1. Introduction

Dr. Fanconi, who noted a family with 3 brothers who had macrocytic anemia and several physical anomalies, first described Fanconi anemia in 1927[1] . He made diagnostic criteria of FA including 1)pancytopenia, 2)skin pigmentation, 3)physical anomalies, 4)short stature, 5) hypogonadism and 6) family history[2] . Schroeder et al reported chromosomal instability in lymphocytes of FA patients[3] , and Sasaki et al found that chromosomal fragility was increased by mitomycin C[4] .

FA patients are at risk for bone marrow failure, myelodysplastic syndrome (MDS), leukemia and solid tumor such as squamous cell carcinoma. The only cure for the hematological complications of FA remains hematopoietic stem cell transplantation (HSCT). In recent years, HSCT from alternative donors have become more popular and have been successfully employed[5][6].

This reference guide for the diagnosis and treatment of patients with FA is the result of a data of several registrations and /or references by Japanese medical professionals for FA.

2. Diagnosis
1) Diagnostic evaluation of FA

FA is an autosomal recessive disorder associated with a high frequency of bone marrow failure, leukemia and solid tumors. FA is a complex disease that can affect many systems of the body. Patients are recognized when they have the combination of aplastic anemia and birth defects, but not restricted to severe birth defects. Current diagnostic criteria are more extensive, and rely on demonstration of chromosomal aberrations in cells cultured with DNA-crosslinking agents such as mitomycin C (MMC) or diepoxybutane (DEB). Indications for chromosome fragility test are below.

- Aplastic anemia in a childhood
- Sibling with FA
- Characteristics birth defects
- Family history consistent with FA or with cancer
- Spontaneous chromosome breaks
- Primary MDS at a young age
- Acute myeloid leukemia (AML)/MDS with chromosomal aberration No1, 3 and 7
- Unusual sensitivity to chemo-radiotherapy
- Cancer typical of FA at an typical age (head and neck squamous cell carcinoma, esophagus and liver

2) Diagnostic strategy for FA (Figure 1.)

If FA is suspected, the patients should be referred to a hematologist to arrange for MMC or DEB chromosome fragility test of blood lymphocytes.   Following DNA damage, the complex of upstream FA gene products (A,B,C,E,F,G,I,L,M) leads to ubiquitination of the product of *FANCD2* on a Western blot with a D2-specific antibody. If diagnostic test results of blood are not conclusive and there is a high probability of FA, skin or bone marrow fibroblast cultures are required to demonstrate sensitivity to DNA-crosslinking agents. FA mutation analysis determines the initial complementation group. Gene sequencing can be performed to determine the relevant mutations.

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Clinical evaluation (bone marrow failure in childhood, physical exam, solid tumor at young age and family history) chromosome fragility test in lymphocytes and/or FANCD2 immunoblot

No breaks
No positive FA signs

Negative tests in blood but positive findings for FA and/or family history

Increased breaks and/or abnormal FACD2 pattern

Negative

Non FA

Positive

FA

Chromosome fragility test, FANCD2 immunoblot and/or FA genes analysis (FA sub-grouping) in fibroblasts

Standard treatment
*Detection of other congenital bone marrow failure syndrome

FA genes analysis (FA sub-grouping)
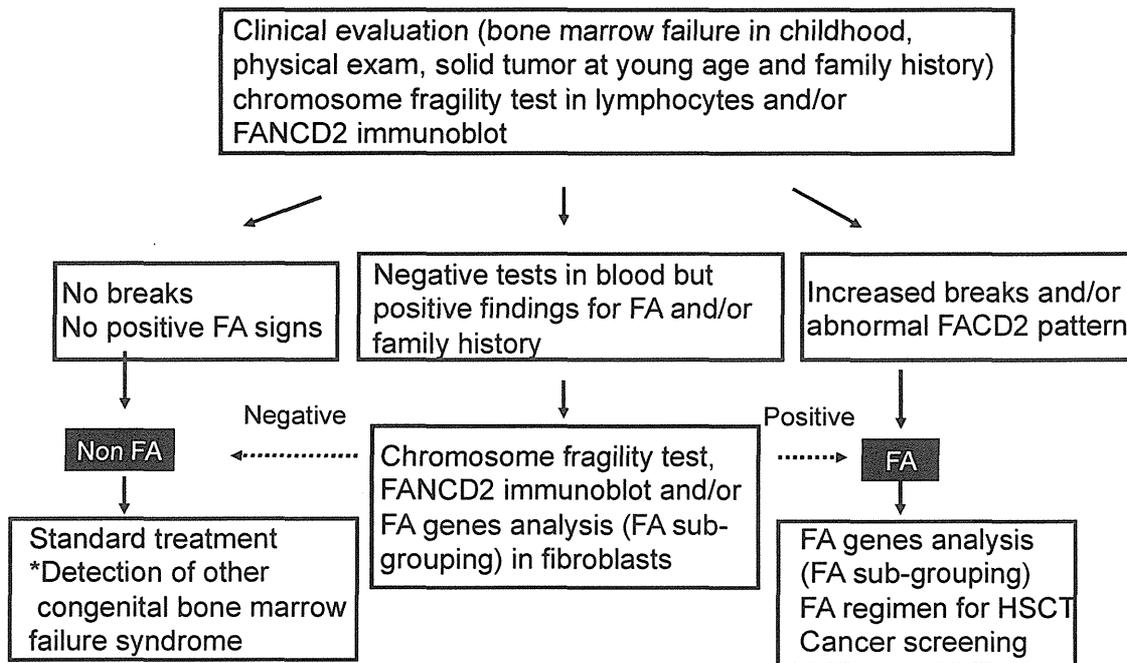FA regimen for HSCT
Cancer screening

Figure 1. Diagnostic strategy for Fanconi anemia
*Shwachman-Diamond syndrome, Dyskeratosis congenita, Congenital amegakaryocytic thrombocytopenia, Pearson syndrome and Diamond-Blackfan anemia

3) Severity of bone marrow failure (Table 1)

Aplastic anemia for FA is also classified on the basis of severity of acquired aplastic anemia in Japan.

