemia risk in ELANE neutropenia: a before-after study from the French Severe Congenital Neutropenia Registry. *Bone Marrow Transplant*. 2020;55(8):1614-1622.
5. Rotulo GA, Plat G, Beaupain B, et al; French Severe Chronic Neutropenia Registry. Recurrent bacterial infections, but not fungal infections, characterise patients with ELANE-related neutropenia: a French Severe Chronic Neutropenia Registry study. *Br J Haematol*. 2021;194(5):908-920.
6. Donadieu J, Beaupain B, Fenneteau O, Bellanné-Chantelot C. Congenital neutropenia in the era of genomics: classification, diagnosis, and natural history. *Br J Haematol*. 2017;179(4):557-574.
7. Horwitz M, Benson KF, Person RE, Aprikyan AG, Dale DC. Mutations in ELA2, encoding neutrophil elastase, define a 21-day biological clock in cyclic haematopoiesis. *Nat Genet*. 1999;23(4):433-436.
8. Boocock GR, Morrison JA, Popovic M, et al. Mutations in SBDS are associated with Shwachman-Diamond syndrome. *Nat Genet*. 2003;33(1):97-101.
9. Bluteau O, Sebert M, Leblanc T, et al. A landscape of germ line mutations in a cohort of inherited bone marrow failure patients. *Blood*. 2018;131(7):717-732.
10. Donadieu J, Beaupain B, Mahlaoui N, Bellanné-Chantelot C. Epidemiology of congenital neutropenia. *Hematol Oncol Clin North Am*. 2013;27(1):1-17, vii.
11. Hsieh MM, Everhart JE, Byrd-Holt DD, Tisdale JF, Rodgers GP. Prevalence of neutropenia in the U.S. population: age, sex, smoking status, and ethnic differences. *Ann Intern Med*. 2007;146(7):486-492.
12. Bejjani N, Beaupain B, Bertrand Y, Bellanné-Chantelot C, Donadieu J. How to differentiate congenital from noncongenital chronic neutropenia at the first medical examination? Proposal of score: a pilot study from the French Severe Chronic Neutropenia registry. *Pediatr Blood Cancer*. 2017;64(12):e26722.
13. Boztug K, Appaswamy G, Ashikov A, et al. A syndrome with congenital neutropenia and mutations in G6PC3. *N Engl J Med*. 2009;360(1):32-43.
14. Veiga-da-Cunha M, Gerin I, Chen YT, et al. A gene on chromosome 11q23 coding for a putative glucose-6-phosphate translocase is mutated in glycogen-storage disease types Ib and Ic. *Am J Hum Genet*. 1998;63(4):976-983.
15. Veiga-da-Cunha M, Chevalier N, Stephenne X, et al. Failure to eliminate a phosphorylated glucose analog leads to neutropenia in patients with G6PT and G6PC3 deficiency. *Proc Natl Acad Sci USA*. 2019;116(4):1241-1250.
16. Wortmann SB, Van Hove JLK, Derks TGJ, et al. Treating neutropenia and neutrophil dysfunction in glycogen storage disease type Ib with an SGLT2 inhibitor. *Blood*. 2020;136(9):1033-1043.
17. Boulanger C, Stephenne X, Diederich J, et al. Successful use of empagliflozin to treat neutropenia in two G6PC3-deficient children: impact of a mutation in SGLT5. *J Inherit Metab Dis*. 2022;45(4):759-768.
18. Grünert SC, Derks TGJ, Adrian K, et al. Efficacy and safety of empagliflozin in glycogen storage disease type Ib: data from an international questionnaire. *Genet Med*. 2022;24(8):1781-1788.
19. Halligan RK, Dalton RN, Turner C, Lewis KA, Mundy HR. Understanding the role of SGLT2 inhibitors in glycogen storage disease type Ib: the experience of one UK centre. *Orphanet J Rare Dis*. 2022;17(1):195.
20. Zuelzer WW. "Myelokathexis"—a new form of chronic granulocytopenia. Report of a case. *N Engl J Med*. 1964;270(April):699-704.
