haematologica
Journal of the Ferrata Storti Foundation

A novel *GATA1* variant linking germline and somatic myelodysplastic syndrome in two patients

by Nuria Revilla, Mireia Atance, Jose María Grasa, Alejandro García-Ortego, Carlos Soto, Socorro María Rodríguez-Pinilla, Teresa Arquero, Sara Perlado, Selene Ledesma, Esperanza Esquinas-Tarifa, Juana Serrano-López, Jose Luis López-Lorenzo, Diego Velasco-Rodríguez, Juan Manuel Alonso-Dominguez, Raquel Mata, Pilar Llamas-Sillero, Rocío N. Salgado and Ana Rio-Machin

Received: September 5, 2025.
Accepted: December 19, 2025.

Citation: Nuria Revilla, Mireia Atance, Jose María Grasa, Alejandro García-Ortego, Carlos Soto, Socorro María Rodríguez-Pinilla, Teresa Arquero, Sara Perlado, Selene Ledesma, Esperanza Esquinas-Tarifa, Juana Serrano-López, Jose Luis López-Lorenzo, Diego Velasco-Rodríguez, Juan Manuel Alonso-Dominguez, Raquel Mata, Pilar Llamas-Sillero, Rocío N. Salgado and Ana Rio-Machin. A novel GATA1 variant linking germline and somatic myelodysplastic syndrome in two patients.
Haematologica. 2025 Dec 24. doi: 10.3324/haematol.2025.289133 [Epub ahead of print]

A novel *GATA1* variant linking germline and somatic myelodysplastic syndrome in two patients

Nuria Revilla[1,2], Mireia Atance[1], Jose María Grasa[3], Alejandro García-Ortego[3], Carlos Soto[1], Socorro María Rodríguez-Pinilla[4], Teresa Arquero[1], Sara Perlado[1], Selene Ledesma[1], Esperanza Esquinas-Tarifa[2], Juana Serrano-López[2], Jose Luis López-Lorenzo[1], Diego Velasco-Rodríguez[1,2], Juan Manuel Alonso-Dominguez[1], Raquel Mata[1], Pilar Llamas-Sillero[1], Rocío N Salgado[1], Ana Rio-Machin[2,5]*

[1]Hematology Department, Hospital Universitario Fundacion Jimenez Diaz, Madrid, Spain

[2]Experimental Hematology Lab, Health Research Institute Fundacion Jimenez Diaz University Hospital, Universidad Autonoma de Madrid (IIS-FJD, UAM), Madrid, Spain

[3]Hematology Department, Hospital Quironsalud Zaragoza, Zaragoza, Spain

[4]Pathology Department, Health Research Institute Fundacion Jimenez Diaz University Hospital, Universidad Autonoma de Madrid (IIS-FJD, UAM), Madrid, Spain

[5]Centre for Haemato-Oncology, Barts Cancer Institute, Queen Mary University of London, United Kingdom

*Correspondence: Ana Rio-Machin, Experimental Hematology Lab, Instituto de Investigación Sanitaria-Fundación Jiménez Díaz University Hospital, Universidad Autónoma de Madrid (IIS-FJD, UAM), Madrid, Spain; email: ana.rmachin@iis-fjd.es

Data sharing statement: The datasets generated and analyzed during the current study are available from the corresponding author on reasonable request.

Disclosures

No conflicts of interest to disclose

Contributions

NR performed the experimental work and data analysis; NR, MA, AR-M, RNS conceived the study and contributed to its design; JMG, AGO, CS, SMR-P, TA, SP, SL, EE-T, JSL, JLL-L, DVR, JMA-D, RM, PLS contributed to diagnostics, acquisition of clinical data, and interpretation of findings; NR and AR-M wrote the manuscript. All authors reviewed and approved the final version of the manuscript.

Funding

This study was predominantly funded by the European Hematology Association through the AR-M´s EHA Junior Research Grant (EHA RG71) and the Miguel Servet Fellowship awarded to AR-M (AES 2023. Carlos III Health Institute. Spanish Ministry of Science, Innovation and Universities).