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Table1.   Severity of bone marrow failure (modified by 2004)

stage 1	mild	except for the following

stage 2   moderate   with more than two of the following
                        Reticulocyte   <60,000/μl
                        Neutrophil   <1,000/μl
                        Platelet   <50,000/μl

stage 3   moderate to
           severe   with more than two of the following and *periodic red cell transfusion
                        Reticulocyte   <60,000/μl
                        Neutrophil   <1,000/μl
                        Platelet   <50,000/μl

stage 4   severe   with more than two of the following
                        Reticulocyte   <20,000/μl
                        Neutrophil   <500/μl
                        Platelet   <20,000/μl

stage 5   very   with neutrophil count<200/μl and more than one of the following
           severe   Reticulocyte   <20,000/μl
                        Platelet   <20,000/μl

* Periodic red cell transfusion needs more than 2 units of red blood cell for a month.

4) Differential diagnosis （Table 2)
   Table 2 shows inherited bone marrow failure syndrome with physical abnormalities. Chromosome
   instability syndrome was a group of inherited conditions associated with chromosomal instability and
   breakage, and the following chromosome instability syndromes are known: xeroderma pigmentosum,
   Ataxia Telangiectasia Bloom syndrome and Nijmegen syndrome.

Table 2.   Inherited bone marrow failure syndrome

	FA	DKC	SDS	CAMT	Pearson
Reports	>1000	>225	>300	>45	>60
Genetics	AR (XL)	XL85%, AD, AR	AR	AR, XL	Sporadic
Mutated genes	16	DKC1 (Xq28) etc.	SBDS (7q11)	c-mpl (1q34)	mt DNS
Age at diagnosis	7.6 y	5~15 y	4 m	9 m	6 m
Physical anomalies	75%	100%	60%	40%	Rare
Pancytopenia	90%	50% by 10 y of age	Neutropenia 95%	40%	Unknown
MDS/AML	>14%	0.4~1.3%	5~33%	5%	0%
Cancer	7%	8~12%	0%	0%	0%
Chromosomal instability	+	-	-	-	-
Prognosis	The average life span 30 y	80% died by 30 y of age	The average life span 36 y	50% died by 3 y of age	80% died by 3 y of age

FA : Fanconi anemia   DKC : Dyskeratosis congenita   SDS : Schwachman-Diamond syndrome
MMC : mitomycin C   CAMT : Congenital amegakaryocytic thrombocytopenia
DEB :   diepoxybutane   AR : autosomal recessive   AD : autosomal dominant   XL : X-linked

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3. Epidemiology

1) A rate of incidence

About 5~10 cases are diagnosed as FA, and the incidence nation-wide is estimated to be 5 in 1000,000 new born babies. The carrier frequency for FA in Japan may be 1 in 200~300 peoples[7].

2) Prognosis

Of the 754 FA patients in the International Fanconi Anemia Registry Study, 601 (80%) experienced the on set of bone marrow failure (BMF) and 173 (23%) had a total of 199 neoplasms. The risk of developing BMF and hematologic and nonhematologic neoplasms increased with advancing age with a 90%, 33%, and, 28% cumulative incidence, respectively, by 40 years of age[8]. Ten-year over-all survival of FA patients without HSCT was 63 % according to the reports of Japanese Society of Pediatric Hematology[9].

