French National Diagnosis and Care Protocol (PNDS)

ACQUIRED AND INHERITED APLASTIC ANEMIA

Version dated 03/01/2023

This PNDS was developed by the **Reference Center for Acquired and Inherited Aplastic Anemia .**
Member of the MaRIH rare diseases health network
(Rare Immuno-Hematological Diseases).

Contents

Abbreviations

AA	Aplastic anemia
ACTH	Adrenocorticotropic Hormone
AD	Autosomal Dominant
AFP	Alfa-fetoprotein
ALT	Alanine Aminotransferase
AMA	Acquired Aplastic Anemia
AML	Acute Myeloid Leukemia
APCs	Apheresis Platelet Concentrates
AR	Autosomal Recessive
AST	Aspartate Aminotransferase
ATG	Anti-thymocyte Globulin
AVK	Antivitamin K
BMT	Bone Marrow Transplant
BSA	Body Surface area
CBC	Complete Blood Count
CBU	Cord Blood Unit
CECOS	Center for the Study and Conservation of Eggs and Human Sperm
CML	Chronic Myeloid Leukemia
CMML	Chronic Myelomonocytic Leukemia
CMV	Cytomegalovirus
CSA	Ciclosporin A
CY	Cyclophosphamide
DBA	Diamond Blackfan Anemia
EBMT	European Society for Blood and Marrow Transplantation
EBV	Epstein Barr virus
ELT	Eltrombopag
EPO	Erythropoietin
FA	Fanconi Anemia
FISH	Fluorescence *in situ* Hybridization
FLU	Fludarabine
G-CSF	Granulocyte Colony-Stimulating Factor
GvHD	Graft Versus Host Disease
GVL	Graft Versus Leukemia (the graft's response to the disease)
HAS	French Health Authority
Hb	Hemoglobin
HbF	Fetal Hemoglobin
HIV	Human Immunodeficiency Virus
HLA	Human Leukocyte Antigen
HSC	Hematopoietic Stem Cells
HT	Hypertension
IBMF	Inherited Bone Marrow Failure
IgG	Immunoglobulin G
IgM	Immunoglobulin M
IUGR	Intrauterine Growth Retardation
IV	Intravenous
IVES	Intravenous Electric Syringe
LBP	Labile Blood Products
LIC	Liver Iron Content
LMWH	Low Molecular Weight Heparin

LTC	Long-term Condition
MA	Marketing Authorization
MAP	Medically Assisted Procreation
MAP	Medically Assisted Procreation
MDPH	Departmental Center for Disabled People
MDS	Myelodysplastic Syndromes
MMF	Mycophenolate Mofetil
MRD	Matched related donor
MRI	Magnetic Resonance Imaging
MDT	Multi Disciplinary Team
NOAC	New Oral Anticoagulants
PC	Platelet Concentrate
PE	Patient Education
FGMP	French Genomic Medicine Plan - 2025 programm
PID	Pre-Implantation Diagnosis
PMN	Polymorphonuclear Neutrophils
PND	Prenatal Diagnosis
PNDS	French National Diagnosis and Care Protocol
PNH	Paroxysmal Nocturnal Hemoglobinuria
PRBC	Unit of packed red blood cells
RC	Reference Center
RLMB	Reference Laboratory of Medical Biology
RP	Ribosomal Proteins
SC	Solid Cancer
SFGM-TC	Francophone Society of Bone Marrow Transplantation and Cellular Therapy
SIA	Search for Irregular Agglutinins
SIV	Slow Intravenous
TBI	Total Body Irradiation
TPE	Therapeutic Patient Education
TPO	Thrombopoietin
TPO-RAs	Thrombopoietin Receptor Agonists

Summary for Physicians and Pediatricians

General information

Aplastic Anemia (AA) is a quantitative bone marrow failure, secondary to the complete or partial disappearance of hematopoietic tissue, without abnormal cell proliferation (aplastic anemia is not a cancer).

The cessation of production of hematopoietic stem cells (HSC) leads to a global failure of hematopoiesis, resulting in cytopenia that will be observed in the CBC.

There are different mechanisms contributing to the occurrence of aplastic anemia: most aplastic anemias are idiopathic (*a priori* autoimmune), 15% to 20% are inherited (genetic), particularly in children, and some are favored by exposure to toxic or medicinal agents (1% to 2%).

AA is a rare disease with an incidence of 2–3 cases per million inhabitants per year in Europe and the United States. Its prevalence is 1 person per 250,000 inhabitants.

The symptoms and severity of AA differ from patient to patient. Aplastic anemia is most often suspected where there is pancytopenia (anemia, neutropenia and thrombocytopenia) with aregenerative anemia: the platelet lineage is almost constantly affected. Neutrophils are sometimes spared. These cytopenias can develop in a few days or more slowly over a few months or years (especially for constitutional aplasias). In general, bicytopenia or pancytopenia always requires specialized hematological advice.

From the treatment point of view, only severe and very severe forms require treatment, as well as moderate AA in case of transfusin requirement. Other forms simply requires monitoring.

The risk of death exists (although it is in sharp decline) and predominates during the first months of the disease or in the case of advanced disease that is refractory to any treatment. Death usually occurs as a result of severe bleeding or serious infection. This disease also exposes the patient to the risk of secondary progression to myelodysplastic syndrome or acute leukemia (myeloid hemopathies, basically). The therapeutic program for a patient with moderate AA begins with the first transfusion, in coordination with the blood transfusion center and a hematologist or pediatric hematologist. It is therefore essential to refer the patient quickly to an expert center (reference center or center of expertise).

In severe idiopathic AA, therapeutic management is an emergency because any delay in treatment has an impact on the prognosis. Bone marrow allograft is the standard treatment for patients under 40 years of age with an HLA-identical intrafamilial donor. More than 80% of cases of severe acquired MA are cured in this situation. In the absence of an intrafamilial donor or if the patient is over 40 years of age, a combination of horse anti-thymocyte globulins (ATG), ciclosporin and eltrombopag is the treatment of choice. In severe and very severe forms in adults, the standard treatment is now a triple therapy combining horse ATG, ciclosporin and eltrombopag (Peffault de Latour et al. 2022); in children, there is no indication for the use of eltrombopag as first-line therapy. This immunosuppressive treatment is effective in 80% of patients with variable quality of response. The median response to this treatment is around 3 months. Transplantation of HSC from an unrelated donor (from anonymous voluntary donors) may be necessary for young refractory or relapsed subjects after immunosuppressive therapy. The results are less good than in the case of transplantation from a compatible family donor, given the risk of graft versus host disease (GvHD), which is the main complication of transplantation in this setting and potentially fatal.

HSC transplantation is the only cure for hematological involvement in constitutional (genetic) forms.

▪ Management in cases where aplastic anemia is suspected and during outpatient follow-up

Assessing clinical signs of severity requiring emergency hospitalization:

► In the case of fever in a neutropenic patient (<500 PMN/mm^3 or 0.5 G/L):
In general, any fever occurring in a neutropenic patient (<0.5 G/L) requires hospital advice.

- Poorly tolerated fever with chills and shock
- Ulcero-necrotic or antibiotic-resistant angina
- Fever resistant to antibiotics after 48 hours
- Fever greater than 38.5°C in a patient with a central venous line

► In the case of poor tolerance of anemia:

- Functional signs of intolerance: dyspnea on the slightest exertion, dizziness, headache, malaise, poorly tolerated tachycardia, angina, confusion, asthenia, anorexia in small children
- Transfusion of units of packed red blood cells is necessary in case of clinical intolerance or systematically if hemoglobin level <7 g/dL (higher threshold in case of comorbidities)

► In case of bleeding risk:

There is usually no spontaneous bleeding risk if platelets are greater than 20 G/L in adults or 10 to 20 G/L in children, except in cases of associated thrombopathy, associated anticoagulant or antiplatelet therapies, or processes promoting bleeding (digestive damage, poorly controlled HT). Platelet transfusion should be considered:

- in the case of patent bleeding (extensive purpura, mucocutaneous hemorrhage, epistaxis, hematuria, etc.)
- as prophylaxis in the case of thrombocytopenia of less than 20 G/L in adults, 10 to 20 G/L in children (it is possible to lower this threshold to 10 G/L in regularly transfused patients with mild symptoms)

▪ **In the event of an emergency**, contact the local hospital service of the national network, available on the List of Centres website

In the event of any problems, contact a doctor from the reference center (RC) or a center of expertise for "Aplastic anemia". The reference center can be called on 01 42 49 96 39 between 8am and 5pm.
Outside of these hours:
- the on-call hematologist at Hôpital Saint-Louis can be contacted via the switchboard on 01 42 49 49 49
- the on-call pediatrician for pediatric hematology at Hôpital Robert-Debré can be contacted on 01 40 03 53 88 or via the switchboard on 01 40 03 20 00

Confirming the diagnosis and assessing disease severity:

This stage of diagnosis requires the patient to be referred to an adult or pediatric hematology center on the list of centers of the national network compiled by the reference center (below).
The assessment of severity and comorbidities and the search for potential underlying genetic diseases will guide the therapeutic approach to be followed and subsequent monitoring.

The attending physician can provide essential information for therapeutic management: **family or personal past medical history, recently initiated treatments or available previous blood counts** (normal or abnormal).

Initial therapeutic management

Initial therapeutic management depends on the severity of aplastic anemia and the patient's age. It requires prolonged hospitalization in the majority of cases. Only moderate Aplastic anemia or

elderly patients will be monitored on an outpatient basis through regular checks on the blood count (alternating between general practitioners and hospital physicians).

Multidisciplinary team(MDT) meetings

Cases can be discussed at the MDT meetings organized by the Aplastic anemia Reference Center (RC). These are usually held on the first and third Wednesdays of each month (dates available on the website www.aplasiemedullaire.com). The case must be presented by a doctor using the MDT meetings case presentation form (available in the section for professionals on the same site), which must be sent by email no later than 48 hours before the MDT meetings to: valerie.quinet@aphp.fr or by fax to 01 42 49 96 36.

French National Registry of Bone Marrow Failures (RIME)

A national registry of acquired and inherited bone marrow failures (RIME), coordinated by the reference center (Prof. Régis Peffault de Latour - Dr. Flore Sicre de Fontbrune - Prof. Thierry Leblanc - Ms. Isabelle Brindel) was set up in early 2017 and allows the anonymized recording of clinical/biological data concerning patients with acquired and inherited Aplastic anemia and paroxysmal nocturnal hemoglobinuria (PNH). The objective is to improve the overall understanding of these pathologies.
To do this, a biobank was set up (Prof. Jean Soulier, Hematology Laboratory, Saint-Louis) to preserve biological samples (blood and marrow) taken at each stage of the management of these patients, as well as fibroblasts for inherited forms.

Emergency forms

These are available via the following links on the Orphanet website "Orphanet Urgence".

- *Good practice in emergencies/PNH*

- Good practice in emergencies/Fanconi Anemia

- *Good practice in emergencies/Blackfan-Diamond Anemia*

Emergency cards

Emergency cards (Aplastic anemia and PNH) developed by the reference center, under the aegis of the MaRIH (rare immuno-hematological diseases) health network in accordance with the National Plan for Rare Diseases, have been disseminated within its national network and must be given to all patients with Aplastic anemia .

Useful information

The PNDS can be found on the website of the French Health Authority by conducting an advanced search for "PNDS Aplastic anemia s" and is also available on the websites of the reference center and the MaRIH health network.

- **Reference Center for Acquired and Inherited Aplastic anemia**
  - Email: cr.aplasiemedullaire.sls@aphp.fr
  - Website: www.aplasiemedullaire.com
  - Tel.: 01 42 49 96 39 / 01 71 20 75 27
  - Organizational chart available on the website

- **Orphanet:** www.orpha.net
  - Orphanet codes

• Acquired Aplastic anemia	• ORPHA 164823 • ORPHA 88 • ORPHA 182040
• Paroxysmal nocturnal hemoglobinuria	• ORPHA 447
• Inherited Aplastic anemia	• ORPHA: 68383
• Fanconi's disease	• ORPHA 84
• Diamond Blackfan anemia	• ORPHA 124
• Telomeropathies	• ORPHA 1775
• *GATA2* syndrome	• ORPHA 228423 • ORPHA 3226 • ORPHA 319465
• Shwachman-Diamond syndrome	• ORPHA 811
• Congenital amegakaryocytosis	• ORPHA 3319
• *SAMD9/SAMD9L*	• Code waiting to be created • Code 494 433 (MIRAGE)
• *TPO*	• Code waiting to be created
• *Myeloproliferative leukemia virus oncogene (MPL)*	• Code waiting to be created
• *ERCC6L2*	• ORPHA 401764
• *MDS1 and EVI1 complex locus (MECOM)*	• ORPHA 71289

➢ **Patient associations**

- French Association of Fanconi's Disease (AFMF): fanconi.com
- Francophone Association of Diamond Blackfan Disease: afmbd.org
- French PNH Association - Aplastic anemia : hpnfrance.com
- TELOMERO ASSO Association for Telomeropathies: telomero-asso.fr

➢ **French Healthcare Network for Rare Immune Hematological Diseases (MaRIH):**

Email: contact@marih.fr
Website: www.marih.fr
Facebook: @Filiere.MaRIH
Twitter: @Filiere_MaRIH
Instagram: filière_sante_marih

Objective of the PNDS

The aim of this National Diagnosis and Care Protocol (PNDS) is to explain to the professionals in charge of patients with AA the current optimal diagnostic and therapeutic management and care pathway of a patient with acquired (AMA) or inherited bone marrow failure (IBMF). AA can be managed as a long-term condition (LTC). The aim of this PNDS is to optimize and harmonize the management and monitoring of AA throughout the country. It can also be used to identify proprietary drugs used for indications not covered by their Marketing Authorization (MA), as well as the proprietary drugs, products or services necessary for patient care but not usually covered or reimbursed.
This PNDS is a practical reference tool for treating physicians (the physician designated by the patient to the French National Health Insurance Office) in consultation with the medical specialist, particularly when establishing the care protocol jointly with the consulting physician and the patient in the case of a request for exemption from co-payment for an off-list condition.
The protocol describes the reference management of a patient with acquired and inherited Aplastic anemia and this update takes into account recent data from the medical literature. However, the PNDS cannot consider all specific cases (comorbidities or complications, details of treatment, hospital care protocols, etc.). It cannot claim to be exhaustive in terms of possible management approaches, nor can it replace the individual responsibility of the physician toward their patient.
This PNDS was developed in accordance with the "Method of developing a national protocol for diagnosis and care of rare diseases" published by the French Health Authority in 2012.

Methodology

The main sources used by the multidisciplinary working group (Annex 1) for the preparation of this guide were:

- the text of the 1st PNDS drawn up in 2009 and the 2nd in 2019, which served as the basis for this update
- the main international recommendations for the management of acquired and inherited Aplastic anemia
- the meta-analyses, clinical trials and cohort studies (non-exhaustive review) published in the literature (PubMed) between 2019 and 2023 concerning the methods of diagnosis, the characteristics and treatment of acquired and inherited Aplastic anemia and paroxysmal nocturnal hemoglobinuria (PNH) in adults and children
- documents from the websites of Orphanet, the French Society of Hematology, the Society of Pediatric Immuno-Hematology (SHIP) and the Pediatric Group of the Francophone Society of Bone Marrow Transplantation and Cellular Therapy (SFGM-TC)
- the recommendations of this new updated version of the PNDS, which were developed and validated by the multidisciplinary working group including adult hematologists and pediatricians and medical biologists, with a critical review by a group of specialists in the management of Aplastic anemia, representatives of patient associations, and diagnostic laboratories associated with the reference center and a treating physician
- for therapeutic aspects, various grades of recommendations were issued, depending on the data from the literature based on the levels of evidence set out in the table below (reference from the French Health Authority - HAS 2013).

Recommendations grade	Level of scientific evidence provided by the literature
A Established scientific evidence	Level 1 - High-potency randomized controlled trials - Meta-analysis of randomized controlled trials - Decision analysis based on well-conducted studies
B Scientific presumption	Level 2 - Low-potency randomized controlled trials - Well-conducted non-randomized controlled trials - Cohort studies
C Low level of scientific evidence	Level 3 - Case studies
	Level 4 - Comparative studies with significant biases - Retrospective studies - Series of cases - Descriptive epidemiological studies (transversal, longitudinal)

This PNDS does not apply to pancytopenia occurring immediately following anti-mitotic chemotherapy, as well as isolated cytopenia (anemia, thrombocytopenia and neutropenia), either acquired or congenital, and refractory cytopenia (myelodysplastic syndromes), which are outside the field of expertise recognized for the "acquired and inherited Aplastic anemia " rare disease reference center.

General information on Aplastic anemia

Definition

Aplastic anemia is a quantitative bone marrow failure secondary to hematopoietic stem cells exhaustion, which causes cytopenia.

Distinction between acquired and inherited Aplastic anemia

Acquired Aplastic anemia is most often acute and of immunological origin, and more rarely of toxic or medicinal origin (below is a table of products that may potentially cause Aplastic anemia). Acquired Aplastic anemia accounts for 80% of cases of Aplastic anemia. These so-called immunological forms include idiopathic Aplastic anemia, post-hepatitis aplastic anemia(hepatitis-aplastic anemiasyndrome) and PNH associated aplastic anemia (Young et al. 2018).

Inherited Aplastic anemia are secondary to a genetic defect, which usually causes an intrinsic defect of the hematopoietic stem cell. These forms are more rare in adults and proportionally much more common in children and adolescents; this relative frequency is not currently established and probably underestimated.

Epidemiology

AA is more common in Asia than in Europe and America. Its incidence is currently in the order of 2 cases per million inhabitants per year in Europe. It reaches 6 cases per million inhabitants in Thailand and 7.4 in China.
The incidence of AA describes a bimodal curve with a first peak in young subjects and a second peak past 50 years (Young et al., 2018).

Professionals involved

The initial, therapeutic and follow-up management of patients with AA systematically involves the treating physician, the adult or pediatric hematologist, and a pediatrician for children.
Depending on the aetiology and the clinical picture, the opinion of other specialists may be necessary: in particular geneticists, radiologists, otorhinolaryngologists (ORL), maxillofacial surgeons, gynecologists, occupational and school physicians, microbiologists, and hemobiologists (responsible for the delivery of labile blood products - LBP).
The involvement of other professionals (psychologists, nurses, psychomotor therapists and physiotherapists, care assistants, home helps, social workers, etc.) may also be necessary.

Diagnosis and initial assessment

The objectives of initial diagnostic management are:

1- To confirm the diagnosis of Aplastic anemia and eliminate differential diagnoses
2- To differentiate between the acquired and inherited forms
3- To assess the degree of severity of the bone marrow failure

Aplastic anemia can be discovered by chance on a blood count prescribed in another medical context or discovered due to symptoms related to cytopenia (anemic or infectious syndromes related to neutropenia, or hemorrhagic syndrome related to thrombocytopenia).
A predominant cytopenia on a single line is possible at the onset of the disease, most often thrombocytopenia. The latter is usually accompanied by other blood count abnormalities (anemia, often macrocytic, or isolated macrocytosis or moderate neutropenia), in which case the tests below should be carried out.

Interviews and physical examinations should look for:

- Signs of severity related to cytopenias that would require emergency hospitalization
- Elements suggesting an underlying inherited pathology
- The absence of tumor syndrome, which would suggest a hematologic malignancy

The process for conducting interviews and clinical examinations is presented in Annex 13.a.

The signs of severity to look for are:

- Extensive or mucosal purpura (intra-oral hemorrhagic bullae), especially if accompanied by bleeding at the eyes fundus or from organs
- An infectious syndrome
- Signs of poor cardiac or neurological tolerance of anemia

Elements to look for in the interview that would suggest a inherited etiology are:

- A personal or family history of bleeding
- A family history of cytopenias or myelodysplastic syndromes and leukemias (risk common to almost all inherited bone marrow failure)
- Personal or family history of symptoms or pathologies affecting the skin and skin appendages (nails and hair), opportunistic infections, pulmonary and/or hepatic pathologies (☞ telomeres related disorders), neurological pathologies, elements specific to *GATA2* syndrome (lymphoedema, pulmonary alveolar proteinosis, etc.)
- A family history of early hair graying (before age 20) or other skin or peripheral disorders (☞ telomeropathies)

- A family history of cancer at an early age (☞ some forms of inherited aplasia including Fanconi anemia)
- Congenital kidney, bone or cardiac malformations, etc. (☞ Fanconi disease, *GATA2* syndrome, -Diamond Blackfan anemia (DBA) and other IBMFs)
- Intrauterine growth retardation (☞ Fanconi disease, Diamond Blackfan anemia, Shwachman-Diamond syndrome and other IBMFs)
- Chronic steatorrhea-like diarrhea or abdominal pain (Shwachman-Diamond syndrome)
- A previous blood count (>6 months) that was already abnormal (suggesting a IBMF, which usually builds up gradually over several years)

Elements to look for in the interview that would suggest an acquired form are:

- A recent or semi-recent episode of jaundice or hepatitis (often cured at the time of aplastic anemia occurence)
- A recent normal full blood count
- Cytopenias that very rapidly become worse
- Clinical signs suggesting associated hemolytic PNH (hemoglobinuria, abdominal pain, dysphagia, history of thrombosis or erectile dysfunction)
- Use of medication that may be responsible for AA (Table 1) and/or occupational exposure (benzene, ionizing radiation, etc.)

Table 1: Main products likely to be responsible for Aplastic anemia (non-exhaustive list)

Medications:
Antibiotics: sulfonamide, co-trimoxazole, linezolid
Anti-inflammatories: indometacin, naproxen, diclofenac, piroxicam, sulfasalazine, penicillamine
Antithyroid drugs: carbimazole, thiouracil
Psychotropic drugs: phenothiazines, dosulepin
Other: chloroquine, mebendazole, allopurinol, thiazide diuretics
Medicinal products not marketed in France: chloramphenicol, phenylbutazone, gold salts, tolbutamide, chlorpropamide
Immunotherapies: nivolumab, pembrolizumab, rituximab
Chemicals
Benzene and other solvents
Pesticides
Oils and other lubricating agents
Recreational drugs: ecstasy, methylene dioxy-methamphetamine (MDMA)

Elements to look for in the clinical examination that would suggest a inherited etiology are:

- Abnormalities suggestive of Fanconi disease: intrauterine growth retardation (IUGR) and height-weight retardation, facial dysmorphia, malformations (thumb, hand, cleft lip and palate, heart, genital tract), skin spots, etc. (Annex 7)
- Abnormalities suggestive of telomeropathy: abnormalities of pigmentation (hypo/hyperpigmentation), nail dystrophy, early hair graying (before 20 years), oral leukoplakia, hepato-splenomegaly, dyspnea on exertion and abnormalities on pulmonary auscultation (pulmonary fibrosis)
- Abnormalities suggestive of *GATA2* syndrome: lymphedema, pulmonary abnormalities, deafness, warts and condylomas, history of thrombosis
- Abnormalities suggestive of Diamond Blackfan anemia: IUGR and height-weight retardation, facial dysmorphia (including cleft lip and palate), other malformations (thumb, hand, heart, genital tract)
- Abnormalities suggestive of Shwachman-Diamond syndrome: IUGR and height-weight retardation, bone abnormalities (metaphyseal dysplasias), abdominal pain and fatty diarrhea
- Abnormalities of the radius suggestive of a TAR syndrome (thrombocytopenia + bilateral radial agenesis)
- Abnormalities suggestive of *SAMD9/SAMD9L* syndrome: neurological and cerebellar involvement in particular, elements of MIRAGE syndrome (MDS-infections-growth retardation-adrenal hypoplasia- A for adrenal-genital abnormalities-enteropathy, see Annex 10)

Other rare inherited pathologies may be associated with Aplastic anemia : the genes involved, clinical and morphological abnormalities and the main manifestations of these syndromes are summarized in the table in Annex 2.

Complementary examinations

The **elements necessary for a diagnosis** of Aplastic anemia are:

- A complete blood count including reticulocytes count that shows pancytopenia (aregenerative anemia, neutropenia and thrombocytopenia)
- A smear that does not find signs of dysplasia or abnormal circulating cells; macrocytosis is common in inherited forms and possible in acquired forms
- A bone marrow differential cell count that confirms the central origin of cytopenias and eliminates an hematological malignancy (myelodysplasia and acute leukemia in particular); most often, the bone marrow smear is poor or inconclusive. Most of the cells observed are lymphocytes or plasma cells; sometimes there is a relative excess of macrophages or mast cells but these remain of normal morphology. Occasionally, an initial bone marrow differential cell count may appear to show normal richness. Perls staining may be necessary in case of diagnostic doubt with myelodysplastic syndrome. Elements of dysplasia are common in inherited forms, such as dyserythropoiesis in Fanconi anemia
- A bone marrow cytogenetical exam medullary karyotype, to eliminate a primary myelodysplastic syndrome or a clonal evolution of AA. Three points worth mentioning: 1) some cytogenetic abnormalities (del(20q), +8 and del(Y)) remain compatible with the diagnosis of AA, 2) karyotype failures are not exceptional in severe forms due to the poverty of the marrow, 3) supplementary interphasic fluorescence *in situ* hybridization (FISH) (probes of 7 and 8) is useful, especially in case of failure of the karyotype. The presence of monosomy 7 at diagnosis, particularly in small children, could indicate the possibility of a *SAMD9/SAMD9L* syndrome (see Annex 10)
- A bone marrow biopsy (richness less than 30% for age): this is the only way to assess marrow richness and eliminate differential diagnoses (myelofibrosis, medullary invasion by malignant pathology, metabolic disease). However, a bone marrow biopsy is not automatically carried out in inherited forms, especially in children
- Acquired somatic mutations search by next-generation sequencing (NGS) to eliminate a myelodysplastic syndrome. However, there are currently no data to recommend an

adaptation of management in the presence of an isolated somatic mutation without other cytological or cytogenetic elements suggesting a myeloid maligancy. This examination must not delay the initiation of treatment in the absence of diagnostic doubt.

The main differential diagnoses are:

1) Hypoplastic myelodysplastic syndromes, mainly in adults, distinguishable by the existence of signs of dysplasia of granulocytic and megakaryocytic lines and/or a cytogenetic abnormality not compatible with the diagnosis of aplasia (an isolated dyserythropoiesis is sometimes observed in acquired aplastic anemia, especially in the presence of a PNH clone)
2) Cytopenias of undetermined significance: richness of the marrow normal; note that a bone marrow differential cell count of normal richness does not rule out Aplastic anemia .

The **etiological examinations to be performed** in all patients are:

- A search for a PNH clone (significant if ≥1%) in granulocytes and monocytes using flow cytometry; its presence suggests an acquired form
- The search for elevated fetal hemoglobin (HbF) before transfusion (above 5% suggests IBMF)
- An alfa-fetoprotein assay (the low αFP elevations present in Fanconi anemia are not detected by all available kits: if possible, this examination should be carried out with the KRYPTOR kit, which has a higher sensitivity)
- An immunoglobulin weight assay and an evaluation of lymphocyte subpopulations (at least CD3, CD4, CD8, CD19)
- Analysis of anti-nuclear factors, anti-native DNA and rheumatoid factor
- Complete liver function tests
- A parvoB19 serology (polymerase chain reaction (PCR) of the blood (marrow) in the case of immune deficiency
- Cardiac and abdomino-pelvic ultrasound to look for congenital malformations
- A chest X-ray (and in case of doubt a CT scan of the chest to look for an exceptional thymoma in adults)

More specialized tests to look for a inherited cause should be carried out in patients under 18 years of age, or in the presence of family history or any clinical elements/elements in the history that could suggest this.
These examinations are:

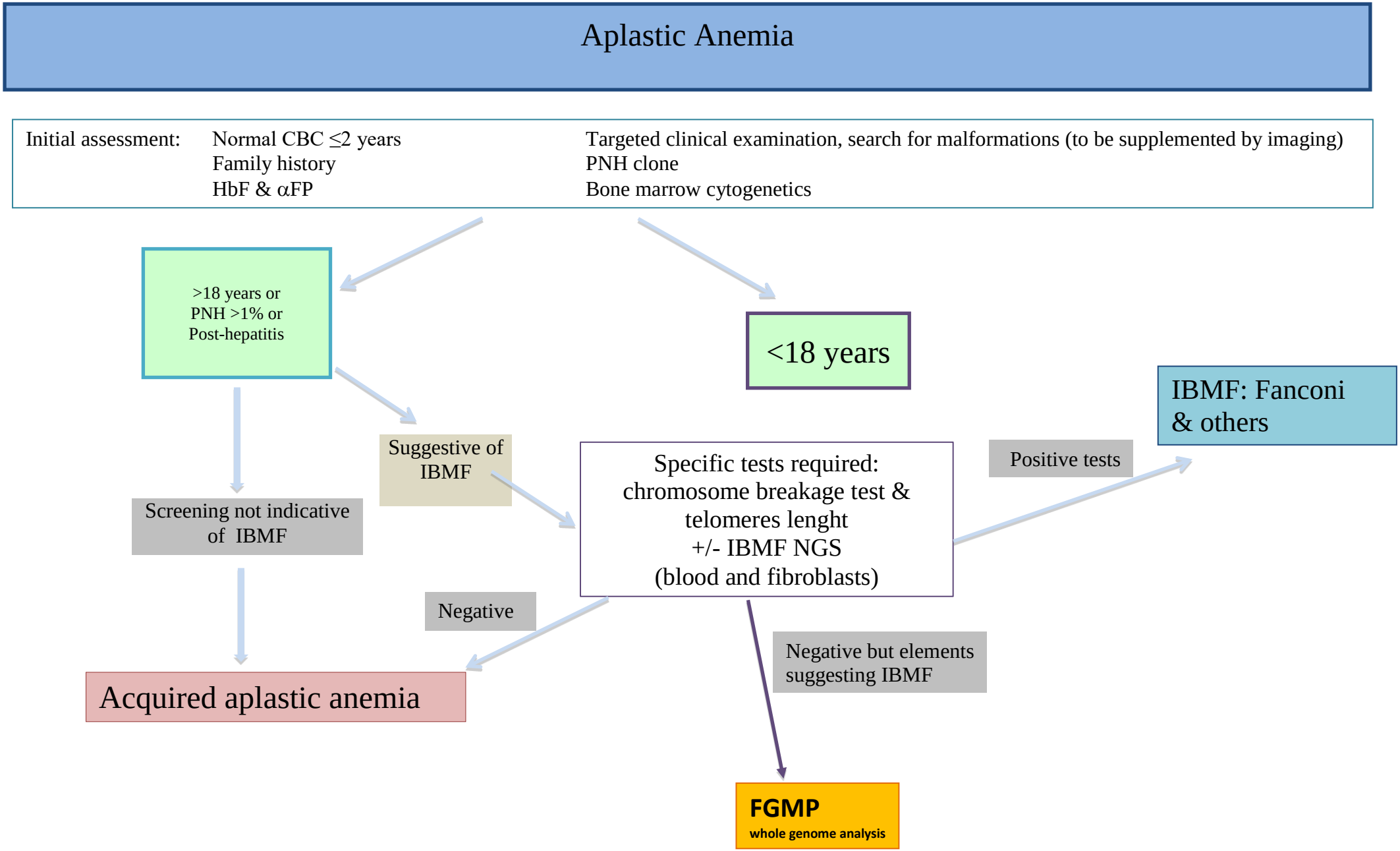
- a chromosomal breakage test to look for Fanconi disease (systematic before 18 years)
- an evaluation of telomere length on blood lymphocytes using the FlowFISH technique (detailed in Annex 8) at the reference medical biology laboratory (LBMR) Robert-Debré in Paris (systematic before 18 years). Telomeres below the 1st percentile suggest telomeropathy, but this is not specific: short telomeres can be seen in other IBMFs and also in acquired forms, especially in hepatitis-aplasia syndromes. Conversely, telomeres between the 1st and 10th percentile can be observed in patients with telomeropathies, especially in 1st generations and the most elderly subjects.
- a search for mutations based on examination of a panel of genes involved in inherited bone marrow failures (Annexes 2 and 3, a non-exhaustive list and likely to evolve), ideally on a non-hematopoietic tissue (fibroblasts obtained by skin biopsy given the possibility of clonal reversion/evolution, which can lead to false negatives on blood samples, Annex 13 b), is recommended systematically before the age of 10 and on a case-by-case basis after that. The samples will be sent first to the LBMR Aplasias (Hôpital Saint-Louis) except where there is potential evidence of telomeropathy, where they will firstly be sent to the LBMR Telomeropathy (Hôpital Bichat). If there is possible evidence of Diamond Blackfan anemia, they will firstly be sent to the LBMR "Diamond Blackfan anemia" (Hôpital Kremlin Bicètre). This examination must not delay treatment if there is no evidence in favor of a inherited cause.

- fat-soluble vitamins, lipase, fecal elastase and, if in doubt, MRI of the pancreas (Shwachman-Diamond syndrome)
- search for elevated erythrocyte adenosine deaminase (eADA) in a patient who has not received a transfusion or has not had a transfusion for over 3 months (Diamond Blackfan anemia)

The degree of exhaustivity and the time spent on the search for a inherited cause will vary from patient to patient:

1) In certain situations where there is obvious evidence of an acquired form (acute aplastic anemia, typical hepatitis-aplastic anemia syndrome or PNH clone greater than 1%), treatment may be started in the absence of these examinations, including in subjects under 18 years of age
2) Where a inherited form is strongly suspected, this assessment may be more exhaustive, but in a child or young adult the specific treatment (immunosuppression or transplantation) should not be delayed by more than 6 weeks in relation to the diagnosis, in the absence of any evidence of a inherited form. This decision is not always simple and can be discussed at the national MDT meetings In this type of situation, a bone marrow transplant with an unrelated donor can be discussed at the MDT meetings.
3) In the absence of genetic diagnosis following conventional genetic analyses (breakage test, NGS IBMF, telomeropathies or Diamond Blackfan anemia), if there is strong evidence of an inherited form or in children in case of failure of immunosuppressive treatment, it is recommended to discuss at the national MDT meetings the possibility of carrying out a genome study in very high throughput sequencing within the framework of the French Genomic Medicine Plan (PFMG) on fibroblast DNA (Seqoia or Auragen platforms, see specific annex)

Decision algorythm for inherited assessment



The **other examinations to be carried out systematically prior to treatment** are:

- Determination of blood group with extended erythrocyte phenotype and search for irregular agglutinins with a view to a possible transfusion
- HLA typing of the patient and their siblings and ideally of their parents as an emergency if aged less than 40 years or if graft planned above 40 years of age
- HIV, hepatitis B, hepatitis C, EBV and CMV viral serologies (herpes simplex virus (HSV) & varicella zoster virus (VZV) in children)
- Investigation of any abnormalities in renal function tests: blood electrolytes, urea, creatinine and calculation of creatinine clearance

Severity degree assessment

Pancytopenia can be life-threatening and is a therapeutic emergency.

The severity of acquired aplasia is assessed according to the Camitta criteria; this is essential to assess the need for commencing treatment (Rovo et al., 2013).

Aplastic anemia is said to be severe according to the Camitta Index if the medullary richness is <30% of hematopoietic cells in a bone marrow biopsy and if there are at least 2 of the following criteria at the periphery:

- Thrombocytopenia <20 G/L
- Neutropenia <0.5 G/L
- A reticulocytopenia of <60 G/L (on an automated machine – threshold used in the RACE study, which differs from the conventionally used threshold of 20 G/L in manual reading) (Peffault de Latour et al., 2022).

The EBMT (European Society for Blood and Marrow Transplantation) distinguishes very severe aplastic anemia with criteria identical to those of severe aplasia but with neutropenia <0.2 G/L.

The existence of aggravating factors (underlying situation, age, associated immune deficiency) must be taken into account in considerations relating to treatment.

Therapeutic management

The therapeutic management of Aplastic anemia depends on the **severity**, its **cause** and the **age** of the patient. In severe forms, it is a therapeutic emergency.

Symptomatic treatment

Transfusion support deserves special attention

- **Anemia**: transfusion of red blood cells (RBC); the transfusion threshold (7 to 9 g/dL) is to be adapted according to age, etiology, cause, clinical tolerance, comorbidities and quality of life. RBCs must be phenotyped (Rh and KeL1) to prevent allo-immunization. To prevent transfusional GvHD, the RBCs should be irradiated for patients who have received anti-thymocyte globulins therapy and/or hematopoietic stem cell transplant from the start of conditioning. Prevention of iron overload by iron chelators should be considered if serum ferritin is >500 µg/l or if the patient has received more than 20 red blood cell transfusions (long transfusion program) (see dedicated section below).

- **Thrombocytopenia**: transfusion of platelet concentrates (PC) in the case of existence of hemorrhagic signs and/or platelets <10–20 G/L depending on hemorrhagic risk factors and clinical tolerance, which is specific to each patient. The significant risk of allo-immunization justifies limiting the number of PC transfusions to a maximum, especially if an allograft is planned. There are no longer any recommendations to transfuse apheresis platelet concentrates (APCs) as a priority for aplastic or allograft patients. PCs are no longer irradiated in France. If transfusions are ineffective and in high-risk subjects (multiparous women), an anti-HLA allo-immunization test should be performed. HLA poly-immunization can make transfusion management complex because of

difficulties in having HLA-compatible APCs. In the absence of HLA-compatible APCs, it may be decided not to carry out prophylactic transfusions. The administration of ATG is associated with an increase in platelet transfusion requirements, which may last for a few days after discontinuation of this treatment.