GATA1 is an X-linked gene encoding a zinc finger (ZF) transcription factor essential for erythroid and megakaryocytic differentiation.[1] Germline mutations in *GATA1* underlie a spectrum of non-malignant hematopoietic disorders, including dyserythropoietic anemia, macrothrombocytopenia, and Diamond-Blackfan anemia (DBA).[1,2] In contrast, somatic *GATA1* mutations are primarily associated with myeloid neoplasms in the context of trisomy 21 (T21), particularly transient abnormal myelopoiesis (TAM) and acute megakaryblastic leukemia (AMKL) in individuals with Down syndrome (DS).[3]

The GATA1 protein contains three functional domains encoded by six exons: (i) an N-terminal transactivation domain (N-TAD, exons 2-3) that drives transcription, (ii) an N-terminal ZF domain (N-ZF, exon 4 and part of 5) mediating cofactor interactions; and (iii) a C-terminal ZF (C-ZF, exon 5 and part of 6) binding consensus DNA motifs to regulate lineage-specific transcription.[1,2]

Pathogenic variants most commonly affect either the N-ZF domain (typically missense) or exon 2 (usually truncating variants). Variants in exon 2 result in the production of the shortened isoform GATA1s, which lacks the N-TAD and are characteristic of Down syndrome–related TAM/AMKL. In contrast, N-ZF missense variants do not generate GATA1s but disrupt DNA or cofactor binding, usually leading to congenital anemia or thrombocytopenia in hemizygous males. Female carriers of N-ZF variants are typically asymptomatic or present mild cytopenias, likely due to random or moderately skewed X-inactivation [1–5]. On the other hand, germline variants in the C-t regions of GATA1, although rare, have been also described: missense variants ~~in~~ affecting the C-ZF have been linked to cytopenias in heterozygous females, particularly when X-inactivation favors the mutant allele[5], and variants located in the more distal C-terminal region have been associated to a distinct form of X-linked congenital hemolytic anemia[5,6]. While *GATA1* variants have primarily been linked to non-malignant hematologic disorders, previous studies have also reported their involvement in myelodysplastic syndromes (MDS) in male patients, both in the germline[7] and somatic[8] contexts. However, germline *GATA1* variants have not been associated with leukemic transformation outside T21[9,10], and to date, no cases of MDS have been reported in female carriers.

Here, we report a novel germline variant in exon 5 of the *GATA1* gene affecting the C-ZF domain (c.808C>T; p.R270W), identified in heterozygosity in two female members of the same family (Figure 1). One of these individuals (Figure 1, II.2), a 52-year-old woman, was initially referred in January 2018 for evaluation of mild thrombocytopenia (134×10^9/L platelets) and macrocytosis (MCV 111 fL) without anemia, in the context of compensated non-immune hemolysis. A PNH clone was excluded in a follow-up sample. She remained asymptomatic and had no relevant bleeding history over the following five years. However, her platelet counts progressively declined to 81×10^9/L, accompanied by the onset of mild anemia and asthenia. In October 2023, bone marrow (BM) aspirate analysis confirmed a diagnosis of myelodysplastic syndrome with excess blasts (MDS-EB) (6% blasts) and normal karyotype. Megakaryocytes displayed a highly distinctive morphology, characterized by reduced size, basophilic cytoplasm and signs of nuclear immaturity, including micromegakaryocyte forms (Figure 2A). A myeloid next-generation sequencing (NGS) panel identified a previously unreported variant of uncertain significance in exon 5 of *GATA1* (c.808C>T; p.R270W), with a variant allele frequency

(VAF) of 46%. Copy number variant (CNV) analysis showed no additional abnormalities. Flow cytometry revealed approximately 5% undifferentiated myeloid blasts with partial CD7 expression. Her disease has remained stable for 20 months under a "watch-and-wait" approach, with a low-risk IPSS-M score.