4. Molecular features

FA is a multigenic disorder with 16 genes currently identified (A, B, C, D1, D2, E, F, G, I, J, L, M, N, O, P, Q). *FANCD1, FANCJ* and *FANCN* were identified as BRCA2, BRIP1 and PALB2, the breast and ovarian cancer susceptibility gene, respectively. These three FA genes are associated with breast cancer in heterozygotes. With the exception of the X-linked *FANCB* gene, the remaining 15 FA genes are autosomal recessive. The most popular gene is *FANCA* (60-70%), and more than 80 % of FA patients belong to *FANCA, FANCC* and/or *FANCG*. These FA proteins cooperate with other gene products, in a common pathway (FA-BRCA pathway), which regulate DNA repair (Figure 2). The FA pathway plays an important role in the proliferation of hematopoietic stem cells and tumor suppression. The FANCD1 and FANCN subtype represents a malignant tumor in early childhood and their prognosis is very poor[10] [11]. On the contrary, clonal expansion of hematopoietic cells with reversion mosaicism can restore normal hematopoiesis[12].
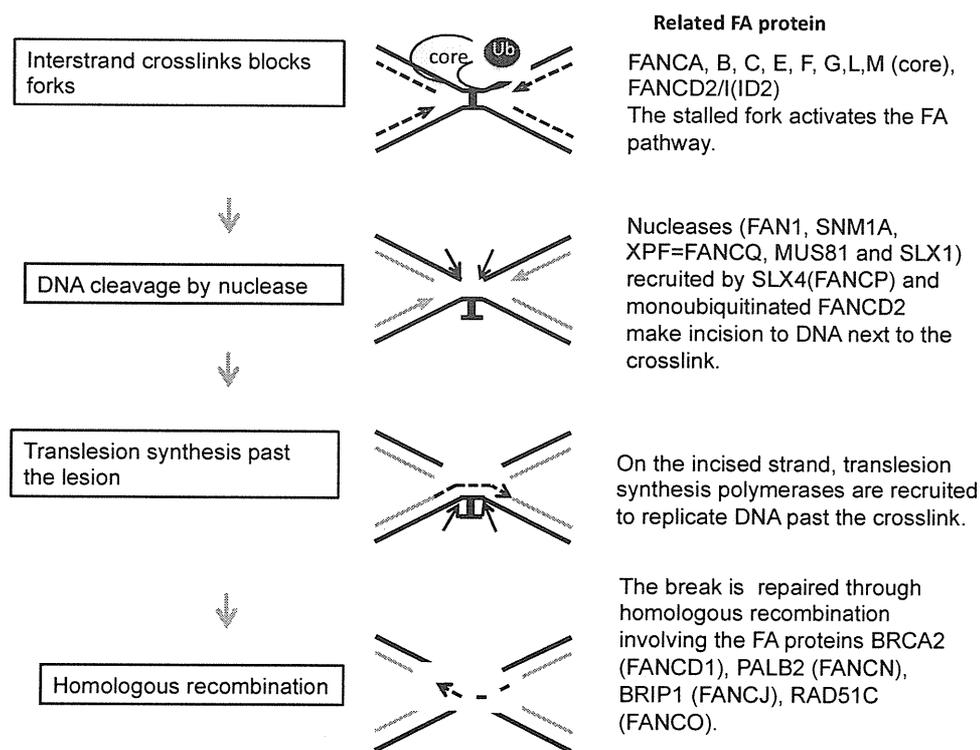

Figure 2. Schema of FA-BRCA pathway and interstrand crosslinks repair

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5. Clinical features

2) Physical appearance (Table 3)

Table 3 lists the physical findings that have been reported in FA in literature and Japanese FA patients. It must be noted, however, that 25% or more of known FA patients have few or none of these features. The most frequently characteristics birth defects in FA are skin hyperpigmentation, *café au lait* spots, short stature, abnormal thumbs and radii[13)14)].

Table 3. Physical finding in Fanconi anemia

Abnormality	Reported in literature	Japan
Skin pigmentation	40%	79%
Short stature	40%	71%
Thumbs	35%	54%

Gonads
  Males
  Females
	
	25%
	2%
		
		Total 8%

Head/Neck	23%	13%
Eyes	20%	10%
Renal	20%	15%
Ears/Deaf	10%	16%
Lower limbs	7%	?
Cardiopulmonary	6%	16%
Gastrointestinal	5%	13%

3) Cancer

Patients with FA are at a particularly high risk of developing MDS/AML and specific solid tumors unusually young ages, including head, neck, esophageal, and gynecological squamous cell carcinoma. In the literature cases, about 15~20% of FA patients had hematologic neoplasms, and 5~10% had nonhematologic neoplasms [8)15) 16)]. Of these neoplasms in Japanese FA patients, 33% were hematologic and 10.4% were nonhematologic[13)]. Solid tumors were observed both after and before HSCT, and developed of a person, the ages between 1 to 39 years old (Table 4).

The types of cancers in FA patients are summarized in Table 5[15)].

Table 4. Reports of Cancer in Fanconi anemia

Report	Alter[15)]	Kulter[8)]	Rosenberg[16)]	Yabe[13)]
Year of observation	1927-2001	1982-2001	-2000	1986-2010
Number	1301	754	145	96
No of HSCT	220 (17%)	219 (24%)	44 (30%)	86 (90%)
Male/Female	1.23	1.05	1.10	1.00
Median age at diagnosis, years (range)	7 (0-48)	NA	5 (0-45)	4.4 (0-24)
MDS/AML	205 (16%)	100 (13%)	32 (22%)	32 (33%)
Cancer	68 (5%)	67 (9%)	13 (9%)	10 (10.4%)

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Table 5. Cancer in Fanconi anemia

Type	Male	Female	Total	Median age, years
Head and Neck	13	13	26	28
Esophagus	1	8	9	27
Cervix	-	3	3	25
Brain	2	4	6	3
Genitourinary	3	3	6	3
Breast	-	4	4	37
Liver	20	14	44	13
Lung	3	0	3	29
Lymphoma	1	1	2	1
Gastric	2	0	2	28
Colon	0	1	1	21
Osteosarcoma	0	1	1	7
Retinoblastoma	0	1	1	0.3

6. Treatment of hematological abnormalities

1) Blood transfusion

FA patients would be transfused to maintain minimal trough hemoglobin of 6g/dl and minimal trough platelet of 5,000/µl. Clinical adequacy of the transfusion regimen should be continuously assessed by patient's subjective symptoms and bleeding episodes.

2) Hematopoietic growth factors

Patients with severe or symptomatic neutropenia (neutrophil count <500/µl) may benefit from a granulocyte-colony stimulating factor (G-CSF). Erythropoietin is not used for anemia in FA patients.