- **Chelation**: iron overload has a negative impact on hematopoiesis and potentially on post-allograft morbidity and mortality. Regular screening must be carried out by measuring ferritin levels, coupled with the transferrin saturation coefficient (the presence of free iron in the blood, which if >60% is associated with higher toxicity on the organs). The iron levels must be measured every 2 to 3 months depending on the frequency of transfusions; they should be measured just before a transfusion and ideally on an empty stomach. Chelation is ideally recommended if the ferritin level is >500 µg/L. In the case of significant overload, hepatic and cardiac MRI are indicated to assess the degree of tissue overload.
There is no optimal chelating agent in patients with MA: each of them involves difficulties with prescription or exposes patients to toxicity. Potential side effects (especially renal failure and agranulocytosis) should be carefully monitored in patients with AA. In the case of renal toxicity in a patient taking ciclosporin, the efficacy of immunosuppressive therapy should be the priority.

Medications	**Problems posed in aplasia**
Deferoxamine - Desféral®	Subcutaneous administration (in practice tolerable with "pin-type" needles) Increased risk for some infections: mucormycosis, *Yersinia enterolytica*
Deferasirox - Exjade®	Major renal toxicity in combination with ciclosporin Rare cases of medullary toxicity
Deferiprone - Ferriprox®	Risk of agranulocytosis, or even Aplastic anemia in the case of predisposition

There are no validated recommendations for the choice of medication. Deferoxamine (Desféral®) administered subcutaneously (8 to 12 hours) can be used as first-line therapy. In a hospitalized patient, the venous route is also possible while maintaining the same duration of infusion. In some patients with major overload, a combination of chelating agents may be justified, for example in the pre-transplant period, to minimize iron overload.

In patients in remission of aplastic anemia or in post-allograft, phlebotomies should be given preference over chelating agents because of their good efficacy and the absence of adverse reactions. The volume and frequency of these phlebotomies will be adapted to clinical tolerance. The total number of phlebotomies will be determined by monitoring the iron balance and iron overload, evaluated by hepatic and cardiac MRI.

<u>Prevention and treatment of infectious complications:</u>
- In the absence of fever and before any etiological treatment, hospitalization is not necessary
- Any fever (defined as a temperature above 38.5°C once or above 38°C twice at 4-hour intervals) in a patient with AA with neutropenia <0.5 G/L requires emergency hospitalization and intravenous antibiotic therapy. Monitoring vital signs is essential and any sign of poor hemodynamic tolerance requires the advice of an intensive care specialist
- In patients with acquired Aplastic anemia and neutropenia <0.5 G/L, anti-fungal prophylaxis with posaconazole, voriconazole or itraconazole is recommended. In inherited forms where the risk of fungal infection appears to be lower, this prophylaxis should be evaluated on a case-by-case basis and according to the severity of neutropenia
- Prophylaxis of pneumocystosis with co-trimoxazole and prophylaxis of HSV and VZV infections with aciclovir or valaciclovir are recommended after the combination of anti-thymocyte globulin and ciclosporin until $CD4^+$ cell counts are above 0.4 G/L (in the case of allergy, prophylaxis with atovaquone may be proposed). A 1^{st} lymphocyte phenotyping can be performed at 3 months.

Treatment with ciclosporin alone does not warrant prophylaxis for pneumocystosis; however, in elderly patients, prophylaxis for herpes zoster with valaciclovir may also be offered

- In patients with inherited AA, the administration of vaccines protecting against bacterial infections (pneumococci, meningococci, *Haemophilus*) and against influenza is recommended, if they have not already been vaccinated as part of the vaccination program recommended by the HAS
- In patients with acquired AA, the recommendations are to follow the general vaccination recommendations of the HAS. Live attenuated vaccines should be reviewed by the referring hematologist and are contraindicated with ciclosporin and/or after treatment with ATG.

  Hematopoietic growth factors:
  - Granulocyte-colony stimulating factor (G-CSF) is not routinely indicated. It can be prescribed as a short-term treatment in case of severe infection or to facilitate a return home.
  - Erythropoietin is generally not useful in the absence of chronic renal failure
  - Thrombopoietin receptor agonists (TPO-RAs) will be discussed in the context of etiological treatment.

Etiological treatments

a) Acquired Aplastic anemia

The indication for treatment depends on the severity: severe Aplastic anemia or a moderate form requiring repeated transfusions are indications for specific treatment.

The treatments available alone or in combination are:

- allogeneic hematopoietic stem cell transplantation
- anti-lymphocyte/thymocyte globulins (horse (with temporary authorization for use) or rabbit ATG)
- ciclosporin
- alemtuzumab
- androgens
- eltrombopag, a thrombopoietin receptor agonist (TPO-RA)

Other immunosuppressive therapies (corticosteroids, azathioprine, mycophenolate, sirolimus, cyclophosphamide) are not indicated for the treatment of Aplastic anemia . Corticosteroids are not effective in aplastic anemia and should not be used in combination except for the prevention of side effects of ATG

1st line treatments

Immunosuppressive therapy of moderate Aplastic anemia in adults and children not eligible for allograft:

The combination of ATG and ciclosporin is the standard immunosuppressive therapy for moderate Aplastic anemia in adults and aplasia in children. Since the study by Scheinberg et al., 2011, horse ATG (ATGAM) has been proposed as a 1st line therapy because of its superiority over rabbit ATG in terms of hematological response and survival rates (grade A). The mean time to hematological response to this therapy can be long, around 3 months according to the median figures (and can be up to 6 months even though moderate Aplastic anemia usually responds more quickly to immunosuppressive therapy than severe forms). Survival at 5 years after treatment with ATG is about 90% for non-severe forms (Peffault de Latour, 2018). ATGAM does not yet have an MA in France and is currently the subject of a temporary authorization for use in a cohort study. A retrospective study conducted in France including patients treated in the context of this temporary authorization for use in a cohort study found the same results (de Latour, 2018).

In children, an early evaluation of ATGAM-ciclosporin therapy can be proposed at 3 months in order to decide rapidly on a matched unrelated transplantin case of complete failure (no response), with the objective that the graft be carried out between 5 and 6 months after ATG if failure is confirmed.
ATGAM is given at a dose of 40 mg/kg daily for 4 consecutive days by slow intravenous administration followed by ciclosporin administration. Therapy with ciclosporin should be prolonged for at least 18 months. Annex 4 details the administration, monitoring and follow-up of ATG-ciclosporin therapy and of patients with non-allografted Aplastic anemia more generally (life-long follow-up).

Immunosuppressive therapy of severe Aplastic anemia in adults

For severe Aplastic anemia in adults, a recent study by the reference center demonstrated a benefit in adding eltrombopag, a thrombopoietin receptor agonist, to the ATG-ciclosporin combination. This is the RACE study: a prospective randomized phase III study comparing ATGAM (horse ATG) + ciclosporin versus ATGAM + ciclosporin + eltrombopag in the first-line treatment of severe Aplastic anemia (Peffault de Latour New England Journal of Medicine (NEJM) 2022) (grade A). Eltrombopag treatment was started from D15 at a dose of 150 mg daily for up to 3 months in the case of complete response or 6 months in the case of partial response at 3 months. This medicine should be taken on an empty stomach (at least 2 hours before and after eating) and should not be consumed at the same time as dairy products. Discontinuation of therapy was abrupt from one day to the next, leading to reintroduction of eltrombopag in around 20% of patients. It is therefore advisable to stop this therapy more gradually after reducing the dose by half for 2 months and if the response remains stable. The addition of eltrombopag to ATG-ciclosporin therapy increased the overall response by approximately 30% at 6 months with a median response at 3 months (versus 9 months with standard therapy). There was no evidence of a difference in toxicity between the 2 arms. The combination of eltrombopag-horse ATG and ciclosporin is therefore becoming the standard treatment for adult patients with severe or very severe Aplastic anemia not eligible for allograft. However, in children the value of this combination (eltrombopag with ATG and ciclosporin) has not been demonstrated (Groarke British Journal of Hematology (BJH) 2021) (grade C).
Following immunosuppressive therapy, patients with a response are at risk of relapse in 15% to 30% of cases, ciclosporin dependence (approximately 30% of cases), progression to myelodysplastic syndrome or acute leukemia (cumulative incidence of 10% at 20 years, without identifiable plateau) and occurrence of hemolytic PNH in 20% of cases (Peffault de Latour et al., 2018).

Particular features of the elderly (over 65 years of age)

It should be noted that in elderly patients, the reference treatment is the combination ATG-ciclosporin-eltrombopag if there are no cardiovascular or renal comorbidities and the risk of loss of autonomy due to prolonged hospitalization is not too high (Contejean et al., 2019) (grade C). In subjects over 65 years of age with comorbidities, in particular cardiovascular comorbidities, making ATG treatment unreasonable, eltrombopag can best be used in combination with ciclosporin or as monotherapy (Lengline et al., 2018) (grade C). Ongoing studies are prospectively evaluating this practice (EMAA study “Efficacy and tolerance of eltrombopag in patients with moderate acquired Aplastic anemia treated with ciclosporin” (National Clinical Trial (NCT) 02773225), SOAR study “Eltrombopag in combination with ciclosporin as first-line treatment in severe MA” (NCT 02998645, presented at the American Society of Hematology (ASH) in 2021) and the study “Eltrombopag in moderate Aplastic anemia ” of the National Institute of Health (NIH) (Fan Xand al., 2020) (grade B).
Data on the use of romiplostim, another TPO-RA, in first-line treatment for Aplastic anemia are not robust at this time (ongoing studies).

Allogeneic hematopoietic stem cell transplantation

Allogeneic hematopoietic stem cell transplantation with a *matched related donor* is the reference first-line treatment in subjects under 40 years of age due to excellent long-term survival (>80%, or even 90% in very young people) (Konopacki et al., 2012) (grade C). In this indication, the source of stell cells must be bone marrow and the conditioning combines cyclophosphamide 200 mg/kg total dose and rabbit ATG (Thymoglobulin®, Sanofi-Genzyme, Gentilly, France) 12.5 mg/kg total dose; prophylaxis for GvHD combines ciclosporin and methotrexate.

Allogeneic stem cell transplant is the only cure for Aplastic anemia. However, due to the significant increase in morbidity and mortality related to treatment after 40 years, including with a matched related donor donor, first-line transplant is reserved for subjects under 40 years of age.
Three particular points deserve attention regarding allograft:

- Primary and secondary graft failure remain a significant problem after a bone marrow transplant (BMT) for acquired AA (10–20%). The incidence of failure decreases significantly if 1) the graft is rich ($CD34^+$ >3 to 5 x 10^6/kg of recipient weight) and not handled, 2) if the patient is not allo-immunized against major histocompatibility antigens (hence the importance of a transfusion policy adapted to these patients), and (3) if the pre-transplant conditioning is sufficiently immunosuppressive. The proposed conditioning in a matched related situation reduces the probability of rejection/failure to less than 5% (randomized trial comparing cyclophosphamide alone to cyclophosphamide + ATG) (Champlin et al., 2007) (grade A). Total body irradiation is abandoned in matched related transplants in 1st line treatment because of its carcinogenic potential.
- Graft versus host disease (GvHD) is the main complication of allograft. The incidence of severe acute GvHD (grades III-IV) is currently about 15%. It is all the more frequent and severe the older the patient. A randomized trial in acquired AA transplant patients comparing ciclosporin alone to the combination of ciclosporin + methotrexate showed a significant reduction in GvHD with combination therapy (Locatelli et al., 2000). The use of bone marrow as a source of cells is essential to reduce the risk of GvHD, as is the duration of ciclosporin prophylaxis (Schrezenmeier et al., 2007, Bacigalupo et al., 2012) (grade A).
- In subjects under 18 years of age, there are currently discussions about offering a *matched unrelated* transplant as first-line treatment if a 10/10 HLA-identical unrelated donor is quickly available and the allograft is feasible in less than 2 months from registration, due to the very good results in this population (Dufour et al., 2015). However, there is no sufficiently substantiated study to recommend this approach systematically. A Hospital Clinical Research Program (upfront matched unrelated donor (MUD), NCT 05419843) will make it possible to carry out a prospective study to evaluate this strategy. These situations must be discussed at the national MDT meetings on a case-by-case basis because the procedures for grafting, particularly conditioning, are very specific. A doubt about a inherited origin can also be an argument to carry out an allograft from the outset.

The follow-up of a patient who has received an allograft for Aplastic anemia is detailed in **Annex 5.**
In certain infectious situations where survival depends on the time taken for hematological reconstitution, a non matched related transplant is sometimes proposed: this strategy cannot be the subject of a recommendation and must systematically be discussed among colleagues at the national MDT meetings.

2nd line treatments

A second treatment with ATG and ciclosporin may be offered in patients over 30 years of age who do not have 10/10 donors, particularly in patients who have relapsed or have partial response. A combination with eltrombopag is logical in this situation (grade C).

Eltrombopag
Eltrombopag may be offered from 3 months of age in patients who are ineligible for allograft and who have failed immunosuppressive therapy, if they have not received it before (Olnes et al., 2012, Desmond et al., 2014, Lengliné et al., 2018) (grade B), see AA therapeutic algorithm below.
In adults, therapy is started at a dose of 75 mg per day and increased to 150 mg on D10–15 if hepatic and digestive tolerance is satisfactory. This medicine should be taken on an empty stomach (at least 2 hours before and after eating) and should not be consumed at the same time as dairy products. Therapy should be continued for at least 4 to 6 months to assess its effectiveness. An increase in the dose to 225 mg/day is possible after 3 to 6 months of treatment in the case of good tolerance (20% additional response in 3 to 6 months). Once the best response is achieved, a

gradual decrease is necessary (50% of the dose every 8 weeks under blood count monitoring) due to the high risk of relapse. A medullary assessment with cytogenetic examination is imperative before therapy and at 6 months.
In children, there is no 1st line indication; use in the case of failure can be discussed at the MDT meetings.

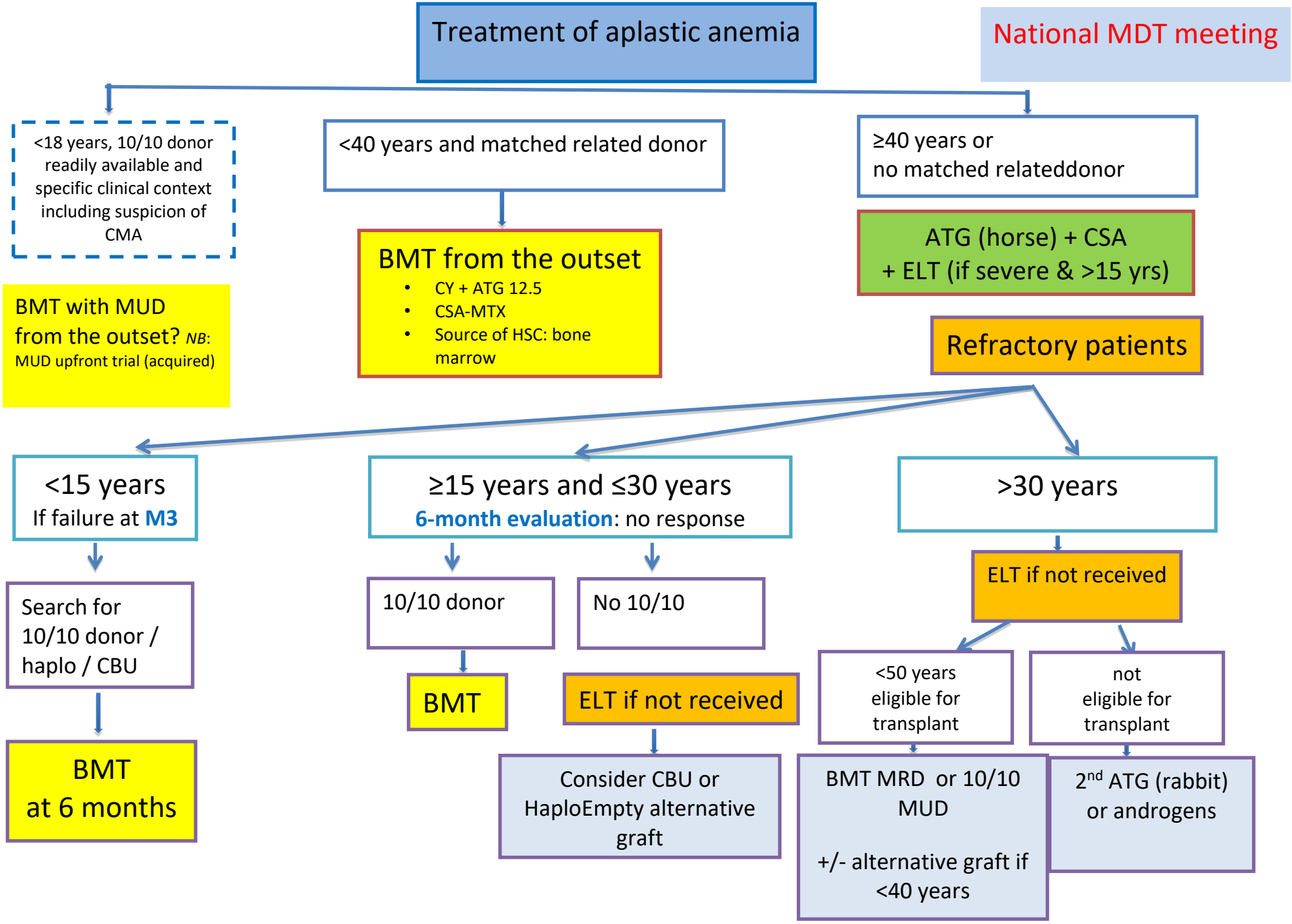
Allogeneic hematopoietic stem cell transplantation

In patients in whom immunosuppressive therapy has failed or who have experienced early relapse, an allograft may be discussed as 2nd line treatment for subjects under 30 years of age (and up to 40 years of age depending on the general condition of the patient) who have a matched unrelated donor (10/10) and patients over 40 years of age who have a matched related donor.
In patients over 50 years of age, allografts will only be offered on a case-by-case basis if alternative therapies fail, due to the high risk of therapy-related mortality in this age group.
The 2 types of conditioning (detailed in Annex 5) proposed for these matched unrelated allografts are:
- For patients under 50 years of age: fludarabine, cyclophosphamide, ATG and total body irradiation (TBI) 2 Gy replaced by double-dose ATG (15 mg/kg) within 14 years
- In patients over 50 years of age: fludarabine, cyclophosphamide, alemtuzumab (Campath®, Sanofi-Genzyme, Gentilly, France, temporary authorization for use) combined with ciclosporin prophylaxis for GvHD
For refractory patients under 30 years of age, an alternative allograft (9/10 donor, haploidentical donor or cord blood transplant should be discussed at the national MDT meetings. A national phase 2 protocol is evaluating haploidentical transplant outcomes for this strategy (HaploEmpty, NCT 05126849).

Other therapies:

Ciclosporin monotherapy should not be used as 1st-line therapy, except in situations where the ATG-ciclosporin combination or allograft is impossible due to comorbidities. This medicine specifically blocks the activation and proliferation of the T lymphocyte. The overall response rate for ciclosporin given alone (without ATG or androgens) is about 30–40%. As with ATG, the response to therapy is all the more significant if the disease is less severe (60% in non-severe forms, 34% in severe forms, and only 25% for very severe forms of the disease). Once the response is obtained, therapy should be continued for one year at optimal dose and then gradually decreased in increments of 10% with blood count monitoring; the experience of the Aplastic anemia RC favors 3-month steps, although some teams propose a faster decrease. It is recommended in the initial phase to obtain plasma trough levels of at least 200 ng/ml and in case of renal intolerance to obtain the maximum tolerated renal dose (urea is a more reliable marker than creatinine).

Androgens: since published data are extremely limited, this treatment should be reserved for refractory or relapsed forms that are not eligible for another treatment. In relapsed or refractory patients not eligible for HSC allograft, androgens obtain a response in about 30% of patients. The only androgen currently available in France is danazol. In adults, it is usually prescribed at a dose of 200 mg x 2 daily and can be increased in 4-week increments up to 800 mg daily. In children, it starts at a dose of 5 mg/kg daily. Once the response has been obtained, the dose should be gradually reduced with blood count monitoring to the minimum effective dose or even to discontinuation. Prolonged treatment of more than one year should be monitored annually by hepatic imaging (ideally CT scan with contrast medium or MRI if no contraindications) because of the risk of tumors; this risk persists after discontinuation of therapy, making prolonged monitoring necessary. In men, prostatic monitoring should be implemented.


Treatment of aplastic anemia
National MDT meeting
<18 years, 10/10 donor readily available and specific clinical context including suspicion of CMA
<40 years and matched related donor
≥40 years or no matched relateddonor
BMT with MUD from the outset? NB: MUD upfront trial (acquired)
BMT from the outset
CY + ATG 12.5
CSA-MTX
Source of HSC: bone marrow
ATG (horse) + CSA + ELT (if severe & >15 yrs)
Refractory patients
<15 years
If failure at M3
≥15 years and ≤30 years
6-month evaluation: no response
>30 years
Search for 10/10 donor / haplo / CBU
10/10 donor
No 10/10
ELT if not received
BMT at 6 months
BMT
ELT if not received
<50 years eligible for transplant
not eligible for transplant
Consider CBU or HaploEmpty alternative graft
BMT MRD or 10/10 MUD
+/- alternative graft if <40 years
2nd ATG (rabbit) or androgens

b) Paroxysmal Nocturnal Hemoglobinuria (PNH)

PNH is closely associated with acquired Aplastic anemia : 30% of adult patients with acquired AA have a PNH clone at diagnosis and 25% of adult patients with PNH will develop Aplastic anemia during follow-up. PNH clones at diagnosis are rarer in children.
The monitoring of patients with acquired AA thus includes regular screening for PNH and its complications, and conversely, the follow-up of a patient with PNH respects the same imperatives as that of a patient with Aplastic anemia regarding the screening and therapy of bone marrow failure but also the risk of clonal evolution toward a myelodysplastic syndrome or acute myeloblastic leukemia.
Annex 6 details the follow-up, therapy and monitoring of PNH patients.
Treatment of hemolytic forms is based on C5 inhibitors, eculizumab or ravulizumab (Sicre de Fontbrune et al., 2018). These patients have a high risk of thrombosis, which requires the same rules of caution as those applied to patients with hereditary thrombophilia.
In the case of associated severe aplasia, the therapeutic algorithm follows the recommendations for aplasia therapy (immunosuppressive therapy or HSC allograft).

c) Inherited bone marrow failure (IBMF)

IBMFs are rare pathologies. However, advances in molecular genetics over the past decade have led to a continuous increase in the number of identified syndromes. As a result, the number of child and adult patients identified as suffering from these syndromes has increased proportionally.
The main IBMFs are:

- Fanconi's disease
- telomeropathies (formerly called congenital dyskeratosis)
- *SAMD9/SAMD9L* syndromes
- congenital amegakaryocytosis (*MPL* mutations), which frequently progresses to Aplastic anemia , and the rarer inherited *TPO* mutations

Other inherited pathologies responsible for central pancytopenia and occasionally presenting as Aplastic anemia are:

- Shwachman Diamond syndrome
- *GATA2* syndrome
- Diamond Blackfan anemia (exceptional and rarely severe aplasia based on blood count data)
- finally, many other recently identified rare genetic entities (*MECOM, ERCC6L2, LIG4*, etc.)

Common elements of the management of patients with these conditions include:

1) Lack of indication for immunosuppressive therapy
2) Discussion of the indication for HSC allograft:
   - some diagnoses have to be made urgently, due to the need to adapt allograft procedures for Fanconi disease and telomeropathies
   - the lack of systematic indication in certain situations such as *SAMD9/SAMD9L* syndromes and some telomeropathies also justifies an early genetic diagnosis
   - for other inherited diagnoses, genetic diagnosis is not necessary to choose conditioning
3) Genetic aspects: genetic counseling and family interviewing (particularly including donor selection)
4) Management of initial and ongoing extra-hematological damage requiring coordinated multidisciplinary care
5) The risk of clonal medullary evolution (myelodysplastic syndrome or acute leukemia) justifying medullary monitoring and also of solid tumors in certain pathologies justifying screening adapted to the genetic pathology

The clinical, biological and management specificities of each type of CMA are detailed in Annexes 7 to 10.

Fanconi anemia (Annex 7)

The management of Fanconi anemia, once the diagnosis is established is dominated by hematological monitoring (appearance of cytopenias and clonal abnormalities) and by screening for squamous cell carcinomas (cancer of the head and neck, respiratory and digestive tracts, pelvis). The occurrence of transfusion requirements or clonal abnormalities with poor prognosis should lead to consideration of bone marrow allograft if a geno-identical or pheno-identical HLA-compatible donor (10/10) is identified (Annex 5). Alternative allografts should be discussed in the national aplasia MDT meetings, given that the results are still preliminary.
In the absence of a compatible donor, supporting care or androgens can be offered.
The existence of growth retardation and malformations must be the subject of specific multidisciplinary follow-up from childhood, adapted on a case-by-case basis. Regular follow-up is necessary to ensure early detection of solid neoplasms (in particular stomatological and ENT) whose only curative treatment is surgery.

Telomeropathies (Annex 8)

Telomeropathies are a set of genetically and phenotypically heterogeneous diseases, characterized by a inherited deficit in telomere maintenance. Telomeropathies can be diagnosed at any age of life, but early symptoms are most often correlated with higher severity.
Hematological manifestations are variable: isolated or macrocytosis-associated thrombocytopenia, medullary hypoplasia, aplastic anemia or myelodysplastic syndrome (MDS), immune deficiency.
Respiratory disorders are most often of the pulmonary fibrosis type.
Hepatic damage is variable and often underestimated, due to the fact that the liver function tests may not show much disturbance despite the severity of the damage. Portal hypertension is often the first manifestation.
Osteoporosis is common and often associated with fractures; systematic screening is necessary.
Androgens can be offered as 1st line treatment in the case of severe hematological impairment.
Allogeneic transplantation may be proposed in the event of failure, if a 10/10 non-carrier donor is identified, with suitable conditioning and after eliminating any contraindications linked to other organ damage. Regular screening should be carried out for extra-hematological disorders.

Diamond Blackfan anemia (Annex 9)

Diamond Blackfan anemia is a bone marrow failure (congenital erythroblastopenia) that develops as a result of a blockage of maturation of erythroid progenitors (BFU-e/CFU-e) and mainly concerns the production of red blood cells. An allelic variation (including a large deletion) in a ribosomal protein gene (23 genes involved) is identified in nearly 75% of patients (Da Costa et al., 2020). The diagnosis is most often made early in life, usually before 1 year of age: a diagnosis after the age of 4 is rare but possible, including in adulthood in forms with mild anemia. More than half of patients have growth retardation and associated malformations, the most common being malformations of the head (cleft lip and palate), thumbs, heart and urogenital tract.

At pediatric age, management is dominated by treatment for anemia (transfusions or corticosteroids >1 year) and associated malformations and by growth monitoring. The 3 possible therapeutic approaches are: 1) corticosteroid therapy, 2) long-term transfusion support and 3) HSC transplantation. Transplantation may be indicated in cortico-resistant patients (or those dependent on high doses of corticosteroids, in excess of 0.3 mg/kg daily); the consensus is for transplants with an HLA-identical sibling donor, in whom a silent form of the disease must be excluded (absence of

macrocytosis, normal eADA and absence of the causal allelic variation), or an unrelated 10/10 donor. The transplant should ideally be carried out before the age of 5. In adulthood, genetic counseling is added in the event of desire for children, along with screening for hematological evolutions (loss of response to corticosteroids, occurrence of myelodysplastic syndrome) and finally screening for solid tumors (in particular osteosarcoma in adolescents and colon cancer (Vlachos A et al., 2018)).

At all ages, prevention of corticosteroid side effects (osteoporosis, in the first instance) and treatment of iron overload are of major importance.

Congenital amegakaryocytosis and disorders related to *TPO* mutations

Although very rare, these two genetic pathologies are described in detail, as they warrant specific therapeutic management due to the high risk of rejection after HSC allograft in the case of the former, which requires appropriate conditioning, and the almost constant risk of graft failure in the case of the latter. The genetic diagnosis of these entities is therefore imperative.
Congenital amegakaryocytosis is a very rare autosomal recessive disease, linked to biallelic mutations of the *MPL* gene (thrombopoietin receptor), which is characterized by neonatal thrombocytopenia, most often symptomatic. In almost all cases, it develops into Aplastic anemia with pancytopenia. In some patients with a less severe form, the diagnosis may be made late in childhood due to pancytopenia. The diagnosis of congenital amegakaryocytosis is based on molecular analysis of the *MPL* gene. Therapeutic management is based on bone marrow transplantation after myeloablative conditioning.
Biallelic mutations in the thrombopoietin (*TPO*) gene have recently been reported in children or adolescents with pancytopenia predominantly on the platelet line. Bone marrow allograft in these patients has been complicated by failures, probably related to thrombopoietin (TPO) deficiency. Eltrombopag appears to be effective for these patients. However, clonal evolutions have been observed. The therapeutic management of these rare pathologies should be systematically discussed in the aplasia or pediatric graft MDT meetings.

SAMD9/SAMD9L syndromes (Annex 10)

These recently described syndromes with similar phenotypes are hematologically characterized by frequent association with monosomy 7 at diagnosis, and by the possibility of spontaneous improvement of aplasia due to genetic compensations. Cytopenias can appear abruptly, sometimes as a result of infection or vaccination, and this acute onset can mimic an acquired form. Extra-hematological involvement is heterogeneous and not consistent with diagnosis.

Patient follow-up

The modalities of the follow-up of patients with AA depend on the underlined etiology and subsequent treatment
In non-transplanted patients, screening for clonal evolution is fundamental regardless of the cause. This is based on regular bone marrow cytology with cytogenetic and molecular studies. The frequency depends on the etiological diagnosis of the aplasia and is detailed in the specific annexes. PNH is subject to the same follow-up recommendations due to a similar risk of long-term clonal evolution.
In patients receiving an allogeneic hematopoietic cell transplantation, follow-up is based on the screening for both post-transplant complications and features associated with AA evolution (Annex 5).
Of note is that, in patients with Fanconi anemia, telomeropathies and to less extent, Diamond Blackfan anemia, screening for secondary malignancies (both solid cancers and hematological neoplasia) is central to patient management (see annexes).

Management of specific situations

Only non-exceptional situations for which recommendations can be made will be discussed here.

The issue of fertility must be addressed before any treatment (Poirot et al., 2014).

The French bioethics law of July 2004, and its subsequent reviews, states that “When medical care is likely to impair fertility, or when fertility is at risk of being prematurely impaired , any person may benefit from the collection and conservation of their gametes or germinal tissue, with their consent and, where appropriate, that of one of the holders of parental authority, or of the tutor when the minor or adult concerned is the subject of a tutorship measure, with a view to the subsequent realization of a medically assisted procreation, .”

Therefore, when an allograft procedure is considered in the treatment of inherited or idiopathic aplastic anemia, the patient must be offered a specialized consultation in reproductive medicine, even if the conditioning most often used in these situations is non-myeloablative and therefore *a priori* unlikely to induce hypofertility or sterility. The benefit/risk balance in patients with AA that are at high risk of bleeding and/or infectious complications should be carefully explained and evaluated, as well as the deleterious impact of delayed transplantation. Fertility preservation techniques can also be proposed after allografting, which warrants specialized follow-up in the absence of prior preservation.

1/ Preservation of fertility in children prior to puberty (Dalle JH et al., 2017).
Before menarche in girls, and before the age of 12–13 and Tanner stage P3–T3 in boys, gamete preservation cannot be considered. Only techniques of cryopreservation of gonadal tissues are conceivable. These may not be possible in cases of profound thrombocytopenia or neutropenia, due to the risk of bleeding or infections.

a. In girls prior to puberty: cryopreservation of the ovarian cortex, the area that contains oocytes, is a technique that has been available for around 20 years. Pregnancies have been reported after heterotopic or orthotopic re-implantation of frozen tissue after allograft for non-malignant disease, at least when the patient was treated during or after puberty. For the youngers, data are not yet available.

b. In boys prior to puberty: it is possible to offer testicular tissue cryopreservation, but as this only contains spermatogonia or spermatozoa from the pre-pubescent period onwards, this is an experimental technique with no practical application to date.

2/ Preservation of fertility in the pubescent subject:

a. In males: it is essential to offer a consultation with CECOS (Center for the Study and Conservation of Eggs and Human Sperm) for the collection and cryopreservation of sperm.

b. In females: it may be possible to carry out stimulation for the removal of follicles and secondary oocytes for vitrification of gametes or *in vitro* fertilization followed by embryo preservation (technique reserved for stable couples). As with cryopreservation of gonadal tissue, the stimulation and trans-vaginal punctures required for these techniques may be contraindicated by thrombocytopenia or neutropenia. In a woman with a stable partner, embryo freezing is in theory possible but rarely compatible with the urgency of care.

3/ Cases of pregnancy (outside the context of bone marrow transplantation):
Pregnancy is a high-risk situation because of the risk of relapse or worsening of aplasia, which may not be reversible after pregnancy. Although there are no large studies on pregnancy and ciclosporin, the existing data on teratogenic risk are reassuring. It is currently the only treatment that can be offered to pregnant women. Ciclosporin crosses the placental barrier and fetal blood levels are about one-third of maternal blood levels.
https://lecrat.fr/spip.php?page=article&id_article=711.

Thrombosis and PNH

Arterial and venous thrombosis are the most serious complications of PNH, and were the main cause of morbidity and mortality before therapy with eculizumab (anti-complement factor 5) became available. This therapy significantly reduced the frequency of thrombosis (a reduction of more than 80% in the number of thrombotic events was confirmed in real life studies (Kelly et al., 2011; Loschi et al., 2016)). Thrombosis remains a complication of PNH that needs to be prevented and detected, particularly in the initial phase of the disease or in a pro-inflammatory context, which can lead to an increase in complement synthesis and a decrease in anti-C5 activity.

1/ Primary prophylaxis

- Prophylaxis with compression stockings should be offered systematically during air travel, in case of immobilization and in pregnant women
- Prophylaxis with low-molecular-weight heparin (LMWH) should be offered in the event of air travel >3 hours, prolonged immobilization, surgery, pregnancy from the end of the 2nd trimester and up to 6 weeks postpartum
- No long-term primary prophylaxis is justified, either with anti-vitamin K (AVK) or LMWH

2/ Curative therapy

The occurrence of thrombosis in a hemolytic PNH patient requires:

- Commencement of immediate effective curative anticoagulation with LMWH and then AVK. This anticoagulation is currently continued for life: a history of thrombosis is actually a risk factor for the occurrence of further thrombosis. In cases of high bleeding risk (e.g. severe thrombocytopenia), the benefit/risk balance should be assessed on a case-by-case basis and based on complement inhibition. New oral anticoagulants (NOACs) have not been evaluated in this population.
- Commencement of therapy with a C5 inhibitor (eculizumab or ravulizumab) if the patient is not already treated, intensification of therapy with eculizumab if the patient is already treated but the complement is poorly blocked or in an acute inflammatory situation (e.g. infection, surgery, postpartum). These situations may require an early infusion of eculizumab 900 mg or an increase in the dose to 1200 mg.

Cerebral thrombophlebitis and splanchnic thrombosis are particularly common in PNH patients. These patients should be referred to specialist centers (hematology and neurology, hematology and hepatology).

Child-adult transition

The child-to-adult transition is a critical period with a high risk of disruption of care and follow-up. It is advisable to implement a transition program, which requires close collaboration between the childrens' and adults' services(with identification of contacts within the adults' services).
This requires several steps:

- A preparatory phase in pediatrics: well before the transition, prepare the adolescents for the change of team. Recognize any resistance to change. Be concerned about their empowerment in the management of their illness. Offer them a first consultation without their parents. Appoint the future referring physician for adults at an early stage and consider a possible gender preference. Find out how the future physician works. Prepare a summary of the case for the patient and the physician
- A transfer phase: if possible, arrange a first[t] joint consultation with the physician for adults. If possible, encourage alternate consultations in the first year. Enlist the help of a transition worker with a visit to the adults' service. Allow for flexibility in transfer (16–20 years) depending on the patient's maturity. If possible, this phase should take place during a calm period of the disease

- A reception phase in the adults' service: provide for an extended consultation during which the names of the various practicioners to whom the patient will be referred will be communicated. If they are not absolutely essential, additional examinations should be avoided during this first consultation. Appreciate knowledge of the disease, learn about lifestyle habits.

Therapeutic Patient education (TPE)

According to the HAS in 2014, TPE aims to help patients manage their lives with a chronic disease as well as possible. It is an ongoing process that should be an integral and permanent part of patient care.
The patient and their close family and friends should have a good understanding of the disease.
Patients, their families and caregivers should be directed to other source of support and advice, for example, peer advocacy support groups (see the list of support groups in the "Useful links" section at the beginning of this document). The "patient days" organized by the support groups affiliated with the reference center and by the Francophone Society of Bone Marrow Transplantation and Cellular Therapy (SFGM-TC) also participate in TPE.
Booklets (Aplastic anemia and PNH) updated in October 2022 are available and distributed by the reference center, providing the patient and their close family and friends with written explanations about aplastic anemia and its management and about PNH.
APLASTIC ANEMIA PATIENT BOOKLET
PNH PATIENT BOOKLET & FAQS
In the case of bone marrow transplantation, SFGM-TC has published a national follow-up booklet (Bull Cancer 2021;108:S87–S89). This follow-up booklet is a support for exchanges between the patients and their caregivers. It is a tool to help the patients, which belongs to them, which they invest in, and which can be personalized.
Consultations with nurses and with clinical pharmacists, whenever they are possible, are important steps in PE.

