In the name of God

HEMATOPOIETIC STEM CELL TRANSPLANTATION (HSCT) IN CONGENITAL NEUTROPHIL DISORDERS.

LEKOCYTE ADHESION DEFICIENCY (LADS)

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Congenital neutrophil disorders

- The list of neutrophil disorders is varied and expanding rapidly with the increasing availability of **next generation gene sequencing.**
- The ability to establish a genetic diagnosis is improved.
- The challenge of linking genotype to phenotype arises, a challenge that is made difficult by the small numbers of patients diagnosed with each individual disease

- Shahrzad Bakhtiar. October 2019 | Volume 7 | Article 436

The Evidence for Allogeneic Hematopoietic Stem Cell Transplantation for Congenital Neutrophil Disorders: A Comprehensive Review by the Inborn Errors Working Party Group of the EBMT.
Shahrzad Bakhtiar. October 2019 | Volume 7 | Article 436.Germany

- Congenital neutrophil disorders as a category of primary immunodeficiency (PID) can be classified in many ways, but a key point of distinction is whether the disorder is

quantitative, or qualitative.

The 2017 International Union of Immunological Societies (IUIS) phenotypic Classification for Primary Immunodeficiencies divides neutrophil disorders into 4 broad categories:

- Congenital neutropenia associated with or without syndromic disease
- Functional neutrophil defects with or without syndromic disease

2017 IUIS Phenotypic Classification
Congenital neutrophil disorders

- **Group 1: Syndrome-Associated Neutropenia**
- **Group 2: Neutropenia Without Syndromic Disease**
- **Group 3: Phagocyte Function With Syndromic Disease**
- **Group 4: Phagocyte Dysfunction Without Syndromic Disease:**

Shahrzad Bakhtiar.October 2019 | Volume 7 | Article 436

Presentation in Congenital neutrophil disorders

- Affected patients can present with variable symptoms including recurrent infections, failure to thrive, and overwhelming septic episodes leading to high morbidity and mortality.
- Early and severe respiratory infections (e.g., Burkholderia cepacia, Aspergillus spp.), visceral abscesses, cellulitis, lymphadenitis, and granulomatous lesions are observed in patients suffering from CGD
- Severe autoinflammatory complications underlining the role of **neutrophils in Autoinflammatory processes** beyond microbial defense
- In many of these diseases there is a recognized risk of progression to **syndrome (MDS) &(AML)**

Treatment Plan Congenital neutrophil disorders

Treatment for neutrophil disorders classically comprises:

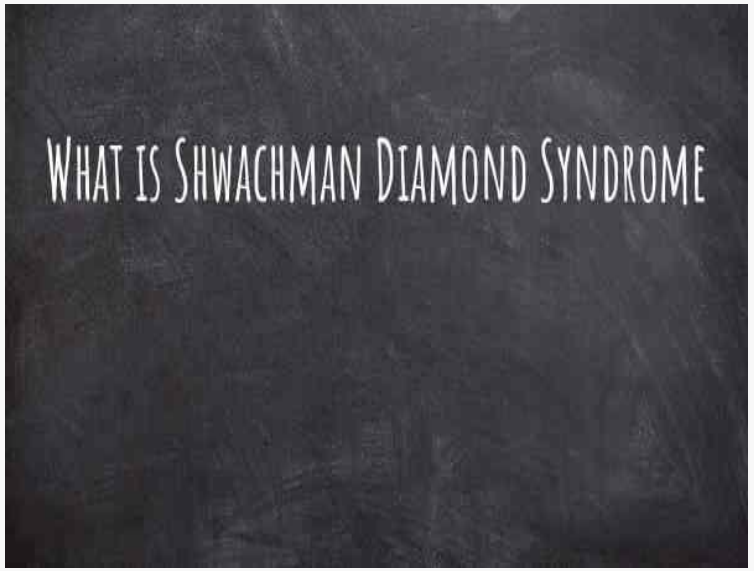
- Anti-microbial therapy
- Granulocyte-colony stimulating factor (G-CSF)
- **Allogeneic hematopoietic stem cell transplantation (alloHSCT)**
- Gene Therapy ???

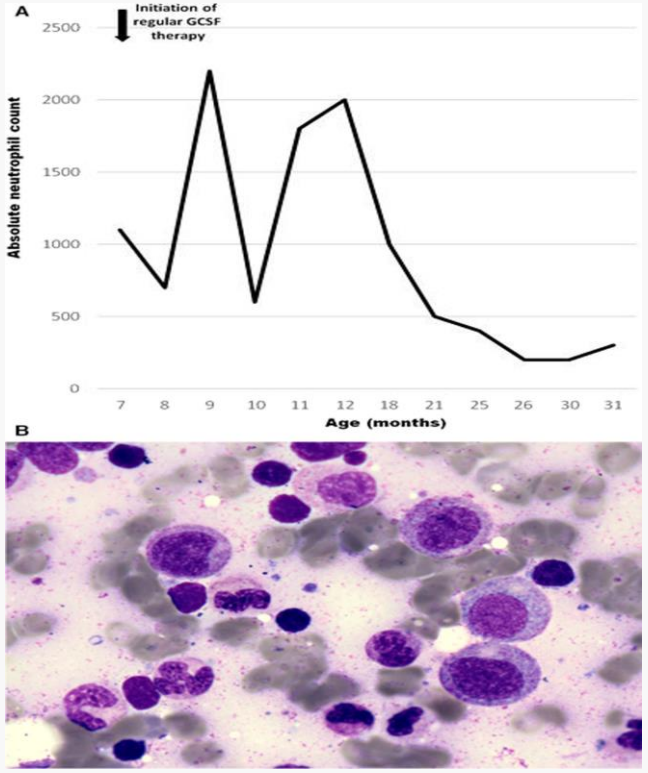
Treatment Plan/ Congenital neutrophil disorders

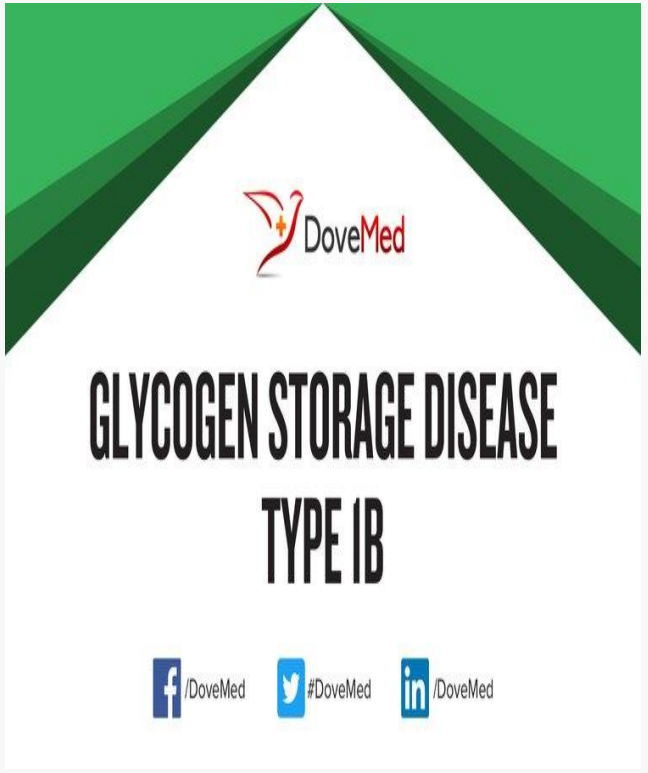
- There is much we still do not understand about the pathomechanism of these diseases, and this makes prognostication and treatment decisions difficult.
- Serious infectious complications and neutropenia are life threatening but can be treated with anti-microbial therapy and G-CSF, respectively, however inflammatory complications of these diseases are likely underappreciated and undertreated
- **Also, the decision to undertake alloHSCT, with all its inheritent risks and potential complications, is made difficult by the lack of published data for most diseases.**