3) Androgen therapy

Immunosuppressive therapy is not recommended for FA patients, because the cause of bone marrow failure should be hematopoietic stem cell damage. Androgens have been widely used for the treatment of cytopenias in FA. The effects of androgens were recognized in half of FA patients, but its effect seems to have lasted only temporarily[17]. Androgen may exacerbate the risk of liver tumors, and prior exposer to androgens is the one of the risk factors affecting survival after unrelated transplant[18]. Metenolone acetate is used in Japan. A clinical trial of danazol, which has less virilizing side effects, is currently under way. The use of steroid hormone should be avoided.

4) Hematopoietic stem cell transplantation

Currently the only cure for the hematological abnormalities of FA remains HSCT. The use of high-dose cyclophosphamide (CY) and radiation in preparative regimens for FA patients often resulted in excessive organ toxicity and death. The use of low dose CY (20-40mg/kg) combined with 4-6 Gy of thoraco-abdominal irradiation (TAI) or total body irradiation (TBI) resulted in reduced toxicity, improved the outcome for the FA patients transplanted from HLA-matched sibling donors [19]. Non-radiation regimens have been increasingly used for FA patients to reduce the second malignancies associated with radiation[20] [21] [22] [23]. HSCT from alternative donors is more complex. It is associated with a higher risk of rejection and acute graft-versus host disease (GVHD), and survival rates was lower than that observed with HLA-matched sibling donors. The 3-year probability of survival in the 69 FA patients transplanted from HLA-matched unrelated donor was 33% by a study on behalf of the European Group for Blood and Marrow Transplantation. Extensive malformation, a positive recipient cytomegalo serology, the use of androgens before transplant, and female donors were associated with a worth outcome[18]. A relevant proportion of FA patients undergoing HSCT can now be dramatically cured, even in the absence of an HLA-identical sibling, especially if the conditioning regimen includes fludarabine[5][25][26](Table 6). In Japanese FA patients transplanted using fludarabine regimen, the 3-year estimate of overall survival (OS) for the 8 patients was 100% when donor was an HLA-matched sibling[27], and 26 patients out of 27 patients transplanted from an alternative donor are alive[6]. Table 7 and Table 8 show the indications of HSCT for FA patients and recommended HSCT methods for FA patients in Japan. We recommend bone marrow for stem cell source. Peripheral blood progenitor cell grafts appeared to contribute to higher chronic GVHD, and chronic GVHD should be the major risk factor of secondary malignancy after HSCT for FA patient[31]. We also do not recommend unrelated cord blood transplantation (CBT) for FA patients, because the risk of graft failure after unrelated CBT was high[32] .

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Table 6. Allogeneic stem cell transplantation for Fanconi anemia

Study	Stem cell source	Conditioning	GVHD prophylaxis	N	Median age at HSCT, years (range)	Rejection (%)	Acute GVHD II-IV (%)	Chronic GVHD (%)	2~3 years OS
Seattle[22]	HLA-identical sibling/BM	CY	CYA/MTX	9	8(4-19)	0	22	0	89
Paris[19]	HLA-identical sibling/BM	CY/TAI	CYA	50	11(4-26)	6	55	70	59
Brazil[23]	HLA-identical sibling/BM	CY	CYA/MTX	10	7(4-21)	0	13	7	88
Italy[28]	HLA-identical sibling/BM	CY CY/TAI (TBI)	CYA/MTX CYA	27	6(2-13)	8	26	13	81
Minnesota[29]	HLA-identical sibling/BM, CB	Flu/CY/ATG	CYA/MP T-cell depletion	11	NA	0	0	0	100
EBM[18]	Alternative	CY/TAI(TBI) ±ATG	CYA/MTX CYA/MP CYA±T-cell depletion	69	11(4-37)	20	43	43	33
Minnesota[30]	Alternative	Flu/CY/ATG/ TAI	CYA/MP T-cell depletion	41	NA	2	19	16	52
Japan[27]	HLA-identical sibling/BM	CY/TAI (TBI)/ATG Flu/CY/ATG	CYA/MTX CYA/MTX	8 7	8(5-24) 6(1-15)	25 0	12 0	38 0	100 100
Japan[6]	Alternative	Flu/CY/ATG/ TAI	FK/MTX±MMF	27	8(2-28)	4	11	31	96

EBMT: European Group for Blood and Marrow Transplantation, CY : cyclophosphamide, TAI : thoraco-abdominal irradiation, TBI : total body irradiation, ATG : antithymocyte globulin, CYA : cyclosporine, MTX : methotrexate, FK : tacrolimus, MMF : mycophenolate mofetil, MP : methylprednisolone, Flu : fludarabine

Table 7. Indication of hematopoietic stem cell transplantation for Fanconi anemia

Disease status	Indication of HSCT
1. Aplastic anemia 　Stage I (mild) 　Stage II (moderate) 　Stage III (moderate to severe) 　Stage IV, V (severe, very severe)	Watch and wait <10 years old: watch and wait ≧10 years old: BMT should be considered if the patient has an HLA-identical sibling. BMT should be considered if the patient has an HLA-identical sibling. BMT should be considered from HLA-matched or one mismatched related or unrelated donor.
2. MDS and AML 　RA 　RAEB and AML	Same as stage IV, V of aplastic anemia BMT should be considered from HLA-matched related or unrelated donor, if the patient has severe dysplasia or cytogenetic clones. BMT should be considered from HLA-matched or one mismatched related or unrelated donor. BMT also should be considered from haploidentical related donor if the patient has life-threatening disease status.