Annexes

Annex 1. Lists of participants in the development of this guide and public conflicts of interest

This work was coordinated by Prof. Régis Peffault de Latour, coordinator of the Reference Center for Acquired and Inherited Aplastic anemia, Hematology Transplant Department, Hôpital Saint-Louis, Paris, in collaboration with Prof. Thierry Leblanc, constituent site of the pediatric reference center of Hôpital Robert Debré in Paris.

Editors 2019–2023 (*no indication if editors in 2019 and 2023*):

Dr. Bénédicte Bruno	Pediatric Hematology Department, Center of expertise of the RC CHU de Lille-Hôpital Jeanne de Flandre, Lille
Prof. Jean-Hugues Dalle	Pediatric Hemato-Immunology Department, Constituent center of the RC Hôpital Robert-Debré, GHU APHP-Nord Université Paris Cité, Paris
Dr. Mony Fahd (2023)	Pediatric Hemato-Immunology Department, Constituent center of the RC Hôpital Robert-Debré, Paris
Dr. Edouard Forcade (2023)	Hematology and Cell Therapy Department

	Center of expertise of the RC CHU de Bordeaux
Dr. Claire Galambrun (2019)	Pediatric Hematology-Oncology Department, Center of expertise of the RC Hôpital de la Timone, Marseille
Prof. Caroline Kannengiesser (2023)	Genetics Laboratory "Inherited Genetics of Telomeropathies" reference laboratory Hôpital Bichat, GHU APHP-Nord Université Paris Cité
Dr. Lise Larcher (2023)	Department of Hematological Biology "Integrative Diagnosis of Aplastic anemia " reference laboratory Hôpital Saint-Louis, Paris
Prof. Thierry Leblanc	Pediatric Hemato-Immunology Department Constituent center of the RC Hôpital Robert-Debré, Paris
Prof. Régis Peffault de Latour	Hematology Transplant Department Coordinating center of the RC Hôpital Saint-Louis, GHU APHP-Nord Université Paris Cité, Paris
Dr. Cécile Renard (2023)	Institute of Pediatric Hematology and Oncology Hospices Civil de Lyon
Dr. Flore Sicre de Fontbrune	Hematology Transplant Department Coordinating center of the RC Hôpital Saint-Louis, Paris
Prof. Jean Soulier	Department of Hematological Biology "Integrative Diagnosis of Aplastic anemia " reference laboratory Hôpital Saint-Louis, GHU APHP-Nord Université Paris Cité, Paris

Reviewers:

Prof. Gérard Socié	Hematology Transplant Department Hôpital Saint-Louis, GHU APHP-Nord Université Paris Cité, Paris.
Prof. Françoise Mechinaud (2023)	Pediatric Hemato-Immunology Department, Hôpital Robert-Debré, Paris

Members of centers of expertise:

Dr. Arthur Sterin (2023)	Pediatric Hematology/Immuno-Oncology Department Hôpital Timone Enfant, Marseille
Dr. Louis Terriou	Internal Medicine Department Hôpital Claude Huriez, Lille
Dr. Marie Angoso (2023)	Pediatric Hematology and Oncology CHU Bordeaux

Reference laboratories associated with the reference center

Prof. Lydie Da Costa	Laboratory of Biological Hematology “Diamond Blackfan Anemia Diagnosis” reference laboratory Hôpital Robert-Debré, GHU APHP-Nord Université Paris Cité, Paris

Physicians and other health professionals not affiliated with the reference center

Prof. Jean Claude Carel	Pediatric Endocrinology and Diabetology Department Hôpital Robert-Debré, Paris
Ms. Anais Ernault (2019)	Neuropsychologist Hôpital Robert-Debré, Paris
Dr. Anne-Claire Leprêtre (2019)	Etablissement Français du Sang [French National Blood Service] Hôpital Saint-Louis, Paris
Prof. Sandrine Passemard (2019)	Neurogeneticist Hôpital Robert-Debré, Paris
Dr. Maxime Eskenazi (2023)	General Practitioner Medical practice, Marseille

Patient association representatives

Mr. Farid Ouabdesselam (2023)	AFMF Association
Mr. Adel Ayad (2023)	French PNH Association-Aplastic anemia
Mr. Marcel Hibert	Association AFMBD

Reference center member:

Ms. Isabelle Brindel	Project Manager at the Reference center

Public Declarations of Interests

All participants in the development of the PNDS completed a public declaration of interests. Public declarations of interests are available online and on the reference center’s website.

Annex 2: Phenotype, genetics and hematological evolution of inherited Aplastic anemia

Syndrome	**Causal Genes (transmission)**	**Clinical phenotype**	**Hematological phenotype, frequency of aplasia**	**Characteristics of MDS/AML**	**Acquired cytogenetic & molecular abnormalities**
Fanconi anemia (See Annex 7)	**22 genes** *FANCA*, *FANCB*, *FANCC*, *FANCD1* (*BRCA2*), *FANCD2*, *FANCE*, *FANCF*, *FANCG* (*XRCC9*), *FANCI*, *FANCJ* (*BRIP1*), *FANCL* (*PHF9*), *FANCM*, *FANCN* (*PALB2*), *FANCO* (*RAD51C*), *FANCP* (*SLX4*), *FANCQ* (*XPF*), *FANCR* (RAD51), *FANCS* (*BRCA1*), *FANCT* (*UBE2T*), *FANCU* (*XRCC2*), *FANCV* (*REV7*/*MAD2L2*), *FANCW* (*RFWD3*). *AR except FANC-B linked to X and FANCR (RAD51) AD.*	Not constant (70%) IUGR, small size Thin and triangular face Café-au-lait or achromic spots (progressive onset) Malformations of bone (thumbs, fingers, forearms), kidney, heart, etc. Early-onset solid cancers : Head&neck squamous cell carcinoma in young adults; embryonic cancers in patients with bi-allelic BRCA2 (FANCD1).	Aregenerative anemia, macrocytosis, thrombocytopenia, pancytopenia (median age 7 years; 90% at 40 years). Hypoplastic/erythroblastic bone marrow. Non-specific dyserythropoiesis.	Hypoplastic MDS et AML. Cumulative incidence of MDS/AML: 30% at 40 years, mainly occurs from 15 years (except group D1/BRCA2 before 5 years)	Frequent unbalanced translocations with 1q and 3q gains, or 7q or 21q/*RUNX1* losses, mutations and translocations.

Telomeropathies/congenital dyskeratosis	*DKC1* (linked to X), *TERC* (AD & AR), *TERT* (AD & AR), *TINF2* (AD), *RTEL1* (AD & AR), *CTC1*(AR), *ACD* (AR & AD), *PARN* (AD & AR), *NOP10*(AR & AD), *WRAP53*(AR), *ZCCHC8*, *NHP2*(AR& AD), *STN1(AR)*, *RPA1(AR)*, *POT1(AR&AD)*, *DCLRE1B/Apollo(AR)*, Unknown in 30% of cases	Early childhood: Hoyeraal-Hreidarsson (HH) syndrome, Reves' syndrome (IUGR, pancytopenia, immune deficiency, ataxia, mental retardation, exudative retinopathy, etc.) and Coats Plus syndrome "Idiopathic" pulmonary fibrosis, pleuroparenchymal fibroelastosis, emphysema. Porto-sinusoidal vascular disease, hepatopulmonary syndrome, steatosis, hepatocellular carcinoma. Severe osteoporosis Cellular immune deficiency Neurological involvement: developmental delay, microcephaly, cerebellar hypoplasia and the spectrum is much broader (Bhala et al., 2019) Progressive skin, hair and nails disorders: hyperpigmentation, reticulate appearance, nail dystrophy and premature graying .	Isolated cytopenias (macrocytic anemia, thrombocytopenia), pancytopenias with poor bone marrow, "idiopathic" Aplastic anemia . Hypoplastic bone marrow.	Hypoplastic or non-hypoplastic MDS. MDS: 3%, probably underestimated but observed/expected ratio at 2663 (median age 35 years). AML secondary to MDS.	Various.
GATA2 or MonoMAC syndrome	*GATA2* (AD)	Immune deficiency: predisposition to infections with HPV and atypical mycobacteria and bacterial infections. Lymphedema. Alveolar proteinosis. Deafness. Vascular malformation. *Erythema nodosum*.	Rare typical aplastic anemia. Cytopenias (particularly neutropenia and monocytopenia). Bone marrow hypoplasia. Frequent moderate dysplasia.	Marked dysplasia. Atypical lipomyelomeningocele (LMMC) and leptomeningeal carcinomatosis (LMC) are described. MDS/AML: cumulative incidence estimated at 70% at 70 years (diagnosis 4 to 60+ years).	Chromosomal abnormalities: 7, 8, del(20q), 3q+. Somatic mutations of *ASXL1* and *SETBP1*
Shwachman-Diamond syndrome	*SDBS* (AR) 90%, *SRP54* (AR), *EFL1* (AR) <10% unknown	Exocrine pancreatic insufficiency (improves after 4 years in 50% of patients) Hepatomegaly Bone dysplasia and osteoporosis	Neutropenia (90%) Anemia (80%) Pancytopenia Hypoplastic bone marrow Minimal frequent dysplasia of the 3 myeloid lineages.	MDS defined only if >5% blasts (frequent granulocytic dysplasia without MDS). MDS/AML: cumulative incidence	*TP53* mutations; Chromosomal abnormalities (iso(7q) and del(20q)).

				between 18% and 36% at 30 years. Male gender subject predominance.	
Congenital amegakaryocytosis	*MPL* (AR)	Rare described malformations of unestablished causality	Most often severe neonatal thrombocytopenia. Absence of megakaryocytes Frequent evolution in the first years of life toward pancytopenia. Very high TPO level.	Not described	
	TPO (AR)	Not described	Moderate to severe thrombocytopenia is diagnosed in childhood or adolescence. Associated anemia and neutropenia of variable severity. The level of TPO decreases sharply. Reported response to eltrombopag.	2 cases of monosomy 7 during evolution (unpublished).	Not described
SAMD9/SAMD9L **(See Annex 10)**	*SAMD9* (AD)/*SAMD9L* (AD) gain of function mutations	Inconsistent. *SAMD9*: MIRAGE syndrome (MDS-infections-growth retardation-adrenal hypoplasia-genital abnormalities-enteropathy). *SAMD9L*: ATXPC (ataxia pancytopenia syndrome). Variable neurological abnormalities with cerebellar atrophy and ataxia, peripheral nervous system impairment, nystagmus. Behavioral disorders.	Cytopenias with poor bone marrow cellularity occurring during the first 2 decades, often associated with monosomy 7, sometimes with sudden onset. Unknown but incomplete penetrance and spontaneous long-term reversion. Variable profile but characterized by somatic mosaicism with uniparental disomy of chromosome 7q or inactivating mutations (loss of function, LOF) of the mutated GL *SAMD9* or *SAMD9L* alleles, causing spontaneous and lasting	MDS or AML. Current incidence unknown.	Monosomy 7, del(7q) or acquired uniparental disomy (UPD) of 7q, acquired loss of function Mutations of *SAMD9* or *SAMD9L*. Acquired somatic mutations of *SETBP1*, *RUNX1*, *ETV6*, *ASXL1* and

			reversion and correction of cytopenias.		classical myeloid genes in (pre)leukemic clones.
MECOM-associated syndrome or RUSAT2 (radio ulnar stenosis and amegakaryocytic thrombocytopenia)	*MECOM/EVI1* (= *MDS1 & EVI1* complex locus), isolated mutations or 3q26 deletion (AD)	Inconsistent. Radio-ulnar synostosis, clinodactyly, brachydactyly, craniosynostosis, facial dysmorphism, cleft palate, dysarthria, cardiac, vascular and renal malformations, Pierre Robin syndrome, and congenital deafness. Immune deficiency B with hypogammaglobulinemia and infections associated with specific mutations (8th zinc finger)	Thrombocytopenia, most often neonatal, of varying severity or severe aplastic anemia in the first years of life. Rare reported cases of moderate thrombocytopenia in adults.	Rare to date. MDS described in adults with moderate thrombocytopenia and extra-hematologic phenotype.	Not described
Diamond Blackfan anemia (See Annex 9)	*RP genes (n = 23): RPS7, RPS10, RPS15A, RPS17, RPS19, RPS20, RPS24, RPS26, RPS27, RPS28, RPS20* *RPL5, RPL8, RPL11, RPL15, RPL18, RPL26, RPL27, RPL31, RPL35, RPL35A* *Other genes: GATA1, TRS2 and HEATR3* (20% to 45% familial forms) Other inherited erythroblastopenia genes: *CECR1/ADA2, EPOR*	Small size (Inta-uterine growth retardation IUGR) Malformations (50% of patients; mainly affecting: thumbs, Ear Nose an Throat (ENT) region (cleft lip and palate), urogenital organs and heart)	Typically early symptomatic macrocytic aregenerative anemia (1st year) with erythroblastopenia. NB: possible *hydrops fetalis* Hematologic response to corticosteroids (60%) or possible spontaneous therapeutic independence. Possible moderate neutropenia or thrombocytopenia. Exceptional typical aplastic anemia.	MDS and AML secondary to MDS. From 30 years old. MDS: O/A ratio = 287 AML: O/A ratio = 28	Few data, most often complex karyotypes; *TP53* mutations

	and *TP53*				
Autosomal dominant AA SRP72	*SRP72* (AD)	Deafness, labyrinthitis.	Pancytopenia, macrocytosis.	MDS with multilineage dysplasia, mainly without excess of blasts. MDS/AML: incidence unknown. 3/6 cases.	Normal karyotype.
DDX41	*DDX41* (AD>AR)	Not present	Rare but described aplastic anemia.	AML or MDS in the second part of life (>40 years). Incomplete penetrance, larger effect on predisposition to myeloid neoplasms in males than in females.	Normal karyotype (50%) or non-specific abnormalities (7, 8, del20q). Somatic mutation of the 2nd allele of *DDX41* in more than50% of cases:.
ERCC6L2	*ERCC6L2* (AR)	Not constant. Microcephaly, facial dysmorphism, developmental delay, learning disabilities, ataxia. Retinal abnormalities. Possible growth retardation. Urinary abnormalities.	Discovery in childhood and adolescence. Moderate to severe thrombocytopenia. Moderate inconsistent neutropenia. Moderate inconsistent anemia. Possible macrocytosis. High HbF. Hypocellular marrow.	MDS or AML	TP53 mutation, complex karyotypes

DNAJC21	*DNAJC21* (AR)	Growth retardation, developmental delay, recurrent infections, abnormalities of the skin, hair (thin and sparse) and teeth (conical or microdontia). More rarely: hearing loss, retinal abnormalities, pancreatic insufficiency, hepatic cirrhosis, osteopenia, skeletal abnormalities, congenital hip dysplasia, ligament hyperlaxity and cryptorchidism.	Bone marrow hypoplasia of varying severity. Various cytogenetic abnormalities (del(17p), del(20q), der(15), trisomies 8, 21, X not associated with signs of dysplasia.	Few cases of AML describeb.	Not described
LIG4	*LIG4* Hypomorphic mutations (AR)	Very common but inconsistent: microcephaly, IUGR, small stature, dysmorphism with bird-like or Seckel syndrome-like facies, hypogonadism, bone marrow hypoplasia, syndactyly, polydactyly, congenital hip dysplasia, photosensitivity, psoriasis, eczema, hypopigmentation, numerous plantar warts. Radiosensitivity. Inconsistent: Combined immunodeficiency with hypogammaglobulinemia and hyperIgM (CID see SCID).	Bone marrow hypoplasia with anemia and thrombocytopenia. Lymphopenia with marked B lymphopenia	Not described	

AD: autosomal dominant; AR: autosomal recessive.

Annex 3: List of searched genes

This non-fixed list represents the genes currently sought in patients with suspected inherited aplastic anemia.

- *ANKRD26*, *RUNX1*,
- *ATG2B*,
- *ATM*, *ATR*, *ATRX*,
- *BRCA1*, *BRCA2* and other Fanconi genes
- *DDX41*,
- *ERCC6L2*,
- *GATA2*,
- *LIG4*, *DNAJC21*,
- *MECOM*,
- *MPL*, *THPO*
- *SAMD9*, *SAMD9L*,
- *SBDS*, *SRP54*
- *SRP72*,
- *DKC1, TERC, TERT, TINF2, RTEL1, CTC1, TPP1, PARN, USB1, NOP10, TCAB1, NHP2 SHQ1, STN1.*

Annex 4: Treatment with anti-thymocyte globulin - ciclosporin and patient follow-up

1- Treatment with ATG, ciclosporin +/- eltrombopag

Immunosuppressive therapy with a combination of:

- **Horse ATG**, ATGAM, Pfizer: 40 mg/kg daily x 4 days. The product is off-label and has a temporary authorization for use.
The recommended infusion time is 12 hours with premedication with dexchlorpheniramine and methylprednisolone (1 mg/kg daily)
Scope and blood pressure monitoring every 15 minutes during ATG (especially on D1).

In case of intolerance:
- Discontinue administration
- Repeat premedication with dexchlorpheniramine and methylprednisolone
- Resume administration as soon as clinical signs improve

If ATGAM is not available and treatment needs to be started, rabbit ATG may be used.

Corticosteroid therapy is continued orally from the day after the last dose of ATG at the dose of 1 mg/kg daily for the prevention of serum sickness and will be reduced by 1/3 on the 10th day and by 2/3 of the starting dose on the 20th day and discontinued on the 30th day in the absence of serum sickness.

Rabbit ATG (Thymoglobulin®, 3.5 mg/kg daily x 5 days) is offered as 2nd line treatment in case of failure or where there is no access to ATGAM. Administration is based on the same recommendations as for horse ATG

Recommended anti-infective prophylaxis in combination with ATG:
- triazole prophylaxis is recommended for the duration of corticosteroid therapy and in patients with neutrophils below 0.5 G/L due to the risk of invasive fungal infection,
- prophylaxis for HSV and VZV infections with valaciclovir and pneumocytosis with co-trimoxazole (atovaquone in case of allergy) is recommended up to 3 months after ATG.

Vaccinations are not contraindicated after ATG, except for live attenuated vaccines. However, it is recommended to wait 3 months because of the risk of loss of efficacy. There is no evidence to support systematic boosters or a new vaccination schedule after ATG-ciclopsorin treatment.
Live attenuated vaccines are contraindicated after ATG and during treatment with ciclosporin: proper T lymphocyte reconstitution should be checked before administering these vaccines.

- **Ciclosporin**:
Administered from D5 of treatment with ATGAM (or rabbit ATG).
Starting dose of 6 mg/kg daily (5 to 10) in two doses per day orally or 3 mg/kg daily intravenously (IVES/24 hours or 2 SIV over 2 hours).
Residual (H+12) recommended ciclosporin target: 200 to 300 ng/ml, which should, however, take into account renal, blood pressure and neurological tolerance. Measurements are carried out on average once a week until dose is balanced. A measurement of the residual level is requested at H12 from the previous dose. Once the response is obtained and the decrease is established, the dose adjustment will only be made according to the clinical and renal tolerance and measurements are no longer carried out systematically. Please note that slightly elevated urea is often a good marker of effective levels. Monitoring of ciclosporin levels is no longer useful once the decrease begins.

In combination with posaconazole or voriconazole, the dose should be halved.
In subjects over 70 years of age or in patients with pre-existing renal insufficiency, it is advisable to start at 3 mg/kg daily, adapting to renal and blood pressure tolerance and gradually increasing the dose.

In case of acute renal failure or other severe complications, temporary discontinuation should be considered pending full recovery and treatment should be recommenced gradually.

- **Eltrombopag:**

In adult patients, eltrombopag is initiated at a dose of 75 mg per day as a single dose starting on D10 of horse ATG-ciclo, informing the patient that the therapy should be taken on an empty stomach (2 hours before and after) and in particular without consuming any dairy products. If tolerance is good, the dose should be increased to 150 mg per day in 1 dose after 10 to 15 days. The dose may be reduced in case of gastrointestinal or hepatic intolerance: it is recommended to seek the maximum tolerated dose in steps of 25 mg.
In case of failure after 3 months at the dose of 150 mg, the dose may be increased to 225 mg if tolerance is satisfactory in the absence of indication for another therapy (especially HSC allograft).

2- Hematologic follow-up

Therapy with ATG and ciclosporin requires hospitalization for approximately 3 to 6 weeks in a protected hematological unit according to the degree of cytopenia and the complications developped. Discharge from the service is considered when the infectious risk is under control and transfusion support with red blood cells or platelets is being provided once a week or less. Follow-up in a day hospital is usually carried out weekly for a period of approximately 2 months (3 months from the start of therapy), which varies according to the frequency of transfusion support and the maintenance of the central catheter.

Once transfusion independence is achieved, follow-up is carried out in consultation at least every 3 months during therapy and then at least twice a year for life.

Ciclosporin tapering schedule: Ciclosporin is usually continued for a period of one year at full dose after the last transfusion and then, in a patient with a good response, gradually decreased by about 10–20% every 3 months and discontinued at the end of the second year (ciclosporin assays are then unnecessary except in case of toxicity). Some responder patients are dependent on ciclosporin and need to maintain long-term therapy.

Eltrombopag tapering schedule: in triple therapy, it is recommended to taper eltrombopag if in complete remission after 3 months, in steps of 75 mg every 2 months. For patients with a partial response, slower dose levels may be achieved (100 mg for 2 months, then 50 mg for 2 months, if the response is maintained, before complete discontinuation).

Eltrombopag has an iron chelating effect. Monitoring of ferritin levels during treatment is recommended, as iron deficiency anemia has been described.

Biological monitoring initially requires a blood count, blood electrolytes, renal function tests (urea and creatinine) and complete liver function tests at day hospitals or during consultations. Longer-term follow-up requires PNH clone testing once a year for asymptomatic patients, or at the time of presenting symptoms (attack of abdominal pain, hemoglobinuria or other warning signs). A bone marrow differential cell count with conventional cytogenetics should be performed every 12 to 18 months, given the risk of clonal evolution over the long-term, with no suggested end date for this systematic follow-up (lifelong follow-up).

3- Non-hematologic follow-up

The risk of renal failure is high and requires adjustment of the dose of ciclosporin combined with abundant hydration. The occurrence of thrombotic microangiopathy is rare in this context but should be discussed in the event of arterial hypertension that is difficult to control, signs of hemolysis and schistocytes in the blood smear. Discontinuation of ciclosporin is the only therapy and no effective

alternative exists. Another calcineurin inhibitor, tacrolimus (Prograf®), has sometimes been reintroduced some time after the acute episode without recurrence of microangiopathy, but its effectiveness has not been scientifically validated in this indication. In the long-term, about 5% to 10% of patients have chronic renal failure.

Another classic complication of ATG-ciclosporin therapy or after BMT is osteonecrosis of the hip. This manifests in the form of mechanical, unilateral hip or knee pain (projected pain), which can be very debilitating. The diagnosis is confirmed by MRI. Initial management consists of relieving the pain, which most of the time recedes in 4 to 6 weeks. The persistence of pain and disability after this period may require a total hip replacement in some patients.

It is also important to monitor patients' skin (squamous cell carcinoma), although this risk has not been clearly identified as related to ciclosporin therapy. Finally, it is necessary to keep an eye out for signs of cataract (corticosteroids at the time of ATG), such as glare at the end of the day in particular.

4- Fertility and pregnancy after ATG-ciclosporin treatment

Outside the context of allogeneic transplantation, pregnancy is a high-risk situation because of the risk of relapse or worsening of aplasia, which may not be reversible after pregnancy. Although there are no large studies on pregnancy and ciclosporin, the existing data on teratogenic risk are reassuring. It is currently the only treatment that can be offered to pregnant women. The main risk is that of maternal-fetal infections. Ciclosporin crosses the placental barrier and fetal blood levels are about one-third of maternal blood levels (https://lecrat.fr/spip.php?page=article&id_article=711). In men, fertility is not generally impaired after immunosuppressive therapy.
There are insufficient teratogenicity data for eltrombopag to suggest initiation or continuation of eltrombopag therapy during pregnancy. In case of life-threatening transfusion-resistant thrombocytopenia, its use in pregnant women can be discussed on a case-by-case basis among colleagues and ideally in the context of the national MDT meetings.

Annex 5: Allogeneic hematopoietic stem cell transplantation and patient follow-up

1- Choice of graft

The source of hemotopoietic stem cells for this type of transplant is bone marrow, whether for acquired or inherited Aplastic anemia . Indeed, there is no need for the graft to react against the disease (graft versus leukemia – GvL) and therefore for the graft to react against the host (graft versus host disease – GvHD), which can only lead to deleterious effects in this disease. Peripheral stem cell transplantation increases this risk due to the greater presence of lymphocytes, which are directly responsible for GvHD. As for placental blood, the lymphocyte count is extremely low, which certainly protects against GvHD, but leads to a prolonged immune deficiency that can result in potentially serious infections, which is why this source of cells is not prioritized in this type of transplant.

2- Conditioning and prophylaxis for graft versus host disease

The currently recommended conditioning and prophylaxis for graft versus host disease are detailed in the table below.

Idiopathic Aplastic anemia	
Geno-identical donor	CY 50 mg/kg daily x 4d (D-5 to D-2) and rATG[a] (12.5 mg/kg) Immunosuppression after allograft combines CSA (3 mg/kg daily IV from D-3) **with methotrexate (15 mg/m² D1, 10 mg/m² on D3, D6 +/- D11)**
	For patients over 40 years of age[b], the alternatives are: - FLU 30 mg/m² daily x 4d (D-6 to D-3), CY 300 mg/m² daily x 4d (D-6 to D-3) and rATG (5–10 mg/kg) (FCA) **or** - FLU 30 mg/m² daily x 4d (D-6 to D-3), CY 300 mg/m² daily x 4d (D-6 to D-3) and alemtuzumab 0.2 mg/kg daily x 5d (D-7 to D-3) (FCC) Immunosuppression after allograft combines CSA (3 mg/kg daily from D-1) with a short regimen of methotrexate (10 mg/m² D1, 8 mg/m² D3 and D6), or CSA alone if alemtuzumab is used
Matched unrelated donor (HLA 10/10) as 2nd line treatment or more	- FLU 30 mg/m² daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3) and rATG 3.75 mg/kg daily x 2d (D-4 to D-3) and TBI 2 gray (D-1) (FCA TBI) **or** - FLU 30 mg/m² daily x 4d (D-6 to D-3), CY 300 mg/m² daily x 4d (D-6 to D-3) and alemtuzumab 0.2 mg/kg daily x 5d (D-7 to D-3) (FCC) Immunosuppression after allograft is similar to that used for geno-identical donors.
	For patients under 14 years of age: - FLU 30 mg/m² daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3) and rATG 3.75 mg/kg daily x 2d (D-2 to D-1) **or** - FLU 30 mg/m² daily x 5d (D-7 to D-3), CY 60 mg/kg daily x 2d (D-3 to D-2) and alemtuzumab 0.3 mg/kg daily x 3d (D-6 to D-4) (FCC, pediatric version). Immunosuppression after allograft is similar to that used for geno-identical donors
Matched unrelated donor (HLA 10/10) in first line treatment	- Upfront MUD protocol: *For patients under 14 years, FLU 30 mg/m² daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3), and rATG 3.75 mg/kg daily x 4d (D-6 to D-3).

	*For patients over 14 years, FLU 30 mg/m^2 daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3), and rATG 3.75 mg/kg daily x 2d (D-4 to D-3) and TBI 2 gray (D-1) Immunosuppression after allograft combines CSA from D-1 + MTX 10 mg/m^2 D1, MTX 8 mg/m^2 D3 & D6 - outside protocol: FLU 30 mg/m^2 daily x 5d (D-7 to D-3), CY 60 mg/kg daily x 2d (D-3 to D-2) and alemtuzumab 0.3 mg/kg daily x 3d (D-6 to D-4) (FCC, pediatric version) Immunosuppression after allograft consists of CSA alone.
Alternative donor	
Unrelated non-compatible donor (HLA 9/10)[c]	- FLU 30 mg/m^2 daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3) and rATG 3.75 mg/kg daily x 2d (D-4 to D-3) and TBI 2 gray (D-1) (FCA TBI) **or** - FLU 30 mg/m^2 daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3) and alemtuzumab 0.2 mg/kg daily x 5d (D-7 to D-3) and TBI 2 gray (D-1) (FCC TBI) Immunosuppression after allograft is similar to that used for geno-identical donors
Cord (CBU)	FLU 30 mg/m^2 daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3) and rATG 2.5 mg/kg daily x 2d (D-4 to D-3) and TBI 2 gray (D-1) (Peffault et al., 2018) Immunosuppression after allograft consists of CSA alone
Haplo-identical graft	rATG 0.5 mg/kg daily on D-9, rATG 2 mg/kg daily x 2d (D-8 to D-7), FLU 30 mg/m^2 daily x 4d (D-6 to D-2), CY 14.5 mg/kg daily x 2d (D-6 to D-5) and TBI 2 gray (D-1) Immunosuppression after allograft consists of CY 50 mg/kg daily x 2d (D3 and D4), tacrolimus (D5) and mycophenolate (D5 to D35) (DeZem et al.; 2015)
Inherited aplastic anemia	
Fanconi's disease	
Geno-identical donor	FLU 30 mg/m^2 daily x 3d (D-4 to D-2), CY 10 mg/kg daily x 4d (D-5 to D-2) Immunosuppression after allograft combines CSA (3 mg/kg daily IV from D-1) and mycophenolate (D+1 to D+45).
Matched unrelated donor (HLA 10/10) in first line treatment	FLU 30 mg/m^2 daily x 4d (D-6 to D-3), CY 10 mg/kg daily x 4d (D-6 to D-3) and rATG 2.5 mg/kg daily x 2d (D-4 to D-3) and TBI 2 gray (D-1) (adults) Patients <14 years of age BU 75% of dose according to SmPC/kg x 4 daily for 2 days (D-7–D-6), FLU 30 mg/m^2 x 3d (D-5 to D-3), CY 10 mg/kg daily x 4d (D-5 to D-2) and rATG 2.5 mg/kg daily x 4d (D-4 to D-1) (children) Immunosuppression after allograft combines CSA (3 mg/kg daily IV from D-1) and mycophenolate (15 mg/kg x 2 daily (D1 to D45).
Haploidentical	With CD34 selection/T depletion αβ in vitro: BU 0.6 to 0.8 mg/kg dose x 2 doses daily for 2 days (D-7 to D-6), FLU 35 mg/m^2 daily from D-5 to D-2, CY 10 mg/kg daily from D-5 to D-2, rATG 2.5 mg/kg daily from D-5 to D-2, GvHD prophylaxis with ciclo With T depletion in vivo: Campath 0.1 mg/kg D-5, 0.2 mg/kg x 2d (D-4 and D-3), FLU 30 mg/m^2 D-6 to D-2, TBI 2Gy on D-1, PTCy 25 mg/kg daily D3 and D4, GvHD prophylaxis with ciclo MMF and Campath 0.1 mg/kg D5

Cord (CBU)	CY 10 mg/kg daily x 4d (D-6 to D-3), FLU 30 mg/m^2 daily x 4d (D-6 to D-3), and rATG 2.5 mg/kg daily x 2d (D-2 to D-3) and TBI 2 gray (D-1) Immunosuppression after allograft combines CSA 3 mg/kg daily (from D-3) and mycophenolate 30 mg/kg daily (D1 to D28).
Telomeropathies	
Geno-identical donor and matched unrelated donor (HLA 10/10)	Bone marrow failure - FLU 30 mg/m^2 daily x 5d (D-7 to D-3), CY 60 mg/kg daily x 2d (D-3 to D-2) and alemtuzumab 0.3 mg/kg daily x 3d (D-6 to D-4) (FCC, **pediatric version**) - FLU 30 mg/m^2 daily x 4d (D-6 to D-3), CY 30 mg/kg daily x 4d (D-6 to D-3) and alemtuzumab 0.2 mg/kg daily x 4d (D-6 to D-3) (FCC, **adult version**); Immunosuppression after allograft consists of CSA alone

[a]-rATG corresponds to rabbit ATG (Thymoglobulin® (Genzyme))
[b]-Alternative conditioning regimens may be used in younger patients with comorbidities
NB: A single injection of rituximab (150 mg/m^2) is given in some centers for preventive reasons if donor or recipient are seropositive for EBV in alternative transplants and if alemtuzumab is used on D5 or D6. (grade D).

Abbreviations: ATG: Anti-thymocyte globulins; D: Day; CSA: ciclosporin; CY: cyclophosphamide; FLU: fludarabine; MTX, Methotrexate; TBI: total body irradiation; HLA: major histocompatibility system

3- Characteristics of post-allograft follow-up in Aplastic anemia

The immediate post-allograft period requires residual ciclosporin levels of approximately 200 to 250 ng/mL for a period of approximately 9 to 12 months post-transplant. This therapy is then decreased gradually in the case of chimerism mainly stable donor until discontinuation over a period of about 6 to 12 months.

It is not uncommon to observe mixed chimerism after transplantation (persistence of recipient cells >5% on whole blood or lymphocyte fraction), especially when using alemtuzumab-based conditioning. The management of mixed chimerism is very specific to Aplastic anemia : If the mixed chimerism is stable and the blood count is satisfactory (Hb >10 g/dL, neutrophils >1 G/L and platelets >100 G/L), it is possible to gradually decrease the immunosuppression every 3 months while monitoring the chimerism and blood count at the same rate. In the case of stability, the reduction can be continued at the same rate and with the same monitoring; if the chimerism or blood count decrease, the ciclosporin must be increased again to hopefully reverse the phenomenon. Tapering will resume once the blood count and chimerism have stabilized.

Graft failure within 30 days of grafting, or graft rejection within 6 months of grafting (exceptional but possible in the longer term), are not uncommon in Aplastic anemia , due to the autoimmune nature of acquired Aplastic anemia and also to the multiple transfusions received prior to grafting, whatever the type of aplasia. In these cases, a second transplant is possible depending on the general condition of the patient and the hematological condition (residual hematopoiesis or severe pancytopenia).

4- Characteristics of post-allograft follow-up in the context of inherited Aplastic anemia

Patients with inherited Aplastic anemia are at high risk of cancer, including after bone marrow allograft. This risk is extremely significant for patients with Fanconi disease and represents the leading long-term cause of death after allograft in these patients. This justifies close multidisciplinary monitoring, including stomatologists and gynecologists, at least twice a year for life. Squamous cell carcinomas are actually the most common in this context and only complete initial surgical management will improve the long-term prognosis. Unfortunately, once there is a local invasion or remote spread, therapy is often only palliative.

5- Preservation of fertility

Refer to paragraph "Management of specific situations" on p. 29.

Annex 6: Diagnostic, therapeutic and follow-up management of PNH patients

1- Diagnosis, evolution and monitoring

Paroxysmal nocturnal hemoglobinuria (PNH), or Marchiafava-Micheli disease, is an acquired clonal hematopoietic stem cell disease characterized by corpuscular hemolytic anemia, Aplastic anemia and frequent occurrence of thrombosis. PNH is a rare disease whose prevalence is estimated at 1/70,000 inhabitants in Europe.
PNH is due to an acquired somatic mutation in the *PIG-A* gene. This results in a blocking of the synthesis of glycophosphatidylinositol (GPI) anchoring molecules, which are responsible for the attachment of numerous proteins to the cell surface. These include CD59 and CD55, which are complement inhibitory proteins that normally prevent the final assembly of the membrane attack complex (MAC). PNH is the consequence of a deficiency of membrane anchoring of these two proteins, manifested clinically by intra-vascular hemolysis.
The classic clinical triad involves a combination of this hemolysis with medullary hypoplasia of varying severity and thromboses (in particular the hepatic, abdominal, cerebral and dermal veins) and signs of dystonia. Generally, anemia manifests in the form of pallor, fatigue and shortness of breath on exertion. Hemoglobinuria can result in darker urine, which is usually more marked in the morning (about 25% of cases), and, in rare cases, renal failure. It is sometimes confused with hematuria, which can lead to urological or nephrological investigations. Jaundice may be present in connection with free hyperbilirubinemia secondary to hemolysis. Depending on its location, thrombosis (which affects 30–40% of patients without therapy) can lead to dyspnea, abdominal pain, hepatomegaly, ascites and headaches. PNH is a chronic disease that progresses through outbreaks of hemolysis. Crisis periods can be triggered by various factors, particularly infections, even common ones, vaccination, surgical intervention, etc. Pregnancy is a high-risk situation for thrombotic or hemorrhagic complications, especially the peripartum period (de Guibert et al., 2011).
PNH is intimately linked to idiopathic Aplastic anemia : at the diagnosis of MA, 20% to 30% of patients have a PNH clone; 20% will develop PNH during follow-up, and conversely, 30% of those with PNH will develop MA during follow-up (Peffault de Latour et al., 2018).
Patients with PNH have a risk of clonal medullary progression with a cumulative incidence of acute myeloblastic leukemia and myelodysplastic syndromes estimated at 10% at 15 years (but which may occur early during follow-up).
PNH can occur at all ages, but it particularly affects young adults (median age of 30 years at diagnosis) and affects slightly more women than men. It is very rare in children under 15 years of age, which justifies discussion of these cases at the Aplastic anemia MDT meetings.
Prognosis is good in the age of eculizumab therapy and survival is close to patients of the same age and gender.
Diagnosis is performed by the reference technique of flow cytometry on blood. Searching for a PNH clone in the marrow or another fluid is not useful (when they mature, cells may be GPI negative). A search for the PIG-A mutation is of no interest because it is not unique and is complex and expensive, and has no place in the management of patients on a daily basis.
A PNH clone search must be performed in the case of:

1) unexplained Coombs-negative hemolytic anemia, especially if it is associated with hemoglobinuria, iron deficiency, other cytopenias or thrombosis
2) splanchnic thrombosis or Budd Chiari syndrome
3) thrombosis associated with Coombs-negative hemolytic anemia
4) a diagnosis of Aplastic anemia , then annually if the patient has not received an allograft
5) a diagnosis of single-line refractory cytopenia

Follow-up of a patient with a PNH clone requires:

- a blood count every 3 months
- a bone marrow differential cell count with cytogenetic study every 12 to 18 months
- an annual evaluation of the PNH clone

- regular assessment of clinical signs of complications: asthenia, dyspnea, headache, abdominal pain, dysphagia, erectile dysfunction and especially frequency of hemolysis episodes
- education regarding situations where there is a risk of thrombosis (plane travel, immobilization, pregnancy, smoking, etc.) and symptoms requiring emergency consultation (fever, abdominal or chest pain, headache, hemorrhagic syndrome), adherence to treatments and proper follow-up of the vaccination schedule

2- Treatment

Non-specific treatment involves a combination of:

- prescription of oral folates to limit the risk of deficiency due to intake
- prescription of non-thrombogenic contraception, if necessary, for women of childbearing age
- a vaccination against influenza to limit the risk of acute hemolysis

C5 inhibitors are the specific reference treatment for PNH: eculizumab (Soliris®) and ravulizumab (Ultomiris®) are now available in France without distinction for adults (ravulizumab is not currently reimbursed (as of December 2022) in children).