Group 1: Syndrome-Associated Neutropenia

Schwachman Diamond syndrome and deficiency of VPS45 protein & Glycogen Storage Disease type 1b







2017 IUIS Phenotypic Classification
Group 1: Syndrome-Associated Neutropenia

- This group of diseases is dominated by conditions defined by:
- Bone marrow failure **(BMF)**
- Predisposition to myelodysplastic syndrome **(MDS)** & acute myeloid leukemia **(AML)**
- As well as additional **non-hematological manifestations** such as **neurodevelopmental delay , skeletal abnormality**

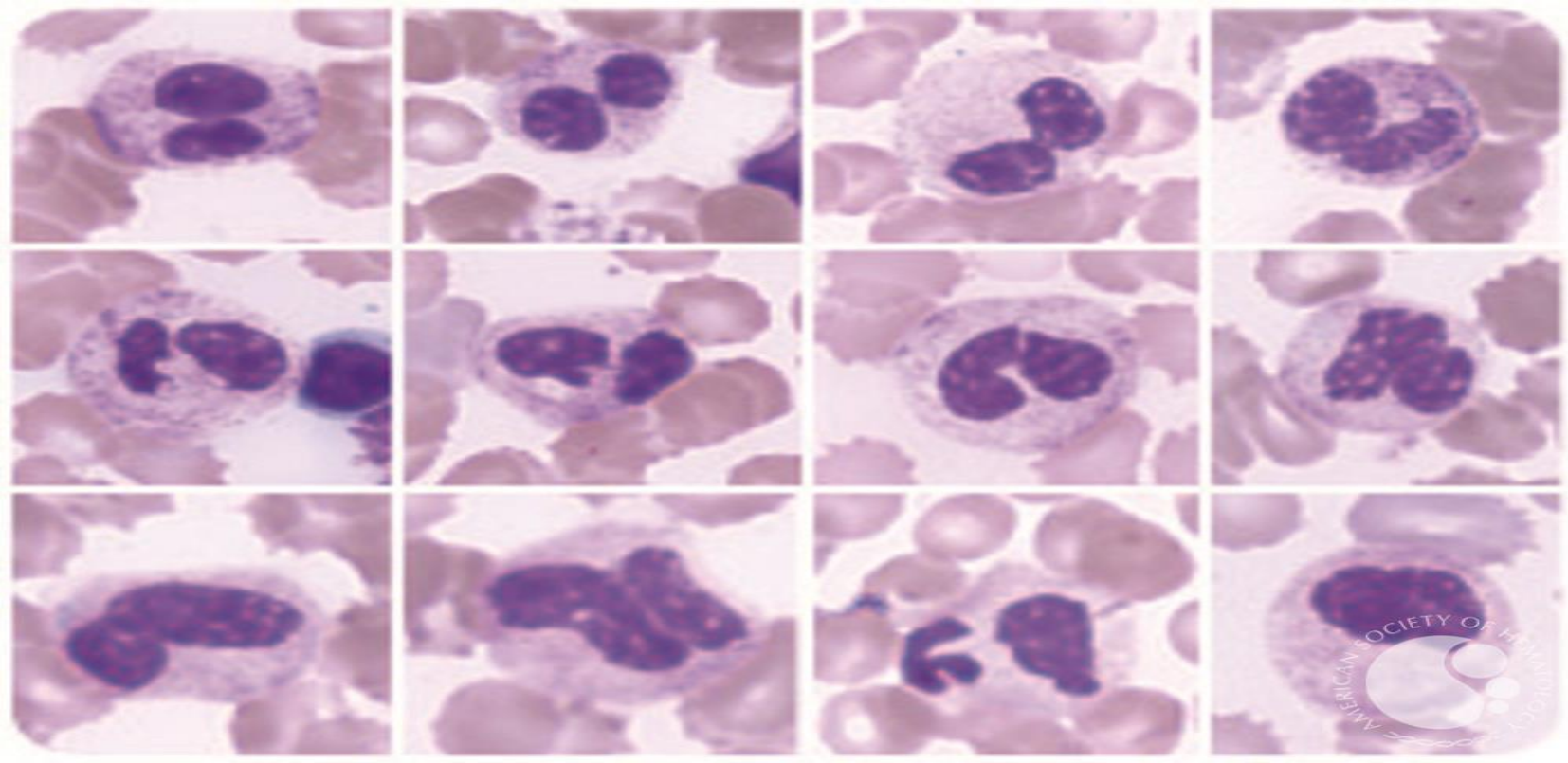
Schwachman Diamond Syndrome
Group 1: Syndrome-Associated Neutropenia

- **Rare AR disorder first described in 1964**
- **Impaired RNA metabolism and ribosomal function. *Ribosomopathy***
- **Mutations in the SBDS gene/Shwachman–Bodian–Diamond syndrome at ch7q11.21. (2002)**
- Pancreatic exocrine insufficiency, immune deficiency, **BM failure, persistent or intermittent neutropenia**,and skeletal abnormalities.somatic development and short stature
- **1/3 of patients develop major hematological complications**

SDS: **Neutropenia is the most common hematologic abnormality.**

BM is often hypocellular and usually contains mild **dysplastic changes in the erythroid, myeloid, and megakaryocytic series, even in the absence of clonal cytogenetic anomalies.**

Jean-François Lesesve.2017



Kasiani Myers. Biol Blood Marrow Transplant. 2020 Aug; 26(8): 1446–1451.USA . **HSCT Group 1: Syndrome-Associated .in Shwachman Diamond Syndrome Neutropenia**

- **52 P/ (SDS) /HSCT/ 2000 - 2017.**
- The median age HSCT 11 years& Median duration of follow-up was 60 mo
- **Indication for HSCT was : BMF; cytopenia or aplastic anemia in 39 p & (MDS)/ (AML) in 13 p**
- The donor type was an HLA-matched sibling for 18 patients, an HLA-matched or mismatched relative for 6 patients, and an HLA-matched or mismatched unrelated donor for 28 patients.
- **Condtioning in BMF : MAC in 13 patients and RIC/ 26.**
- At the time of this report, **29 /39 P BMF were alive**, **5-year overall survival was 72%**
- **Graft failure and GVHD : predominant causes of death.**
- **Only 2 of 13 patients in group of MDS/ AML :alive (15%), Relapse the predominant cause of death.**

Group 1: Syndrome-Associated Neutropenia

Deficiency of VPS 45 Protein

- Impaired trafficking of endosomes and lysosomes, **impaired degranulation, release of inflammatory mediators, and neutrophil migration**
- **Neutropenia, thrombasthenia, myelofibrosis, and progressive bone marrow failure.**
- Very early in life (before 1 year of age)
- NO response to G-CSF therapy.
- HSCT; in severe neutropenia unresponsive to G-CSF and for recurrent, severe infections