MDS: myelodysplastic syndrome, AML: acute myeloid leukemia, RA: refractory anemia, RAEB: refractory anemia with excess of blasts

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Table 8. Conditioning and GVHD prophylaxis for Fanconi anemia

Disease status	HLA –identical sibling donor	Alternative donor
Aplastic anemia and RA Conditioning	Flu 25mg/m² × 6days CY 10mg/kg × 4days ATG 1.25mg/kg × 4days	Flu 25mg/m² × 6days CY 10mg/kg × 4days ATG 1.25mg/kg × 4days TLI/TAI 3Gy (3gy X1)
GVHD prophylaxis	<10 years old: CyA (1.5mg/kg ×2/day) ≧10 years old: CyA (1.5mg/kg×2/day) + MTX (day1; 10 mg/ m²,3 and 6; 7 mg/m²)	FK (0.02 – 0.03mg/kg/day) + MTX (day1; 15 mg/ m²,3,6 and 11; 10mg/m²)
RAEB and AML Conditioning	Flu 25mg/m² × 6days CY 10mg/kg × 4days ATG 1.25mg/kg × 4days TBI 4.5Gy (1.5Gy X 3)	Flu 25mg/m² × 6days CY 10mg/kg × 4days ATG 1.25mg/kg × 4days TBI 4.5Gy (1.5Gy X 3)
GVHD prophylaxis	<10 years old: CyA (1.5mg/kg ×2/day) ≧10 years old: CyA (1.5mg/kg×2/day) + MTX (day1; 10 mg/ m²,3 and 6; 7 mg/m²)	FK (0.02 – 0.03mg/kg/day) + MTX (day1; 15 mg/ m²,3,6 and 11;10mg/m²)

Flu : fludarabine,   CY : cyclophosphamide,   ATG : antithymocyte globulin
TAI : thoraco-abdominal irradiation,   TLI : total lymphoid irradiation
TBI : total body irradiation, CyA : cyclosporine,   MTX : methotrexate,   FK : tacrolimus

5.   Future view

Patients with FA usually are diagnosed in childhood, and their registrations have been managed in the Japanese Pediatric Hematology/Oncology society in Japan. Although adult FA population is small, it is important to understand transplanted/non-transplanted adult FA patients, who have or not have developed bone marrow failure, hematological malignancy and solid tumors. Fludarabine-based preconditioning regimen can be used satisfactorily in alternative HSCT for FA; however, long-term observation of secondary cancers and other late effects will be required to determine the therapeutic utility of this approach.

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遺伝性鉄芽球性貧血診療の参照ガイド

先天性骨髄不全症候群の診療の参照ガイドWG
遺伝性鉄芽球性貧血：東北大学血液免疫科　張替秀郎

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1．緒　言

　鉄芽球性貧血は、骨髄における環状鉄芽球の出現を特徴とする貧血であり、環状鉄芽球はミトコンドリアへの鉄の異常蓄積により形成される。鉄芽球性貧血は、遺伝性鉄芽球性貧血と、骨髄異形成症候群(MDS)およびアルコールや薬剤による二次性鉄芽球性貧血からなる後天性鉄芽球性貧血に大別される。遺伝性鉄芽球性貧血はまれな疾患で、ヘム合成不全、鉄-硫黄クラスター形成不全などにより、ミトコンドリアにおける鉄代謝に異常が生じ発症する難治性貧血である。1945年にCooleyがX連鎖性小球性低色素性貧血を呈する家族性貧血症を報告したが、1946年にRundlesとFallsがこの家系を含む2家系を報告したことで、このX連鎖性小球性低色素性貧血はRundles and Falls症候群と名づけられた(1)。後にこの貧血は赤血球におけるヘム合成系の初発酵素であるδ－アミノレブリン酸合成酵素（*ALAS2*）の変異によるX連鎖性鉄芽球性貧血（XLSA）であることが証明された(2)。現在、遺伝性鉄芽球性貧血の原因としてこの*ALAS2*の変異がもっとも多く報告されているが、その他にも鉄－硫黄クラスター合成・輸送に関わる遺伝子、ミトコンドリアDNA遺伝子、ミトコンドリアトランスポーター遺伝子、ミトコンドリアtRNA関連遺伝子など複数の遺伝子の変異が報告されている。表1に主な遺伝性鉄芽球性貧血とその原因遺伝子を示す。ただし、原因遺伝子が同定されない遺伝性鉄芽球性貧血も多く、既報の遺伝子以外にも原因となる遺伝子が存在すると考えられている。遺伝性鉄芽球性貧血は、原因遺伝子の機能の多様性から、貧血以外に神経・筋など他の臓器に異常を認める場合が多く、また貧血の重症度もさまざまである。多くの遺伝性鉄芽球性貧血では特異的治療法がないものの、XLSAのように適切な診断・治療がなされれば、貧血の改善が期待できるみられるタイプも存在するため、遺伝子診断による確定診断が重要である。