Following confirmation of the germline nature of the *GATA1* variant through analysis of cultured skin fibroblasts, segregation analysis was performed. One sister (Figure 1, II.4) was found to carry the same *GATA1* p.R270W variant and exhibited stable, isolated mild thrombocytopenia (110–120 × 10^9/L) over a 10-year period of routine monitoring, without associated anemia or macrocytosis. Interestingly, the family history included a maternal uncle who had died from a hematologic malignancy. Six additional relatives (Figure 1, II.3, II.6, II.7, III.1, III.2, III.3) showed no hematologic abnormalities and were confirmed wild-type (WT) for *GATA1*.

To investigate whether the *GATA1* mutation affects platelet function, we performed functional studies on primary samples from both carriers (II.2 and II.4) and one WT relative (III.2). Platelet function was evaluated through multiple complementary readouts: surface glycoprotein expression (GPIb/IX, GPIIb/IIIa, GPIa/IIa, GPVI), agonist-induced fibrinogen binding (PAC-1), and α- and δ-granule secretion (CD62P and CD63, respectively), all assessed by flow cytometry as previously described.[11] Expression levels of platelets glycoproteins (with the exception of GPVI), as well as of CD62P and CD63 were comparable between carriers, the WT relative and healthy controls. Both carriers showed a modest reduction in GPVI expression (≈20%) compared with WT relatives and controls (Figure 3A). More notably, agonist-induced PAC-1 binding in response to ADP and TRAP stimulation was markedly reduced in both carriers, with a more pronounced defect in the sister with MDS showing up to an 80% reduction upon TRAP stimulation (Figure 3D). This indicates impaired activation of the αIIbβ3 integrin upon agonist stimulation, potentially reducing the capacity of platelets to bind fibrinogen and form stable aggregates. Mutations in *GATA1* can impair megakaryocytic maturation and lead to reduced expression and activation of the αIIbβ3 integrin, encoded by *ITGA2B* and *ITGB3* genes. This consistent with evidence that *ITGA2B* is a direct transcriptional target of GATA1[12]. PAC-1 is a monoclonal antibody that specifically recognizes the activated conformation of αIIbβ3; therefore, the reduced PAC-1 binding observed in our carriers supports the presence of defective integrin activation and impaired platelet function, both hallmarks of *GATA1*-related dysfunction.

While platelet aggregation studies in response to collagen could not be directly assessed due to thrombocytopenia in our family, the reduced GPVI expression may further contribute to defective αIIbβ3 activation. Altogether, these functional defects are consistent with previous reports of C-ZF *GATA1* variants that compromise platelet function and contribute to bleeding tendency.[13] Moreover, this presentation of compensated hemolysis without anemia, later evolving to mild anemia, has also been reported in association with other *GATA1* variants [6]. As certain *GATA1* variants have been associated with thrombocytopenia and the rare Lutheran-null blood phenotype[14], we assessed Lutheran antigen expression in our family. Both carriers (II.2 and II.4) and WT relatives (II.3, II.6, II.7, III.1 and III.3) showed a Lu(a–), Lub(+) phenotype, indicating no alteration associated with our novel variant. Interestingly, variant classification tools yield variable interpretations of *GATA1* c.808C>T: *Varsome* (https://varsome.com/)

considers it *likely pathogenic* (applying PP3, PM1, PM2), while *Franklin* (https://franklin.genoox.com) classifies it as a variant of uncertain significance (VUS) based on PP3 and PM2 criteria. According to the American College of Medical Genetics and Genomics and the Association for Molecular Pathology guidelines, the incorporation of our functional data allows the application of the PS3 criterion (supportive functional evidence), which would support reclassification of this variant as *likely pathogenic*.

Interestingly, an unrelated male patient was incidentally found to harbor the same *GATA1* variant (p.R270W) with a VAF of 84.4%. However, the absence of this variant in cultured skin fibroblasts confirmed the somatic origin in this case.