Volume 121, Issue 25, 20 June 2013, Pages 5078-5087

Group 1: Syndrome-Associated Neutropenia

GSD type Ib

- Case reports are available, such as in the case of successful transplant of Glycogen Storage Disease type 1b using reduced intensity conditioning

2017 IUIS Phenotypic Classification
Congenital neutrophil disorders

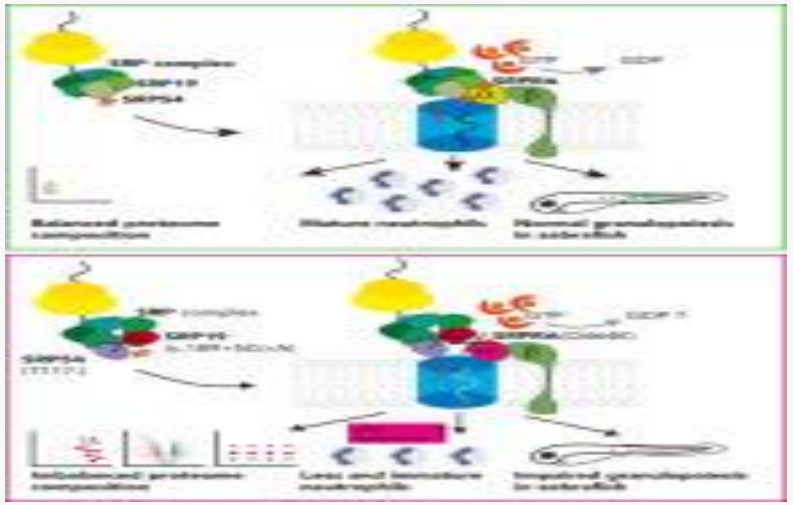
- **Group 2: Neutropenia Without Syndromic Disease, mutations in:**

- ELANE- AD, NO MDS / AML, ch (19p13.3), ALLOHSCT
- HAX1 – AR, MDS/AML , ch (1p22.1) , ALLOHSCT
- WAS (xP11.23 ,GOF –X linked) , some times MDS, NO HSCT
- GFI1- mild to moderate neutropenia, and there are no reports on alloHSCT
- CSF3R: tendency to MDS/ AML
- SRP54 genes

Genetic defects in **SRP19 and SRPRA** cause **severe congenital neutropenia**

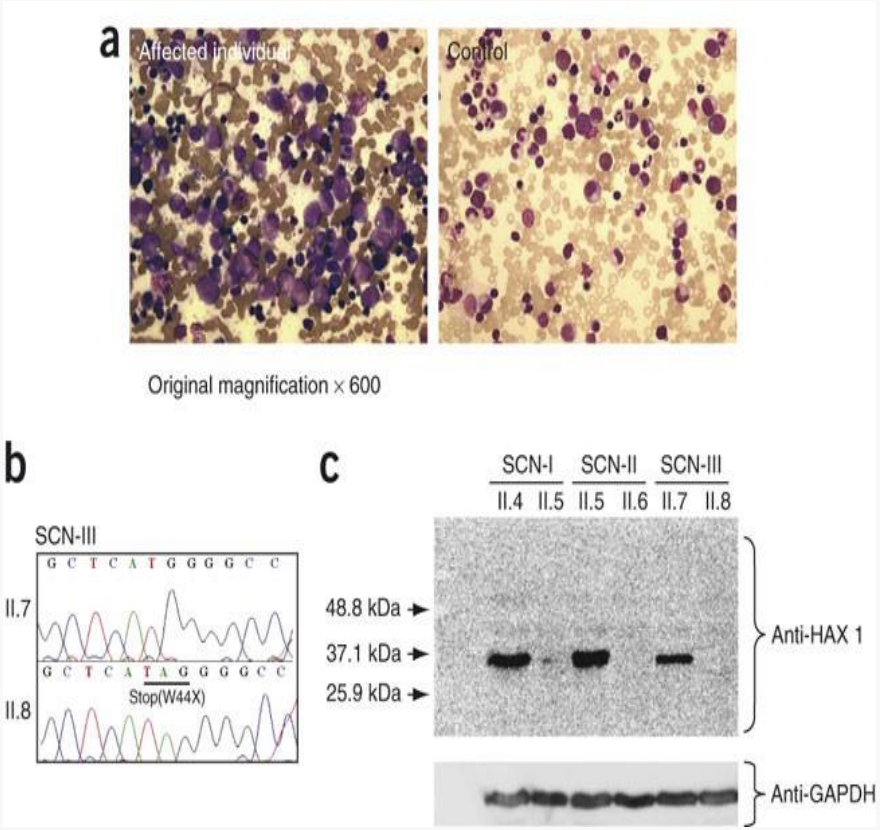
Blood . 9FEBRUARY **2023** | VOLUME 141, NUMBER 6 645.Germany

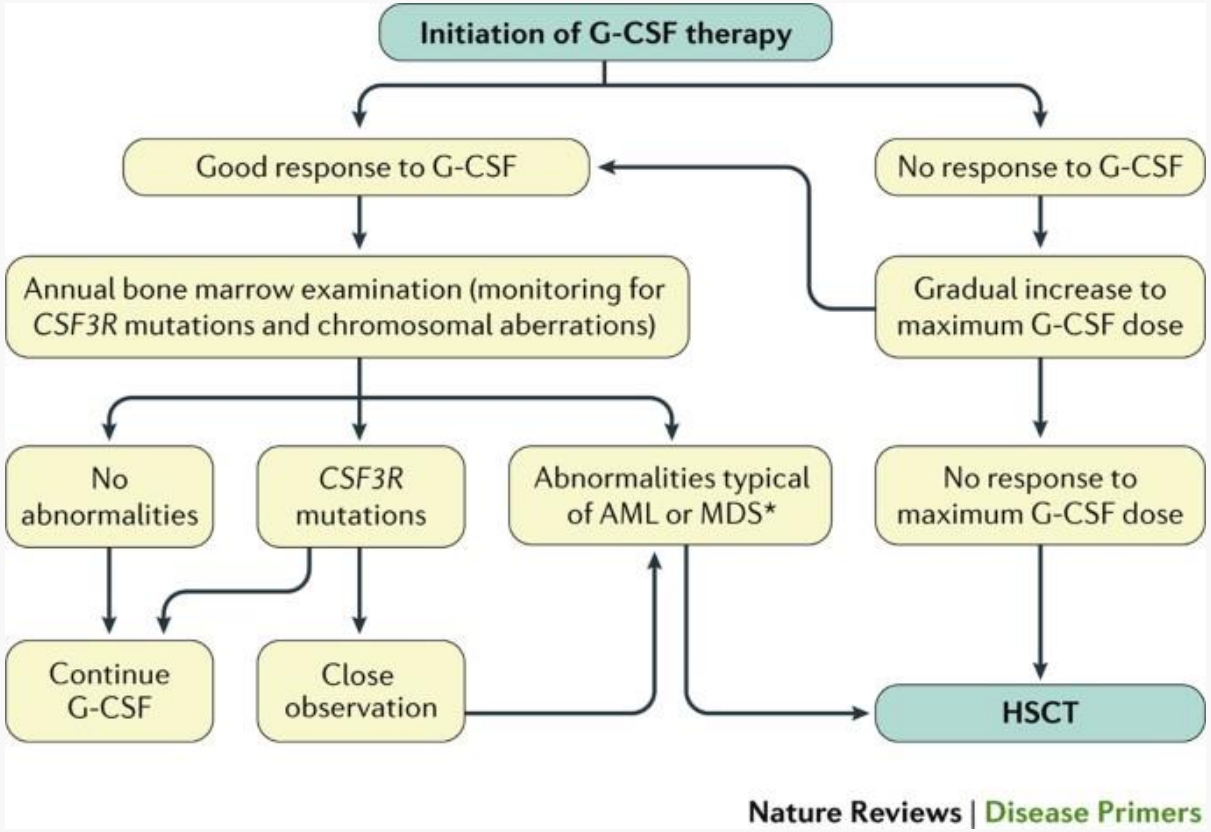
- **SRP19 & SRPRA:**
- **Novel genes affected in congenital neutropenia and essential for granule protein processing**