	遺伝形式	遺伝子座	遺伝子	治療
XLSA*	X連鎖性	Xp11.21	ALAS2	Vit B6
XLSA / A**	X連鎖性	Xq13.1	ABC7	—
SA /GLRX5	常染色体劣性？	14q32.13	GLRX5	？
SA /SCL25A38	常染色体劣性？	3p22.1	SCL25A38	？
PMPS***	母性	ミトコンドリア	ミトコンドリア	—
TRMA****	常染色体劣性？	1q23.3	SCL19A2	Vit B1
MLASA*****	常染色体劣性？	12q24.33	PUS1	—

　*X-連鎖性鉄芽球性貧血　　　　**小脳失調を伴うX-連鎖性鉄芽球性貧血
***Pearson Marrow-Pancreas症候群　　****チアミン反応性巨赤芽球性貧血
*****ミトコンドリア筋症を伴う鉄芽球性貧血

表1　遺伝性鉄芽球性貧血の責任遺伝子

2．　診　断

1）疾患概念
　　骨髄における環状鉄芽球の出現を特徴とする貧血である。

2）診断基準
　　環状鉄芽球が骨髄総赤芽球の15％を超える（FAB分類）
　　血清フェリチンの増加、不飽和鉄結合能減少を認める。
　　上記に加えて遺伝子変異が確認できたものが、遺伝性鉄芽球性貧血の確定診断となる。家族歴は遺伝性鉄芽球性貧血を強く疑う所見である。

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遺伝性で最も頻度の高いXLSAは小球性低色素性の貧血で男児発症を特徴とする。

環状鉄芽球の定義：核周囲1/3以上にわたって10個以上の鉄顆粒が存在（新WHO分類）

3）診断のフローチャート
　遺伝性鉄芽球性貧血は、まず鉄芽球の存在、若年発症、遺伝性により疑い、遺伝子解析により診断を確定する。家系の中での遺伝性が明らかでない場合は、造血細胞以外の組織で遺伝子の変異を確認し、胚細胞変異であることを確認する。遺伝性鉄芽球性貧血の中では*ALAS2*変異によるXLSAの頻度が最も高いため、男児で、臨床上ビタミンB6に反応性を認めた場合は積極的に遺伝子解析を行う。XLSAの場合は変異ALAS2たんぱく質の活性低下をin vitroで確認することも可能である。

4）鑑別診断
　　　以下に挙げる後天性鉄芽球性貧血を除外する必要がある。
　　　　　後天性鉄芽球性貧血
　　　　　　　　薬剤性、中毒性： 抗結核薬、鉛等
　　　　　　　　アルコール性：ヘム合成酵素障害、VitB6欠乏
　　　　　　　　骨髄異形成症候群
　通常、後天性鉄芽球性貧血は発症年齢、遺伝性から鑑別が可能であるが、成年発症のXLSA症例も報告されていることから(3)、時に遺伝性との鑑別を必要とする。アルコール性、薬剤性の後天性鉄芽球性貧血については、生活歴、治療歴から鑑別する。薬剤性はVit B6に対する拮抗作用を原因として発症することが多い。Vit B6はALAS2の補酵素であるため、その欠乏により、ALAS2活性が低下し鉄芽球性貧血の発症に至る。抗結核薬のINHはその代表的な薬剤である。多系統の血球に異常が認められる場合や染色体異常が認められる場合は骨髄異形成症候群の診断となるが、貧血のみで染色体異常がなく、ビタミンB6に反応する場合は、遺伝子解析を考慮するべきである。

3. 疫学
　3) 発生頻度
　　　発症頻度は極めて稀で詳細な疫学データはない。最も頻度の高い遺伝性鉄芽球性貧血はXLSAで、現在までに94家系、57種類の*ALAS2*の変異が確認されている（未発表を含める。表2）。83例の遺伝性鉄芽球性貧血症例を解析した米国の報告では、*ALAS2*、*SLC25A38*、mitochondria DNA、*PUS1*に変異を認めた頻度はそれぞれ37%, 15%, 2.5%, 2.5%であった(4)。厚生労働省研究班（遺伝性鉄芽球性貧血の診断分類と治療法の確立班）にて本邦の遺伝性鉄芽球性貧血の実態を調査したところ、変異遺伝子が確定した症例はすべて*ALAS2*遺伝子変異によるXLSAであり、、*SLC25A38*、*PUS1*、*ABCB7*、*GLRX5*、*SLC19A2*遺伝子変異による遺伝性鉄芽球性貧血は認められなかった（表3, 4）(5)。

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Ex.	substitution		No. of pedigree	Ex.	substitution		No. of pedigree	Ex.	substitution		No. of pedigree
4	L107P		1	6	S251P		1	9	R452	C	9 (2)
5	M154I		1	7	D263N		2			G	1
	K156E		1		C276W		1			S	2
	D159	N	1		I289T		1			H	8 (2)
		Y	1		G291S		1		R458H		1
	T161A		1		K299Q		1		I476N		1
	F165L		2		V301A		1	10	Y506-fs		1
	R170	S	1	8	P339L		1		T508S		1
		C	2 (1)		D351R		1		R517	C	1
		L	3 (2)		R375C		1			G	1
		H	2		T388S		1		P520L		3
	A172T		1	9	C395Y		1		H524D		1
	D190V		1		G398D		1	11	K535del		1
	Y199H		1		R411	C	6 (2)		R559H		1
	R204	Q	1			H	4		R560H		2
		stop	1		G416D		1		V562A		1
6	R218H		1		M426V		1		M567I		1
	R227C		1		R436W		1		S568G		2 (1)
	E242K		1		R448Q		3		R572H		2