This 65-year-old male patient was referred from primary care in February 2023 for evaluation of macrocytic anemia (hemoglobin 10.6 g/dL) and thrombocytopenia (platelets 88 $\times 10^9$/L). A review of prior laboratory results revealed these cytopenias were already present in December 2022, and earlier blood count from July 2021 showed isolated mild thrombocytopenia (hemoglobin 13.1 g/dL, platelets 110 $\times 10^9$/L). He was subsequently diagnosed with trilineage MDS characterized by dysgranulopoiesis with neutrophil hyposegmentation, erythroid precursors with occasional mitoses and binucleation, 2% blast cells, and immature micromegakaryocytes with basophilic cytoplasm, morphologically resembling those observed in the female patient with germline *GATA1* mutation (Figure 2B). Cytogenetics showed a normal male karyotype (46,XY[20]), and NGS analysis revealed pathogenic somatic variants in *SRSF2* (c.284C>A, p.P95H, VAF 41.6%), *NF1* (c.4339del, p.Q1447Rfs*22, VAF 41.4%), and *TET2* (c.3965T>C, p.L1322P, VAF 33.0%), in addition to the *GATA1* variant. Unfortunately, no NGS panel was available at the time when isolated thrombocytopenia was first detected. The patient underwent allogeneic hematopoietic stem cell transplantation (HSCT) from an HLA-identical sister donor in August 2024. Fourteen months after HSCT, he remains in complete remission, with hemoglobin 13.9 g/dL, white blood cells 4.9 $\times 10^9$/L, and platelets 156 $\times 10^9$/L.

The phenotypic similarity between the male patient with the somatic *GATA1* variant and the female patient with the germline *GATA1* variant was striking (Figure 2A-B). In both, bone marrow displayed a predominance of small, immature, basophilic megakaryocytes. In the male, these aberrant megakaryocytes expressed CD34 (Figure 2C), consistent with findings in murine models harboring leukemogenic GATA1 mutations.[15] Indeed, *Gata1^low* mouse models demonstrate comparable abnormal megakaryocytic phenotype with impaired maturation, excessive proliferation, and progression to myelofibrosis.[16,17] These observations support the notion that GATA1 function is highly dosage-sensitive, and that disruption of its C-ZF DNA-binding domain—whether germline or somatic—can severely impair megakaryocyte differentiation and promote clonal evolution. In the female carrier with MDS-EB (Figure 1, II.2), no bone marrow biopsy was performed, consistent with clinical practice in isolated cytopenias, which may explain why this megakaryocytic phenotype has not been described before in germline carriers.

The only other reported germline *GATA1* variant associated with myeloid disease in females is p.T263M, affecting a residue near the variant described here (p.R270W).[5] Two proband sisters showed thrombocytopenia, neutrophilia, and anemia, with marrow resembling cellular-

phase myelofibrosis. As in our case, megakaryocytic hyperplasia with atypical morphology and varying degrees of fibrosis was observed. Both sisters also had additional lesions—monosomy 7, a deleterious JAK3 variant, and other potentially pathogenic *NOTCH1* and *ATM* variants—complicating genotype–phenotype interpretation.

Our male patient with somatic *GATA1* p.R270W also carried high-risk *SRSF2*, *NF1* and *TET2* mutations and underwent allogeneic transplantation. By contrast, the female germline carrier had no additional abnormalities. These findings support that *GATA1* variants disrupt megakaryopoiesis and predispose to clonal hematopoiesis, even without cooperating mutations. However, a family history of hematologic malignancy—particularly a maternal uncle who died of a hematologic malignancy—suggests incomplete penetrance or sex-related differences in expression.

All patients involved in the study provided written informed consent. The study was approved by the Ethics Committee of the Fundación Jiménez Díaz University Hospital (CEI code: PIC287-24_FJD).

In summary, we describe a novel germline *GATA1* C-ZF variant associated with thrombocytopenia and MDS in female patients. The abnormal megakaryocytic phenotype mirrored that of a second male patient with the same variant acquired somatically. Functional assays confirmed impaired platelet function in primary samples of carriers. This represents the first report of a *GATA1* variant linked to myeloid neoplasm in the absence of T21, thereby expanding the clinical spectrum of *GATA1*-related disorders. This case also underscores the pathogenic potential of *GATA1* mutations in females, challenges the traditional X-linked inheritance paradigm, and supports the inclusion of *GATA1* in diagnostic NGS panels for MDS, particularly in cases with megakaryocytic dysplasia or family history of hematological disease. Further studies are needed to clarify the molecular mechanisms underlying C-ZF domain *GATA1* variants and to define their long-term clinical impact.