Group 2:**Severe Congenital Neutropenia**

Elane , Hax1, G6pc3





Francesca Fioredda.*Blood* (2017) 130 (Suppl_1) : 2267.

Severe Congenital Neutropenia with ELANE G-CSF *Vs* HSCT in Severe Mutation

- ❑162 SCN *ELANE* mutated patients
- ❑Established indications for (HSCT) in SCN include:
  - Transformation to acute leukemia (AL) and/or (MDS)
  - G-CSF resistance (need for >20mcg/kg/day).
  - Poor response to G-CSF (10-20 mcg/kg/d) and, (a high incidence of infectious events)

➢ Indication is less clear in cases requiring a G-CSF doses up to 10 mcg/kg but with good control of infections.

Francesca Fioredda. Blood (2015) 126 (16): 1885–1892

- 2015, the EBMT & SCETIDE / largest retrospective cohort / severe congenital neutropenia / Elane , Hax1, G6pc3 Allo HSCT:
- Median age at HSCT was 4.7 years (range, 0.23-43.1).
- Study 136 p- ; OS 3 y 82%
- TRM- Transplant related mortality ; 17%.
- Better outcomes :HSCT < 10 Y of age & Before the development of MDS/AML.
- HSCT : patients with severe infections or unresponsiveness to G-CSF or requiring high doses of G-CSF (over 8 mcg/kg/day to maintain an absolute neutrophil count over 0.5 × 109/L).
- Both MAC and RIC conditioning was effective.

2017 IUIS Phenotypic Classification.Congenital neutrophil disorders
Shahrzad Bakhtiar. October 2019 | Volume 7 | Article 436.Germany

Group 3: Phagocyte Function With Syndromic Disease:

- Defective neutrophil function with **normal or elevated neutrophil numbers** that is part of a broader syndrome:

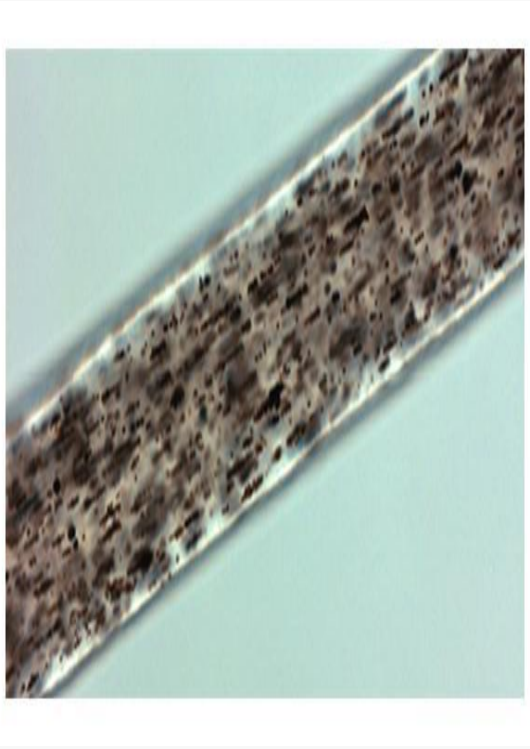
- **Leucocyte adhesion deficiencies I(AR), II(AR),III(AR), IV(AR)**
- **CEDIAK-HIGASHI SYNDROME**

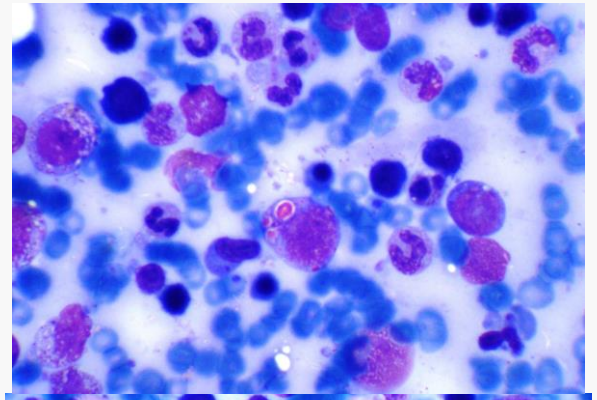
- **Localized juvenile periodontis (AR). NO HSCT**
- **ß-ACTIN: (AD), No HSCT**

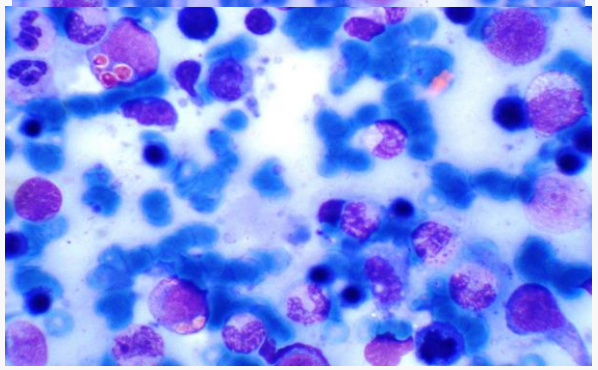
A 8 y old girl : MRD , BM , RIC -ALLO HSCT ; 4 y old age.

CEDIAK-HIGASHI SYNDROME (rare AR disorder):Lysosomal trafficking regulator gene (LYST 1)

Variable degrees of oculocutaneous albinism, recurrent pyogenic infections, a tendency for mild bleeding, and late neurologic dysfunction. **The 'accelerated phase' of CHS, (HLH) .**