表2. これまでに確認されている XLSAにおける*ALAS2*遺伝子変異。Pyridoxineに反応する変異は網掛けで示す。

Case number	Age at diag	Gender	Position of ALAS2 mutation	*SF3B1* mutation	Hb at onset (g/dl)	MCV at onset (fl)	Increment of Hb by Vit B6 treatment (g/dl)	In vitro enzymatic activity of mutant protein*	
								w/o PLP	with PLP
1	0	M	R170C	N/D	4.8	52.5	1.7	64.1%	72.5% **
2	20	M	R411C	N/D	4.8	52.5	5.2	11.9%	25.1% [ref 19]
3	68	M	R452C	-	6.0	67.3	No effect	99.9%	94.0% [ref 21]
4	17	M	D190V	N/D	8.9	66.9	No effect	98.6%	98.5% [ref 20]
5	36	M	R452C	-	7.4	70.0	No effect	99.9%	94.0% [ref 21]
6	36	M	M567I	N/D	6.5	64.4	3.4	38.1%	25.2% [ref 21]
7	14	M	V562A	-	8.1	61.2	4.7	150.6%	116.9% [ref 21]
8	31	M	R170L	-	4.1	50.8	8.1	31.1%	60.8% **
9	3	M	R452C	-	5.4	54.4	2.9	11.9%	25.1% [ref 19]
10	62	M	R170L	N/D	8.0	73.9	No effect	31.1%	60.8% **

*% of WT, ** present study

表3. 遺伝性鉄芽球性貧血の診断分類と治療法の確立班の調査研究により確認された本邦の遺伝性鉄芽球性貧血（XLSA）

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Case number	Age at diag (y.o)	Gender	Family history	Gene mutation							Hb (g/dl)	MCV (fl)	Response to Vit B6
				ALAS2	SLC 25A38	GLRX5	ABCB7	SLC 19A2	PUS1	SF3B1			
11	14	M	-	Ex11 dup .	-	-	-	-	-	-	7.1	60.0	-
12	19	M	-	intron 1	-	-	-	-	-	-	7.8	73.9	-
13	4	M	-	Intron 1	-	-	-	-	-	-	6.6	73.6	-
14	0	M	+	Intron 1	-	-	-	-	-	-	3.9	65.0	-
15	20	M	+	Intron 1	-	-	-	-	-	-	7.6	82.0	+
16	0	M	-	-	-	-	-	-	-	-	6.8	88.1	N/D*
17	32	M	-	N/D	N/D	N/D	N/D	N/D	N/D	N/D	11.2	69	+
18	36	M	-	N/D	N/D	N/D	N/D	N/D	N/D	N/D	10.8	67.3	+
19	18	F	+	-	-	-	-	-	-	-	9.3	96.2	+

N/D: not done, *Vit B6 was not administered due to PMPS

表4. 遺伝性鉄芽球性貧血の診断分類と治療法の確立班の調査研究により確認された本邦の遺伝性鉄芽球性貧血（XLSA以外）

　2）自然歴・予後
　　　極めて稀な疾患のため、疫学、病態解析に関してまとまった報告がなく、不明である。

4．病因・病態
　遺伝性鉄芽球性貧血の原因となる遺伝子は複数あり、それぞれの機能は異なっている。ヘム合成はミトコンドリアにおいてグリシンとスクシニルCoAが重合し、δ－アミノレブリン酸が合成されるステップから始まるが、ALAS2は赤血球において特異的にこの重合を触媒する酵素であり、本遺伝子の変異によりヘム合成が不全となり、ミトコンドリアでの鉄利用障害が起こるものと考えられている。SLC25A38はミトコンドリア内膜に存在するトランスポーターであり、グリシンの輸送に関与していると考えられており、鉄芽球性貧血の発症機序はALAS2変異と同様であることが予想される(6)。一方、thiamine transporterである SLC19A2遺伝子の変異によるミトコンドリア鉄沈着は、thiamine欠乏によるスクシニルCoAの不足が原因と考えられている(7)。ただし、SLC19A2の変異による鉄芽球性貧血はXLSAと異なり、血中プロトポルフィリンレベルの低下が認められず、また大球性であるため、XLSA同様のヘム合成障害が原因であるかどうか疑問である。Pearson-marrow-pancreas症候群はmitochondria DNAの欠失によるものであり、神経・筋・外分泌機能に障害が認められ、多くは乳児期に死亡する(8)。鉄芽球の形成機序は明らかとなっていないが、呼吸鎖遺伝子の異常によって鉄の還元障害が起こり、フェロケラテーゼによるプロトポルフィリンへの鉄挿入が不全となっている可能性が考えられる。GLRX5はヘムと並ぶ鉄利用分子である鉄—硫黄クラスターの合成に関わる遺伝子であり(9)、ABCB7はこの鉄—硫黄クラスターのミトコンドリアからの排出を担うトランスポーターである(10)。いずれも、鉄—硫黄クラスターの障害を通じてミトコンドリアにおける鉄の利用障害を誘導すると考えられているが、その機序は共通でない。すなわち、GLRX5の変異による鉄着はIRPを介したALAS2活性低下によるものと考えられているが、ABCB7の変異においては、これらの所見は確認されていない。PUS1はtRNAの修飾に関与する遺伝子であり、本遺伝子の変異により、ミトコンドリアでのたんぱく質の翻訳に障害が生じるものと考えられているが、鉄利用障害に至る直接的な関与については明らかとなっていない(11)。いずれにおいても、ミトコンドリアでの鉄利用障害により、過剰な鉄がミトコンドリアに沈着し、環状鉄芽球が認められるようになる。この鉄過剰状態は細胞内の酸化還元反応を障害し、アポトーシスを誘導し貧血の発症に至ると考えられている(12)。