1) eculizumab (Soliris®) at a dose of 600 mg intravenously once weekly for 4 weeks, followed by 900 mg every 2 weeks from week 5 of therapy. This treatment must not be abruptly interrupted due to the risk of severe hemolytic disease. Infusions may be delayed by 48 hours from the theoretical date, but this period must not be exceeded.

2) ravulizumab (Ultomiris®) may be used with new patients or those receiving eculizumab. A weight-dependent loading dose is administered intravenously at the 1st injection, followed by a 1st maintenance dose, also adapted to weight, on D15. Subsequent infusions are given at the maintenance dose every 8 weeks with a tolerance of +/- 7 days. Follow-up recommendations and prophylaxis are similar to those for eculizumab.

Indications to start specific treatment (eculizumab or ravulizumab) in a patient with a PNH clone greater than 10% in size are:

- hemolysis responsible for anemia requiring transfusion support
- arterial or venous thrombosis
- a significant PNH clone (>50%) associated with clinical signs of dystonia or biological signs of active hemolysis
- pregnancy, due to the risk of severe complications for mother and child

The introduction of a specific therapy automatically requires 100% management.

Patients on complement inhibitors are at risk of meningococcal infections.

At least 15 to 21 days before initiation of therapy, patients must be given a tetravalent conjugate vaccine against meningococcal ACWY135 serotypes and a meningococcal B vaccine. A booster of meningococcal B vaccine should be provided 1 month after the first injection. The Reference Center (CR) for Rare Diseases recommends that boosters of the 2 vaccines should be provided every 3 years thereafter for the entire duration of treatment.

In cases where anti-C5 therapy must be commenced urgently, particularly in the case of thrombosis threatening the vital or functional prognosis, effective treatment of meningococcal infection must be started (with beta-lactam antibiotics or quinolones) and continued for up to 14 days after the first vaccinations.

Daily preventive treatment with penicillin is recommended for the duration of therapy (oracillin or amoxicillin in case of macrolide allergy), due to the persistence of a risk of meningococcal infection.

Vaccinations against pneumococcal disease and *Haemophilus influenzae* are also recommended, as well as vaccination against seasonal influenza, the latter being a risk factor for meningococcal infection and causing outbreaks of hemolysis.

An emergency card must be given to the patient and they must be informed of the risks and situations requiring emergency consultation (fever, chills, unusual headaches, malaise, etc.).

There are no published data relating to the use of ravulizumab in the acute phase of suprahepatic vein thrombosis: Budd Chiari syndrome in the acute phase requires close monitoring of the efficacy of C5 inhibitors (CH50), probably due to an increase in the volume of distribution (Plessier et al., 2022). As this monitoring is not possible with ravulizumab, the CR supports the use of eculizumab in this situation. It is recommended to seek hepatological advice from the CR for vascular hepatic diseases.

Therapy efficacy cannot be evaluated for 6 months; if hemolytic anemia persists during this period, simple monitoring and transfusions at a level appropriate to the patient's clinical tolerance are recommended.

The persistence of symptomatic hemolytic anemia after 6 months of therapy requires verification of

1) the absence of chronic inflammation responsible for an increase in the synthesis of complement proteins and an incomplete blockage of the latter
2) the absence of additional causes of hemolysis (medicines)
3) with eculizumab, the CH50 assessment, which should theoretically be less than 10% (this lab test does not assess the efficacy of terminal complement blockade with ravulizumab)

In patients with chronic persistent symptomatic hemolysis and CH50 above 10% without associated inflammation, an increase in the eculizumab dose to 1200 mg may be proposed. The latter should be reassessed after 6 months. Ravulizumab at the proposed doses is equivalent to the 1200 mg dose of eculizumab, therefore dose increases are not proposed.

In the event of lack of efficacy, these patients should be discussed at the national MDT meetings in order to refer them to other therapeutic protocols (proximal inhibitors) or to early access to pegcetacoplan (Aspaveli®).

The development of non-hemolytic anemia, neutropenia and/or thrombocytopenia should undergo a medullary assessment to look for Aplastic anemia or clonal evolution. The management of Aplastic anemia occurring in a patient with PNH follows that of idiopathic Aplastic anemia point by point and thus depends on its severity, possible transfusion needs, the age of the patient and the existence of a geno-identical donor.

If possible, scheduled surgery should be performed within 48 to 72 hours after eculizumab injection and within two weeks following ravulizumab injection. The CBC and LDH should be monitored for the following 2 weeks and an additional eculizumab injection should be discussed in the event of active hemolysis (LDH >3N or hemolytic anemia requiring transfusion in the presence of signs of hemolysis). In the event of emergency surgery, an additional pre- or postoperative eculizumab injection should be provided depending on the time since the last eculizumab or ravulizumab injection, thrombotic risk and hemolysis markers. Thrombosis prophylaxis with LMWH should be initiated.

Discontinuation of therapy: discontinuation of therapy should only be considered after allogeneic hematopoietic stem cell transplantation or in exceptional cases of complete disappearance of the clone (to be confirmed on 2 samples taken 6 months apart). In all other cases, discontinuation of therapy exposes the patient to the risk of severe acute hemolysis and potentially serious complications.

C3 inhibitor, pegcetacoplan (Aspaveli®): this therapy, which is administered subcutaneously twice a week, has been approved for early access for PNH patients with hemoglobin of less than 10.5 g/dl in connection with persistent hemolysis who have been taking C5 inhibitors for at least 3 months. A phase 3 study randomizing pegcetacoplan and eculizumab in patients with persistent hemolysis responsible for anemia demonstrated the superiority of this therapy in these patients (increase of almost 4 points in hemoglobin in the pegcetacoplan arm compared to the control group with transfusion independence in nearly 85% of patients (versus 15% in the control group) after 16 weeks of therapy). However, this therapy has been associated, on the one hand, with the occurrence of severe hemolytic episodes complicated by organ failure, mainly during infectious episodes, as a result of which prescription is subject to validation by the CR, and, on the other hand, with the occurrence of interstitial pneumopathies that require special monitoring. Initiation of therapy requires prior vaccinations against pneumococcus and *Haemophilus influenzae*. The first two months of therapy should be carried out in a hospital setting: the first month of therapy consists of dual C5 and C3 inhibitor therapy followed by the discontinuation of the C5 inhibitor. This

discontinuation may be associated with a relapse of hemolysis. Advice for the implementation and follow-up of this therapy was published in May 2022 by the CR (prescription treatment guide).
Medical care must be associated with:

- psychological care if the patient so wishes,
- social care, because of the difficulties inherent in the pathology and the impact of the therapy on the professional and social life of the patient: a part-time therapist and a referral to the MDPH may be justified,
- information on the existence of patient associations and professional/patient meeting days within the framework of the reference center and the MaRIH rare diseases health network.

3- Monitoring of infections

The risk of meningococcal infection is not zero, despite vaccines and antibiotic prophylaxis. Any fever should prompt consideration of this infection.
The etiological assessment includes a complete clinical examination and systematic blood cultures. In case of febrile headaches, a lumbar puncture should be performed. If this is not possible (AVK), treatment with IV meningeal dose C3G should be initiated. Similarly, in case of signs of severe sepsis, a C3G injection should be provided at the slightest doubt, pending the results of blood cultures. In case of contact with a person infected with meningococcus, prophylactic treatment with rifampicin or ciprofloxacin is mandatory.
Apart from these mandatory vaccinations against meningococcal serotypes ACWY135 and B before therapy and then every 3 years, the recommended vaccinations are: *Haemophilus influenzae*, pneumococcus and seasonal influenza. They are mandatory for patients receiving proximal inhibitors (C3, factor B and factor D inhibitors).
Live attenuated vaccines (yellow fever, measles, mumps and rubella (MMA), etc.) are not contraindicated with eculizumab but are contraindicated with immunosuppressants (ciclosporin). Advice from a specialist is recommended.
Other vaccines are allowed.

4- Pregnancy

Pregnancy is a high-risk situation in patients with PNH and should be discussed in advance with patients of childbearing age. The risks are multiple:

- Worsening of aplasia during pregnancy, which may require transfusions of red blood cells and platelets to maintain a hemoglobin threshold greater than 10 g/dl and a platelet threshold greater than 25 G/L.
- Worsening of aplasia may or may not be reversible after delivery and may require aplasia therapy after delivery
- Risk of thrombotic and hemorrhagic complications for the mother, especially during the last trimester of pregnancy and 6 weeks postpartum
- Risk of fetal complications with increased risk of intrauterine growth retardation, preterm delivery and fetal loss

The management of pregnancy in a patient with hemolytic PNH requires:

1- Initiation of eculizumab treatment if the patient is not already being treated, and dose escalation to 1200 mg at the end of the second trimester of pregnancy and until delivery
2- Initiation of antithrombotic prophylaxis with LMWH at the end of the 2nd trimester of pregnancy and up to 6 weeks after delivery
3- Care in a level 3 maternity unit and notification of the obstetric team
4- Administration of an early infusion of 600 mg eculizumab prior to delivery if the last infusion was more than 7 days previously

Breast-feeding is not contraindicated with eculizumab.

In the absence of data on the use of ravulizumab in pregnant women, the CR recommends the use of eculizumab in pregnant women. The switch will be made on the theoretical date of the next course of ravulizumab, but no later than the 30th week of amenorrhea.
From a regulatory perspective, there are no studies to conclude that eculizumab is safe during pregnancy, although no additional risk of malformations for the unborn child or complications for the future mother has been reported to date. (product-information/soliris-epar-product-information_fr.pdf). For the reference center, the benefit/risk balance is in favor of eculizumab therapy (Kelly et al., 2015).
In women, estroprogestational contraceptives are contraindicated due to the risk of thrombosis.
Permitted contraceptives are:

- intrauterine devices, if there is a low risk of bleeding and no neutropenia
- female and male condoms
- the progestin-only pill (except in the event of thrombocytopenia, because of the risk of spotting) or the high-dose progestin pill
- luteinizing hormone-releasing hormone (LHRH) agonists

Emergency contraception based on oral progestins taken within 72 hours is allowed.

Annex 7: Fanconi's disease

EPIDEMIOLOGY

Fanconi anemia is the most common form of inherited aplastic anemia. The frequency of heterozygous carriers has been estimated at 1/300 in the United States and Europe. Transmission is autosomal recessive, except the very rare X-linked (*FANCB* gene) and autosomal dominant inheritances (linked to the *FANCR/RAD51* gene). All ethnic groups are affected.

GENETICS

Twenty-two Fanconi genes have been identified (Bogliolo et al., 2015). In Europe, the most frequently mutated genes (90%) are *FANCA* (2/3 cases), then *FANCG, FANCD2* and *FANCC.* The products of these 22 genes interact in a unique biological pathway named FA/BRCA, involved in maintaining genome integrity through the control of DNA damage repair blocking replication, in particular interstrand crosslinks (ICL). When one of the genes is bi-allelically mutated , the FA/BRCA pathway is inactivated.

Among the groups of genes described above, it is necessary to note a particular group of genes called "downstream genes", encoding FANC proteins acting downstream to the mono-ubiquitination of *FANCD2*, at the level of the nuclear foci of DNA repair. These genes (including *BRCA2* and *PALB2*, initially identified as genes associated with familial predisposition to ovary and breast cancer) are bi-allelically mutated in fewer than 5% of Fanconi patients, but they are associated with a very high risk of cancer and leukemia, especially for *BRCA2* before the age of 5 years (Radulovic et al., 2023).

A fraction of the patients, with a frequency which depends on the age, is reported to present a state of somatic mosaicism (correction of a mutation on one of the 2 alleles of the *FANC* gene involved). If it occurs in a hematopoietic stem cell, the implementation of clonal hematopoiesis allows an improvement or even a normalization of cytopenias. Only fibroblast tests will then be characteristic of Fanconi anemia (Waisfisz et al., 1999; Soulier et al., 1995).

CLINICAL MANIFESTATIONS

Clinical expression of Fanconi anemia only partially reflects genetic heterogeneity (Shimamura et al., 2010). The classic picture combines:

- consistent growth retardation (present at birth and almost constant),
- characteristic facial dysmorphia (triangular appearance of the face, marked saddle nose, microphthalmia with pseudo-hypertelorism and thin features)
- skin abnormalities (pigmented spots known as "café-au-lait", achromic spots and areas of melanoderma that become more pronounced with age, predominantly located on the trunk and neck)
- thumb abnormalities (50% of cases)
- pancytopenia of secondary onset that worsens with age

The associated malformations are inconsistent and highly variable and are reported below.

Location	Clinical signs
Skin	- >3 café-au-lait spots - hypopigmentation - hyperpigmentation, melanoderma
Morphotype and growth	- intrauterine growth retardation - small size - endocrine abnormalities - typical Fanconi facies
Ophthalmic	- short palpebral fissures - ptosis - hypertelorism - hypotelorism - strabismus

	- cataract - microphthalmia
Thumb/radius	- hypoplasia of the thenar eminence - agenesis or hypoplasia of the radius - agenesis or hypoplasia of the thumb - floating thumb - bifid thumb
Skeleton, other	- agenesis or dysplasia of the ulna - micrognathia - domed forehead - spina bifida - fusion of cervical vertebrae - other vertebral abnormalities - agenesis of the clavicle - dysplasia/malposition of the scapula - osteonecrosis - hip dysplasia/dislocation - rib abnormalities - clubfoot - agenesis/hypoplasia of the sacrum - asymmetry in leg length - kyphosis - brachydactyly - arachnodactyly - humeral abnormalities - craniosynostosis
Kidneys and urinary tract	- ectopic kidney - horseshoe kidney - renal malrotation - renal agenesis or hypoplasia - renal dysplasia - hydronephrosis - hydroureter - urethral stenosis - vesicoureteral reflux
Ears and deafness (conduction)	- agenesis or abnormalities of the pinna - prominent ears - ear malposition (low or posterior) - agenesis or atrophy of the ear canal - tympanic membrane agenesis - fusion of ossicles - microtia
Genital organs	- micropenis - hypospadias - testicular atrophy or agenesis - testicular ectopy - chordee - phimosis - azoospermia - bicornuate uterus - aplasia or hypoplasia of the vagina and uterus - vaginal atresia - hypoplastic uterus - agenesis or hypoplasia of the ovaries

	- hypoplastic or fused labia
Cardiopulmonary	- patent ductus arteriosus - agenesis of the ventricular septum - aortic or pulmonary stenosis - coarctation of the aorta - double aortic arch - cardiomyopathy - tetralogy of Fallot - pulmonary atresia
Gastrointestinal	- esophageal atresia - duodenal atresia - anal atresia - tracheoesophageal fistula - annular pancreas - intestinal malrotation - intestinal obstruction - biliary atresia - intestinal cystic dilatation
Central nervous system	- microcephaly - hydrocephalus - facial paralysis - arterial malformation - abnormality of the pituitary stalk - agenesis of the *septum pellucidum/corpus callosum* - hyperreflexia - neural tube defect - Arnold-Chiari malformation - Moyamoya disease - single ventricle

Hematological abnormalities are virtually constant and the median age of detection of cytopenias is 7 years (0–36 years). At 40 years, the cumulative incidence reaches 98% (Butturini et al., 1995; Sebert et al., 2023). The rare forms of the FANCM group may be spared bone marrow failure (Bogliolo et al., 2018).

It involves a combination of normocytic or macrocytic aregenerative anemia, neutropenia and thrombocytopenia, but may begin with isolated macrocytosis. This then gradually progresses toward severe bone marrow failure. The bone marrow differential cell count shows poor, erythroblastic or hypoplastic bone marrow. Isolated erythroid dysplasia has no prognostic value.

Medullary karyotype may reveal clonal abnormalities (Butturini et al., 1995). Their frequency increases with age: 15% at 10 years, 37% at 20 years, 67% at 30 years. Cytogenetic abnormalities are varied and most often affect chromosomes 1, 3, 7, 11 or 21 (Quentin et al., 2011; Sebert et al., 2023). The prognostic value of these abnormalities is variable: isolated 1q abnormalities are associated to clonal hematopoiesis and have unclear prognosis, whereas 3q abnormalities, 5q abnormalities, monosomies 7 or 7q should raise concerns about rapid progression to leukemia (Tonnies et al., 2003; Quentin et al., 2011; Peffault de Latour et al., 2016; Sébert et al., Cell Stem Cell 2023). Specialized advice from the Fanconi reference laboratory (Hôpital Saint-Louis) is generally recommended in case of clonal evolution. The classic karyotype should be coupled with FISH and molecular studies (CGH, NGS); translocations or mutations of *EVI1* or *RUNX1* have been reported and have poor prognosis (Quentin et al., 2011). The risk of progression to myelodysplastic syndrome or acute myeloblastic leukemia also increases with age: 7% at 10 years, 27% at 20 years, 43% at 30 years. Their prognosis is poor. This evolution typically occurs during the 2nd decade in non-allografted patients; more rarely, the diagnosis will be later.

PREDISPOSITION TO SOLID CANCERS

Fanconi anemia is a syndrome that has been demonstrated to predispose to solid cancers (Rosenberg et al., 2005; Alter et al., 2010). The most common cancers are cancers of the head and neck, esophagus, and vulva. These are squamous cell carcinomas, often preceded by leukoplakia that must be monitored and treated before an aggressive course. The cumulative incidence at 40 years is 46%.
Allograft, particularly when followed by chronic graft-versus-host reaction, increases the risk of cancer occurrence and of early onset.
More rarely, although these cases are being identified with increasing frequency with age, diagnosis is made when solid cancer is found in an adult patient with normal or subnormal blood cell counts, and in whom a diagnosis of Fanconi anemia had never been made before. Very poor tolerance of oncological treatments is therefore often what leads to a diagnosis of Fanconi disease.

DIAGNOSIS

Chromosomal breaks

The reference test is the study of the number of chromosomal breaks per cell after exposure to DNA bridging agents. Cells show an increase in the number of breaks, reflecting hypersensitivity to alkylating agents such as di-epoxybutane (DEB) or mitomycin C (MMC) (Oostra AB et al., 2012). The test must be performed in a reference laboratory (the Cytogenetics Laboratories of the national network, including Hôpital Saint-Louis in Paris, Institut Gustave-Roussy in Villejuif, Hôpital Saint-Vincent in Lille, Hôpital La Timone in Marseille, Hôpital Pellegrin in Bordeaux, Hôpitaux Civils de Lyon).
The karyotype carried out on blood lymphocytes may be normal or may spontaneously show a level of chromosomal breaks, which is considerably higher after exposure to DNA-bridging agents (alkylating and related agents). It is normal in heterozygous subjects (who, moreover, have no sign or predisposition to hemopathies or solid cancers except for the rare downstream groups such as D1/BRCA2).
Other tests that can be performed on blood or fibroblasts are:

- the study of the cell cycle by flow cytometry (significant increase in the rate of cells blocked in the G2/M phase after addition of an alkylating agent). Currently, this test is not much practiced.
- the study of the mono-ubiquitination of FANCD2 by Western blot, carried out only at Hôpital Saint-Louis: this test makes it possible to identify more than 90% of affected patients, and more importantly, to classify them between FA core and downstream patients (Soulier et al., 2005). It helps also in the interpretation of FANC gene variants (Variants of unknown significance – VUS).

The combination of the different tests helps to confirm or exclude the diagnosis, including for forms with suspected somatic mosaicism in blood when they are performed on skin fibroblasts (Soulier et al., 2005; Pinto et al., 2009). In fact, a spontaneous reversion of the mutation by homologous recombination, mitotic crossing over or additional mutation can correct a constitutional mutation. The corrected progenitors acquire a selective advantage and can repopulate the bone marrow. This is called somatic mosaicism, and blood tests can be ambiguous or negative. A diagnosis can be made in these situations on non-hematopoietic cells in culture and skin fibroblasts.
For the propositus, genetic tests are not practiced from the outset (too many genes and variants of unknown significance, problem of reversions), but once the diagnosis is confirmed on phenotypic tests (breaks and FANCD2 test).
The molecular genetic diagnosis of Fanconi disease is now carried out in a standard way in France, at the Genetics Laboratory of the Institut Curie. This test can be performed on blood or, ideally, on fibroblast DNA, to avoid the clonal drift that is common in blood or bone marrow, even if it appears normal: its main clinical interest is to enable genetic testing as part of a family investigation, notably to intra-familial allogeneic transplantation, or genetic counseling for possible MAP with pre-implantation or prenatal diagnosis. Knowing a patient's gene only has an impact on management for those rare patients (<5%) with mutations in downstream genes, who are at very high risk of cancer, and whose parents are also at risk, which should lead to an oncogenetic consultation. Genotype/phenotype correlations are currently being evaluated as part of the research.

FOLLOW-UP AND THERAPEUTIC MANAGEMENT

Treatment is multidisciplinary. The American association for patients with Fanconi anemia(FARF) has made proposals for monitoring various types of organ damage. This document has been

translated and adapted under the aegis of the AFMF with the participation of the Aplastic anemia RC and can be consulted for information purposes on the AFMF website Fanconi Guide.

a) Monitoring and management of hematological aspects

A **full blood count** with reticulocyte count should be performed every 3 months, and a bone marrow differential cell count with karyotype (and additional analyses) every year to detect any clonal changes.

Transfusions should be used sparingly, to reduce the risk of allo-immunization. The emergence of transfusion requirements is an indication for hematopoietic stem cell transplantation if a geno- or pheno-identical, HLA-compatible donor is available. In the absence of an HLA-compatible 10/10 donor, it is recommended to discuss the case at the national Aplastic anemia MDT meetings. The appearance of cytogenetic or molecular abnormalities with poor prognosis must lead to the discussion of an HSC allograft.

Allogeneic hematopoietic stem cell transplantation
HSC allograft is the only cure for hematological disorders, but it does not correct other abnormalities or malformations.

- *Indications*

An allograft with a 10/10 geno-identical or pheno-identical donor is indicated in case of severe cytopenias suggesting the need for systematic transfusion support or in the event of clonal evolution (appearance of a cytogenetic clone with an abnormality that has a poor prognosis or aspect of MDS/AML).
A clonal evolution (appearance of a cytogenetic clone with an abnormality that has a poor prognosis or aspect of MDS/AML) in a patient with no contraindication to a graft should lead to discussion of a haploidentical allograft in the absence of an HLA-compatible donor. Profound cytopenias requiring regular transfusions in young patients may also prompt discussion of this alternative.

- *Conditioning*

Graft conditioning must take into account the high sensitivity to breaking and oxidative agents that contraindicate certain chemotherapies. Most teams agree that no radiotherapy should be carried out in conditioning. Low doses (2 to 4 Gy) are of interest only in the context of unrelated allograft conditioning in adults.
The different types of conditioning are described in Annex 5.

In a geno-identical situation, overall survival reaches 80% at 2 years in the most recent series (Peffault de Latour et al., 2013).
Where there is an alternative donor, recent results have demonstrated a marked improvement in survival after these transplants compared with historical data (overall survival of 40% after mismatch transplants in the years 1990–2000): several publications over the last decade of cohorts of allograft patients with new haploidentical or pheno-identical 9/10 grafting protocols with unmodified grafts and GvHD prophylaxis with post-transplant cyclophosphamide or *ex vivo* T depletion adapted to the particular sensitivity of patients with Fanconi disease have shown a reduction in the incidence of these severe complications. Overall survival in recent studies varies from 65% to 85% depending on patient age and hematological status. A retrospective European study that compared 9/10 pheno-identical and haploidentical grafts in this indication showed a survival benefit in patients who had received transplants from a haploidentical donor. The results of haploidentical transplants recently published by different teams using an *in vivo* (Bonfim et al., 2017) or *ex vivo* (Strocchio et al., 2021) T-depletion strategy are in favor of these haploidentical transplant strategies; however, strict comparison of these different transplant methods is made difficult due to the heterogeneity of patients included in the different cohorts (age, underlying pathology). At the same time, data from the English team (Bernard et al., 2021) support the use of alemtuzumab to reduce the incidence of GvHD.

These alternative grafts remain difficult indications that must be decided on a case-by-case basis and should be discussed at the MDT meetings. A prospective observational protocol for the evaluation of haploidentical grafts in patients with a progression to myeloid hemopathy or severe pancytopenia will be implemented in 2023.

- *Graft-versus-host disease (GvHD)*

At a comparable degree of compatibility, Fanconi patients have more severe acute or chronic graft-versus-host disease (GvHD) lesions than other patients. Bone marrow transplant are recommended over peripheral stem cell grafts. Fludarabine, which induces a significant T lymphocyte depletion, decreasing the risk of GvHD at the cost of lower toxicity than alkylating agents for these patients, is recommended.
GvHD is responsible for an increase in the frequency of secondary tumors, which occur on average from 10 years after allograft.

Other treatments
Hematopoietic growth factors have no place in the correction of cytopenias but can be used occasionally in the absence of therapeutic alternatives (G-CSF for threatening infections). The role of TPO-RAs has not been established, which means that they cannot be recommended.
In the absence of the possibility of transplantation, androgens are regularly effective and can prevent the need for transfusions. Side effects (virilization and advancing bone age, lipid and liver abnormalities, and liver tumors – adenomas and adenocarcinomas) should be closely monitored (MRI or CT scan with a low dose of contrast medium annually for monitoring liver tumors). Danazol is used in the first instance as it is less virilizing, at a dose of 5 mg/kg in children (without exceeding the adult dose) and 800 mg per day in adults: once a response has been achieved, the minimum effective dose should be sought.
Several teams are conducting gene therapy trials in a research setting on newly diagnosed FANCA patients. The difficulty is to efficiently transduce the patient's stem cells in sufficient numbers to allow their expansion. Encouraging results have been published for 4 patients (Rio et al., 2019), but these therapies remain in the research domain (clinical trials) and do not apply to the majority of patients.

b) Follow-up and management of extra-hematological aspects (excluding post-allograft complications)

The follow-up of these patients is most often multidisciplinary and will involve pediatricians and adult doctors from different specialties as well as members of paramedical professions. In pediatrics, this follow-up is dominated by the management of congenital malformations, growth, and neurosensory and developmental aspects. In adulthood, it is dominated by screening for solid tumors. These patients should be monitored for life, including the rare non-allograft patients.

Genetic monitoring
1. Genetic diagnosis
A genetic diagnosis makes it possible to confirm the diagnosis, to verify that the 2 parents are each carrying a mutation (which makes it possible to confirm that the 2 mutations identified in the patient are indeed *in trans*) and thus to specify the risk of recurrence in the family, to detect a still silent form in a sibling as part of the donor search, and to allow the proposal of a prenatal diagnosis (PND) or pre-implantation diagnosis (PID) in the event of a desire for a new child.

2. Genetic counseling
This can be offered to all parents and adult patients if they wish. It is essential if parents wish to have another child by assisted reproduction or if they are carriers of a variant of a downstream gene (see above). This implies that the mutation(s) has/have been identified and that it/they can be analyzed on DNA extracted from a single embryo cell. The practice of selecting a non-affected embryo that is HLA-identical to a sick sibling (double selection) is no longer practiced in France at present.

Malformations and other congenital disorders: diagnosis and monitoring
Systematic testing must be carried out for these, at least for those that are the most common and for those that may have an impact on the development of the child. The diagnostic assessment will be directed by the clinic. As a minimum, and to avoid unnecessary irradiation, it must include:

- an abdominopelvic ultrasound
- an echocardiogram, a neurosensory assessment with neurological, ophthalmological and ENT evaluation including an audiogram, and if necessary an assessment by a psychomotor therapist
- an evaluation by a psychologist

The management of initial malformations and disorders will be ensured by organ specialists. It should anticipate the development of bone marrow failure; in particular, if surgical interventions are necessary it is logical to perform them before the occurrence of severe thrombocytopenia.

Sensory and neurological disorders, psychomotor development
Children with Fanconi disease may have:

- neurosensory disorders: vision or hearing disorders
- motor difficulties due to congenital malformations of the forearms and hands
- academic difficulties that may be related to specific disorders and the seriousness of the disease but may also reveal specific learning disorders.

This warrants systematic neurological, ophthalmological and auditory evaluation.
The neuropediatrician (or neurologist) will evaluate the impact of musculoskeletal abnormalities of the radial ray (abnormalities of the thumb and radius, hypoplasia of the thenar eminence) on fine motor skills. A number of children have difficulties with handwriting, fatigue and slowness in writing, which harms their written work at school and does not reflect their good level of knowledge and understanding of a subject. The neuropediatrician will recommend a neuropsychological, psychomotor or occupational therapy assessment (depending on the age of the child) to identify specific disorders, which will lead to specific recognition by the MDPH, and will prescribe suitable care, as well as make recommendations regarding school (see Annex 12).

Monitoring of growth and puberty
Fanconi patients have a risk of small stature in adulthood. The causes are multiple:

- disease-related (small stature due to syndrome) and associated with IUGR
- endocrine deficiency including growth hormone deficiency, usually due to malformation of the pituitary region with ectopic post hypophysis
- chronic anemia
- treatment sequelae (corticosteroids, graft conditioning)

This requires systematic monitoring of the growth of these children and systematic consultations in pediatric endocrinology from the diagnosis, during the pre-pubertal and pubertal periods.
Delayed puberty may occur, which needs to be detected and possibly treated with hormone replacement therapy.
In agreement with the family, growth hormone therapy is usually indicated in the case of growth hormone deficiency with malformation of the pituitary region. It is more debatable in other situations, due to the lower effectiveness and the less clear benefit/risk ratio. In the case of pituitary malformation, there is also a risk of other pituitary deficiencies, in particular, adrenocorticotropic hormone (ACTH) (risk of acute adrenal insufficiency).

Reproduction and pregnancy
Fanconi patients have reduced fertility due to the possible combination of urogenital malformations, hypogonadism, risk of early menopause and sterility induced by bone marrow allograft. However, spontaneous pregnancies are not uncommon, even after bone marrow allograft. Fertility preservation measures should be implemented whenever possible (see the section on fertility on p. 28). Patients should be able to have consultations on assisted reproduction.

Immunological monitoring
Rare non-allograft Fanconi patients, including revertant patients, may develop a predominantly B immune deficiency in adulthood. This requires annual monitoring of immunoglobulin levels (IgG, IgA, IgM).

Solid cancers and Fanconi anemia
There are few large published series analyzing the therapeutic management of solid cancers in Fanconi anemia.
The risk of solid tumors is particularly increased:
- for Fanconi patients with damage to so-called downstream genes, i.e. those coding for FANC proteins acting downstream from FANCD2 mono-ubiquitination, at the nuclear foci of DNA repair. These genes (including *BRCA2, BRCA1* and *PALB2*, initially identified as genes associated with breast cancers) are bi-allelically mutated in fewer than 5% of Fanconi patients but are associated with a very high risk of cancer (and leukemia) before the age of 5 years,
- in chronic GvHD after bone marrow allograft,
- in patients who have received androgens for prolonged periods for liver tumors.

Cancers appear from the age of 20 years in non-grafted patients: their incidence increases continuously with age to an incidence of 4.4% per year at the age of 40 years. In allograft patients, these cancers occur earlier (10 years earlier): their incidence reaches 4.7% per year, 10 years after transplantation. Median survival after cancer diagnosis is similar in both populations: data from the literature report a median survival of 13 months. Acute GvHD and chronic GvHD are risk factors for solid cancers in allograft patients. (Rosenberg et al., 2005)
Although many tumors affecting various organs are described, the vast majority are squamous cell carcinomas of the head and neck (Alter et al., 2018). A recent review of the literature concerning oral cavity cancers in patients with Fanconi's disease shows that all areas are likely to be affected, although the tongue is the most frequently affected site (Furquim et al., 2018).
Anogenital cancers are the only ones described as induced by human papillomavirus (HPV) in Fanconi disease (Van Zeeburg et al., 2008).
Cancer of these regions appearing in an unusually young person and/or without risk factors, especially when associated with unusual toxicity of radiotherapy and/or chemotherapy, should prompt investigations for Fanconi anemia (revertant patients in particular).

▪ ***Prevention***

Patients should be informed of this risk and encouraged to learn how to examine themselves: any suspicious lesions in the mouth that do not disappear within 3 weeks should lead to a consultation with a specialist. The preventive measures on which there is currently a consensus are:
- having good oral hygiene
- reducing consumption of alcohol, especially spirits. Mouthwashes containing alcohol should also be avoided
- avoiding smoking tobacco and cannabis and passive exposure to smoke
- getting vaccinated against HPV from the age of 9 for boys and girls as recommended by pediatric societies around the world to prevent the occurrence of HPV-associated squamous cell carcinoma

In practice, however, screening for solid tumors must cover all patients, whether they have received a transplant or not. It should be started in pre-adolescence for types of cancer that can be found in children and continued for life.
The general measures are to avoid known carcinogens (tobacco, alcohol, UV exposure and exposure to occupational carcinogens). In general, Fanconi patients should at least follow the recommendations made for the general population. Because Fanconi patients have increased sensitivity to radiation, the treating physician must also limit their exposure as much as possible by favoring non-irradiating examinations whenever possible (ultrasound, MRI).

- ***Monitoring methods must be adapted to each organ or location***

1. ENT region

From the age of 10, the patient must have a thorough examination every 6 to 12 months with an ENT specialist, a maxillofacial surgeon or any other doctor who is experienced in the detection of head and neck cancer and is familiar with Fanconi's disease. The examination should include careful exploration of the nasopharynx and oropharynx and if there is the slightest doubt, a biopsy should be performed.

To date, systematic screening for esophageal cancer by fibroscopy and for hypopharyngeal or laryngeal cancer by nasofibroscopy is not recommended. Furthermore, in the experience of the aplastic anemia RC, these cancers tend to occur after a first cancer in the oral cavity: endoscopic follow-up is therefore recommended, including the hypopharynx, larynx and esophagus.

2. Gynecology

Patients have an increased risk of squamous cell cancers of the lower genital tract: the cervix, vagina and vulva.

Vaccination against HPV should be routine.

From the age of 13, patients should be monitored by a gynecologist for visual inspection of the external genitalia. From the age of 18, women should receive a comprehensive annual gynecological examination with a cervical smear. A colposcopy and biopsy should be carried out if lesions are identified during the examination or if the smear is abnormal. The examination must also include a search for dysplastic lesions of the anogenital region.

3. Dermatology

Patients should be informed of the risk of skin cancer. The general measures are the limitation of UV exposure and the use of high-strength sunscreens (SPF 30 to 50). A dermatological examination must be carried out once a year. In men, it must be combined with a thorough examination of the anogenital region.

4. Liver

Systematic examinations should be carried out for liver tumors using ultrasound or, ideally, MRI. The risk is particularly high in patients who have been treated with androgens, even after discontinuing such therapy, and these patients must be monitored annually. In patients who have not received androgens, an ultrasound or MRI is also recommended annually after the age of 30 years. It is not easy to distinguish between adenoma and hepatocarcinoma in X-rays, and any lesion must be biopsied.

- ***Therapies***

1. Surgery

The only curative therapy for solid cancers in patients with Fanconi anemiais surgery. The earlier the lesion is managed, the higher the probability that surgery will be able to remove it completely, which warrants close monitoring for life.

The margins of the excised tissue should be histologically negative: a margin of 1 cm macroscopically is recommended during surgery with iterative repeats, if necessary, if histology does not confirm this.

The examination should be repeated regularly, given the risk of recurrence or occurrence of a new cancer.

Repeated biopsies are necessary in the case of diffuse superficial lesions.

Systematic lymph node dissection is performed in this indication. It should be bilateral if the lesion is near the midline.

If excision of cancer is not possible, therapeutic management should be discussed among colleagues.