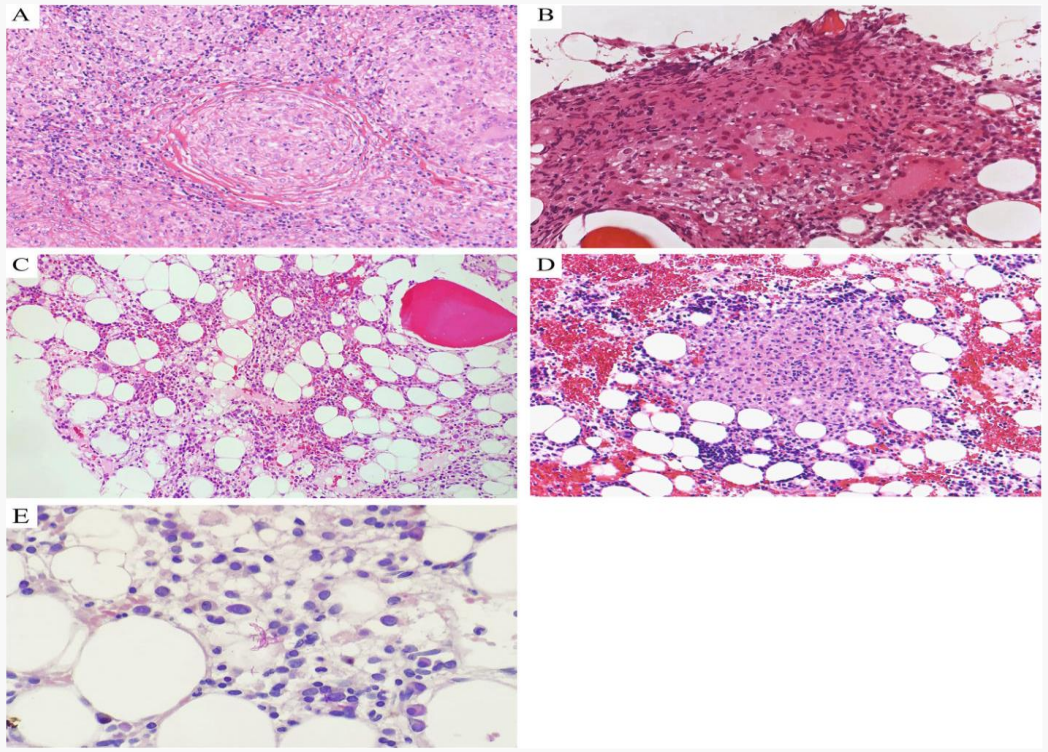






Group 4 : Phagocyte Dysfunction Without Syndromic Disease

CGD & MONO-MAC (Gata2 mutation)



***Blood* (2020) 136 (10): 1201–1211.**
atopoietic cell transplantation in chronic granulomatous disease: a study of 712 children and adults

- **EBMTstudy:** Retrospective multicenter/CGD-p /HSCT 1993 -2018.
- 635/712 children (aged <18 years) and 77 adults.
- Median age at transplantation was 7 years (**range, 0.1-48.6).**
- (OS) & (EFS) / 3 Y: 85.7% & 75.8%, respectively.
- **Older patients and Recipients of 1-antigen-mismatched grafts** had a **less favorable outcome.**
- **HSCT : strongly considered at a younger age and particularly in the presence of a well-matched donor.**

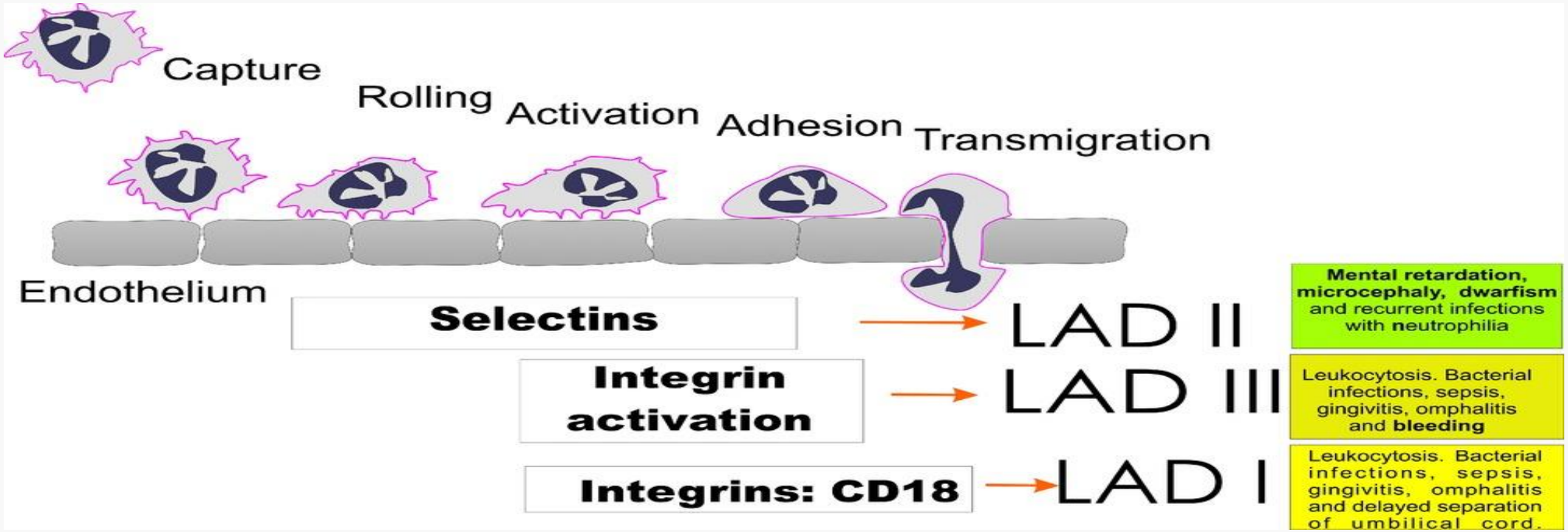
2017 IUIS Phenotypic Classification

Congenital neutrophil disorders

- **Mono-MAC ; Infections,Cytopenia (including monocytopenia), Predisposition to mycobacteria, Lymphedema,Pulmonary alveolar proteinosis,Deafness**
- **GATA2 -3q21.3, AD**
- **No response to GCSF**
- **Risk of MDS/ AML**
- **Treat : Allo-HSCT**

Group 3: Phagocyte Function With Syndromic Disease

Leukocyte adhesion deficiencies (LADs) ; AR PID characterized by a blockage in the process of leukocyte emigration to sites of inflammation



Leukocyte adhesion deficiency (LAD): Genes & Diseases (2020) 7, 107e114.

- Leukocyte adhesion deficiency (LAD) / as a group **of rare PID.**
- **Initial cases of LAD were described in 1970s in 6 infants from 2 families who had delayed separation of cord along with severe recurrent bacterial infections**

LAD Classification.

Blood advances. Shahrzad Bakhtiar. 12 JANUARY 2021 x VOLUME 5, NUMBER 1

- **LAD-I (AR)with mutations in ITGB2,ch 12q13.13, causing dysfunctional b2 integrins (CD11/CD18)**
- **LAD-II (AR) , ch11p11.2, , mutations in SLC35C1, a gene that encodes a GDP fucose transporter of the Golgi system, resulting in syndromic features, mental retardation, and typical Bombay blood type.**
- **LAD-III (LAD-I variant) (AR) is caused by mutations in, ch11q13.1 ,FERMT3 resulting in an activation defect of all b-integrins on immune cells and thrombocytes.**
  - **(LAD-IV) : AR , CFTR gene , ch7q31.2, Clinical features of cystic fibrosis**

Leukocyte adhesion defect: Where do we stand circa 2019 ? J humki Das.