21. Beaussant Cohen S, Fenneteau O, Plouvier E, et al. Description and outcome of a cohort of 8 patients with WHIM syndrome from the French Severe Chronic Neutropenia Registry. *Orphanet J Rare Dis*. 2012;7(September):71.
22. Heusinkveld LE, Yim E, Yang A, et al. Pathogenesis, diagnosis and therapeutic strategies in WHIM syndrome immunodeficiency. *Expert Opin Orphan Drugs*. 2017;5(10):813-825.
23. Hernandez PA, Gorlin RJ, Lukens JN, et al. Mutations in the chemokine receptor gene CXCR4 are associated with WHIM syndrome, a combined immunodeficiency disease. *Nat Genet*. 2003;34(1):70-74.
24. Badolato R, Donadieu J; WHIM Research Group. How I treat warts, hypogammaglobulinemia, infections, and myelokathexis syndrome. *Blood*. 2017;130(23):2491-2498.
25. Dale DC, Bolyard AA, Kelley ML, et al. The CXCR4 antagonist plerixafor is a potential therapy for myelokathexis, WHIM syndrome. *Blood*. 2011;118(18):4963-4966.
26. McDermott DH, Liu Q, Velez D, et al. A phase 1 clinical trial of long-term, low-dose treatment of WHIM syndrome with the CXCR4 antagonist plerixafor. *Blood*. 2014;123(15):2308-2316.
27. McDermott DH, Pastrana DV, Calvo KR, et al. Plerixafor for the treatment of WHIM syndrome. *N Engl J Med*. 2019;380(2):163-170.
28. Dale DC, Firkin F, Bolyard AA, et al. Results of a phase 2 trial of an oral CXCR4 antagonist, mavorixafor, for treatment of WHIM syndrome. *Blood*. 2020;136(26):2994-3003.
29. Beekman R, Valkhof MG, Sanders MA, et al. Sequential gain of mutations in severe congenital neutropenia progressing to acute myeloid leukemia. *Blood*. 2012;119(22):5071-5077.
30. Kennedy AL, Myers KC, Bowman J, et al. Distinct genetic pathways define pre-malignant versus compensatory clonal hematopoiesis in Shwachman-Diamond syndrome. *Nat Commun*. 2021;12(1):1334.
31. West RR, Calvo KR, Embree LJ, et al. ASXL1 and STAG2 are common mutations in GATA2 deficiency patients with bone marrow disease and myelodysplastic syndrome. *Blood Adv*. 2022;6(3):793-807.
32. Revy P, Kannengiesser C, Fischer A. Somatic genetic rescue in Mendelian haematopoietic diseases. *Nat Rev Genet*. 2019;20(10):582-598.
33. Zambetti NA, Bindels EM, van Strien PM, et al. Deficiency of the ribosome biogenesis gene SBDS in hematopoietic stem and progenitor cells causes neutropenia in mice by attenuating lineage progression in myelocytes. *Haematologica*. 2015;100(10):1285-1293.
34. Golomb L, Volarevic S, Oren M. p53 and ribosome biogenesis stress: the essentials. *FEBS Lett*. 2014;588(16):2571-2579.
35. Xia J, Miller CA, Baty J, et al. Somatic mutations and clonal hematopoiesis in congenital neutropenia. *Blood*. 2018;131(4):408-416.
36. Tan S, Kermasson L, Hilcenko C, et al. Somatic genetic rescue of a germline ribosome assembly defect. *Nat Commun*. 2021;12(1):5044.
37. Jaako P, Faille A, Tan S, et al. eIF6 rebinding dynamically couples ribosome maturation and translation. *Nat Commun*. 2022;13(1):1562.
38. Desplantes C, Fremond ML, Beaupain B, et al. Clinical spectrum and long-term follow-up of 14 cases with G6PC3 mutations from the French severe congenital neutropenia registry. *Orphanet J Rare Dis*. 2014;9(December):183.