2. Chemotherapy

Specialist hematological advice is required before chemotherapy in a patient with Fanconi anemia. In the context of Fanconi anemia (allograft or non-allograft patient), any chemotherapy – even at reduced doses – can have the following effects:

- unusual hematological toxicity, due to the low capacity for bone marrow regeneration
- unusual extra-hematological toxicity, especially mucosal

If chemotherapy is not formally contraindicated and is being considered, it should be performed under close hematological supervision (blood count x 2 per week) and during hospitalization if significant neutropenia or thrombocytopenia is observed. In these patients, hemorrhagic and infectious complications are a common cause of death after a diagnosis of solid cancer. Combinations of anti-vascular endothelial growth factor (VEGF) antibodies and chemotherapy with Taxol, with administration methods adapted to hematological toxicity, are being evaluated. It is recommended to seek advice from the CR.
New targeted therapies need to be evaluated in a coordinated manner, especially PARP inhibitors, which appear to be of interest in this indication.

3. Radiotherapy

Conventional curative radiotherapy is generally contraindicated in patients suffering from Fanconianemia in the context of the treatment of solid cancers:, at an oncological dose, it is associated with unacceptable toxicity. Reduced doses are associated with significant mucosal toxicity without being effective.
Low doses (2 to 4 Gy) are of interest only in unrelated allograft conditioning in adults (Peffault de Latour et al., 2016).

Everyday life

Fanconi anemia, like any chronic disease, has repercussions in everyday life, whether family, social, school or professional.
This impact must be considered as soon as the diagnosis is announced and over the long-term.
Sick people and their families may face a variety of challenges:

- Accepting the diagnosis and mourning a healthy child
- Learning to manage difficult periods: bone marrow transplantation, waiting for results, anxiety, isolation, etc.
- Managing the various impacts of the disease on family (brothers and sisters, partners, etc.), professional life, and a child's schooling
- Making life plans despite uncertainties about life expectancy

For all these reasons, families need to find a space to express their emotions, doubts and hopes. This space can be found with close family and friends but also, above all, with a professional or within the association for Fanconi anemia (AFMF).

Annex 8: Telomeropathies (formerly known as congenital dyskeratosis)

Telomeropathies are a heterogeneous group of pathologies characterized by a deficiency in telomere maintenance, causing a range of clinical symptoms that are mainly hematological, respiratory, hepatic, cutaneomucous, immunological, osseous, digestive, neurological and ophthalmological. For a long time it was referred to as " dyskeratosis congenita ", but the existence of telomere shortening in blood cells, the identification of a growing number of genes involved in this pathology and the wide variability of the clinical phenotype have led to a preference for the term "telomeropathy".

The clinical presentation (number of organs involved and nature and severity of organ damage, age at occurrence, prognosis) depends on several factors: the gene involved, the nature of the genetic abnormality, whether the mutation is heterozygous or homozygous, the generation affected and environmental factors (exposure to toxic substances in particular).

To date, 17 genes have been identified as responsible for telomeropathies. These genes are: *TERT*, *TERC*, *DKC1*, *RTEL1*, *PARN*, *TINF2*, *WRAP53*, *NOP10*, *NHP2, ACD*, *CTC1*, *NAF1*, *ZCCHC8*, *STN1, POT1*, *RPA1,* and *DCLRE1B/Apollo*, and other genes that are sometimes considered by some as telomeropathy genes, although their involvement in telomeropathy is debated (*ERCC6L2*, *USB1, MDM4*, *NPM1*, *SON* and *SHQ1*).

The most severe phenotype, called Hoyeraal-Hreidarsson syndrome, is characterized by neonatal or very early impairment combining pancytopenia, severe immune deficiency, neurological involvement with microcephaly, retinal involvement and variable organ involvement (liver, digestive, bone involvement). The prognosis is extremely poor. It is most often related to bi-allelic mutations and is associated with extremely short telomeres.

Hematological disorders of the Aplastic anemia type most often occur in young individuals (children, adolescents and young adults) of the 2nd or 3rd generation and are often associated with organ damage (hepatic or pulmonary) and premature greying hair. Cutaneomucous involvement is inconsistent and/or has delayed onset. The first event may be a myelodysplastic syndrome, in a patient with or without previously identified organ damage: these more recently identified forms most often occur between 30 and 50 years of age, later than the classic hematological disorders but at an early age compared to myelodysplastic syndromes where there is no underlying predisposition. The systematic search for genetic factors predisposing to the occurrence of these myelodysplastic syndromes in young subjects or those with a family history of hematological, hepatological or pulmonary disease is responsible for a rapid increase in the number of cases identified.

Isolated pulmonary involvement occurs at a later age (between 50 and 60 years) and is most often identified in investigations for idiopathic or familial pulmonary fibrosis. A significant proportion of these patients have hematological abnormalities (macrocytosis and thrombocytopenia in particular) and a number will develop hematological disorders during pulmonary follow-up (cytopenia or myelodysplastic syndrome). Liver damage is often difficult to diagnose and requires a combination of systematic monitoring of laboratory (often falsely reassuring) and imaging tests: porto-sinusoidal vascular diseases, nonalcoholic steatohepatitis (NASH) and liver cirrhosis are the most frequently found forms when a liver biopsy is performed. They are associated with an increased risk of hepatocellular carcinoma.

The risk of solid tumors is also increased in this syndrome: the main tumors described are squamous cell carcinomas of the oral cavity and hepatocarcinomas.

Other disorders commonly described in combination with those described above are 1) severe and early osteoporosis, including men, 2) retinal involvement, 3) gastrointestinal angiodysplasias, sometimes responsible for severe hemorrhagic complications, 4) venous thrombosis and 5) increased risk of lymphoma.

Developments in molecular genetic techniques have made it possible to search for these constitutional mutations in France since 2010, leading to an exponential increase in the number of cases identified over the period 2010–2017. Thus, the number of patients diagnosed annually increased from 8 in 2010 to 84 in 2016. The prevalence of telomeropathies is therefore currently estimated at 0.6 per 100,000 inhabitants, distributed equally between the mainly hematological and mainly pulmonary forms. The prevalence of mainly hepatic forms is currently unknown.

Indications for diagnostic tests in hematology

Hematological involvement most often presents as a bi- or pancytopenia with hypo-cellular marrow. The involvement of the platelet line is the most constant and is often associated with macrocytosis. This progresses to severe pancytopenia or secondary myelodysplastic syndrome (sometimes progressing to acute leukemia). The search for a constitutional etiology (study of telomere length and genetic analyses) in subjects under 40 years of age with severe or moderate aplastic anemia is now one of the recommendations that most often allows these patients to be identified at an early stage. Telomere length is almost consistently below the 1st percentile in these patients; however, the positive predictive value is not 100%: Patients with other types of inherited hypoplasia (for example, Fanconi anemia, Shwachman-Diamond syndrome or GATA2) or idiopathic Aplastic anemia may have telomeres below the 1st percentile. In patients over 60 years of age, telomeres between the 1st and 10th percentile can probably be considered pathological.

The identification of a deleterious variant, a gene responsible for telomeropathies or the existence of an extra-hematological clinical phenotype (pulmonary, hepatic or cutaneomucous involvement or greying hair before the age of 20 years) are mandatory to confirm the diagnosis of telomeropathy. Thrombocytopenia or unexplained macrocytosis associated with pulmonary fibrosis and/or hepatic impairment should lead to the same investigations.

Treatment of hematologic disorders

For patients with severe aplastic anemia related to telomeropathy (the mechanism of which is not immunological), immunosuppressive therapy is ineffective and may aggravate cytopenias and the risk of infectious, hemorrhagic and bone complications.

For these patients, the only potentially curative therapy is allogeneic bone marrow transplantation from an unmutated HLA-compatible intrafamilial donor or an unrelated HLA-compatible donor. However, bone marrow transplantation does not correct the genetic deficit in extra-hematopoietic cells and exposes organs to infectious, toxic and immunological complications. The prognosis for bone marrow transplantation in telomeropathies is poor, which is why many teams have not performed transplants in this indication for many years (Rocha et al., 1998). Thus, in a recent review of the literature, overall survival in a cohort of 109 patients was estimated at 57% and 23% at 5 and 10 years respectively, due to hepatic, pulmonary and graft dysfunction complications (Barbaro et al., 2016). Improved early survival for transplant patients after 2000 has not resulted in a long-term survival benefit in this population, due to middle- and long-term liver and lung complications. New conditioning regimens have been proposed in recent years to limit extra-hematopoietic toxicity in patients with telomeropathies. However, these have not been in use for long enough to determine whether there is any benefit in terms of survival in the middle- and long-term. Rapid worsening of lung and liver damage that is asymptomatic or causes few symptoms before transplantation has been observed after non-myeloablative conditioning, especially when total body irradiation is used, even at low doses (Sorge et al., 2017, and unpublished personal data).

Indications for bone marrow transplantation are therefore currently limited to patients with severe aplasia (PNN <0.5 G/L, platelets <20 G/L, reticulocytes <20 G/L and/or transfusion requirements or infectious complications) or high-risk myelodysplastic syndrome with an HLA-compatible donor and no significant hepatic or pulmonary organ dysfunction (grade C).

Androgens are the main therapeutic alternative to bone marrow transplantations in severe hematological disease (grade B). In the only published prospective study, Townsley et al, (2016) observed a hematological response in 79% of patients at 3 months (24 assessed) and 83% of patients at 24 months (12 assessed). 12 of the 13 patients who received transfusions before therapy were independent of transfusions during therapy. The median increase was 3.3 g/dL for hemoglobin, 0.3 G/L for PNN and 14 G/L for platelets. One patient experienced a rapid worsening of their moderate aplasia under therapy. The reduction in telomere length loss observed in this study was not confirmed by other retrospective studies over longer follow-up periods. This study also suggests a benefit in terms of reduction of the loss of forced vital capacity in patients with respiratory impairment. In this study, 37% of patients stopped therapy early before 24 months. The main side effects reported were:

- hepatic: elevation of transaminases in 41% of cases, severe hepatic dysfunction requiring the creation of a portosystemic shunt
- muscular: cramps in a third of patients
- endocrinologic: abnormalities of lipid profile in a quarter of patients

- headache in 15% of patients
- weight gain in 15% of patients
- one patient had a severe thrombotic event and another was diagnosed with hemangioblastoma during therapy.

Danatrol is given at a dosage of 400 mg x 2 daily for at least 3 months prior to evaluation. In the event of poor tolerance, the dose may be reduced to 200 mg x 2 daily. Once the response is stable, the dose is gradually reduced at intervals of at least 6 months. Annual monitoring with MRI or a contrast-enhanced CT scan of the liver is necessary due to the increased risk of liver tumors.
There are no data in the literature on the efficacy of thrombopoietin receptor agonists in these patients. In the experience of the aplastic anemia RC, none of the treated patients achieved even a partial hematological response (unpublished personal data). Transfusion support poses the middle- and long-term problem of the risk of allo-immunization, leading to loss of efficacy, iron overload and the absence of effect on the infectious risk.

Follow-up
- A blood count is recommended every 3 to 6 months
- A bone marrow aspiration with cytogenetic analysis is recommended for diagnosis and then every 18 months or in case of worsening of hematological abnormalities
- Osteomedullary biopsy is of interest only in the event that it is impossible to carry out aspiration of bone marrow
- Monitoring the length of telomeres is unnecessary

A cellular immune deficiency is possible and should be evaluated in case of opportunistic or repeated infections. Hypergammaglobulinemia is common.

Screening for and prevention of extra-hematologic involvement
- An initial pulmonary assessment by CT scan and respiratory function exploration (RFE) is recommanded. In the absence of abnormalities, the CT scan will be repeated every 3 to 5 years and the RFE every 2 to 3 years. In the majority of cases of respiratory involvement (55% to 90% depending on the series), an additional risk factor is found: smoking, occupational exposure to toxic substances or a context of hypersensitivity pneumopathy. The focus must be on the removal of toxic substances and tobacco especially. In the absence of CT scan abnormalities and in the presence of dyspnea, a search for hepatopulmonary syndrome should be performed (Borie et al., 2016).
- Hepatic involvement is currently the least well characterized and the prevalence of hepatic involvement, estimated at less than 10% in cohorts of patients with telomeropathies, is probably significantly underestimated. The clinico-pathological description is heterogeneous: porto-sinusoidal vascular disease, cryptogenic fibrosis or cirrhosis, non-cirrhotic portal hypertension, regenerative nodular hepatopathy or steatosis. Histological damage is most often underestimated when evaluation is based on laboratory tests and/or imaging tests. Histology often reveals severe hepatocyte damage with portal fibrosis lesions. Portal hypertension is frequently found at an early stage and is responsible for hemorrhagic complications. Central hematological involvement is often underestimated in these patients, whose cytopenias are associated with portal hypertension. Hepatocellular carcinomas and angiosarcomas can occure in the settings of hepatopathy. Abdominal ultrasound with a search for portal hypertension should be performed at diagnosis and in the event of any disruption in the liver function tests. Specialized hepatological advice is recommended in the event of any abnormalities in the laboratory tests or imaging tests. Alcohol must be avoided.
- Osteoporosis fractures are common and bone densitometry and adequate vitamin D supplementation are routinely recommended.
- Follow-up by a maxillofacial surgeon is recommended in case of leukoplakia, to screen for precancerous lesions.
- thelomeropathy do not seem to be associated with a fertility decrease in both men and women. Fetal complications and HELLP syndromes (hemolysis, elevated liver enzymes and low platelets) are more common in women with telomeropathy (Giri, et al., 2021).

Genetic counseling and screening of relatives

For all patients with mutations, overall survival is difficult to predict because it in particular depends on the affected generation. In a retrospective study involving mainly patients with a classic severe form, the median survival was 42 years. This does not allow us to extrapolate to all telomeropathies, due to the later age of onset of the first manifestations in the predominantly pulmonary and hepatic forms and in myelodysplastic syndromes. However, when a patient has symptomatic hematological, pulmonary or hepatic involvement, life expectancy is less than 5 years and median survival is probably 2 to 3 years

The early diagnosis of sub-clinical damage (promoted by the access of relatives to molecular genetic diagnosis at the pre-symptomatic stage) makes it possible to carry out preventive work by limiting exposure to toxic substances that contribute to the development of organ damage. To date, however, no treatment can prevent the disease from getting worse.

The diagnosis of non-symptomatic relatives must be offered to everyone as part of a genetic or mixed consultation. For children who are minors, a clinical and biological evaluation without genetic diagnosis can be proposed in the first instance until they reach the age where they can choose to do this test. In fact, screening should only be carried out if a direct benefit is expected, in accordance with the law of bioethics.

Note that genetic counseling is especially complex in these diseases and must be done by doctors who have a good knowledge of them: for some genes, both mono-allelic and bi-allelic forms are described and rare phenocopies have also been described, i.e. patients who may have telomeropathy-like manifestations linked to the fact that they have inherited short telomeres even though they are not carriers of a mutation.

Prenatal or pre-conceptional diagnosis is difficult because of the dual heritability of the mutation and the size of the telomeres. A non-mutation-carrying relative may have inherited short telomeres and thus be exposed to complications similar to those observed in individuals carrying the mutation. Although a small number of cases have been described, this limits the negative predictive value of genetic diagnosis in this context. This does not contraindicate the latter, but clear information has to be given to families.

Psychological support should be proposedto patients and relatives because of the severity of the disease.

Annex 9: Diamond Blackfan anemia (DBA)

Diamond Blackfan anemia is an inherited bone marrow failure syndrome characterized typically by erythroblastopenia, secondary to a blockage of erythroid progenitor maturation (BFU-e/CFU-e stages). The disease is mostly associated with a defect in the production of red blood cells. Due to the absence of anemia in some patients, the name Diamond Blackfan disease or Diamond Blackfan syndrome is currently preferred (L Da Costa, T Leblanc et al., 2020).

EPIDEMIOLOGY

DBAis a rare disease with an estimated incidence in several European registries of between 5 and 7 cases per 1,000,000 live births. It is likely that this corresponds to the classic forms and that the actual incidence is higher.

GENETICS

To date, 23 genes encoding ribosome structural proteins have been identified, either *RPS* (encoding small subunit proteins) or *RPL* genes (encoding large subunit proteins). The most frequent identified gene is *RPS19* , involved in 25% of patients. Six genes (*RPS19, RPL5*, *RPL11*, *RPS26*, *RPS24* and *RPL35a*) are involved in approximately 90% of patients of known genotype (L Da Costa et al. 2017; JC Ulirsch et al., 2019). For these genes, the transmission is autosomal dominant (haplo-insufficiency). Functionally, these mutations are associated with a defect in ribosome biogenesis (Leger-Sylvestre et al. 2005; Choesmel et al., 2007).

To these *RPL* and *RPS* genes, we can add 2 genes encoding chaperone proteins involved in the biosynthesis of ribosomes identified in very rare patients:

- *TSR2,* which codes for a chaperone protein of RPS26 (X-linked transmission).
- *HEATR3*, which codes for a protein involved in the nuclear import of RPL5 in men (autosomal recessive transmission) (MF O'Donohue et al., 2022)

There is a consensus on another two genes whose pathogenic variants are responsible for phenotypes very close to DBA:

- *GATA1*, which codes for a transcription factor involved in erythropoiesis; the gene is located on the X chromosome and the transmission is linked to gender; mutations associated with a DBA-type picture are specific, resulting in the loss of the long isoform
- *TP53*, with monoallelic gain of function mutation.

There are two other genes involved in other constitutional erythroblastopenia that are different from DBA (differential diagnosis)

- *EPO*, which codes for erythropoietin (autosomal recessive transmission, loss of function mutation, 1 family described)
- *CECR1/ADA2* (autosomal recessive transmission) involved in DADA2 syndrome; some patients may present with erythroblastopenia mimicking DBA.

Currently, a mutation is identified in 80% of DBA patients.
It should be noted that 7 other genes that code for RP proteins have been involved in some patients but are currenly under investigation.
In the absence of an identified mutation, an exome- or genome-type study (French national PFMG 2025 program) can be carried out in a research setting.

CLINICAL MANIFESTATIONS

Diagnosis is classicaly made early in life, usually before 1 year of age (median age at diagnosis of 2 months); diagnosis after 4 years was considered to be rare but possible. Actually more and more cases are diagnosed in adults, in forms with mild anemia or in patients without anemia in whom DBA is diagnosed due to other manifestations (see below).

In small children, the clinical picture involves a combination of isolated pallor and dyspnea that causes difficulties during breastfeeding.
Anemia is typically isolated without splenomegaly, signs of hemolysis or manifestations related to the involvement of other hematopoietic lineages.
More than half of patients have growth retardation and associated malformations, the most common being malformations of the thumb (including the classic triphalangeal thumb), cephalic area (cleft lip and palate), heart and urogenital organs, although many other malformations have been reported (Willig & al., 1999).
Apart from the anemia assessment, a diagnosis may also be based on:

- Assessment of congenital malformations or polymalformative syndrome with or without anemia (value of an isolated macrocytosis)
- Assessment of minor CBC abnormalities, including isolated macrocytosis or neutropenia; the platelet line is usually preserved
- Assessment of B lymphocyte deficiency with hypogammaglobulinemia
- Myelodysplastic syndrome occurring in a "young" subject (before 60 years of age)
- Cancer unusual in age or characteristics and unexpected hematological intolerance to chemotherapy (transfusion requirements)
- Family investigation with diagnosis of DBA in an asymptomatic carrier at the time of the study

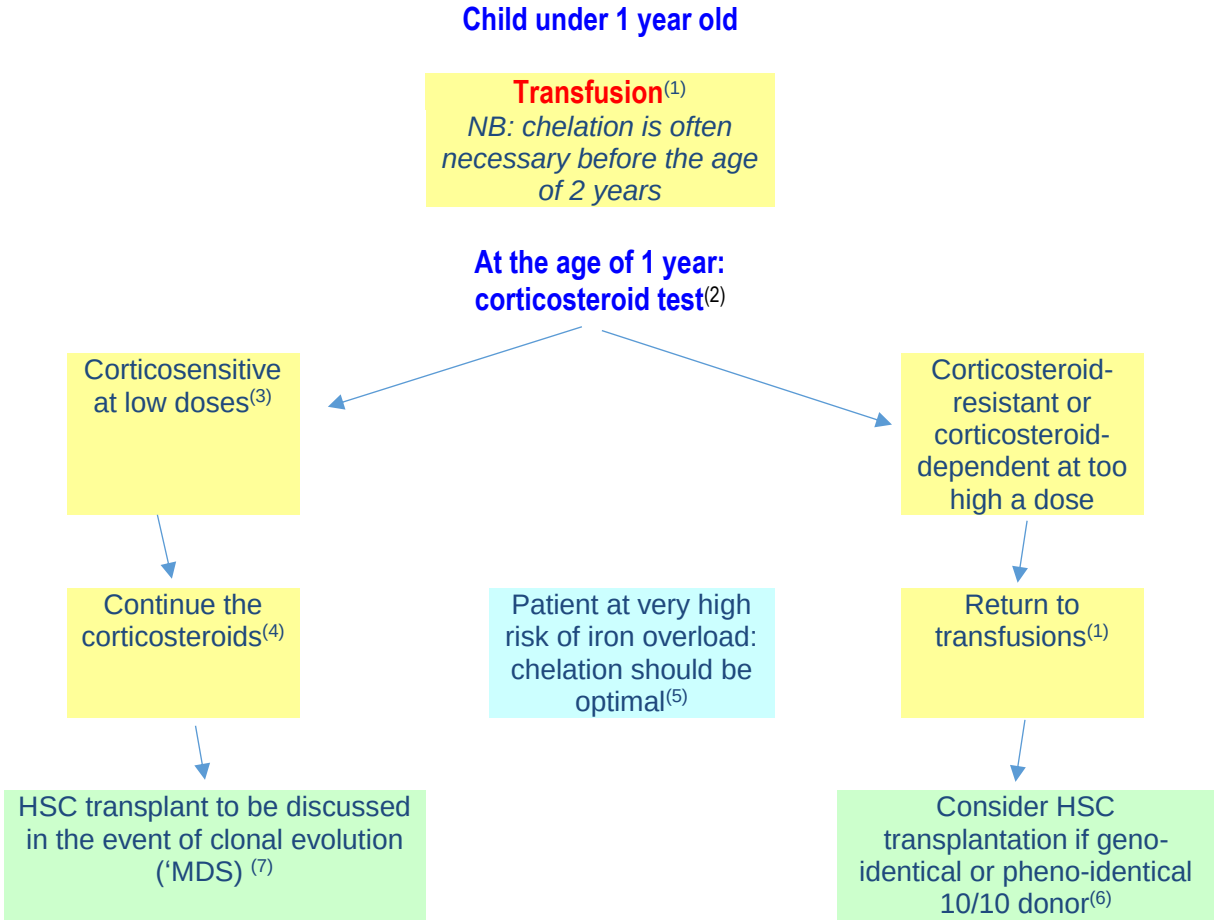
The diagnosis of DBA is based on a set of clinical and biological arguments.
In a clinical context that suggests DBA, the biological elements in favor of DBA are mainly:

- The blood count shows an isolated, macrocytic anemia with little or no regeneration (<20 G/L); reticulocytopenia can be severe in forms requiring transfusion, with no reticulocytes in the peripheral blood. Conversely, some patients will have normal or subnormal hemoglobin, present reticulocytes and simply macrocytosis, or even a normal blood count.
- Damage to other lines without clinical impact may be present: thrombocytosis (newborns only), moderate thrombocytopenia, leukoneutropenia, lymphopenia (frequent also in patients on corticosteroid therapy). In exceptional cases, neutropenia can be severe (*RPL35*a gene)
- Evidence of erythroblastopenia in the bone marrow differential cell count (absence or <5% of erythroblastic precursors within a normocellular marrow) without any sign of dyserythropoiesis; it may be absent in non-anemic patients or in forms with non-severe anemia. Otherwise, the marrow is usually of normal richness. It may appear poor in adults.
- An increased activity of fetal hemoglobin F beyond 6 months of life

- A high level of a red blood cell enzyme: erythrocyte adenosine deaminase (eADA), which is assayed in very few laboratories and must be evaluated before any transfusion of red blood cells (or after 3 months without transfusion).
- Ribosomal RNA maturation profiling is currently used in research to validate the pathogenicity of a mutation; it is not yet used in clinical practice but could be a functional test in the future.

Genetic diagnosis confirms the presence of a mutation in the majority of patients (80%). Diagnosis is most often made by targeted NGS. If this is negative, it is necessary to look for large deletions (10-20% of cases of DBA) using an appropriate technique such as customized comparative genomic hybridization (CGH) array or multiplex ligation dependent probe amplification (MLPA).

Management of anemia in a patient with DBA



(1) : maintain hemoglobin >9 g/dL
(2) : the test can be delayed if there is a significant growth retardation and when transfusions are well tolerated without too much iron overload. This test can be done regardless of age in the case of late diagnosis and also in adulthood in patients who had a phase of therapeutic independence after being treated in childhood
(3) : maximum tolerable dose: 0.30 mg/kg daily according to the international consensus but ideally a dosage of <0.15 mg/kg daily
(4) : long-term corticosteroid therapy should be discussed according to its efficacy and toxicity. A return to transfusions may be preferable for the patient. To be discussed especially in the pre-pubertal period to optimize growth and in adulthood, where a relative loss of corticosensitivity is regularly observed.
(5) : chelation objectives: ferritin between 300 and 500 µg/L and no iron overload in hepatic MRI (LIC <36 µmol/g)
(6) : the current consensus considers only these types of transplant. The transplant should ideally be done between 3 and 5 years of age.
(7) : these patients develop MDS early compared to the general population and are therefore potential candidates for an allograft.

Abbreviations: LIC : Liver Iron Content, HSC: hematopoietic stem cells, Hb: hemoglobin, MDS: myelodysplastic syndrome

:

THERAPEUTIC ASPECTS (1): treatment of anemia and follow-up

1) Patients on transfusion support:

The current consensus is not to use corticosteroids in children under 1 year old.

Transfusion support must allow normal growth and psychomotor development in children and must allow a reasonable quality of life in adults. The recommended transfusion threshold is 9.0 to 9.5 g/dL; tolerance to chronic anemia decreases rapidly with age and some adult patients may require more support.

The interval between 2 transfusions is 3 to 4 weeks; some patients who still have significant reticulocytosis may receive transfusions less often.
As in any regularly transfused patient, a serological assessment should be carried out at least once a year. Vaccination against hepatitis B is recommended if the patient is not immunized.
Monitoring of iron overload must be particularly rigorous: it is established that DBA patients are at higher risk of iron overload with equal transfusion support than other transfused patients (Porter JB & al., Br J Haematol 2014). This is particularly marked in small children, who can very quickly develop a severe tissue overload, including at the cardiac level, considering that the chelators are difficult to handle at this age.
Chelation must therefore be optimal and discussed as soon as the ferritin exceeds 500 µg/L, which is often the case before the age of 2 years.
The consensual objectives in these patients are to keep ferritin below 500 µg/l and hepatic (LIC; determined by hepatic MRI) and cardiac (myocardial T2* measured by MRI) iron load within the normal range.
The 2 most commonly used chelators are:

- Deferoxamine: dosage of 40 to 60 mg/kg daily in subcutaneous infusions and with a minimum passage time of 8 hours, ideally 12 hours. This therapy is often prescribed 5 days out of 7 to ensure the patient's adherence. In children younger than 2 years of age this is the only chelator that can be used; the maximum dose in infants and toddlers (up to 3 years) is 30 mg/kg daily.
- Deferasirox, which is used as first-line treatment in children over 2 years of age and adults at the usual doses (14 to 28 mg/kg daily)

Deferiprone is not usually recommended in DBA patients due to a proven higher risk of agranulocytosis (estimated at 10%): its prescription can only be considered in 3rd line treatment in a patient with uncontrolled iron overload, after discussion with expert physicians (MDT meeting) and after educating the patient and their relatives about the risk of infection (medical treatment for any febrile episode with written recommendations provided to the patient). It should be given in 3 doses at a maximum dose of 75 mg/kg daily (Lecornec et al., 2022).
The intensity of iron overload in these patients often requires the use of a combination of 2 chelators. This also often limits the side effects of each drug.
It should be noted that one of the objectives of chelating treatment is also to control the non-transferrin bound iron (NTBI) present as soon as transferrin saturation exceeds 60% to 70% (which is the case in practically all transfused DBA patients). This in theory requires daily administration of a chelator and it is possible, for example, to give 2 days of deferasirox to a patient treated with deferoxamine 5 days out of 7.
Evaluation of iron overload must be carried out:

- By monitoring ferritin levels: these should be evaluated just before a transfusion, in the same lab, and monthly if possible. If it is not possible to estimate the tissue overload based on an isolated ferritin level, consideration of the development of levels, the prescribed chelators and the patient's history usually allows adequate monitoring

- Monitoring of the transferrin saturation coefficient is especially useful either at the beginning of the transfusion program to judge the buildup of overload following the transfusion or in a patient with very good chelation levels, in order to adapt the therapy; however, it is very rare to succeed in bringing the saturation coefficient down to an appropriate level (<60%).

- Hepatic MRI should be performed at the age of 5 to 6 years at the latest and possibly earlier in case of uncontrolled overload, and then every 12 to 24 months depending on the degree of overload. Cardiac MRI should also be performed regularly, at least during the first evaluation and then depending on the results and the development of iron overload.

It should be noted that in patients with ferritin levels of around 500 µg/L, the risk of chelating agent toxicity is greater. It must be ensured that control is also good at the tissue level, but usually it is necessary to reduce the dosage of chelators, particularly deferasirox, to avoid toxicity problems.

Other systematic follow-ups in transfused patients:

- The toxicity of chelating agents should be monitored according to the usual recommendations.
- Regular assessments (1 per year) to detect endocrine, hepatic and cardiac damage from hemochromatosis

2) Patients on corticosteroids:

Corticosteroid test: corticosteroid therapy should be evaluated at the age of 1 year. This delay allows the diagnosis to be confirmed and limits the impact on growth and final size in adulthood.

However, this can be modulated: in the event of significant growth retardation, and if transfusions are well tolerated in a child without uncontrolled iron overload, the test may be delayed to 18 months; on the other hand, in a country where transfusion support is not reliable, an earlier trial can be discussed.

The starting dosage in children (as in adults) is 2 mg/kg daily in 2 doses without exceeding 80 mg.

Responder patients have a reticulocyte crisis around D10: if this is confirmed, corticosteroids can be lowered from D15 and then every 15 days provided that reticulocytes are maintained and Hb levels normalize. This can initially be done in steps of 0.5 mg/kg, down to a dosage of 0.5 mg/kg daily. From this threshold, more gradual steps of 0.1 mg/kg every 2 to 4 weeks will be taken to determine the minimum dose needed to maintain hemoglobin levels. According to the consensus, 0.30 mg/kg daily is the maximum tolerable long-term dosage. The ideal, often achieved with “true” corticosensitive patients, is to have a dosage of less than 0.15 mg/kg daily, knowing that for some patients erythropoiesis can be maintained with a dosage that can appear “homeopathic”.

Once the minimum dosage is defined, it may possibly be doubled for children and given 1 day out of 2, although the benefit of this in terms of growth is currently being rediscussed and this arrangement may be associated with a certain degree of adrenal insufficiency 1 day out of 2.

In the event of reticulocyte count relapse after the initial reduction, it must be concluded, depending on the case, either that the dose has been reduced below the minimum dose, and it will be necessary to go back, or (if this relapse is early) that the patient has developed corticosteroid dependence at too high a dose, and it will be necessary to return to transfusions.

Patients who do not respond at D15 should receive another 15 days of corticosteroids at 2 mg/kg before concluding that there is primary corticosteroid resistance (20% of patients); in the case of non-response, discontinuation of corticosteroids will be carried out quickly (one week). A second corticosteroid test can be done at a later stage, but a more favorable result is rare.

DBAis the only example of human disease where corticosteroids can be prescribed from the age of 1 year for an indefinite period (“lifetime” treatment) and their side effects must be regularly evaluated, in particular for bone involvement, which routinely occurs even at very low doses and warrants monitoring with bone densitometry. Vitamin D supplementation should be systematic.

Continued corticosteroid therapy should be regularly discussed depending on its effectiveness (some DBA patients lose their corticosensitivity over time and do not maintain a hemoglobin level compatible with a good quality of life) and its side effects. In these patients with a “double sentence” (toxicity and insufficient Hb level), it is necessary to return to transfusion support.

Temporary discontinuation of effective corticosteroid therapy may be indicated in various circumstances:

- Pre-pubertal period: to optimize pubertal growth spurt and final size. Even very low doses (<0.15 mg/kg daily) do not allow a normal pubertal growth spurt. This should be discussed between the pediatric endocrinologist and the patient, bearing in mind that the withdrawal of corticosteroids should be for 2 to 3 years.
- Growth hormone therapy (see below).

• Pregnancy: in the case of corticosteroid therapy at a relatively high dose and particularly since the physiological anemia of pregnancy can "decompensate" anemia; to be discussed with the obstetric team

“Remissions” (these tend to be referred to as therapeutic independence periods) may occur after a period of corticosteroid therapy. In a French registry study, 42 out of 125 patients who responded to corticosteroids were in therapeutic independence during the analysis (Willig & al., 1999). These remissions occur either in small children (before the age of 2 years) or during adolescence. In very rare cases (2 published), a somatic reversion in a hematopoietic stem cell has been demonstrated, but the current consensus is that this mechanism occurs in only a tiny minority of patients.
These untreated patients have minor blood count abnormalities (borderline Hb, macrocytosis, elevated HbF, tendency to leukoneutropenia).
They should be regularly monitored since a “relapse” of anemia can occur in adulthood and they also have the same risk of solid tumors or hypogammaglobulinemia as all DBApatients.

3) Bone marrow transplantation:
An indication for bone marrow transplantation should be discussed:
- In any corticosteroid-resistant child that is dependent on transfusions. The results of the transplant are better in children under 10 years and the current recommendations are to do this before the age of 5 years if possible, and ideally without too much iron overload. It is generally accepted that transplants are too risky for adult patients.

The new consensus for this indication is that we can consider 2 types of donor:

- Either a geno-identical donor: assuming that the disease has been excluded in them, which can be difficult in the absence of identified mutation; at the slightest doubt, it will be necessary to discuss choosing an unrelated donor
- Or an unrelated (10/10) donor, given the current good results (Strahm & al., Blood Adv 2020)

- In any patient presenting a clonal evolution, regardless of age or graft type.

NB: Very rarely, other clinical situations may lead to the discussion of an allograft: severe neutropenia (*RPL35a* gene) or uncontrollable iron overload.

Methods of transplant:

Given the results reported in the literature, the transplant should preferably be carried out between the ages of 2 and 10 years. Before the age of 2, the risk of toxicity is increased, and except in the case of early myelodysplastic syndrome (very rare) , earlier transplantation is not justified. Beyond the age of 10, the results of transplants are significantly less good in terms of disease-free survival and survival without GvHD (GRFS), with an excess of GvHD possibly related to more transfusions in the history and greater allo-immunization.

The recommended conditioning is myeloablative as these patients have a rich bone marrow:
- BUSULFAN adapted to the weight of the child and in accordance with the SmPC (associated with pharmacokinetics with standard area-under-the-curve (AUC) targets of 900–1350 µMolxmin) in 4 days from D-5 to D-2, FLUDARABINE 40 mg/m^2 daily x 4 days from D-6 to D-3 and serotherapy (Thymoglobulins® 10 mg/kg, total dose).
- or reduced toxicity conditioning with TREOSULFAN and FLUDARABINE +/- THIOTEPA and serotherapy (Thymoglobulins® 10 mg/kg, total dose). The SmPC for treosulfan provides for adjustment of the dose of this medication to body surface area (BSA): 10 g/kg daily for children with a BSA of less than 0.5 m^2, 12 g/kg daily for children with a BSA of 0.4 to 1 m^2 and 14 g/kg daily for patients with a BSA of greater than 1 m^2. It should be noted that this medication currently does not have an MA in the treatment of non-malignant pathologies in pediatrics.
There is no indication for total body irradiation

- Prophylaxis for graft versus host disease is standard, based on a calcineurin inhibitor combined with "short" methotrexate or mycophenolate mofetil when using a related placental blood graft.
- The preferred cellular source is bone marrow, to reduce the risk of chronic GvHD.
- The use of an alternative donor, a non-related voluntary donor of HLA compatibility <10/10 or haplo-identical donor, should only be discussed in case of myelodysplastic syndrome. The use of unrelated placental blood does not lead to satisfactory results, but studies reporting these disappointing results are old (Mugishima H et al., 2007 and Bizzoto et al., 2011).

4) Other treatments for anemia

There is currently no other drug treatment for anemia in DBA patients other than corticosteroids.

Erythropoietin, sotatercept or ciclosporin are not active. Eltrombopag is not a validated therapy and should not be used outside of a possible clinical trial.

Only leucine has been associated with very rare hematological responses; although the impact on height-weight growth is relatively common and may justify its use in children, hematological responses are nevertheless very rare (Vlachos Lipton & al, Pediatr Blood Cancer 2021).

THERAPEUTIC ASPECTS (2): other disorders

The follow-up of these patients must be multidisciplinary and will involve pediatricians and adult doctors from different specialties.

Management of congenital malformations
Variable depending on type.