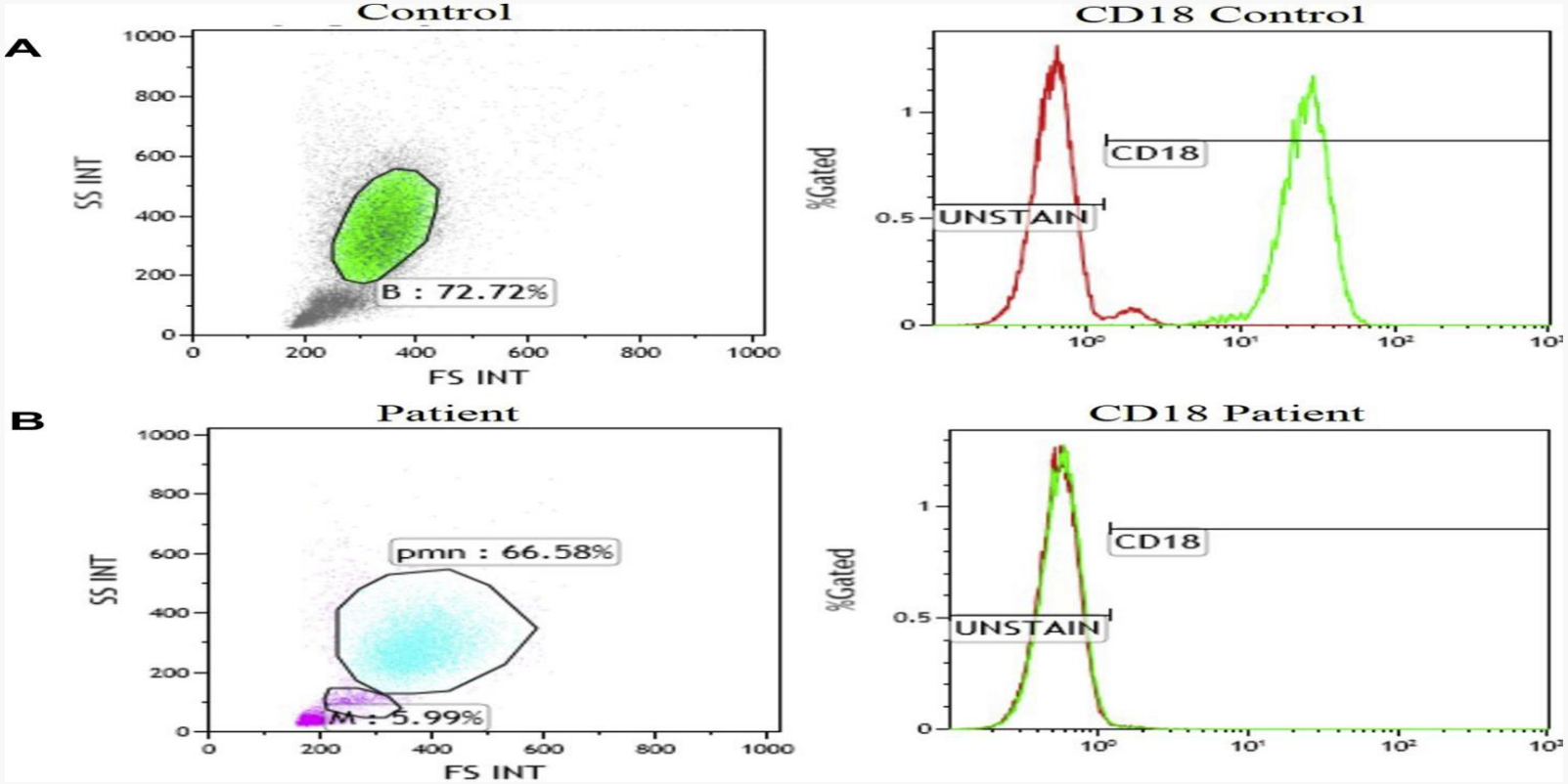
.Genes & Diseases (2020) 7, 107e114.

Integrins in LAD I

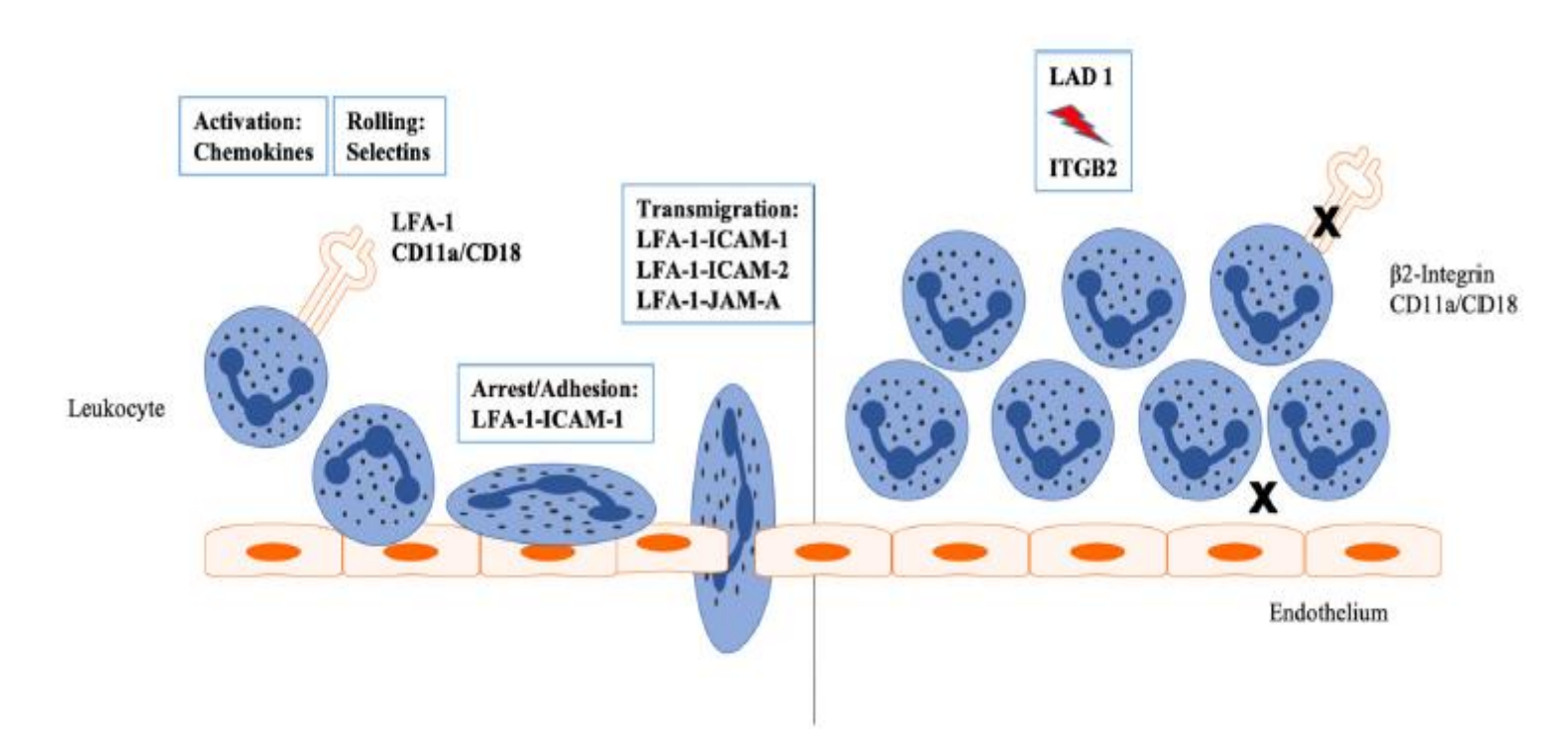
- **Defects in membrane expression of leukocyte adhesion glycoproteins of integrin superfamily**
- **The function of b2 integrin, CD18 is lost in LAD I.**
- **Integrins** are noncovalently associated, **heterodimeric cell surface receptors**, consisting of one a subunit (CD11a, CD11b, or CD11c) and a common b chain (CD18), which helps in surface expression of the CD11 chains.
- **These proteins facilitate leukocyte adhesion to endothelium.**
- **Patients with LAD I have defective polymorphonuclear cell adherence, leading to defective chemotaxis and trafficking, as well as low natural killer (NK) and cytotoxic T-lymphocyte (CTL) activity**

Flow-CytometricImmunophenotyping for CD18 Leukocytes on peripheral blood: neutrophil gated using SSC/FSC & CD18 expression is measured in (A) healthy control against (B) LAD patient.