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13. 臨床症状、検査所見

1) 貧血
　　病型により軽度～中等度まで認められる。原因遺伝子が同じであっても、変異によって重症度が異なる。

2) ヘモクロマトーシス
　　病型と輸血量によりその程度は異なる。
　　*HFE*遺伝子に変異を認めるとヘモクロマトーシスの進行速度が速いが、日本人ではその遺伝子の変異の頻度は少ないといわれている。

3)その他の合併症
　　*ALAS2*および*SLC25A38*以外の遺伝子変異による遺伝性鉄芽球性貧血の場合、ミトコンドリア機能異常などによりにより、造血不全以外の臓器障害（Ataxia、代謝性アシドーシス、膵外分泌不全、インスリン依存性糖尿病、神経症状など）を認めることがある。

4)各病型の特徴
　　XLSA:小球性低色素性貧血、全身の鉄過剰状態を認める。XLSAの多くの症例において、ALAS2たんぱく質の構造変化により、補酵素であるビタミンB6との親和性が低下することが貧血の原因となっていると考えられており、実際に半数以上でビタミンB6の投与にて貧血の改善を認める。
　　GLRX5変異による遺伝性鉄芽球性貧血：*Glutaredoxin5*の変異でFe-S clusters合成が障害される結果、ミトコンドリアに鉄が沈着する。骨髄での環状鉄芽球は少ないが、中等度の貧血、肝脾腫、鉄過剰を認める。
　　Ataxiaを伴うXLSA (XLSA/A)：早期より(通常1歳以内より)ataxiaを認める。Ataxiaは進行しないか、進行しても緩徐である。貧血は小球性低色素性である。貧血は軽度でpyridoxineに反応しない。ミトコンドリアの膜輸送蛋白である*ABCB7*遺伝子の変異が原因である。
　　SLC25A38変異による遺伝性鉄芽球性貧血：*SLC25A38*はglycineを輸送するミトコンドリアの膜蛋白遺伝子と考えられている。常染色体劣性遺伝で、前述の通り、*ALAS2*についで、頻度が高い遺伝性鉄芽球性貧血と考えられている。多くは重度の小球性低色素性貧血を呈し、鉄過剰状態にあり、XLSAと同様の臨床症状を呈するため、XLSAを疑う症状を呈するもののALAS2の変異が認められない場合、本遺伝子の変異検索が必要である。
　　Pearson marrow pancreas syndrome：代謝性アシドーシス、ataxia、膵外分泌不全を伴う。通常乳児期に死亡する。貧血は正球性で好中球減少と血小板減少を時に伴う。ミトコンドリアDNAの欠損が原因で、通常孤発性で*de novo*の発症例が多い。
　　Thiamine-responsive megaloblastic anemia(TRMA)：インスリン依存性糖尿病、神経性難聴を伴う全身性の疾患。稀な常染色体劣性遺伝で通常幼少期に診断される。貧血は巨赤芽球を伴う大球性の貧血である。Thiamineの投与に反応するが、葉酸やVitB12、pyridoxineには反応しない。Thiamine transporterである*SCL19A2*遺伝子の異常が原因である。
　　Mitochondrial myopathy and sideroblastic anemia (MLASA)：極めて稀な常染色体劣性遺伝疾患。筋症、乳酸アシドーシス、鉄芽球性貧血を特徴とする。*Pseudouridyate synthase 1 gene (PUS1)*の欠損により発症する。

6. 治療法

1)薬物療法
　　（ア）　ビタミン補充療法
　pyridoxine投与
　　XLSAでは半分以上の患者がpyridoxineの経口投与に反応する(50- 100mg/day)。表2にXLSAにおける遺伝子変異を示す。Pyridoxineに反応する変異は網掛けで示す。
　Thiamine投与
　　TRMAでビタミンB1 (25 – 75mg/day)の投与で反応を示す。

その他の疾患では特異的な薬物療法はない。

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(イ) 鉄キレート療法

特に輸血依存状態となった症例では、鉄過剰症によるヘモクロマトーシスのリスクが高く、フェリチン値、臓器障害の有無により、鉄キレート療法を行う。

2) 輸血療法

必要に応じて施行する。

3) 造血幹細胞移植

これまでに 3 例の報告がある(13)。いずれも造血能の回復を認めており、造血幹細胞移植は効果があると考えられる。ただし、ヘモクロマトーシスを伴っている症例が多く、その他の合併症が致命的となる可能性もあるため、前処置等に配慮が必要と考えられる。

7. 問題点・将来展望

遺伝性鉄芽球性貧血は、ビタミンB6等で治療が可能なことがあり、遺伝子の変異の同定が重要である。しかしながら、希少疾患であるため、症例の把握と、遺伝子解析のセンター化が必要である。さらに、今後は既知の遺伝子変異を有さない症例における変異遺伝子の同定が課題であり、同様の課題を持つ他の遺伝性造血不全グループと共同で新規遺伝子同定システムを構築する必要がある。

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