Growth monitoring
DBA patients have a high risk of small stature in adulthood. The causes are multiple:

- Small stature due to syndrome and IUGR
- Rare growth hormone deficiencies (with or without abnormality of the pituitary region in imaging)
- Effects of corticosteroids
- Effects of iron overload
- Delayed puberty
- Chronic anemia

This justifies a precise monitoring of the growth of these children and at least one systematic consultation in pediatric endocrinology in order to assess the impact of the disease on growth. The impact of possible corticosteroid therapy will be discussed (see above).
Growth hormone therapy has shown short-term efficacy in catching up with the growth curve, but no adult height data are available (Howel & al., Pediatr Blood Cancer 2015). Such therapy may be offered to children with the most severe growth retardation, within the framework of the usual indications for these therapies. The indication should be discussed with a pediatric endocrinologist and will take into account expected height in adulthood and the expected gain in cm. It should be noted that the initiation of GH therapy must be associated with discontinuation or reduction of corticosteroid therapy and the establishment of transfusion support. More recently, leucine therapy has been proposed, which could improve the height-weight growth of DBA children (Vlachos & al, Pediatr Blood Cancer 2020).

Immunological monitoring
DBA patients are often lymphopenic and this can be accentuated by corticosteroid therapy. Changes of the CVD type (common variable deficiency) have been reported and this requires annual measurement of Ig G, A and M.

Oncological risk
DBA patients have an increased risk of malignant hemopathies and cancers.

1) MDS and AML

The risk is very high for myelodysplastic syndromes, which occur at a younger age than in the general population; it is considered that there is premature aging of the bone marrow in DBA patients, and MDS can occur most often from the age of 50–60 (Vlachos & al, Blood 2012). However, there are no published descriptive data on these MDS.

In all patients, a FBC with reticulocyte count should be carried out every 3 months; any unusual changes in the FBC should lead to discussion of a bone marrowaspiration and biopsy. To date, there are no recommendations regarding the practice of systematic bone marrow differential cell counts.

2) Solid tumors

The risk of solid cancers is also increased and this has been demonstrated in 2 successive studies of the American registry (Vlachos & al, Blood 2012, Vlachos & al, Blood 2018). The median age for the first tumor is 36 years. The types of cancers observed are variable; the 2 cancers appearing significantly more frequently than in the general population are osteosarcoma and colon cancer.

To date, there are no “official” recommendations regarding the practice of screening tests for these tumors.

However, the American registry recently recommended the practice of systematic colonoscopies: every 5 years from the age of 20 (Lipton & al, Genes 2022) and this recommendation will be included in the next guidelines (to be published in 2024)

Patients and their treating physicians should be informed of the increased risk of cancer.

Patients should be aware of the main symptoms requiring specialist consultation and they and their physicians should at a minimum follow the recommendations given for cancer prevention in the general population.

DBA annual patient follow-up summary:

In all patients	FBC with reticulocyte count at least quarterly; bone marrow differential cell count if FBC abnormality Ig G, A, M levels: Once a year Colonoscopy: every 5 years from the age of 20
In patients taking corticosteroids	Annual metabolic assessment Vitamin D level: Once a year Bone densitometry every 2 to 3 years
In transfused patients	Viral serology (HIV, HBV & HCV, HTLV): Once a year Ferritin: before each transfusion Hepatic and myocardial MRI: every 12 to 24 months depending on degree of overload ECG & cardiac ultrasound: every 12 to 24 months; if myocardial overload on MRI: Holter Endrocrinologic follow-up once a year; sex hormones, T4 and TSH, PTH Monitoring for toxicity & side effects of chelating agents: Fundus of the eye, lens examination (deferasirox): once every 1 to 2 years Audiogram (deferasirox, deferoxamine): once every 1 to 2 years Renal ultrasound: search for lithiasis (deferasirox, deferoxamine): once every 2 to 3 years Zinc deficiency: once a year

<u>Genetic counseling and medically assisted procreation</u>

DBA tis most often sporadic (55%) (dominant neomutation); in familial forms; its transmission is autosomal dominant for RP genes. No genetic anticipation has been described inDBA . The severity of the phenotype (hematological and extra-hematological) of an unborn child cannot be

predicted, either by the gene or mutation identified, or by the phenotype of the parent or other affected siblings.
Parents should therefore be informed that in case of familial forms with one of the two parents suffering from or carrying a mutation and/or a high eADA, the risk of birth of an affected child is 50% each time and that the birth of a severely affected child cannot be excluded.
This calls for genetic counseling, which must be offered to all parents and adult patients. It is essential if parents wish to have another child by assisted reproduction.
This assumes that the mutation has been identified and confirmed as pathological and that it allows an analysis of DNA extracted from a single embryo cell.
The practice of selecting an embryo that is both unaffected and HLA-identical with a sick brother or sister (double selection) is no longer practiced in France at present but can be done in some EU countries

Pregnancy
Women with DBAhave a high risk of complicated pregnancies (Faivre and Leblanc & al, Haematologica 2006).
This warrants follow-up in level 3 maternity units.
The required dose of corticosteroids may be too high for fetal development and should be discussed with the obstetrical team; this may involve the provision of transfusion support during pregnancy. This may also be necessary for a woman in therapeutic independence but with a hemoglobin level that is too low. For a woman undergoing transfusion support, transfusion intake will normally need to be increased.
In all cases, the objective during pregnancy is to maintain hemoglobin above 10.5 g/dl, in line with the data obtained in women with thalassemia.
Chelating agents are contraindicated during pregnancy and this should ideally be anticipated, with a prior period of chelation intensification.
Ultrasound monitoring must be vigilant for risks of:

- fetal damage:
    - screening for severe malformations by morphological fetal ultrasound
    - screening for severe fetal anemia with hydrops fetalis (very rare in DBA but possible and underestimated), which may require in utero transfusions.
    - a CBC with a reticulocyte count and a genetic test carried out on cord blood or even on a fetal blood sample before transfusion are justified.
- maternal complications of the vasculo-placental type

The level of iron overload should be re-evaluated at the end of pregnancy to discuss how to resume chelation.

Annex 10: *SAMD9* & *SAMD9L* syndrome

This is an entity of aplastic anemia of inherited origin identified in 2016 (Chen & al, Am J human Genet 2016, Narumi & al, Nat Genet 2016): knowledge about this syndrome remains limited to date, particularly for data on long-term follow-up.

EPIDEMIOLOGY

Epidemiological data are limited, but this is a non-exceptional IBMF, especially for forms in young children (before 5 years of age). In the CRMR study, *SAMD9/SAMD9L* mutations accounted for 9% of patients in a cohort of 179 patients composed mostly of children (median age: 11 years; age <2 years: 21%) presenting with aplasia that appeared to be inherited (Fanconi disease and other classic forms of IBMF excluded). This proportion rises to 18% if we consider only the cases (48% of the cohort) for which a genetic origin is confirmed (Bluteau & al, Blood 2018). In the EWOG and Saint Jude study, these cases represent 8% of a cohort of 669 patients classified as “myelodysplastic syndromes in children” (Sahoo & al, Nat Med 2021). It should be noted that a significant proportion of undiagnosed cases (pancytopenia that resolves rapidly) cannot be ruled out.
Transmission is autosomal dominant and all ethnic groups are, *a priori,* affected.

GENETICS

Two genes are involved: *SAMD9* and *SAMD9L*. They are juxtaposed gene on long arm of chromosome 7 (7q21.2). The functions of the proteins encoded by these genes remain poorly understood. They have been linked to inflammation, antiviral responses, organogenesis and tumor suppressionFinally, at the cellular level, they are involved in endosomal fusion and protein translation. Transmission is autosomal dominant and the mutations reported are monoallelic gain of function mutations.
In this syndrome, a high proportion of events have been reported to enable "somatic genetic rescue", i.e. correction of aplasia through the emergence of clones carrying monosomy 7, somatic mutations of the gene in question (correcting or attenuating the effect of the germline mutation) or uniparental disomy of the “wild-type” allele. It should be noted that mutations of these genes have been described as associated with other phenotypes:

- Normophosphatemic familial tumoral calcinosis: "loss-of-function" and bi-allelic germinal of *SAMD9 mutations*
- Auto-inflammatory disease with excessive interferon-type response: acquired "gain-of-function" of *SAMD9L mutations*

CLINICAL MANIFESTATIONS

Initially 2 distinct clinical syndromes were reported: ataxia-aplasia syndrome (linked to *SAMD9L* mutations) and MIRAGE syndrome (linked to *SAMD9* mutations); MIRAGE is an acronym for the main elements of the phenotype: Myelodysplasia, Infections, Restriction of growth, Adrenal hypoplasia, Genital phenotype and Enteropathy (Chen & al, Am J human Genet 2016, Narumi & al, Nat Genet 2016).
With the increase in the number of cases identified, it has become evident that these initial descriptions of 2 different entities do not reflect clinical reality; it is not accepted today that any of these clinical signs is specific to a given gene, and we prefer to talk about *SAMD9/SAMD9L* syndrome. Clinical expression remains very heterogeneous and some patients do not have an extra-hematological phenotype when diagnosed with aplastic anemia (although it may be revealed by thorough examinations or appear over time), while other patients have had a clinical picture

initially dominated by extra-hematological damage, sometimes very severe or even leading to early death (MIRAGE syndrome).
The exact extent of the extra-hematological phenotype remains to be defined; the predominant extra-hematological features are reported below.

Organ & location	Clinical signs
Morphotype and growth	- intrauterine growth retardation - short stature
Endocrine system	- Adrenal hypoplasia and adrenal insufficiency
Genital system	- Hypogonadism
Lungs	- Alveolar proteinosis
Gastrointestinal	- Enteropathy with chronic diarrhea and malabsorption
Central nervous system	- Ataxia (which may only appear in adulthood or in the elderly) - White matter abnormalities on MRI (children)

Hematological features are hypoplasia or aplastic anemia with a risk of clonal evolution.
The clinical and hematological features of aplastic anemia in children that may suggest *SAMD9/SAMD9L* syndrome are:

- Family history
- Young age (< 5 years), although a later revelation is possible
- No history of neonatal thrombocytopenia (which would suggest amegakaryocytosis), of severe phenotype of of the Hoyeraal-Hreidarsson syndrome (which would suggest telomeropathy) or of MECOM syndrome, i.e. the other 3 IBMFs entities that can be seen in small children
- A relatively acute onset (whereas it is a progressive onset that typically suggests a inherited form), sometimes in the course of an infectious episode or a vaccination
- Presence of a monosomy 7 from the diagnosis
- Quick and spontaneous improvement of cytopenia

DIAGNOSIS

As in all IBMFs, HbF can be abnormally elevated at diagnosis.
The bone marrow aspirate will suggest aplastic anemia or medullary hypoplasia; dysmegakaryopoiesis typical of monosomy 7 may be present. An OMB may be indicated to confirm hypocellular marrow, although it is not systematically performed, particularly in small children, in whom this procedure is performed under general anesthesia.
The presence of a monosomy 7 in the medullary karyotype may suggest these syndromes, and FISH must also be carried out to look for this. The medullary karyotype can also reveal the presence of other clonal abnormalities, and this should then raise concerns about progression to leukemia. This risk should be specified by means of “myeloid” NGS to look for acquired mutations of genes involved in MDS and AML.
Genetic diagnosis, most often done by NGS, will identify the presence of a “gain-of-function” mutation of the *SAMD9* or *SAMD9L* gene. It should be noted that the reported mutations are often classified as “variant of unknown significance” (class 3 according to the nomenclature of the ACMG) and can be difficult to interpret. NGS on the blood can also reveal the presence of acquired mutations of the same gene, suggesting the establishment of a somatic genetic rescue phenomenon (these mutations are most often in cis). The mutation can be completely lost in the blood (mononuclear cells) via monosomy 7 or acquired uniparental isodisomy, which then requires analysis on non-hematopoietic cells such as skin fibroblasts.

Finally, a family investigation must be carried out. It can identify a suggestive phenotype in the carrier parent or a member of their lineage. The genetic test can also identify pauci-clonal hematopoiesis in the carrier parent, attesting that they have had aplastic anemia (whether identified as such or not) and that they have "corrected" their hematopoiesis. Here too, the germinal mutation of a carrier parent can go unnoticed because of a complete correction by acquired uniparental isodisomy, which can lead to the erroneous belief that the index case mutation is *de novo*.

FOLLOW-UP AND THERAPEUTIC MANAGEMENT

Care must be multidisciplinary, although there are no established recommendations for the moment.

I - Follow-up and management of hematological aspects

The possibility of spontaneous correction of aplastic anemia makes this entity a very specific situation. The same is true of the frequency of monosomy 7, which is typically a poor prognostic factor, indicating an emergency allograft, and which here can be seen as a favorable event, allowing correction of hematopoiesis for a time.
The situation is further complicated by the absence of:

- Accurate data about the proportion of patients who will "correct" their hematopoiesis
- Criteria to predict development, either toward complete correction of medullary impairment (with loss of monosomy 7) or toward progression to leukemia.

Management of these patients should therefore be discussed at the MTM.
The key elements for deciding on the therapeutic strategy are:

- The monitoring of blood counts (once a week or more according to clinical signs in a patient transfused with platelets to once a month in a patient with moderate pancytopenia): evidence of an improvement or not in the counts. Improvement can be "paroxysmal" very soon after diagnosis or much more progressive. It can be complete (normal FBC) or partial (persistence of moderate cytopenias).
- Cytological analysis of the bone marrow smear (once every 3 months in case of monosomy 7): worsening of myelodysplastic aspects, appearance of blasts; the opinion of an expert cytologist can be requested from the hematologists at the CRMR.
- Monitoring of medullary karyotype: appearance of other chromosomal abnormalities known to be associated with leukemic progression.
- Molecular monitoring in two respects:
    - Search for adverse elements: appearance of somatic mutations of genes known to be involved in leukemic progression
    - Search for favorable elements: appearance of somatic mutations of SAMD9 or SAMD9L suggesting genetic correction (several different clones can be identified and will be quantified and monitored).

In all cases, the search for a family or unrelated donor must be initiated in principle in order to anticipate a potential indication for transplantation

Depending on the evolution, 3 situations can be distinguished:

1) Rapid impression of counts improving in the FBC: the patient can then be monitored regularly according to the methods described above. It should be noted that this improvement can be very gradual and is not always complete. Monosomy 7 in particular can persist for more than a year.

The "reassuring" elements here include:

- Continuous improvement of FBC
- The absence of other chromosomal abnormalities and, after a certain time, the progressive decrease of monosomy 7 (to be quantified in FISH)
- The absence of abnormalities of myeloid genes in NGS and the appearance of "revertant" clones with increasing VAF

In case of normalization of the blood count and disappearance of monosomy 7, we propose (subject to data, which are still limited) monitoring including a blood count every 3 months and a bone marrow check every 12 to 18 months.

2) Presence of elements suggesting leukemic transformation: indication for allograft

3) Intermediate situation: no obvious correction but no worrying elements either. Again, there are no data currently available to specify the proportion of patients who will actually experience a "genetic rescue" or to determine a reasonable "wait time". The indication for a transplant should then be discussed at the MTM, especially if pancytopenia is severe requiring transfusions, and with risk of severe infections.

Allogeneic hematopoietic stem cell transplantation

HSC allograft is the only cure for aplastic anemia in the absence of spontaneous correction and also the only treatment for a characterized pre-leukemic condition.
When selecting a geno-identical sibling donor, it must be ensured that the donor is not a carrier of the mutation, bearing in mind that normal FBCs are not sufficient to provide reassurance and that genetic research must systematically be conducted; in case of doubt in an emergency, the preference for a 10/10 pheno-identical donor can be discussed.

The prognosis for these grafts seems standard, other than for children with severe extra-hematological involvement (MIRAGE syndrome). There are no reports of any particular sensitivity to graft conditioning or of any particular risk of GvHD.

Methods of transplant:
Few data are currently available in the literature to issue strong recommendations regarding transplantation.
Nevertheless, the following should be remembered:

- A definite indication for transplantation when there are karyotypic abnormalities or clonal events arise in addition to monosomy 7, or increasing signs of myelodysplasia in the bone marrow and/or transfusion dependence
- The use of standard or even reduced toxicity myeloablative conditioning :
  - Weight-based Busulfan in accordance with the summary of product characteristics (in combination with pharmacokinetics with standard AUC targets of 900–1350 µMolxmin) in 4 days from D-5 to D-2, fludarabine 40 mg/m^2 daily x 4 days from D-6 to D-3 and serotherapy.
  - Or thiotepa 8 mg/m^2 on D-7, treosulfan 14 g/m^2 daily from D-6 to D-4, fludarabine 40 mg/m^2 daily from D-6 to D-3 and serotherapy. It should be noted that this conditioning, proposed by the German EWOG-MDS group, does not provide for the adaptation of doses of treosulfan according to the body surface area (BSA), which is not in accordance with the summary of product characteristics, which states that the dose is 10 g/m^2 daily for children with a BSA of less than 0.4 m^2, 12 g/m^2 daily for children with a BSA between 0.4 and 1 m^2 and 14 g/m^2 daily for patients with a BSA greater than 1 m^2. It should be noted that treosulfan currently has no marketing authorization for the the treatment of non-malignant pathologies in pediatrics.
- There is no reason to use total body irradiation
- The existence of severe comorbidities, particularly in patients with MIRAGE syndrome or prolonged severe aplasia without any sign of myelodysplasia, may prompt discussion of the use of reduced intensity conditioning at the national MTM.
- Prophylaxis for graft versus host disease is standard, based on a calcineurin inhibitor combined with "short" methotrexate or mycophenolate mofetil when using a cord blood.
- The preferred cellular source is bone marrow (to reduce the risk of chronic extensive GvHD).
- The use of an alternative donor (non-related voluntary donor of HLA compatibility <10/10 or haplo-identical donor) should be discussed at the aplasia MTM, especially in case of myelodysplastic syndrome or exacerbation or prolonged severe aplastic anemia without tendency to correction.

Hematologic supportive care

Complementary care will be the same as for any aplasia. Transfusion support will be adapted to the FBC and, as with all IBMFs, transfusions will be used sparingly to avoid allo-immunization.
Hematopoietic growth factors have no role in the correction of cytopenias but can be used occasionally, especially G-CSF, which can be prescribed in case of severe infection. The role of TPO-RAs has not been established, which means that they cannot be recommended.

II - Follow-up and management of extra-hematological aspects (excluding post-allograft complications)

The extent of the extra-hematological phenotype has yet to be described. The search for organ damage must be systematic, at least for the most common types and for those types that may have an impact on the development of the child.
A diagnostic assessment should be based on the clinical findings and should include, at a minimum:

- An abdomino-pelvic ultrasound (search for adrenal hypoplasia in particular, but low diagnostic value of ultrasound)
- An echocardiography (rare cardiac malformations have been described, although it has not been confirmed to date that they are part of the phenotype)
- An endocrine assessment (search for adrenal insufficiency to be done in case of suspicion of *SAMD9 & SAMD9L* syndrome because of the risk of acute adrenal insufficiency)
- A neurological and neurosensory assessment with neurological evaluation and brain MRI
- An assessment by a psychologist

The management of initial disorders will be ensured by organ specialists. It should anticipate the development of bone marrow failure; in particular, if surgical interventions are necessary it is logical to perform them before the occurrence of severe thrombocytopenia.

The follow-up of these patients is most often multidisciplinary and will involve pediatricians and doctors for adults from different specialties as well as members of paramedical professions.
At pediatric age, this follow-up is dominated by the management of the most severe extra-hematological disorders: enteropathy (which may require parenteral nutrition), hypogonadism, adrenal insufficiency, growth and neurosensory and developmental aspects.
Few data are available for adult patients; neurological follow-up is still necessary (possible late development of ataxia or pseudo-parkinsonian syndrome). There does not appear to be, based on the current state of knowledge, a risk of MDS/AML in adulthood or an increased risk of solid tumors. On the other hand, hypogammaglobulinemia may develop and should be managed according to the recommendations available for common variable immunodeficiencies.
These patients should be monitored throughout their lives, including non-allograft patients.

a) Sensory and neurological disorders, psychomotor development

Patients with *SAMD9/SAMD9L* syndrome may have different types of neurological damage:

- Ataxia syndrome (with cerebellar hypotrophy) or pseudo-parkinsonian syndrome
- Spinocerebellar ataxia
- White matter disorders that may be associated with developmental abnormalities.

This requires an evaluation and systematic neurological follow-up in children and adults (possible late development of ataxia or pseudo-parkinsonian syndrome).

b) Endocrinological and gastroenterological monitoring, including monitoring of growth and puberty

SAMD9/SAMD9L patients are at risk of small stature in adulthood. The causes are potentially multiple:

- Related to the disease (small stature due to syndrome), associated with IUGR
- Chronic anemia (rare situation here)
- Sequelae of therapies received (corticosteroids, graft conditioning)

They may also have adrenal insufficiency related to adrenal hypoplasia.
Pubertal delays are possible; screening will need to be carried out for these, and management with hormone replacement therapy may be necessary. Growth hormone therapy may be indicated.

This requires systematic follow-up by a pediatric endocrinologist.
Children may develop enteropathy, which may require parenteral nutrition.

c) Solid cancers

A “spontaneous” increased risk of solid tumors has not been reported to date in these patients. However, this may exist for allograft patients.
In general, there may be concerns that these patients could have a reduced bone marrow reserve (pauci-clonal hematopoiesis or grafted patient) and poor hematological tolerance to chemotherapy.

d) Immunological monitoring

Like all non-transplanted IBMF patients, these patients are at risk of developing a predominantly B immune deficiency in adulthood, although few data are currently available.
This requires annual monitoring of immunoglobulin levels (IgG, IgA, IgM).

e) Reproduction and pregnancy

These patients may have hypogonadism and reduced fertility, especially if there are urogenital malformations and a risk of sequelae induced by bone marrow allograft.
Patients should be able to have consultations in a fertility center.

f) Genetic monitoring

1. Genetic diagnosis

A genetic diagnosis makes it possible to confirm the diagnosis, to check if one of the 2 parents is a carrier and thus specify the risk of recurrence in the family, to detect a still-silent form in a sibling as part of the donor search, and to offer a prenatal diagnosis (PND) or pre-implantation diagnosis (PID) in the event of a desire for a new child.

2. Genetic counseling

This can be offered to all parents and adult patients if they wish. It is essential if parents wish to have another child by assisted reproduction. This assumes that the mutation has been identified and confirmed as pathological and that it allows an analysis of DNA extracted from a single embryo cell. The practice of selecting a non-affected embryo that is HLA-identical to a sick sibling (double selection) is no longer used in France.

Annex 11: iron chelation

1- Screening and indication

Screening for secondary hemochromatosis
Determination of serum ferritin and transferrin saturation coefficient every 3 months
Just before a transfusion, if possible on an empty stomach

Chelation if ferritin >500 µg/L

In the event of high overload (serum ferritin >1000 µg/l): MRI measurement of intrahepatic and intracardiac iron concentration.

Refer to Annex 9 “Diamond Blackfan anemia” for chelation in this disease.

2- Chelating drugs

There is no optimal chelating agent: each of them exposes patients to prescription difficulties or toxicity.
Monitoring renal function and CBC.
In case of renal toxicity in a patient taking ciclosporin, immunosuppressive therapy has priority.

Medications	Problems posed in aplasia
Deferoxamine - Desféral®	Subcutaneous administration (in practice tolerable with "pin-type" needles) Risk of infection: *Mucor, Yersinia enterolytica*
Deferasirox - Exjade®	Major renal toxicity in combination with ciclosporin Rare cases of medullary toxicity Hepatic toxicity
Deferiprone - Ferriprox®	Risk of agranulocytosis and neutropenia, or even Aplastic anemia in the case of predisposition

Deferoxamine (Desféral®):
40 to 60 mg/kg per day subcutaneously over 8 to 12 hours (ideally 12 hours), 5 days out of 7 for better patient adherence.
Requires a home health provider to provide the pump or infuser. The subcutaneous infusions can be placed by a nurse or the patient themselves.
In pediatrics: only chelator available before 2 years. Maximum dose of 30 mg/kg daily for up to 3 years.

Deferasirox (Exjade®):
14 to 28 mg/kg once daily
In children over 2 years of age and in adults

Deferiprone (Ferriprox®):
25 mg/kg 3 times daily

3- Choice of chelating agents

There are no validated recommendations for the choice of medication.

Deferoxamine (Desféral®) is proposed as a first-line treatment in patients with AA or on CSA..

Deferiprone (Ferriprox®) should be avoided in the context of aplasia.

In case of major overload, a combination of chelating agents is possible, for example in the pre-transplant period to minimize iron overload.

4- Indication for phlebotomies

Patients in remission of their aplasia or in post-allograft.
Good efficacy and no side effects.

10 to 20 ml/kg/phlebotomy (maximum 400–500 ml) every 15 days
Volume and frequency to be adapted to clinical tolerance.

CBC at each phlebotomy with hemoglobin maintained >10 g/dl.

Possibility of using a health provider to carry out phlebotomies at home, after carrying it out in a hospital setting the first time.

The total number of phlebotomies is determined by monitoring the iron balance and iron overload, evaluated by hepatic and cardiac MRI.

Annex 12: Proprietary drugs used outside their indications or conditions for off-label use - Procedures, products or services not reimbursed at 100% by health insurance

- **Proprietary drugs used outside their indications or conditions for off-label use**

Proprietary drugs	**Possible temporary authorization for use Early access authorization (EAA)**	**Indications** Refer to therapeutic algorithms
ATGAM® (horse ATG)	EAA	Acquired AA
CAMPATH® (Anti-CD52 monoclonal Ab)	Temporary authorization for use in cohort study	Acquired AA
DANATROL® (Danazol)	-	Aquired AA IBMF
ASPAVELI® (pegcetacoplan)	EAA	PNH
TRECONDI® (treosulfan)	-	Transplant conditioning

- **Procedures, products or services not reimbursed by health insurance (at 100%)**

Fanconi's disease

Dental care:

Recommended product: Mouth guard

Management of specific learning disorders:

1. Psychomotor education for preschool children (<6 years)
   - Implement a left-to-right and top-to-bottom search strategy (reading type) using cross-reference exercises (such as Ortho edition's "Watch my eyes")
   - Strengthen visuo-constructive capabilities through complex figure reproduction games (such as Ortho edition's "COMPOX") and the manipulation of construction objects (cube games, Tangrams, puzzles, etc.) with a construction strategy that goes from the most general to the smallest detail
   - Consolidate praxic abilities (through gestural imitation games), manual dexterity and visuo-motor coordination (exercises such as labyrinth games, coloring, mandalas, etc.)
   - Develop prerequisites for handwriting skills
   - Work on tonico-emotional regulation

2. Occupational therapy for school-age children (>6 years)
   - Evaluate fine motor skills and graphomotor skills
   - Improve/adapt the hold of the pencil
   - Improve the fluidity of graphomotor movements
   - Offer material aids if necessary to help children get used to IT tools with a view to integrating them into the classroom

Recognition of specific learning disorders with the teaching team (1) **or with the MDPH** (2):

(2) For simple adjustments, a simple Personalized Welcome Project may suffice: the PWP is a simple mechanism for organizing adjustments to the schooling of children suffering from Learning Disabilities: it is circular no. 2003–135 of 8–9–2003 (insert in the B.O. of 18–9–2003) on the reception of children and adolescents suffering from health disorders evolving over a long period in public and private educational establishments under contract to the primary and secondary levels.

This PWP is a contract that must be signed by the school doctor, head teacher and the child's family (as well as members of the educational team).

(2) The **Personalized Schooling Project** is developed by the educational team in collaboration with parents and must be validated by the Commission on the Rights and Autonomy of Persons with Disabilities (CDAPH), based at the Departmental Center for Disabled People (MDPH). This project makes it possible to report on the severity of the repercussions of the learning disorders identified, requiring the implementation of human assistance (AVS-I) or material aids (integration of IT tools in the classroom).
In addition to specific measures that schools can take to accommodate children and recommendations for accommodative measures in schools, the MDPH file allows also to plan for a possible educational orientation, to lead to recognition of the degree of disability and to financial compensation to cover disability-related expenses, etc. The child's doctor fills out the medical certificate for this purpose, which must be attached to the MDPH file completed by the family.

3. Depending on the learning difficulties identified in children treated for Fanconi anemia, examples of recommended accommodative measures that schools can take are:

- Attentional fatigue/psychomotor agitation

- Placing the student in the first row, away from any distraction (door, window) and near the adult, if possible alone or next to a quiet classmate
- Setting up tutoring. Another student helping them repeat an instruction and return to the task
- Splitting long tasks into short steps
- Planning times when the student can move without disturbing others (distributing notebooks, cleaning the board, etc.)
- Avoiding withdrawals from recess, as children need them for exercise
- Because of their tiredness and inattention, students make more careless mistakes. It is therefore important to teach them to re-read their work systematically

- Graphomotor disorders

- Introducing a one-third time slot for carrying out checks, to take account of students' slowness and fatigue. In other words, focusing on the quality of work over the quantity
- During assessments, implement either:
  - one third extra time
  - one third fewer exercises
  - multiplication of the mark by 1.33
- Encouraging the use of pencil adapters (also called writing guides) to make it easier to handle the writing tool. On a daily basis, it is also advisable to use pens that predefine the location of the fingers (e.g. “Velocity” by Bic or “Griffix” by Pelikan) or architect’s pens (by Gibert or Graphigro) that slide easily and have a very solid tip
- Avoiding copying and writing in large quantities
- Whenever possible, asking to present knowledge orally, or proposing the completion of gap-fill and/or multiple-choice texts.
- If the goal is to promote speed, accepting a less accurate product. Allowing the student to type documents for homework assignments

If necessary, using a computer:

- Allow the use of a computer tool during lessons and in all national exams, once the preparatory sessions have been carried out in occupational therapy. The computer may have the following software:
  - Microsoft Office Word
  - And if necessary, Dragon Naturally Speaking (voice recognition)
  - Using a sliding ruler to easily copy a document (classmate’s lesson, etc.)

Annex 13: Practical sheets for the management of Aplastic anemia

Sheets are available on the website of the Reference Center for Aplastic anemia (section for professionals)

Annex 13a: interviewing and clinical examination in Aplastic anemia

Physician's name:
Date:

patient label

INTERVIEW	Y/N	Comments
Personal or family history suggesting IBMF		
Hemorrhagic syndrome		
Cytopenias, leukemia, myelodysplasia		
Cancer at an early age		
Skin and skin appendages, premature canities		
Opportunistic infections		
Pulmonary fibrosis		
Liver		
Unexplained early deaths		
Personal history suggesting IBMF		
Congenital malformations: Bone, heart, urogenital organs, etc. (Fanconi, BDA)		
IUGR (Fanconi, BDA, Shwachman)		
Chronic diarrhea, abdominal pain (Shwachman)		
Abnormal FBCs for more than 6 months		
Thrombosis (GATA2)		
Suggesting an acquired form		
Recent episode of jaundice or hepatitis		
CBC normal for <6 months		
Hemoglobinuria, abdominal pain, dysphagia, thrombosis, erectile dysfunction (PNH)		
Drugs (see PNDS Table 1)		

CLINICAL EXAMINATION	Y/N	Comments
Height-weight retardation (Fanconi, BDA, Shwachman)		
Facial dysmorphia (Fanconi, BDA)		
Malformations (Fanconi, BDA)		
Café-au-lait spots (Fanconi)		
Pigmentation, nail abnormalities, canities, oral leukoplakia (telomeropathies)		
Hepatosplenomegaly (telomeropathy: portal hypertension)		
Respiratory involvement (telomeropathies, GATA2)		
Lymphedema, warts, condylomas (GATA2)		
Deafness (GATA2)		
Radius abnormalities (TAR)		

Annex 13b: Evaluation of Aplastic anemia at diagnosis

Additional examinations recommended for the diagnosis of Aplastic anemia

For all patients:

patient label

- ☐ CBC + blood smear + reticulocytes
- ☐ Bone marrow differential cell count + medullary karyotype + FISH cen(7)
- ☐ Bone marrow biopsy
- ☐ Emergency HLA typing of the patient if age <60 years (and siblings if age of the recipient <40 years)
- ☐ T B NK lymphocyte phenotype (in immunology)
- ☐ Electrophoresis of proteins in adults and IgG, IgA, IgM weight assay
- ☐ Search for a PNH clone
- ☐ Hemoglobin electrophoresis (or HbF assay) before transfusion, if possible
- ☐ Alpha fetoprotein
- ☐ Anti-nuclear and anti-DNA antibodies
- ☐ Abdominal ultrasound (renal ++)
- ☐ Echocardiogram
- ☐ Blood electrolytes, urea, creatinine

- ☐ Blood calcium, phosphorus, magnesium
- ☐ Complete liver function tests
- ☐ Complete hemostasis
- ☐ Chest X-ray F+P
- ☐ HIV 1+2, hepatitis B and C, HTLV1, EBV, CMV serologies.

Depending on the context (see algorithm):

- ☐ Inherited blood karyotype with search for spontaneous chromosomal breaks and after MMC
- ☐ FANCD2 & NGS blood and fibroblasts test (skin biopsy)
- ☐ Telomere length study in patients under 60 years of age (Dr. E. Lainey, Laboratory of Hematology, Hôpital Robert Debré, Paris) on appointment, Monday and Tuesday
- ☐ Systematically associated search for mutations in telomeropathies (Prof. C. Kannengiesser, Molecular Genetics at Hôpital Bichat)
- ☐ Measurement of erythrocyte adenosine deaminase (eADA) activity (Dr. S. Sanquer, Biochemistry Laboratory, Hôpital Necker, Paris)
- ☐ Search for Diamond Blackfan anemia mutations (Prof. L. Da Costa, Laboratory of Hematology, Hôpital Robert Debré, Paris)
- ☐ X-rays of the bones of the forearms and hands
- ☐ CT scan of the chest to look for thymoma in adults

Annex 14. Bibliographic references

Ades L, Mary JY, Robin M, Ferry C, Porcher R, Esperou H, et al. Long-term outcome after bone marrow transplantation for severe aplastic anemia. Blood 2004;103(7):2490-7.

Agence nationale d'accréditation et d'évaluation en santé. Guide d'analyse de la littérature et gradation des recommandations, Paris: ANAES; 2000.

Ahmed IA, Farooqi MS, Vander Lugt MT, Boklan J, Rose M, Friehling ED, Triplett B, Lieuw K,et al. Outcomes of HSCT in patients with germline SMAD9/SAMD9L mutations, Biol Blood Marrow Transplant. 2019 Nov;25(11):2186-2196. doi: 10.1016/j.bbmt.2019.07.007. Epub 2019 Jul 12.

Alder JK, Hanumanthu VS, Strong MA, DeZern AE, Stanley SE, Takemoto CM, Danilova L, Applegate CD, Bolton SG, Mohr DW, Brodsky RA, Casella JF, Greider CW, Jackson JB, Armanios M. Diagnostic utility of telomere length testing in a hospital-based setting Proc Natl Acad Sci U S A. 2018 Mar 6;115(10):E2358-E2365. doi: 10.1073/pnas.1720427115. Epub 2018 Feb 20. Erratum in: Proc Natl Acad Sci U S A. 2018 Apr 23.https://www.ncbi.nlm.nih.gov/pubmed/29686093

Alter BP, Giri N, Savage SA, Peters JA, Loud JT, Leathwood L et al, Malignancies and survival patterns in the National Cancer Institute inherited bone marrow failure syndromes cohort study. Br J Haematol . 2010 Jul;150(2):179-88. doi: 10.1111/j.1365-2141.2010.08212.x. Epub 2010 Apr 30.

Alter BP, Giri N, Savage SA, Rosenberg PS. Cancer in the National Cancer Institute inherited bone marrow failure syndrome cohort after fifteen years of follow-up. Haematologica. 2018 Jan;103(1):30-39. doi: 10.3324/haematol.2017.178111. Epub 2017 Oct 19.

Appelbaum FR, Barrall J, Storb R, Ramberg R, Doney K, Sale GE, et al. Clonal cytogenetic abnormalities in patients with otherwise typical aplastic anemia. Exp Hematol 1987;15(11):1134-9.

Bacigalupo A, Hows J, Gordon-Smith EC, Gluckman E, Van Lint MT, Congiu M, et al. Bone marrow transplantation for severe aplastic anemia from donors other than HLA identical siblings : a report of the BMT Working Party. Bone Marrow Transplant 1988;3(6):531-5.

Bacigalupo A, Hows J, Gluckman E, Nissen C, Marsh J, Van Lint MT, et al. Bone marrow transplantation (BMT) versus immunosuppression for the treatment of severe aplastic anaemia (SAA) : a report of the EBMT Sworking party. Br J Haematol 1988;70(2):177-82.

Bacigalupo A, Figari O, Tong J, Piaggio G, Miceli S, Frassoni F, et al. Long-term marrow culture in patients with aplastic anemia compared with marrow transplant recipients and normal controls. Exp Hematol 1992;20(4):425-30.

Bacigalupo A, Chaple M, Hows J, Van Lint MT, McCann S, Milligan D, et al. Treatment of aplastic anaemia (AA) with antilymphocyte globulin(ALG) and methylprednisolone (MPred) with or without androgens: a randomized trial from the EBMT SAA working party. Br J Haematol 1993;83(1):145-51.

Bacigalupo A. Guidelines for the treatment of severe aplastic anemia. Working Party on Severe Aplastic Anemia (WPSAA) of the European Group of Bone MarrowTransplantation (EBMT). Haematologica 1994;79(5):438-44.