CD18 expression is normal in control while its absent in the patient.



The leukocyte adhesion cascade. Inflammation leads to an activation of the by endogenous and exogenous stimuli.
Int. J. Mol. Sci. 2022



LAD1- Pediatr 2020;9(1):34-42

- Incidence of 1/1,000,000
- **Ulcerative deep tissue infections, leukocytosis with impaired pus formation, delayed detachment of the umbilical cord, and omphalitis**
- **Severity of the infectious complications correlates with the level of <u>CD11/CD18 expression on leukocytes .</u>**
- Patients with CD18-expressing leukocytes at **levels lower than 2% /severe LAD-I.**
- In severe LAD-I do not undergo HSCT, **death is 75% by the age of 2 years.**
- Patients with CD18-expressing leukocytes at **levels of 2–30% moderate LAD-I**
- Most patients with moderate LAD-I survive childhood, but often experience recurrent infections and usually have poor long-term outcomes.

Leukocyte Adhesion Deficiency III: Report of Two Siblings

Pediatrics and Neonatology (2017) 58, 99e100.Turkey. Deniz Aygun

- **LAD III is an AR disorder & an immunodeficiency syndrome**
- **Less than 40 LAD-III cases have been reported.**
- **Abnormal integrin activation**
- **Impair integrin function, that results from deficiency of b1, b2, and b3 integrin activation by physiological stimuli in platelets and leukocytes**
- **Bleeding tendency due to dysfunctional platelet aggregation, as in Glanzmann's thrombasthenia.** Melena, and Petechial lesions of the mucosa, although life-threatening cerebral hemorrhage may also occur.
- **Osteopetrosis-like bone lesions** attributed **to defective osteoclast**

LADS Type 1 & 3

- **LAD Type I, ITGB2 , 12q13.13, AR**
- Severe leukocytosis,Severe recurrent bacterial infections, impaired pus formation,Delayed wound healing, Delayed umbilical cord detachment
- G-SCF responsive: No
- Risk of progression to MDS/AML: No
- **Evidence of successful HSCT: Yes.**

- **LAD type III,FERMT3, 11q13.1 ,AR**
- Severe leukocytosis ,Recurrent bacterial infection,Bleeding tendency.
- GCSF responsive : No
- Risk of progression to MDS/AML: No
- **Evidence of successful HSCT: Yes.**

TREATMENT OF LAD I & LAD III

- **LAD III: Recombinant factor VIIa might be beneficial for preventing bleeding in LAD-III**

- **Gene therapy :may provide a possible alternative to allo-HSCT and is currently being explored in a phase 1/2 study with a retroviral based gene therapy (RP-L201) for LAD-I patients (Rocket Pharma).**

Shahrzad Bakhtiar. October 2019 | Volume 7 | Article 436.Germany

Haematologica 2018; 103:e264(France)

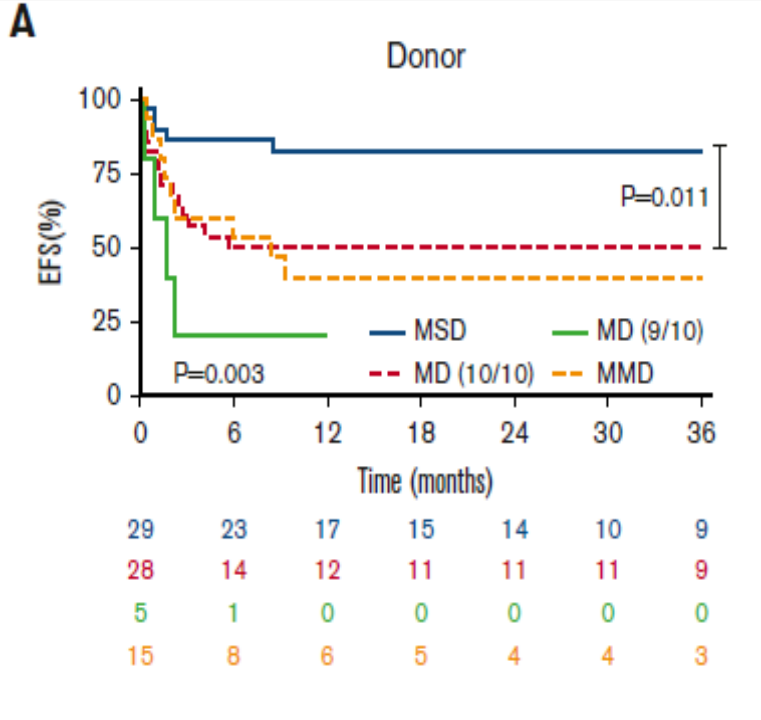
12 JANUARY 2021 x VOLUME 5, NUMBER 1. ALLO HSCT in leukocyte adhesion deficiency type I and III. Shahrzad Bakhtiar

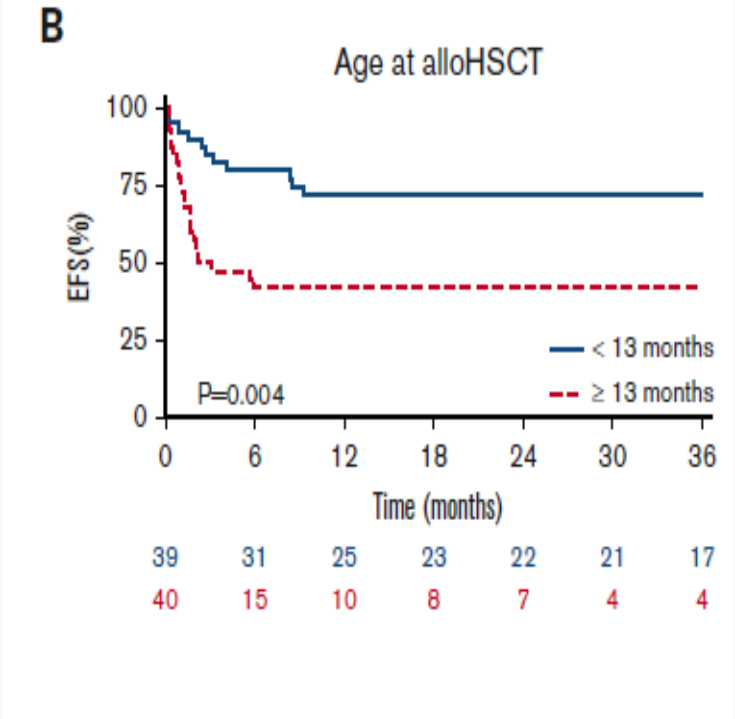
- Retrospective multicenter **EBMT study**
- Data from **84 LAD patients from 33 centers**, / allo-HSCT **2007 - 2017.**
- **3-year overall survival estimate : 83% (**74-92) for the **entire Cohort**
- **OS 3 - Y: 84%** (75-94) & **75% (50-100) for LAD-I and LAD-III**, respectively.
- **3 Y(EFS) : 56% (46-69) for the entire cohort;** 58% (46-72) and 56% (23-88) for LAD-I and LAD-III
- Incidences of **Graft failure (GF) at 3 years of 17%** (9%-26%)
- Grade II to IV (aGVHD) at 100 days of 24% (15%-34%).

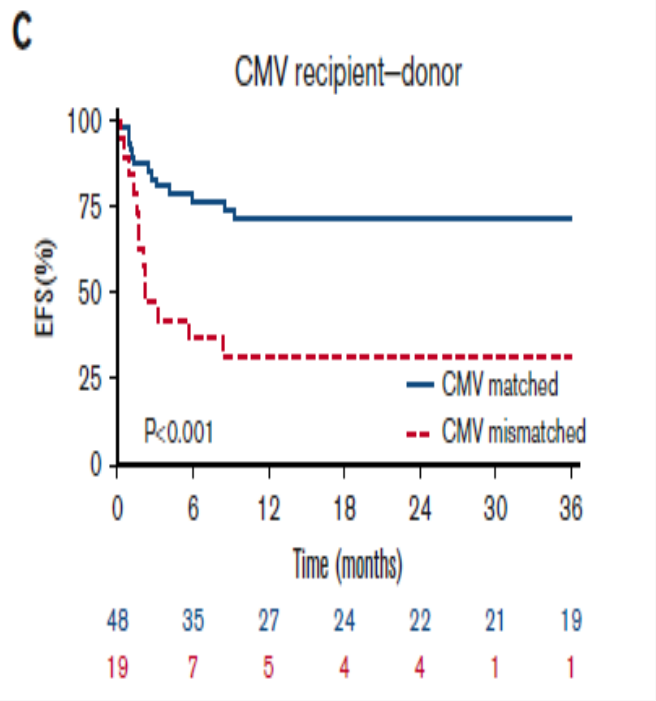
12 JANUARY 2021 x VOLUME 5, NUMBER 1. ALLO HSCT in Shahrzad Bakhtiar.leukocyte adhesion deficiency type I and III

- **Risk factors in HSCT : Associated with severe acute GVHD and inferior EFS include:**
- **Patients' age at transplant >13 months**
- **Transplantation from a non-sibling donor**
- **Any serological cytomegalovirus mismatch in donor-recipient pairs were significantly**
- **The choice of busulfan- or treosulfan-based conditioning, type of GVHD prophylaxis, and serotherapy did not impact overall survival, EFS, or aGVHD.**
-

EFS. 12 JANUARY 2021 x VOLUME 5, NUMBER 1







Successful umbilical cord blood transplantation in children with leukocyte adhesion deficiency type I . IXiaowen Qian. Transl Pediatr 2020;9(1):34-42

- **5 children / LAD-I /UCBT 2016 -2018** . (2 boys & 3 girls)
- Median age at UCBT was **9 months (range, 8 to 32 mo**).
- **MAC** /each patient and included **busulfan, fludarabine, & cyclophosphamide**.
- HLA matching : 8/10 to 10/10.
- The median dose of total nucleated cells (TNC) infused was **10.2×107 /kg** (range, 4.5×107 to 20.6×107 /kg)
- Median dose of **CD34+ cells was 3.2×10 5 /kg** (range, 1.9×105 to 5.7×105 /kg).
- Median time of neutrophil engraftment ; 20 days (range, 13 to 28 days).

Successful umbilical cord blood transplantation in children with leukocyte adhesion deficiency type I . IXiaowen Qian. Transl Pediatr 2020;9(1):34-42

- The median time of platelet engraftment was 36 days (range, 32 to 56 days).
- **All patients received complete donor chimerism (CDC**).
- 4/5 p : grade II–IV acute GVHD.
- The median follow-up time HSCT: 19 months (range, 8 to 38 months).
- **4 :complete clinical remission.**
- **1 Death : Bronchiolitis obliterans 8 months after UCBT.**
- **Conclusions: UCBT is an effective treatment method for LAD-I patients.**

Case report: HLA-haploidentical HSCT with posttransplant cyclophosphamide in a patient with LAD type I .Motoi Yamashita. Frontiers in Immunology. 2022

- **A case of LAD-I successfully treated with HLA-haploidentical HSCT with PT-CY** Donor :mother , The Source : PB
- Anti-HLA antibodies in the patient :Negative
- RIC ; **Alemtuzumab** 0.8 mg/k g IV (0.16 mg/kg for 5 days; days –14 to –10)+ **Fludarabine** 180 mg/m2 IV (45 mg/m2 for 4 days; days –9 to –6), (AUC)-targeted **busulfan** IV (65 mg*h/L; days –5 to –2)
- **GVHD-P :PT-CY 50 mg /kg IV on days +3 and +4, then Tacrolimus IV & po (MMF) day +5. Filgrastim 300 mg/m2 IV ;on days +6 - +12.**

- **Engraftment :on day +13 HSCT** **Complete Chimerism**
- Complication ; mild GVHD, CMV reactivation and VOD/ SOS.

Understanding the Role of LFA-1 in LAD I :Moving towards Inflammation? Julia Fekadu Int. J. Mol. Sci. 2022, 23, 3578. Management of inflammatory symptoms

- **ROLE :LFA-1** (Lymphocyte function-associated antigen-1), **a heterodimeric integrin (CD11a/CD18) present on the surface of all leukocytes;**
- **A distinct hyperinflammatory component of this disease involving the interleukin-12 (IL-12)/IL-23 pathway**
- **Recently;**
- **Ustekinumab (a monoclonal antibody binding the common P40 unit of IL-12 and IL-23) ameliorated inflammatory symptoms in an adult LAD-I patient. Decreased IL-17 levels**

Leukocyte Adhesion Deficiency III: Report of Two Siblings.Pediatrics and Neonatology (2017) 58, 99e100.Turkey. Deniz Aygun. Turkey

- **2 Siblings, a 5-y girl & a 3-y boy .**
- **Delayed separation of the umbilical cord** , FTT, bleeding disorders, **several blood transfusions**, Rrecurrent pulmonary infections
- Marked leukocytosis, Nl platelet counts..
- **Expression CD18, CD11a, and CD11b integrins : Nl** NO **LAD I syndrome.**
- Molecular analysis ;: Homozygous substitution of FERMT3 gene
- HSCT from fully matched foreign donors
- **Death : older sibling due to severe bacteremia and GVHD during the 3th mo post HSCT**
- **HSCT successfully inother young sibling & alive.**

CONCLUSION

- Characterization of the molecular mechanisms of PID is important for confirming the diagnosis as well as guiding genetic consultation .
- **potential complications of HSCT in congenital neutrophil disorders should be considered**
- **However ,HSCT is the curative treatment** of some patients with congenital neutrophil disorders include ; LAD I & III, CGD
- **SO:**
- **Early diagnosis** & **performing HSCT** in **early infancy** using an **appropriate conditioning regimen** ; **improve the outcome -P**

THANK

YOU