Bacigalupo A, Broccia G, Corda G, Arcese W, Carotenuto M, Gallamini A, et al. Antilymphocyte globulin, cyclosporin, and granulocyte colony-stimulating factor in patients with acquired severe aplastic anemia (SAA): a pilot study of the EBMT SAA Working Party. Blood 1995;85(5):1348-53.

Bacigalupo A. A etiology of severe aplastic anaemia and outcome after allogeneic bone marrowtransplantation or immunosuppression therapy. Working Party on Severe Aplastic Anaemia of the European Blood and Marrow Transplantation Group. Eur J Haematol Suppl 1996;60:16-9.

Bacigalupo A, Oneto R, Bruno B, Socié G, Passweg J, Locasciulli A, et al. Current results of bone marrow transplantation in patients with acquired severe aplastic anemia. Report of the European Group for Blood and Marrow transplantation. On behalf of the Working Party on Severe Aplastic Anemia of the European Group for Blood and Marrow Transplantation. Acta Haematol 2000;103(1):19-25.

Bacigalupo A, Bruno B, Saracco P, Di BE, Locasciulli A, Locatelli F, et al. Antilymphocyte globulin, cyclosporine, prednisolone, and granulocyte colony-stimulating factor for severe aplastic anemia: an update of the GITMO/EBMT study on 100 patients. European Group for Blood and Marrow Transplantation (EBMT) Working Party on Severe Aplastic Anemia and the Gruppo Italiano Trapianti di Midolio Osseo (GITMO). Blood 2000;95(6):1931-4.

Bacigalupo A, Brand R, Oneto R, Bruno B, Socié G, Passweg J, et al. Treatment of acquired severe aplastic anemia: bone marrow transplantation compared with immunosuppressive therapy--The European Group for Blood and Marrow Transplantation experience. Semin Hematol 2000;37(1):69-80.

Bacigalupo A, Socié G, Schrezenmeier H, Tichelli A, Locasciulli A, Fuehrer M, Risitano AM, Dufour C, Passweg JR, Oneto R, Aljurf M, Flynn C, Mialou V, Hamladji RM, Marsh JC; Aplastic Anemia Working Party of the European Group for Blood and Marrow Transplantation (WPSAA-EBMT). Bone marrow versus peripheral blood as the stem cell source for sibling transplants in acquired aplastic anemia: survival advantage for bone marrow in all age groups. Haematologica. 2012 Aug;97(8):1142-8. Epub 2012 Feb 7.

Bhala S and Savage S.A. CNS manifestations in patients with telomere biology disorders. Neurol Gen et. 2019; Oct 29; 5(6):370.

Barrett J, Sauntharajah Y, Molldrem J. Myelodysplastic syndrome and aplastic anemia: distinct entities or diseases linked by a common pathophysiology? Semin Hematol 2000;37(1):15-29.

Barrett J. Myelodysplastic syndrome and aplastic anemia--diagnostic andconceptual uncertainties. Leuk Res 2000;24(7):595-6.

Barone A. Diagnosis and management of acquired aplastic anemia in childhood. Guidelines from the Marrow Failure Study Group of the Pediatric Haemato-Oncology Italian Association (AIEOP).Blood Cells Mol Dis. 2015 Jun;55(1):40-7. doi: 10.1016/j.bcmd.2015.03.007. Epub 2015 Mar 31.

Baumelou E, Guiguet M, Mary JY. Epidemiology of aplastic anemia in France: a case-control study. I. Medical history and medication use. The French Cooperative Group for Epidemiological Study of Aplastic Anemia. Blood 1993;81(6):1471-8.

Bogliolo M, Surrallés J.Fanconi anemia: a model disease for studies on human genetics and advanced therapeutics. Curr Opin Genet Dev 2015 Aug;33:32-40 : 10.1016/j.gde.2015.07.002. Epub 2015 Aug 6.

Fanette Bernard, Chakradhara Rao S Uppugunduri, Stefan Meyer, Michelle Cummins, Katharine Patrick, Beki James, Roderick Skinner, Sanjay Tewari, Ben Carpenter, Robert Wynn, Paul Veys, Persis Amrolia, the UK Paediatric BMT group. Excellent overall and chronic graft-versus-host-disease-free event-free survival in Fanconi anaemia patients undergoing matched related- and unrelated-donor bone marrow transplantation using alemtuzumab–Flu–Cy: the UK experience. British Journal of Haematology, 2021, 193, 804–813

Bizzetto R, Bonfim C, Rocha V, Socié G, Locatelli F, Chan K et al.; Eurocord and SAA-WP from EBMT. Outcomes after related and unrelated umbilical cord blood transplantation for hereditary bone marrow failure syndromes other than Fanconi anemia. Haematologica. 2011 Jan;96(1):134-41. doi: 10.3324/haematol.2010.027839. Epub 2010 Nov 11.

Bluteau O, Sebert M, Leblanc T, Peffault de Latour R, Quentin S, A landscape of germ line mutations in a cohort of inherited bone marrow failure patients. Blood. 2018 Feb 15;131(7):717-

Bogliolo M, Bluteau D, Lespinasse J, Pujol R, Vasquez N, d'Enghien CD,Biallelic truncating FANCM mutations cause early-onset cancer but not Fanconi anemia. Genet Med. 2018 Apr;20(4):458-463. doi: 10.1038/gim.2017.124. Epub 2017 Aug 24.

Bonfim C, Ribeiro L, Nichele S, Loth G, Bitencourt M, Koliski A, Kuwahara C and al. Haploidentical Bone Marrow Transplantation with Post-Transplant Cyclophosphamide for Children and Adolescents with Fanconi Anemia. Biol Blood Marrow Transplant. 2017 Feb;23(2):310-317. doi: 10.1016/j.bbmt.2016.11.006. Epub 2016 Nov 7.

Borie, R, Tabèze L, Thabut G, Nunes Hilario, Cottin V, Marchand-Adam S, Prevot G, et al. 2016. "Prevalence and Characteristics of TERT and TERC Mutations in Suspected Genetic Pulmonary Fibrosis." The European Respiratory Journal 48 (6): 1721–31. doi:10.1183/13993003.02115-2015.

British Committee for Standards in Haematology, Marsh JC, Ball SE, Darbyshire P, Gordon-Smith EC, Keidan AJ, et al. Guidelines for the diagnosis and management of acquired aplastic anaemia. Br J Haematol 2003;123(5):782-801.

Brodsky RA, Sensenbrenner LL, Jones RJ. Complete remission in severe aplastic anemia after high-dose cyclophosphamide without bone marrow transplantation. Blood 1996;87(2):491-4.

Brodsky RA, Jones RJ. High-dose cyclophosphamide in aplastic anaemia. Lancet 2001;357(9262):1128-9.

Butturini A, Gale RP, Verlander PC, Adler-Brecher B, Gillio AP, Auerbach AD.Hematologic abnormalities in Fanconi anemia: an International Fanconi Anemia Registry study. Blood. 1994 Sep 1;84(5):1650-5.

Camitta B, O'Reilly RJ, Sensenbrenner L, Rappeport J, Champlin R, Doney K, et al. Antithoracic duct lymphocyte globulin therapy of severe aplastic anemia. Blood 1983;62(4):883-8.

Camitta B, Deeg HJ, Castro-Malaspina H, Ramsay NKC. Unrelated or mismatched bon marrow transplants for aplastic anemia: Experience at four major centers. Ontogeny of hematopoiesis aplastic anemia. International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235.

Camitta BM. What is the definition of cure for aplastic anemia? Acta Haematol 2000;103(1):16-8.

Chen DH, Below JE, Shimamura A, Keel SB, Matsushita M, Wolff J et al. Ataxia-Pancytopenia Syndrome Is Caused by Missense Mutations in SAMD9L. Am J Hum Genet. 2016 Jun 2;98(6):1146-1158. doi: 10.1016/j.ajhg.2016.04.009.

Champlin RE, Ho WG, Feig SA, Winston DJ, Lenarsky C, Gale RP. Do androgens enhance the response to antithymocyte globulin in patients with aplastic anemia? A prospective randomized trial. Blood 1985;66(1):184-8.

Champlin RE, Horowitz MM, van Bekkum DW, Camitta BM, Elfenbein GE, Gale RP, et al. Graft failure following bone marrow transplantation for severe aplastic anemia: risk factors and treatment results. Blood 1989;73(2):606-13.

Champlin RE, Nimer SD, Ireland P, Oette DH, Golde DW. Treatment of refractory aplastic anemia with recombinant human granulocyte-macrophage-colony-stimulating factor. Blood 1989;73(3):694-9.

Champlin RE, Perez WS, Passweg JR, Klein JP, Camitta BM, Gluckman E, et al. Bone marrow transplantation for severe aplastic anemia: a randomized controlled study of conditioning regimens. Blood 2007;109(10):4582-5.

Choesmel V, Bacqueville D, Rouquette J, Noaillac-Depeyre J, Fribourg S and al. Impaired ribosome biogenesis in Diamond-Blackfan anemia. Blood. 2007 Feb 1;109(3):1275-83. doi: 10.1182/blood-2006-07-038372. Epub 2006 Oct 19.

Collopy LC, Walne AJ, Cardoso S, de la Fuente J, Mohamed M, Toriello H, Tamary H, Ling AJ, Lloyd T, Kassam R, Tummala H, Vulliamy TJ, Dokal I. Triallelic and epigenetic-like inheritance in

human disorders of telomerase. Blood. 2015 Jul 9;126(2):176-84. doi: 10.1182/blood-2015-03-633388. Epub 2015 May 29.

Contejean A, Resche-Rigon M, Tamburini J, Alcantara M, Jardin F, et al. Aplastic anemia in the elderly: a nationwide survey on behalf of the French Reference Center for Aplastic Anemia. 2019 Feb;104(2):256-262. doi: 10.3324/haematol.2018.198440. Epub 2018 Sep 27.

Da Costa L, Leblanc T, Mohandas N. Diamond-Blackfan anemia. Blood. 2020 Sep 10;136(11):1262-1273. doi: 10.1182/blood.2019000947.

Da Costa L, O'Donohue MF, van Dooijeweert B, Albrecht K, Unal S et al. Molecular approaches to diagnose Diamond-Blackfan anemia: The EuroDBA experience. Eur J Med Genet. 2018 Nov;61(11):664-673. doi: 10.1016/j.ejmg.2017.10.017. Epub 2017 Oct 26.

Da Costa L, Crétien A, Marie I, Tchernia G, Leblanc T. L'anémie de Diamond-Blackfan, depuis la découverte du gène de protéine ribosomique S19 (rps 19). Hématologie 2005;11(6):385-96.

Dalle JH, Lucchini G, Balduzzi A, Ifversen M, Jahnukainen K, Macklon KT, Ahler A et al. State-of-the-art fertility preservation in children and adolescents undergoing haematopoietic stem cell transplantation: a report on the expert meeting of the Paediatric Diseases Working Party (PDWP) of the European Society for Blood and Marrow Transplantation (EBMT) in Baden, Austria, 29-30 September 2015.Bone Marrow Transplant. 2017 Jul;52(7):1029-1035).

de Guibert S, Peffault de Latour R, Varoqueaux N, Labussière H, Rio B, Paroxysmal nocturnal hemoglobinuria and pregnancy before the eculizumab era: the French experience. Haematologica. 2011 Sep;96(9):1276-83.

De Planque MM, Bacigalupo A, Wursch A, Hows JM, Devergie A, Frickhofen N, et al. Long-term follow-up of severe aplastic anaemia patients treated with antithymocyte globulin. Severe Aplastic Anaemia Working Party of the European Cooperative Group for Bone Marrow Transplantation (EBMT). Br J Haematol 1989;73(1):121-6.

de Latour RP, Mary JY, Salanoubat C, Terriou L, Etienne G, Paroxysmal nocturnal hemoglobinuria: natural history of disease subcategories. Blood. 2008 Oct 15;112(8):3099-106.

Dearden C, Foukaneli T, Lee P, Gordon-Smith EC, Marsh JC. The incidence and significance of fevers during treatment with antithymocyte globulin for aplastic anaemia. Br J Haematol 1998;103(3):846-8.

Deeg HJ, Lum LG, Sanders J, Levy GJ, Sullivan KM, Beatty P , et al. Severe aplastic anemia associated with chronic mucocutaneous candidiasis. Immunologic and hematologic reconstitution after allogeneic bone marrow transplantation. Transplantation 1986;41(5):583-6.

Deeg HJ, Self S, Storb R, Doney K, Appelbaum FR, Witherspoon RP, et al. Decreased incidence of marrow graft rejection in patients with severe aplastic anemia: changing impact of risk factors. Blood 1986;68(6):1363-8.

Deeg HJ, Anasetti C, Petersdorf E, Storb R, Doney K, Hansen JA, et al. Cyclophosphamide plus ATG conditioning is insufficient for sustained hematopoietic reconstitution in patients with severe aplastic anemia transplanted with marrow from HLA-A, B, DRB matched unrelated donors. Blood 1994;83(11):3417-8.

Deeg HJ, Doney K, Anasetti C, Storb R. Transplant and nontransplant therapy for patients with severe aplastic anemia: A update of the Seattle experience. Ontogeny of hematopoiesis aplastic anemia. International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235:312.

Deeg HJ, Socié G, Schoch G, Henry-Amar M, Witherspoon RP, Devergie A, et al. Malignancies after marrow transplantation for aplastic anemia and fanconi anemia: a joint Seattle and Paris analysis of results in 700 patients. Blood 1996;87(1):386-92.

Deeg HJ, Leisenring W, Storb R, Nims J, Flowers ME, Witherspoon RP, et al. Long-term outcome after marrow transplantation for severe aplastic anemia. Blood 1998;91(10):3637-45.

Deeg HJ, O'Donnell M, Tolar J, Agarwal R, Harris RE, Feig SA, et al. Optimization of conditioning for marrow transplantation from unrelated donors for patients with aplastic anemia after failure of immunosuppressive therapy. Blood 2006;108(5):1485-91.

Delanoy. N. Haematological immune-related adverse events induced by anti-PD-1 or anti-PD-L1 immunotherapy: a descriptive observational study. Lancet Haematol. 2019 Jan;6(1):e48-e57. doi: 10.1016/S2352-3026(18)30175-3. Epub 2018 Dec 4.

Desmond R, Townsley DM, Dumitriu B, Olnes MJ, Scheinberg P, Bevans M, Parikh AR, Broder K, Calvo KR, Wu CO, Young NS, Dunbar CE. Eltrombopag restores trilineage hematopoiesis in refractory severe aplastic anemia that can be sustained on discontinuation of drug. Blood. 2014 Mar 20;123(12):1818-25. doi: 10.1182/blood-2013-10-534743. Epub 2013 Dec 17.

DeZern AE, Zahurak M, Symons H, Cooke K, Jones RJ, Brodsky RA.Alternative Donor Transplantation with High-Dose Post-Transplantation Cyclophosphamide for Refractory Severe Aplastic Anemia. Biol Blood Marrow Transplant. 2017,Mar;23(3):498-504.doi: 10.1016/j.bbmt.2016.12.628. Epub 2016 Dec 21

Doney K, Storb R, Appelbaum FR, Buckner CD, Sanders J, Singer J, et al. Recombinant granulocyte-macrophage colony stimulating factor followed by immunosuppressive therapy for aplastic anaemia. Br J Haematol 1993;85(1):182-4.

Doney K, Storb, R,Lilleby, K,AppelbaumFR. Recombinant interleukin-1 followed by immunosuppressive therapy for aplastic anemia. Am J Hematol 1996;52(1):61-2.

Doney K, Leisenring W, Storb R, Appelbaum FR. Primary treatment of acquired aplastic anemia: outcomes with bone marrow transplantation and immunosuppressive therapy. Seattle Bone Marrow Transplant Team. Ann Intern Med 1997;126(2):107-15.

Dufort G, Pisano S, Incoronato A, Castiglioni M, Carracedo M, Pagés C et al. Feasibility and outcome of haploidentical SCT in pediatric high-risk hematologic malignancies and Fanconi anemia in Uruguay. Bone Marrow Transplant. 2012 May;47(5):663-8. doi: 10.1038/bmt.2011.148. Epub 2011 Jul 18.

Dufour C, Veys P, Carraro E, Bhatnagar N, Pillon M, Wynn R, Similar outcome of upfront-unrelated and matched sibling stem cell transplantation in idiopathic paediatric aplastic anaemia. A study on behalf of the UK Paediatric BMT Working Party, Paediatric Diseases Working Party and Severe Aplastic Anaemia Working Party of EBMT. Br J Haematol. 2015 Nov;171(4):585-94

Eapen M, Davies SM, Ramsay NK. Late graft rejection and second infusion of bone marrow in children with aplastic anaemia. Br J Haematol 1999;104(1):186-8.

Eapen M, Ramsay NK, Mertens AC, Robison LL, Defor T, Davies SM. Late outcomes after bone marrow transplant for aplastic anaemia. Br J Haematol 2000;111(3):754-60.

Guidelines for Hematopoietic Stem Cell Transplantation (HSCT) in Childhood MDS and JMML for Patients enrolled in EWOG-MDS Studies.(https://ewog-mds-saa.org/fileadmin/mediapool/10_andere/ewog.mds/pdf/protocoldocs/other/ConsensusGuidelines HSCTinMDSandJMML_v1.34.pdf)

Fan X, Desmond R, Winkler T, Young DJ, Dumitriu B, Townsley DM, Gutierrez-Rodrigues F, Lotter J, Valdez J, Sellers SE, Barranta ME, Shalhoub RN, Wu CO, Albitar M, Calvo KR, Young NS, Dunbar CE. Eltrombopag for patients with moderate aplastic anemia or uni-lineage cytopenias. Blood Adv. 2020 Apr 28;4(8):1700-1710. doi: 10.1182/bloodadvances.2020001657.

Fioredda F, Iacobelli S, Korthof ET, Knol C, van Biezen A, Outcome of haematopoietic stem cell transplantation in dyskeratosis congenita. Br J Haematol. 2018 Oct;183(1):110-118

Formankova R, Kanderova V, Rackova M, Svaton M, Brdicka T, Riha P et al. Novel SAMD9 Mutation in a Patient With Immunodeficiency, Neutropenia, Impaired Anti-CMV Response, and

Severe Gastrointestinal Involvement. Front Immunol. 2019 Sep 18;10:2194. doi: 10.3389/fimmu.2019.02194. eCollection 2019.

Frickhofen N, Schreznmeier H, Bacigalupo A. Result of European trials of immunosuppression for treatment of aplastic anemia. Ontogeny of hematopoiesis aplastic anemia. International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235:343.

Frickhofen N, Rosenfeld SJ. Immunosuppressive treatment of aplastic anemia with antithymocyte globulin and cyclosporine. Semin Hematol 2000;37(1):56-68.

Frickhofen N, Heimpel H, Kaltwasser JP, Schrezenmeier H, German Aplastic Anemia Study Group. Antithymocyte globulin with or without cyclosporin A: 11-year follow-up of a randomized trial comparing treatments of aplastic anemia. Blood 2003;101(4):1236-42.

Fuhrer M, Rampf U, Baumann I, Faldum A, Niemeyer C, Janka-Schaub G, et al. Immunosuppressive therapy for aplastic anemia in children: a more severe disease predicts better survival. Blood 2005;106(6):2102-4.

Furquim CP, Pivovar A, Amenábar JM, Bonfim C, Torres-Pereira CC. Oral cancer in Fanconi anemia: Review of 121 cases. Crit Rev Oncol Hematol. 2018 May;125:35-40. doi: 10.1016/j.critrevonc.2018.02.013. Epub 2018 Mar 6

Geary CG, Harrison CJ, Philpott NJ, Hows JM, Gordon-Smith EC, Marsh JC. Abnormal cytogenetic clones in patients with aplastic anaemia: response to immunosuppressive therapy. Br J Haematol 1999;104(2):271-4.

Giri N, Alter BP, Savage SA, Stratton P. Gynaecological and reproductive health of women with telomere biology disorders. Br J Haematol. 2021 Jun;193(6):1238-1246. doi: 10.1111/bjh.17545. Epub 2021 May 21.

Gluckman E, Socié G, Devergie A, Bourdeau-Esperou H, Traineau R, Cosset JM. Bone marrow transplantation in 107 patients with severe aplastic anemia using cyclophosphamide and thoraco-abdominal irradiation for conditioning:long-term follow-up. Blood 1991;78(9):2451-5.

Gluckman E, Horowitz MM, Champlin RE, Hows JM, Bacigalupo A, Biggs JC, et al. Bone marrow transplantation for severe aplastic anemia: influence of conditioning and graft-versus-host disease prophylaxis regimens on outcome. Blood 1992;79(1):269-75.

Gluckman E, Rokicka-Milewska R, Hann I, Nikiforakis E, Tavakoli F, Cohen-Scali S, et al. Results and follow-up of a phase III randomized study of recombinant human-granulocyte stimulating factor as support for immunosuppressive therapy in patients with severe aplastic anaemia. Br J Haematol 2002;119(4):1075-82.

Gluckman E, Rocha V, Ionescu I, Bierings M, Harris RE, Wagner J et al. Results of unrelated cord blood transplant in fanconi anemia patients: risk factor analysis for engraftment and survival. Biol Blood Marrow Transplant. 2007 Sep;13(9):1073-82. doi: 10.1016/j.bbmt.2007.05.015. Epub 2007 Jul 20.

Gordon-Smith EC, Rutherford TR. Fanconi anaemia--constitutional, familial aplastic anaemia. Baillieres Clin Haematol 1989;2(1):139-52.

Gordon-Smith EC, Rutherford TR. Fanconi anemia: constitutional aplastic anemia. Semin Hematol 1991;28(2):104-12.

Gordon-Smith EC, Marsh JC, Geary CG. Is it time to stop using chloramphenicol on the eye? Prospective study of aplastic anaemia should give definitive answer. BMJ 1995;311(7002):451.

Griscelli-Bennaceur A, Gluckman E, Scrobohaci ML, Jonveaux P, Vu T, Bazarbachi A, et al. Aplastic anemiaand paroxysmal nocturnal hemoglobinuria: search for a pathogenetic link. Blood 1995;85(5):1354-63.

Groarke Emma M , Bhavisha A. Patel, Fernanda Gutierrez-Rodrigues, Olga Rios, Jennifer Lotter, Daniela Baldoni, et al. Eltrombopag added to immunosuppression for children with treatment-naïve severe aplastic anaemia. British Journal of Haematology, 2021,192,605–614doi.org/10.1111/bjh.17232

Guidelines for Hematopoietic Stem Cell Transplantation (HSCT) in Childhood MDS and JMML for Patients enrolled in EWOG-MDS Studies.(https://ewog-mds-saa.org/fileadmin/mediapool/10_andere/ewog.mds/pdf/protocoldocs/other/ConsensusGuidelines HSCTinMDSandJMML_v1.34.pdf)

Guiguet M, Baumelou E, Mary JY. A case-control study of aplasticanaemia: occupational exposures. The French Cooperative Group for Epidemiological Study of Aplastic Anaemia. Int J Epidemiol 1995;24(5):993-9.

Hattori H, Machii T, Ueda E, Shibano M, Kageyama T, Kitani T. Increased frequency of somatic mutations at glycophorin A loci in patients with aplastic anaemia, myelodysplastic syndrome and paroxysmal nocturnal haemoglobinuria. Br J Haematol 1997;98(2):384-91.

Haute Autorité de Santé - Agence française de sécurité sanitaire du médicament et des produits de santé. Transfusion de globules rouges homologues : produits, indications alternatives. Recommandations. Novembre 2014.

Haute Autorité de Santé - Agence française de sécurité sanitaire du médicament et des produits de santé. Transfusion de plaquettes : produits, indications. Recommandations. Octobre 2015.
Hillmen P, Young NS, Schubert J, Brodsky RA, Socié G, The complement inhibitor eculizumab in paroxysmal nocturnal hemoglobinuria. N Engl J Med. 2006 Sep 21;355(12):1233-43.

Hillmen P, Szer J, Weitz I, Röth A, Höchsmann B, Panse J et al. Pegcetacoplan versus Eculizumab in Paroxysmal Nocturnal Hemoglobinuria**.**N Engl J Med. 2021 Mar 18;384(11):1028-1037. doi: 10.1056/NEJMoa2029073. PMID: 33730455

Hillmen P, Muus P, Röth A, Elebute MO, Risitano AM, Long-term safety and efficacy of sustained eculizumab treatment in patients with paroxysmal nocturnal haemoglobinuria.Br J Haematol. 2013 Jul;162(1):62-73.

Hinterberger W, Rowlings PA, Hinterberger-Fischer M, Gibson J, Jacobsen N, Klein JP, et al. Results of transplanting bone marrow from genetically identical twins into patients with aplastic anemia. Ann Intern Med 1997;126(2):116-22.

Hoffman TW, van Moorsel CHM, Borie R, Crestani B. Pulmonary phenotypes associated with genetic variation in telomere-related genes. Curr Opin Pulm Med. 2018 May;24(3):269-280.

Horowitz MM, Passweg JR, Sobocinski KA, Nugent M, Klein JP. Bone marrow transplantation for severe aplastic anemia. A report from the International Bone Marrow Transplant Registry. Ontogeny of hematopoiesis aplastic anemia. International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235:317.

Horowitz MM. Currentstatus of allogeneic bone marrow transplantation in acquired aplastic anemia. Semin Hematol 2000;37(1):30-42.Hugel B, Socié G, Vu T, Toti F, Gluckman E, Freyssinet JM, et al. Elevated levels of circulating procoagulant microparticles in patients with paroxysmal nocturnal hemoglobinuria and aplastic anemia. Blood 1999;93(10):3451-6.

Howell JC, Joshi SA, Hornung L, Khoury J, Harris RE, Rose SR.. Growth hormone improves short stature in children with Diamond-Blackfan anemia. Pediatr Blood Cancer. 2015 Mar;62(3):402-8. doi: 10.1002/pbc.25341. Epub 2014 Dec 9.

Hughes WT, Armstrong D, Bodey GP, Bow EJ, Brown AE, Calandra T, et al. 2002 guidelines for the use of antimicrobial agents in neutropenic patients with cancer. Clin Infect Dis 2002;34:730-51.

Issaragrisil S, Sriratanasatavorn C, Piankijagum A, Vannasaeng S, Porapakkham Y, Leaverton PE, et al. Incidence of aplastic anemia in Bangkok. The Aplastic Anemia Study Group. Blood 1991;77(10):2166-8.

Issaragrisil S, Kaufman DW, Anderson TE, Chansung K, Thamprasit T, Sirijirachai J, et al. An association of aplastic anaemia in Thailand with low socioeconomic status. Aplastic Anemia Study Group. Br J Haematol 1995;91(1):80-4.

Issaragrisil S, Kaufman DW, Anderson T. Incidence and non-drug aetiologies of aplastic anaemia in Thailand. The Thai Aplastic Anaemia Study Group. Eur J Haematol Suppl 1996;60:31-4.

Issaragrisil S, Kaufman DW, Anderson T, Chansung K, Thamprasit T, Sirijirachai J, et al. Low drug attributability of aplastic anemia in Thailand. The Aplastic Anemia Study Group. Blood 1997;89(11):4034-9.

Issaragrisil S, Chansung K, Kaufman DW, Sirijirachai J, Thamprasit T, Young NS. Aplastic anemia in rural Thailand: its association with grain farming and agricultural pesticide exposure. Aplastic Anemia Study Group. Am J Public Health 1997;87(9):1551-4.

Issaragrisil S, Leaverton PE, Chansung K, Thamprasit T, Porapakham Y, Vannasaeng S, et al. Regional patterns in the incidence of aplastic anemia in Thailand. The Aplastic Anemia Study Group. Am J Hematol 1999;61(3):164-8.

Jin JY, Tooze JA, Marsh JC, Matthey F, Gordon-Smith EC. Myelodysplasia following aplastic anaemia paroxysmal nocturnal haemoglobinuria syndrome after treatment with immunosuppression and G-CSF: evidence for the emergence of a separate clone. Br J Haematol 1996;94(3):510-2.

Jin JY, Tooze JA, Marsh JC, Gordon-Smith EC. Glycosylphosphatidyl-inositol (GPI)-linked protein deficiency on the platelets of patients with aplastic anaemia and paroxysmal nocturnal haemoglobinuria: two distinct patterns correlating with expression on neutrophils. Br J Haematol 1997;96(3):493-6.

Josten KM, Tooze JA, Borthwick-Clarke C, Gordon-Smith EC, Rutherford TR. Acquired aplastic anemia and paroxysmal nocturnal hemoglobinuria: studies on clonality. Blood 1991;78(12):3162-7.

Kaito K, Kobayashi M, Katayama T, Masuoka H, Shimada T, Nishiwaki K, et al. Long-term administration of G-CSF for aplastic anaemia is closely related to the early evolution of monosomy 7 MDS in adults. Br J Haematol 1998;103(2):297-303.

Kami M, Machida U, Hirai H. Late clonal complications in older patients receiving immunosuppressive therapy for aplastic anemia. Ann Intern Med 1999;131(8):633-4.

Karakantza M, Cavenagh JD, Gordon-Smith EC, Gibson FM. Adhesion molecule expression on CD34+ progenitor cells from normal and aplastic anaemia bone marrow. Br J Haematol 1995;91(4):800-3.

Kaufman DW, Kelly JP, Jurgelon JM, Anderson T, Issaragrisil S, Wiholm BE, et al. Drugs in the aetiology of agranulocytosis and aplastic anaemia. Eur J Haematol Suppl 1996;60:23-30.

Kaufman DW, Issaragrisil S, Anderson T, Chansung K, Thamprasit T, Sirijirachai J, et al. Use of household pesticides and the risk of aplastic anaemia in Thailand. The Aplastic Anemia Study Group. Int J Epidemiol 1997;26(3):643-50.

Kawaguchi K, Wada H, Mori A, Takemoto Y, Kakishita E, Kanamaru. Detection of GPI-anchored protein-deficient cells in patients with aplastic anaemia and evidence for clonal expansion during the clinical course. Br J Haematol 1999;105(1):80-4.

Kelly RJ1, Hill A, Arnold LM, Brooksbank GL, Richards SJ, Cullen M, Mitchell LD, Cohen DR, Gregory WM, Hillmen P. Long-term treatment with eculizumab in paroxysmal nocturnal hemoglobinuria: sustained efficacy and improved survival. Blood 2011 Jun 23;117(25):6786-92. doi: 10.1182/blood-2011-02-333997. Epub 2011 Apr 1

Kelly JP, Jurgelon JM, Issaragrisil S, Keisu M, Kaufman DW. An epidemiological study of aplastic anaemia: relationship of drug exposures to clinical features and outcome. Eur J Haematol Suppl 1996;60:47-52.

Kelly RJ, Höchsmann B, Szer J, Kulasekararaj A, de Guibert S. Eculizumab in Pregnant Patients with Paroxysmal Nocturnal Hemoglobinuria. N Engl J Med. 2015 Sep 10;373(11):1032-9.

Killick SB, Marsh JC. Aplastic anaemia: management. Blood Rev 2000;14(3):157-71.

Khincha PP, Bertuch AA, Gadalla SM, Giri N, Alter BP, Savage SA. Similar telomere attrition rates in androgen-treated and untreated patients with dyskeratosis congenita. Blood Adv. 2018 Jun 12;2(11):1243-1249.

Khincha PP, Wentzensen IM, Giri N, Alter BP, Savage SA. Response to androgen therapy in patients with dyskeratosis congenita. Br J Haematol. 2014 May;165(3):349-57.

Knight SW, Heiss NS, Vulliamy TJ, Aalfs CM, McMahon C, Richmond P, et al. Unexplained aplastic anaemia, immunodeficiency, and cerebellar hypoplasia (Hoyeraal-Hreidarsson syndrome) due to mutations in the dyskeratosis congenita gene, DKC1. Br J Haematol 1999;107(2):335-9.

Kodera Y, Morishima Y, Kato S, Akiyama Y, Sao H, Matsuyama T, et al. Analysis of 500 bone marrow transplants from unrelated donors (UR-BMT) facilitated by the Japan Marrow Donor Program: confirmation of UR-BMT as a standard therapy for patients with leukemia and aplastic anemia. Bone Marrow Transplant 1999;24(9):995-1003.

Kojima S, Inaba J, Kondo M, Kato K, Matsuyama T, Horikoshi Y, et al. Unrelated donor marrow transplantation for severe acquired aplastic anemia using cyclophosphamide, antithymocyte globulin, and total body irradiation. Blood 1995;85(1):291-2.

Kojima S. Use of hematopoietic growth factors for treatment of aplastic anemia. Bone Marrow Transplant 1996;18 Suppl 3:S36-S38.

Kojima S, Horibe K, Inaba J, Yoshimi A, Takahashi Y, Kudo K, et al. Long-term outcome of acquired aplastic anaemia in children: comparison between immunosuppressive therapy and bone marrow transplantation. Br J Haematol 2000;111(1):321-8.

Kojima S, Hibi S, Kosaka Y, Yamamoto M, Tsuchida M,Mugishima H, et al. Immunosuppressive therapy using antithymocyte globulin, cyclosporine, and danazol with or without human granulocyte colony-stimulating factor in children with acquired aplastic anemia. Blood 2000;96(6):2049-54.

Kojima S. Evidence-based medicine for treatment of aplastic anemia in children. Rinsho Ketsueki 2005;46(6):394-8.

Kojima S, Frickhofen N, Deeg HJ, Okamoto S, Marsh J, Teramura M, et al. Aplastic anemia. Int J Hematol 2005;82(5):408-11.

Konopacki J, Porcher R, Robin M, Bieri S, Cayuela JM, Larghero et al. Long-term follow up after allogeneic stem cell transplantation in patients with severe aplastic anemia after cyclophosphamide plus antithymocyte globulin conditioning. Haematologica. 2012. May;97(5):710-6.doi: 10.3324/haematol.2011.050096. Epub 2011 Dec 16.

Kosaka Y, Yagasaki H, Sano K, Kobayashi R, Ayukawa H, Kaneko T, et al. Prospective multicenter trial comparing repeated immunosuppressive therapy with stem-cell transplantation from an alternative donor as second-line treatment for children with severe and very severe aplastic anemia. Blood 2008;111(3):1054-9.

Kruse EB, Naumann F, Schwarzer G, Borchmann P, Peinemann F, Bohlius J, et al. First-line stem cell transplantation from related donors compared to immunosuppressive treatment for acquired severe aplastic anaemia (Protocol). The Cochrane Database of Systematic Reviews 2007;Issue 1.

Kulasekararaj AG, Hill A, Rottinghaus ST, Langemeijer S, Wells R, et al. Ravulizumab (ALXN1210) vs eculizumab in C5-inhibitor-experienced adult patients with PNH: the 302 study. Blood. 2019 Feb 7;133(6):540-549. doi: 10.1182/blood-2018-09-876805. Epub 2018 Dec 3

Lanneaux J, Poidvin A, Soole F, Leclerc G, Grimaud M, Dalle JH.Fanconi anemia in 2012: diagnosis, pediatric follow-up and treatment]. Arch Pediatr. 2012 Oct;19(10):1100-9. doi: 10.1016/j.arcped.2012.07.023. Epub 2012 Sep 7.

Langston AA, Sanders JE, Deeg HJ, Crawford SW, Anasetti C, Sullivan KM, et al. Allogeneic marrow transplantation for aplastic anaemia associated with dyskeratosis congenita. Br J Haematol 1996;92(3):758-65.

Lecornec N, Castex MP, Réguerre Y, Moreau P, Marie I, Garçon L, Da Costa L, Leblanc T. Agranulocytosis in patients with Diamond-Blackfan anaemia (DBA) treated with deferiprone for post-transfusion iron overload: A retrospective study of the French DBA cohort. Br J Haematol. 2022 Oct;199(2):285-288.

Léger-Silvestre I, Caffrey JM, Dawaliby R, Alvarez-Arias DA, Gas N, Bertolone SJ et al. Specific Role for Yeast Homologs of the Diamond Blackfan Anemia-associated Rps19 Protein in Ribosome Synthesis. J Biol Chem. 2005 Nov 18;280(46):38177-85.
Faivre L, Meerpohl J, Da Costa L, Marie I, Nouvel C, Gnekow A, Bender-Götze C, Bauters F, Coiffier B, Peaud PY, Rispal P, Berrebi A, Berger C, Flesch M, Sagot P, Varet B, Niemeyer C, Tchernia G, Leblanc T. High-risk pregnancies in Diamond-Blackfan anemia: a survey of 64 pregnancies from the French and German registries. Haematologica. 2006 Apr;91(4):530-3. Epub 2006 Mar 15.

Lee JW, Sicre de Fontbrune F, Wong Lee Lee L, Pessoa V, Gualandro S, et al. Ravulizumab (ALXN1210) vs eculizumab in adult patients with PNH naive to complement inhibitors: the 301 study. Blood. 2019 Feb 7;133(6):530-539. doi: 10.1182/blood-2018-09-876136. Epub 2018 Dec 3.

Lengline E, Drenou B, Peterlin P, Tournilhac O, Abraham J, Berceanu A, Dupriez B, Guillerm G, Raffoux E, de Fontbrune FS, Ades L, Balsat M, Chaoui D, Coppo P, Corm S, Leblanc T, Maillard N, Terriou L, Socié G, de Latour RP. Nationwide survey on the use of eltrombopag in patients with severe aplastic anemia: a report on behalf of the French Reference Center for Aplastic Anemia. Haematologica. 2018 Feb;103(2):212-220. doi: 10.3324/haematol.2017.176339. Epub 2017 Nov 23.

Lipton JM, Molmenti CLS, Desai P, Lipton A, Ellis SR, Vlachos A. Early Onset Colorectal Cancer: An Emerging Cancer Risk in Patients with Diamond Blackfan Anemia. Genes (Basel). 2021 Dec 26;13(1):56. doi: 10.3390/genes13010056.

Lipton JM, Molmenti CLS, Hussain M, Desai P, Florento M, Atsidaftos E, Vlachos A. Colorectal cancer screening and surveillance strategy for patients with Diamond Blackfan anemia: Preliminary recommendations from the Diamond Blackfan Anemia Registry. Pediatr Blood Cancer. 2021 Aug;68(8):e28984. doi: 10.1002/pbc.28984. Epub 2021 Jun 5.

Locasciulli A, van't VL, Bacigalupo A, Hows J, Van Lint MT, Gluckman E, et al. Treatment with marrow transplantation or immunosuppression of childhood acquired severe aplastic anemia: a report from the EBMT SAA Working Party. Bone Marrow Transplant 1990;6(3):211-7.

Locasciulli A, Arcese W, Locatelli F, Di BE, Bacigalupo A, Italian Aplastic Anaemia Study Group. Treatment of aplastic anaemia with granulocyte-colony stimulating factor and risk of malignancy. Italian Aplastic Anaemia, Study Group. Lancet 2001;357(9249):43-4.

Locasciulli A, Bruno B, Rambaldi A, Saracco P, Dufour C, Finelli C, et al. Treatment of severe aplastic anemia with antilymphocyte globulin, cyclosporine and two different granulocyte colony-stimulating factor regimens: a GITMO prospective randomized study. Haematologica 2004;89(9):1054-61.

Locatelli F, Porta F, Zecca M, Pedrazzoli P, Maccario R, Giani S, et al. Successful bone marrow transplantation in children with severe aplastic anemia using HLA-partially matched family donors. Am J Hematol 1993;42(3):328-33.

Locatelli F, Bruno B, Zecca M, Van-Lint MT, McCann S, Arcese W, et al. Cyclosporin A and short-term methotrexate versus cyclosporin A as graft versus host disease prophylaxis in patients with severe aplastic anemia given allogeneic bone marrow transplantation from an HLA-identical sibling: results of a GITMO/EBMT randomized trial. Blood 2000; 96(5):1690-7.

Lorand-Metze I, Meira DG, Lima CS, Vassallo J, Metze K. The differential diagnosis between aplastic anemia and hypocellular myelodysplasia in patients with pancytopenia. Haematologica 1999;84(6):564-5.

Loschi M, Porcher R, Barraco F, Terriou L, Mohty M, Impact of eculizumab treatment on paroxysmal nocturnal hemoglobinuria: a treatment versus no-treatment study. Am J Hematol. 2016 Jun;91(4):366-70.

Maciejewski JP, Sato T, Selleri C, Anderson S, Young NS. Long term culture-initiating cell number in aplastic anemia. Blood 1995;86(suppl 1):477a.

Maciejewski JP, Selleri C, Sato T, Anderson S, Young NS. Increased expression of Fas antigen on bone marrow CD34+ cells of patients with aplastic anaemia. Br J Haematol 1995;91(1):245-52.

Maciejewski JP, Selleri C, Sato T, Anderson S, Young NS. A severe and consistent deficit in marrow and circulating primitive hematopoietic cells (long-term culture-initiating cells) in acquired aplastic anemia. Blood 1996;88(6):1983-91.

Maciejewski JP, Sloand EM, Sato T, Anderson S, Young NS. Impaired hematopoiesis in paroxysmal nocturnal hemoglobinuria/aplastic anemia is not associated with a selective proliferative defect in the glycosylphosphatidylinositol-anchored protein-deficient clone. Blood 1997;89(4):1173-81.

Maciejewski JP, Kim S, Sloand E, Selleri C, Young NS. Sustained long-term hematologic recovery despite a marked quantitative defect in the stem cell compartment of patients with aplastic anemia after immunosuppressive therapy. Am J Hematol 2000;65(2):123-31.

Manz CY, Nissen C, Wodnar-Filipowicz A. Deficiency of CD34+ c-kit+ and CD34+38- hematopoietic precursors in aplastic anemia after immunosuppressive treatment. Am J Hematol 1996;52(4):264-74.

Manz CY, Dietrich PY, Schnuriger V, Nissen C, Wodnar-Filipowicz A. T-cell receptor beta chain variability in bone marrow and peripheral blood in severe acquired aplastic anemia. Blood Cells Mol Dis 1997;23(1):110-22.

Margolis D, Camitta B, Pietryga D, Keever-Taylor C, Baxter-Lowe LA, Pierce K, et al. Unrelated donor bone marrow transplantation to treat severe aplastic anaemia in children and young adults. Br J Haematol 1996;94(1):65-72.

Margolis DA, Casper JT. Alternative-donor hematopoietic stem-cell transplantation for severe aplastic anemia. Semin Hematol 2000;37(1):43-55.Marmont AM. Who really discovered aplastic anemia? Haematologica 1995;80(3):294.

Marsh J, Schrezenmeier H, Marin P, Ilhan O, Ljungman P, McCann S, et al. Prospective randomized multicenter study comparing cyclosporin alone versus the combination of antithymocyte globulin and cyclosporin for treatment of patients with non severe aplastic anemia: a report from the European Blood and Marrow Transplant (EBMT) Severe Aplastic Anaemia Working Party. Blood 1999;93(7):2191-5.

Marsh JC, Chang J, Testa NG, Hows JM, Dexter TM. The hematopoietic defect in aplastic anemia assessed by long-term marrow culture. Blood 1990;76(9):1748-57.

Marsh JC, Geary CG. Is aplastic anaemia a pre-leukaemic disorder? Br J Haematol 1991;77(4):447-52.

Marsh JC, Chang J, Testa NG, Hows JM, Dexter TM. In vitro assessment of marrow 'stem cell' and stromal cell function in aplastic anaemia. Br J Haematol 1991;78(2):258-67.

Marsh JC, Socié G, Schrezenmeier H, Tichelli A, Gluckman E, Ljungman P, et al. Haemopoietic growth factors in aplastic anaemia: a cautionary note. European Bone Marrow Transplant Working Party for Severe Aplastic Anaemia. Lancet 1994;344(8916):172-3.

Marsh JC. Long-term bone marrow cultures in aplastic anaemia. Eur J Haematol Suppl 1996;60:75-9.

Marsh JC, Gordon-Smith EC. Treatment options in severe aplastic anaemia. Lancet 1998;351(9119):1830-1.

Marsh JC. Treatment of aplastic anaemia: first do no harm. Lancet 2000;356(9241):1536-7.

Marsh JC. Hematopoietic growth factors in the pathogenesis and for the treatment of aplastic anemia. Semin Hematol 2000;37(1):81-90.

Marsh JC. Results of immunosuppression in aplastic anaemia. Acta Haematol. 2000;103(1):26-32.

Marsh JC. Treatment of acquired aplastic anemia. Haematologica 2007;92(1):2-5.

Marsh JC, Ball SE, Cavenagh J, Darbyshire P, Dokal I, Gordon-Smith EC, Keidan J, Laurie A, Martin A, Mercieca J, Killick SB, Stewart R, Yin JA; British Committee for Standards in Haematology. Br J Haematol. 2009 Oct;147(1):43-70. doi: 10.1111/j.1365-2141.2009.07842.x. Epub 2009 Aug 10.

Mary JY, Baumelou E, Guiguet M. Epidemiology of aplastic anemia in France: a prospective multicentric study. The French Cooperative Group for Epidemiological Study of Aplastic Anemia. Blood 1990;75(8):1646-53.

Mary JY, Guiguet M, Baumelou E. Epidemiology of aplastic anemia: The French experience. Ontogeny of hematopoiesis aplastic anemia. International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235:303.

Mary JY, Guiguet M, Baumelou E. Drug use and aplastic anaemia: the French experience.French Cooperative Group for the Epidemiological Study of Aplastic Anaemia. Eur J Haematol Suppl 1996;60:35-41.

Maury S, Balere-Appert ML, Chir Z, Boiron JM, Galambrun C, Yakouben K, et al. Unrelated stem cell transplantation for severe acquired aplastic anemia: improved outcome in the era of high-resolution HLA matching between donor and recipient. Haematologica 2007;92(5):589-96.

Mayo Foundation for medical Education and Research. Aplastic anemia <http://www.mayoclinic.com> [consulté le 21-11-2007].

Merk B, Hildebrand A, Rojewski M, Raghavachar A, Schrezenmeier H. Heterogeneous PIG-A mutations in aplastic anemia, paroxysmal nocturnal hemoglobinuria and non Hodkin´s lymphoma. Blood 1997;90:434a-5a.

Mehta PA, Davies SM, Leemhuis T, Myers K, Kernan NA, Prockop SE et al. Radiation-free, alternative-donor HCT for Fanconi anemia patients: results from a prospective multi-institutional study. Blood. 2017 Apr 20;129(16):2308-2315. doi: 10.1182/blood-2016-09-743112. Epub 2017 Feb 8.

Mortazavi Y, Tooze JA, Gordon-Smith EC, Rutherford TR. N-RAS gene mutation in patients with aplastic anemia and aplastic anemia/ paroxysmal nocturnal hemoglobinuria during evolution to clonal disease. Blood 2000;95(2):646-50.

Mortazavi Y, Chopra R, Gordon-Smith EC, Rutherford TR. Clonal patterns of X-chromosome inactivation in female patients with aplastic anaemia studies using a novel reverse transcription polymerase chain reaction method. Eur J Haematol 2000;64(6):385-95.

Moustacchi E. Fanconi's anemia.Paris: Encyclopédie Orphanet; 2003.

Mugishima H, Ohga S, Ohara A, Kojima S, Fujisawa K, Tsukimoto I; Aplastic Anemia Committee of the Japanese Society of Pediatric Hematology. Hematopoietic stem cell transplantation for Diamond-Blackfan anemia: a report from the Aplastic Anemia Committee of the Japanese Society of Pediatric Hematology. Pediatr Transplant. 2007 Sep;11(6):601-7. doi: 10.1111/j.1399-3046.2006.00616.x.

Najean Y, Haguenauer O. Long-term (5 to 20 years) Evolution of nongrafted aplastic anemias. The Cooperative Group for the Study of Aplastic and Refractory Anemias. Blood 1990;76(11):2222-8.

Nakao S, Yamaguchi M, Takamatsu H, Shiobara S, Matusda T. Expansion of a paroxysmal nocturnal hemoglobinuria (PNH) clone after cyclosporine therapy for aplastic anemia/PNH syndrome. Blood 1992;80(11):2943-4.

Nakao S, Takamatsu H, Chuhjo T, Ueda M, Shiobara S, Matsuda T, et al. Identification of a specific HLA class II haplotype strongly associated with susceptibility to cyclosporine-dependent aplastic anemia. Blood 1994;84(12):4257-61.

Nakao S, Takamatsu H, Yachie A, Itoh T, Yamaguchi M, Ueda M, et al. Establishment of a CD4+ T cell clone recognizing autologous hematopoietic progenitor cells from a patient with immune-mediated aplastic anemia. Exp Hematol 1995;23(5):433-8.

Nakao S, Yamaguchi M, Takamatsu H, Takami A, Chuhjo T, Ueda M, et al. Relative erythroid hyperplasia in the bone marrow at diagnosis of aplastic anaemia: a predictive marker for a favourable response to cyclosporine therapy. Br J Haematol 1996;92(2):318-23.

Nakao S, Takami A, Sugimori N, Ueda M, Shiobara S, Matsuda T, et al. Response to immunosuppressive therapy and an HLA-DRB1 allele in patients with aplastic anaemia: HLA-DRB1*1501 does not predict response to antithymocyte globulin. Br J Haematol 1996;92(1):155-8.

Nakao S, Takami A, Takamatsu H, Zeng W, Sugimori N, Yamazaki H, et al. Isolation of a T-cell clone showing HLA-DRB1*0405-restricted cytotoxicity for hematopoietic cells in a patient with aplastic anemia. Blood 1997;89(10):3691-9.

Nalepa G, Clapp DW. Fanconi anaemia and cancer: an intricate relationship. Nat Rev Cancer. 2018 Mar;18(3):168-185. doi: 10.1038/nrc.2017.116. Epub 2018 Jan 29. Review.

Narayanan MN, Geary CG, Freemont AJ, Kendra JR. Long-term follow-up of aplastic anaemia. Br J Haematol 1994;86(4):837-43.

Narumi S, Amano N, Ishii T, Katsumata N, Muroya K, Adachi M, Toyoshima K et al. SAMD9 mutations cause a novel multisystem disorder, MIRAGE syndrome, and are associated with loss of chromosome 7. Nat Genet. 2016 Jul;48(7):792-7. doi: 10.1038/ng.3569. Epub 2016 May 16.

Nissen C. The pathophysiology of aplastic anemia. Semin Hematol 1991;28(4):313-8.

Nistico A, Young NS. gamma-Interferon gene expression in the bone marrow of patients with aplastic anemia. Ann Intern Med 1994;120(6):463-9.

Marie-Françoise O'Donohue, Lydie Da Costa, Marco Lezzerini, Sule Unal, Clément Joret et al. HEATR3 variants impair nuclear import of uL18 (RPL5) and drive Diamond-Blackfan anemia. Blood (2022) 139 (21): 3111–3126.

Olnes MJ, Scheinberg P, Calvo KR, Desmond R, Tang Y, Dumitriu B, Parikh AR, Soto S, Biancotto A, Feng X, Lozier J, Wu CO, Young NS, Dunbar CE. Eltrombopag and improved hematopoiesis in refractory aplastic anemia. N Engl J Med. 2012 Jul 5;367(1):11-9. doi: 10.1056/NEJMoa1200931. Erratum in: N Engl J Med. 2012 Jul 19;367(3):284.

Oostra AB, Nieuwint AW, Joenje H, de Winter JP.Diagnosis of fanconi anemia: chromosomal breakage analysis. Anemia. 2012;2012:238731. doi: 10.1155/2012/238731. Epub 2012 May 24.

Paquette RL, Tebyani N, Frane M, Ireland P, Ho WG, Champlin RE, et al. Long-term outcome of aplastic anemia in adults treated with antithymocyte globulin: comparison with bone marrow transplantation. Blood 1995;85(1):283-90.

Passweg JR, Socié G, Hinterberger W, Bacigalupo A, Biggs JC, Camitta BM, et al. Bone marrow transplantation for severe aplastic anemia: has outcome improved? Blood 1997;90(2):858-64.

Peffault de Latour R, Kulasekararaj A, Iacobelli S, Terwel SR, Cook R, Griffin Met al. Eltrombopag Added to Immunosuppression in Severe Aplastic Anemia. N Engl J Med . 2022 Jan 6;386(1):11-23. doi: 10.1056/NEJMoa2109965.

Peffault de Latour R, Tabrizi R, Marcais A, Leblanc T, Lamy T, Mohty M, Tavitian S, Jubert C, Pasquet M, Galambrun C, Nguyen S, Cahn JY, Braun T, Deconinck E, Bay JO, Sicre de Fontbrune F, Barraco F, Socié G. Nationwide survey on the use of horse antithymocyte globulins (ATGAM) in patients with acquired aplastic anemia: A report on behalf of the French Reference Center for Aplastic Anemia. Am J Hematol. 2018 May;93(5):635-642. doi: 10.1002/ajh.25050. Epub 2018 Feb 14.

Peffault de Latour R, Chevret S, Jubert C, Sirvent A, Galambrun C, Ruggeri A, Gandemer V, Cornillon J, Rialland F, Dalle JH, Forcade E, Bruno B, Paillard C, Rorlich PS, Salmon A, Fürst S, Sicre de Fontbrune F, Rubio MT, Bay JO, Mohty M, Larghero J, Gluckman E, Socié G; Francophone Society of Bone Marrow Transplantation and Cellular Therapy.Unrelated cord blood transplantation in patients with idiopathic refractory severe aplastic anemia: a nationwide phase 2 study. Blood. 2018 Aug 16;132(7):750-754. doi: 10.1182/blood-2018-01-829630. Epub 2018 May 14.

Peffault de Latour R. Transplantation for bone marrow failure: current issues. Hematology Am Soc Hematol Educ Program. 2016 Dec 2;2016(1):90-98.

Peffault de Latour R, Schrezenmeier H, Bacigalupo A, Blaise D, de Souza CA. Allogeneic stem cell transplantation in paroxysmal nocturnal hemoglobinuria. Haematologica. 2012 Nov;97(11):1666-73.

Peffault de Latour R, Soulier J. How I treat MDS and AML in Fanconi anemia. Blood. 2016 Jun 16;127(24):2971-9. doi: 10.1182/blood-2016-01-583625. Epub 2016 Mar 28.

Peffault de Latour R, Porcher R, Dalle JH, Aljurf M, Korthof ET, Svahn J, Willemze R, Barrenetxea C, Mialou V, Soulier J, Ayas M, Oneto R, Bacigalupo A, Marsh JC, Peters C, Socie G, Dufour C. Allogeneic hematopoietic stem cell transplantation in Fanconi anemia: the European Group for Blood and Marrow Transplantation experience. Blood.2013 Dec 19;122(26):4279-86.

Pinto FO, Leblanc T, Chamousset D, Le Roux G, Brethon B, Cassinat B,Diagnosis of Fanconi anemia in patients with bone marrow failure. Haematologica . 2009 Apr;94(4):487-95. doi: 10.3324/haematol.13592. Epub 2009 Mar 10.

Poirot C, Yakouben K, Martelli H, Sitbon L, Prades M, Fertility preservation for children and adolescents before gonadotoxic treatment. Rev Prat ; 2014, 64 :169-73.

Quentin S, Cuccuini W, Ceccaldi R, Nibourel O, Pondarre C, Pagès MP. Myelodysplasia and leukemia of Fanconi anemia are associated with a specific pattern of genomic abnormalities that includes cryptic RUNX1/AML1 lesions. Blood . 2011 Apr 14;117(15):e161-70.
doi: 10.1182/blood-2010-09-308726. Epub 2011 Feb 16.

Radulovic I, Schündeln MM, Müller L, Ptok J, Honisch E, Niederacher D, Wiek C, Scheckenbach K, Leblanc T, Larcher L, Soulier J et al. A novel cancer risk prediction score for the natural course of FA patients with biallelic BRCA2/FANCD1 mutations. Hum Mol Genet . 2023 Jan 31;ddad017.
doi: 10.1093/hmg/ddad017. Online ahead of print

Raghavachar A, Vogt HG, Herrmann F, et al. Treatment of aplastic anemia with antilymphocyte globulin and methylprednisolone with or without cyclosporine. The German Aplastic Anemia Study Group. N Engl J Med 1991;324(19):1297-304.

Reddy V, Khan S, Wingard JR, Mehta P. Treatment results in aplastic anemia trials need to be analyzed separately for pediatric and adult populations. Blood 1999;94(5):1833-4.

Revy P, Kannengiesser C, Bertuch AA. Genetics of human telomere biology disorders. Nat Rev Genet. 2022 Sep 23. doi: 10.1038/s41576-022-00527-z. Epub ahead of print.
Río P, Navarro S, Wang W, Sánchez-Domínguez R, Pujol RM, Segovia JC et al. Successful engraftment of gene-corrected hematopoietic stem cells in non-conditioned patients with Fanconi anemia. Nat Med. 2019 Sep;25(9):1396-1401.doi: 10.1038/s41591-019-0550-z. Epub 2019 Sep 9.

Rosenberg PS, Alter BP, Bolyard AA, Bonilla MA, Boxer LA, Cham B, et al. The incidence of leukemia and mortality from sepsis in patients with severe congenital neutropenia receiving long-term G-CSF therapy. Blood 2006;107(12):4628-35.

Rosenberg PS, Socié G, Alter BP, Gluckman E. Risk of head and neck squamous cell cancer and death in patients with Fanconi anemia who did and did not receive transplants. Blood. 2005 Jan 1;105(1):67-73. Epub 2004 Aug 26.

Rosenfeld S, Follmann D, Nunez O, Young NS. Antithymocyte globulin and cyclosporine for severe aplastic anemia: association between hematologic response and long-term outcome. JAMA 2003;289(9):1130-5.

Rosenfeld SJ, Kimball J, Vining D, Young NS. Intensive immunosuppression with antithymocyte globulin and cyclosporine as treatment for severe acquired aplastic anemia. Blood 1995;85(11):3058-65.

Rovo A, Tichelli A, Dufour C. Diagnosis of acquired aplastic anemia. BMT 2013 48,162–167.

Sahoo S.S. et al. Clinical evolution, genetic landscape and trajectories of clonal hematopoiesis in SAMD9/SAMD9L syndromes. Nature Medicine (2021)Savage S.A. Beginning at the ends: telomeres and human disease. Version 1. F1000Res. 2018; 7: F1000 Faculty Rev-524.

Schratz KE, Haley L, Danoff SK, Blackford AL, DeZern AE, Gocke CD et al. Cancer spectrum and outcomes in the Mendelian short telomere syndromes. Blood. 2020 May 28;135(22):1946-1956. doi: 10.1182/blood.2019003264. PMID: 32076714

Scheinberg P, Nunez O, Weinstein B, Scheinberg P, Biancotto A, Wu CO, Young NS. Horse versus rabbit antithymocyte globulin in acquired aplastic anemia. N Engl J Med. 2011 Aug 4;365(5):430-8. doi: 10.1056/NEJMoa1103975

Schrezenmeier H, Hertenstein B, Wagner B, Raghavachar A, Heimpel H. A pathogenetic link between aplastic anemia and paroxysmal nocturnal hemoglobinuria is suggested by a high frequency of aplastic anemia patients with a deficiency of phosphatidylinositol glycan anchored proteins. Exp Hematol 1995; 23(1):81-7.

Schrezenmeier H, Jenal M, Herrmann F, Heimpel H, Raghavachar A. Quantitative analysis of cobblestone area-forming cells in bone marrow of patients with aplastic anemia by limiting dilution assay. Blood 1996;88(12):4474-80.

Schrezenmeier H, Passweg JR, Marsh JC, Bacigalupo A, Bredeson CN, Bullorsky E, et al. Worse outcome and more chronic GVHD with peripheral blood progenitor cells than bone marrow in HLA-matched sibling donor transplants for young patients with severe acquired aplastic anemia. Blood 2007;110(4):1397-400.

Sebert M, Gachet S, Leblanc T, Rousseau A, Bluteau O, Kim R, Ben Abdelali R, Sicre de Fontbrune F, Clonal hematopoiesis driven by chromosome 1q/MDM4 trisomy defines a canonical route toward

leukemia in Fanconi anemia. Cell Stem Cell. 2023 Feb 2;30(2):153-170.e9. doi:10.1016/j.stem.2023.01.006.

Shimamura A, Alter BP. Pathophysiology and management of inherited bone marrow failure syndromes. Blood Rev. 2010 May;24(3):101-22. doi: 10.1016/j.blre.2010.03.002. Epub 2010 Apr 24.

Smith TJ, Khatcheressian J, Lyman GH, Ozer H, Armitage JO, Balducci L,et al. 2006 update of recommendations for the use of white blood cell growth factors. An evidence-based clinical practice guideline. J Clin Oncol 2006;24(19):3187-205.

Socié G, Henry-Amar M, Cosset JM, Devergie A, Girinsky T, Gluckman E. Increased incidence of solid malignant tumors after bone marrow transplantation for severe aplastic anemia. Blood 1991;78(2):277-9.

Socié G, Henry-Amar M, Bacigalupo A, Hows J, Tichelli A, Ljungman P, et al. Malignant tumors occurring after treatment of aplastic anemia. European Bone Marrow Transplantation-Severe Aplastic Anaemia Working Party. N Engl J Med 1993;329(16):1152-7.

Socié G. Is aplastic anemia a preleukemic disorder ? Facts and hypotheses. Ontogeny of hematopoiesis aplastic anemia. International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235:222.

Socié G. Could aplastic anaemia be considered a pre-pre-leukaemic disorder? Eur J Haematol Suppl 1996;60:60-3.

Socié G, Rosenfeld S, Frickhofen N, Gluckman E, Tichelli A. Late clonal diseases of treated aplastic anemia. Semin Hematol 2000;37(1):91-101.

Socié G, Mary JY, Schrezenmeier H, Marsh J, Bacigalupo A, Locasciulli A, et al. Granulocyte-stimulating factor and severe aplastic anemia: a survey by the European Group for Blood and Marrow Transplantation (EBMT). Blood 2007;109(7):2794-6.

Soulier J, Leblanc T, Larghero J, Dastot H, Shimamura A, Guardiola P, Esperou H et al. Detection of somatic mosaicism and classification of Fanconi anemia patients by analysis of the FA/BRCA pathway. Blood . 2005 Feb 1;105(3):1329-36. doi: 10.1182/blood-2004-05-1852. Epub 2004 Sep 21.

Storb R, Doney KC, Thomas ED, Appelbaum F, Buckner CD, Clift RA, et al. Marrow transplantation with or without donor buffy coat cells for 65 transfused aplastic anemia patients. Blood 1982;59(2):236-46.

Storb R, Prentice RL, Banaji M, Witherspoon RP, Sullivan KM, Stewart P, et al. Marrow transplantation from HLA-identical siblings for treatment of aplastic anemia: is exposure to marrow donor blood products 24 hours before high-dose cyclophosphamide needed for successful engraftment? Blood 1983;61(4):672-5.

Storb R, Deeg HJ, Farewell V, Doney K, Appelbaum F, Beatty P, et al. Marrow transplantation for severe aplastic anemia: methotrexate alone compared with a combination of methotrexate and cyclosporine for prevention of acute graft-versus-host disease. Blood 1986;68(1):119-25.

Storb R, Deeg HJ, Pepe M, Doney K, Appelbaum F, Beatty P, et al. Graft-versus-host disease prevention by methotrexate combined with cyclosporin compared to methotrexate alone in patients given marrow grafts for severe aplastic anaemia: long-term follow-up of a controlled trial. Br J Haematol 1989;72(4):567-72.

Storb R, Sanders JE, Pepe M, Anasetti C, Appelbaum FR, Buckner CD, et al. Graft-versus-host disease prophylaxis with methotrexate/cyclosporine in children with severe aplastic anemia treated with cyclophosphamide and HLA-identical marrow grafts. Blood 1991;78(4):1144-5.

Storb R, Etzioni R, Anasetti C, Appelbaum FR, Buckner CD, Bensinger W, et al. Cyclophosphamide combined with antithymocyte globulin in preparation for allogeneic marrow transplants in patients with aplastic anemia. Blood 1994;84(3):941-9.

Storb R, Leisenring W, Deeg HJ, Anasetti C, Appelbaum F, Bensinger W, et al. Long-term follow-up of a randomized trial of graft-versus-host disease prevention by methotrexate/cyclosporine versus methotrexate alone in patients given marrow grafts for severe aplastic anemia. Blood 1994;83(9):2749-50.

Storb R, Leisenring W, Anasetti C, Appelbaum FR, Buckner CD, Bensinger WI, et al. Long-term follow-up of allogeneic marrow transplants in patients with aplastic anemia conditioned by cyclophosphamide combined with antithymocyte globulin. Blood 1997;89(10):3890-1.

Strahm B, Loewecke F, Niemeyer CM, Albert M, Ansari M, Bader P et al. Favorable outcomes of hematopoietic stem cell transplantation in children and adolescents with Diamond-Blackfan anemia. Blood Adv. 2020 Apr 28;4(8):1760-1769. doi: 10.1182/bloodadvances.2019001210.

Strocchio L, Pagliara D, Algeri M, Li Pira G, Rossi F, Bertaina V and al. HLA-haploidentical TCRαβ+/CD19+-depleted stem cell transplantation in children and young adults with Fanconi anemia. Blood Adv. 2021 Mar 9;5(5):1333-1339. doi: 10.1182/bloodadvances.2020003707.

Stucki A, Leisenring W, Sandmaier BM, Sanders J, Anasetti C, Storb R. Decreased rejection and improved survival of first and second marrow transplants for severe aplastic anemia (a 26-year retrospective analysis). Blood 1998;92(8):2742-9.

Tchernia G, Delaunay J. Anémie de Blackfan-Diamond. Paris: Encyclopédie Orphanet; 2003.

Teramura M, Kobayashi S, Iwabe K, Yoshinaga K, Mizoguchi H. Mechanism of action of antithymocyte globulin in the treatment of aplastic anaemia: in vitro evidence for the presence of immunosuppressive mechanism. Br J Haematol 1997;96(1):80-4.

Teramura M, Kimura A, Iwase S, Yonemura Y, Nakao S, Urabe A, et al. Treatment of severe aplastic anemia with antithymocyte globulin and cyclosporin A with or without G-CSF in adults: a multicenter randomized study in Japan. Blood 2007;110(6):1756-61.

Tichelli A, Gratwohl A, Wursch A, Nissen C, Speck B. Late haematological complications in severe aplastic anaemia. Br J Haematol 1988;69(3):413-8.

Tichelli A, Gratwohl A, Nissen C, Signer E, Stebler GC, Speck B. Morphology in patients with severe aplastic anemia treated with antilymphocyte globulin. Blood 1992;80(2):337-45.

Tichelli A, Gratwohl A, Nissen C, Speck B. Late clonal complications in severe aplastic anemia. Leuk Lymphoma 1994;12(3-4):167-75.

Tichelli A, Passweg J, Nissen C, Bargetzi M, Hoffmann T, Wodnar-Filipowicz A, et al. Repeated treatment with horse antilymphocyte globulin for severe aplastic anaemia. Br J Haematol 1998;100(2):393-400.

Tichelli A, Socié G, Henry-Amar M, Marsh J, Passweg J, Schrezenmeier H, et al. Effectiveness of immunosuppressive therapy in older patients with aplastic anemia. European Group for Blood and Marrow Transplantation Severe Aplastic Anaemia Working Party. Ann Intern Med 1999;130(3):193-201.

Tichelli A, Socié G, Marsh J, Barge R, Frickhofen N, McCann S, et al. Outcome of pregnancy and disease course among women with aplastic anemia treated with immunosuppression. Ann Intern Med 2002;137(3):164-72.

Tisdale JF, Dunn DE, Geller N, Plante M, Nunez O, Dunbar CE, et al. High-dose cyclophosphamide in severe aplastic anaemia: a randomised trial. Lancet 2000;356(9241):1554-9.

Tisdale JF, Dunn DE, Maciejewski J. Cyclophosphamide and other new agents for the treatment of severe aplastic anemia. Semin Hematol 2000;37(1):102-9.

Tisdale JF, Maciejewski JP, Nunez O, Rosenfeld SJ, Young NS. Late complications following treatment for severe aplastic anemia (SAA) with high-dose cyclophosphamide (Cy): follow-up of a randomized trial. Blood 2002;100(13):4668-70.

Tönnies H, Huber S, Kuhl JS, Gerlach A, Ebell W, Neitzel H.Clonal chromosomal aberrations in bone marrow cells of Fanconi anemia patients: gains of the chromosomal segment 3q26q29 as an adverse risk factor. Blood . 2003 May 15;101(10):3872-4.
doi: 10.1182/blood-2002-10-3243. Epub 2003 Jan 2.

Tooze JA, Saso R, Marsh JC, Papadopoulos A, Pulford K, Gordon-Smith EC. The novel monoclonal antibody By114 helps detect the early emergence of a paroxysmal nocturnal hemoglobinuria clone in aplastic aemia. Exp Hematol 1995;23 (14):1484-91.

Townsley DM, Dumitriu B, Liu D, Biancotto A, Weinstein B, Danazol Treatment for Telomere Diseases. N Engl J Med. 2016 May 19;374(20):1922-31.

Townsley, Danielle M., Bogdan Dumitriu, and Neal S. Young. 2016. “Danazol Treatment for Telomere Diseases.” The New England Journal of Medicine 375 (11): 1095–96. doi:10.1056/NEJMc1607752.

Townsley DM, Scheinberg P, Winkler T, Desmond R, Dumitriu B et al. Eltrombopag Added to Standard Immunosuppression for Aplastic Anemia. N Engl J Med. 2017 Apr 20;376 (16):1540-1550.

Ulirsch JC, Verboon JM, Kazerounian S, Guo MH, Yuan D, et al. The Genetic Landscape of Diamond-Blackfan Anemia. Am J Hum Genet. 2018 Dec 6;103(6):930-947. doi: 10.1016/j.ajhg.2018.10.027. Epub 2018 Nov 29.

Uwai M, Kanai N, Hatake K, Saito K, Miura Y. Effects of haemopoietic growth factors for aplastic anaemia. Lancet 1994;344 (8924):756-7.

Van Zeeburg HJ, Snijders PJ, Wu T, Gluckman E, Soulier J, Surralles J, Castella M, van der Wal JE, Wennerberg J, Califano J, Velleuer E, Dietrich R, Ebell W, Bloemena E, Joenje H, Leemans CR, Brakenhoff RH. Clinical and molecular characteristics of squamous cell carcinomas from Fanconi anemia patients. J Natl Cancer Inst. 2008 Nov 19;100(22):1649-53. doi: 10.1093/jnci/djn366. Epub 2008 Nov 11.

Vlachos A, Rosenberg PS, Atsidaftos E, Alter BP, Lipton JM. Incidence of neoplasia in Diamond Blackfan anemia: a report from the Diamond Blackfan Anemia Registry. Blood. 2012 Apr 19;119(16):3815-9. doi: 10.1182/blood-2011-08-375972. Epub 2012 Feb 23

Vlachos A, Rosenberg PS, Atsidaftos E, Kang J, Onel K, Sharaf RN, Alter BP, Lipton JM. Increased risk of colon cancer and osteogenic sarcoma in Diamond-Blackfan anemia. Blood. 2018 Nov 15;132(20):2205-2208. doi: 10.1182/blood-2018-05-848937. Epub 2018 Sep 28.

Vlachos A, Atsidaftos E, Lababidi ML, Muir E, Rogers ZR, Alhushki W et al. L-leucine improves anemia and growth in patients with transfusion-dependent Diamond-Blackfan anemia: Results from a multicenter pilot phase I/II study from the Diamond-Blackfan Anemia Registry. Pediatr Blood Cancer. 2020 Dec;67(12):e28748. doi: 10.1002/pbc.28748. Epub 2020 Oct 6.

Waisfisz Q, Morgan NV, Savino M, de Winter JP, van Berkel CG, Hoatlin ME,Spontaneous functional correction of homozygous fanconi anaemia alleles reveals novel mechanistic basis for reverse mosaicism. Nat Genet. 1999 Aug;22(4):379-83. doi: 10.1038/11956. `

Willig TN, Niemeyer CM, Leblanc T, Tiemann C, Robert A, Budde J, Lambiliotte A, Kohne E, Souillet G, Eber S, Stephan JL, Girot R, Bordigoni P, Cornu G, Blanche S, Guillard JM, Mohandas N, Tchernia G. Identification of new prognosis factors from the clinical and epidemiologic analysis of a registry of 229 Diamond-Blackfan anemia patients. DBA group of Société d'Hématologie et d'Immunologie Pédiatrique (SHIP), Gesellshaft für Pädiatrische Onkologie und Hämatologie (GPOH), and the European Society for Pediatric Hematology and Immunology (ESPHI). Pediatr Res. 1999 Nov;46(5):553-61.

Young NS. The problem of clonality in aplastic anemia: Dr Dameshek's riddle, restated. Blood 1992;79(6):1385-92.

Young NS, Alter BP. Aplastic anemia acquired and inherited. Philadelphia: Saunders WB; 1994.

Young NS, Barrett AJ. The treatment of severe acquired aplastic anemia. Blood 1995;85(12):3367-77.

Young NS, Maciejewski JP, Selleri C, Sato T, Nistico A, Brown KE, et al. The pathophysiology of acquired aplastic anemia. Ontogeny of hematopoiesis aplastic anemia.

International workshop Paris April 3-6, 1995. Colloques INSERM 1995;235:201.

Young NS. Aplastic anaemia. Lancet 1995;346(8969):228-32.

Young NS. Immune pathophysiology of acquired aplastic anaemia. Eur J Haematol Suppl 1996;60:55-9.

Young NS, Maciejewski J. The pathophysiology of acquired aplastic anemia. N Engl J Med 1997;336(19):1365-72.

Young NS. Autoimmunity and its treatment in aplastic anemia. Ann Intern Med 1997;126(2):166-8.

Young NS. Acquired aplastic anemia. JAMA 1999;282(3):271-8.

Young NS. Hematopoietic cell destruction by immune mechanisms in acquired aplastic anemia. Semin Hematol 2000;37(1):3-14.

Young NS. Introduction: acquired aplastic anemia. Semin Hematol 2000;37(1):2.

Young NS. Acquired aplastic anemia. Ann Intern Med 2002;136(7):534-46.

Young NS, Calado RT, Scheinberg P. Current concepts in the pathophysiology and treatment of aplastic anemia. Blood 2006;108(8):2509-19.

Zeng W, Nakao S, Takamatsu H, Yachie A, Takami A, Kondo Y, et al. Characterization of T-cell repertoire of the bone marrow in immune-mediated aplastic anemia: evidence for the involvement of antigen-driven T-cell response in cyclosporine-dependent aplastic anemia. Blood 1999;93(9):3008-16.