References

1. Crispino JD, Horwitz MS. GATA factor mutations in hematologic disease. Blood. 2017;129(15):2103-2110.

2. Chang AN, Cantor AB, Fujiwara Y, et al. GATA-factor dependence of the multitype zinc-finger protein FOG-1 for its essential role in megakaryopoiesis. Proc Natl Acad Sci U S A. 2002;99(14):9237-9242.

3. Garnett C, Cruz Hernandez D, Vyas P. GATA1 and cooperating mutations in myeloid leukaemia of Down syndrome. IUBMB Life. 2020;72(1):119-130.

4. Hollanda LM, Lima CSP, Cunha AF, et al. An inherited mutation leading to production of only the short isoform of GATA-1 is associated with impaired erythropoiesis. Nat Genet. 2006;38(7):807-812.

5. Svidnicki MCCM, Filho MAF, Brandão MM, et al. New germline GATA1 variant in females with anemia and thrombocytopenia. Blood Cells Mol Dis. 2021;88:102545.

6. Ludwig LS, Lareau CA, Bao EL, et al. Congenital anemia reveals distinct targeting mechanisms for master transcription factor GATA1. Blood. 2022;139(16):2534-2546.

7. Kratz CP, Niemeyer CM, Karow A, Volz-Fleckenstein M, Schmitt-Gräff A, Strahm B. Congenital transfusion-dependent anemia and thrombocytopenia with myelodysplasia due to a recurrent GATA1(G208R) germline mutation. Leukemia. 2008;22(2):432-434.

8. Freson K, De Waele L, Thys C, et al. A Myelodysplastic Syndrome Associated with a Clonal GATA1 Mutation. Blood. 2008;112(11):1243-1243.

9. Hasle H, Kline RM, Kjeldsen E, et al. Germline GATA1s-generating mutations predispose to leukemia with acquired trisomy 21 and Down syndrome-like phenotype. Blood. 2022;139(21):3159-3165.

10. Camargo R, Sahoo SS, Córdoba JC, Magalhães IQ. Germline GATA1 exon 2 mutation associated with chronic cytopenia and a non-down syndrome transient abnormal myelopoiesis with clonal trisomy 21. Leukemia. 2022;36(9):2347-2350.

11. Palma-Barqueros V, Revilla N, Zaninetti C, et al. Src-related thrombocytopenia: a fine line between a megakaryocyte dysfunction and an immune-mediated disease. Blood Adv. 2022;6(17):5244-5255.

12. Songdej N, Rao AK. Hematopoietic transcription factor mutations: important players in inherited platelet defects. Blood. 2017;129(21):2873-2881.

13. Bastida JM, Malvestiti S, Boeckelmann D, et al. A Novel GATA1 Variant in the C-Terminal Zinc Finger Compared with the Platelet Phenotype of Patients with A Likely Pathogenic Variant in the N-Terminal Zinc Finger. Cells. 2022;11(20):3223.

14. Singleton BK, Roxby DJ, Stirling JW, et al. A novel GATA1 mutation (Stop414Arg) in a family with the rare X-linked blood group Lu(a-b-) phenotype and mild macrothrombocytic thrombocytopenia. Br J Haematol. 2013;161(1):139-142.

15. Arkoun B, Robert E, Boudia F, et al. Stepwise GATA1 and SMC3 mutations alter megakaryocyte differentiation in a Down syndrome leukemia model. J Clin Investigat. 2022;132(14):e156290.

16. Vyas P, Ault K, Jackson CW, Orkin SH, Shivdasani RA. Consequences of GATA-1 deficiency in megakaryocytes and platelets. Blood. 1999;93(9):2867-2875.

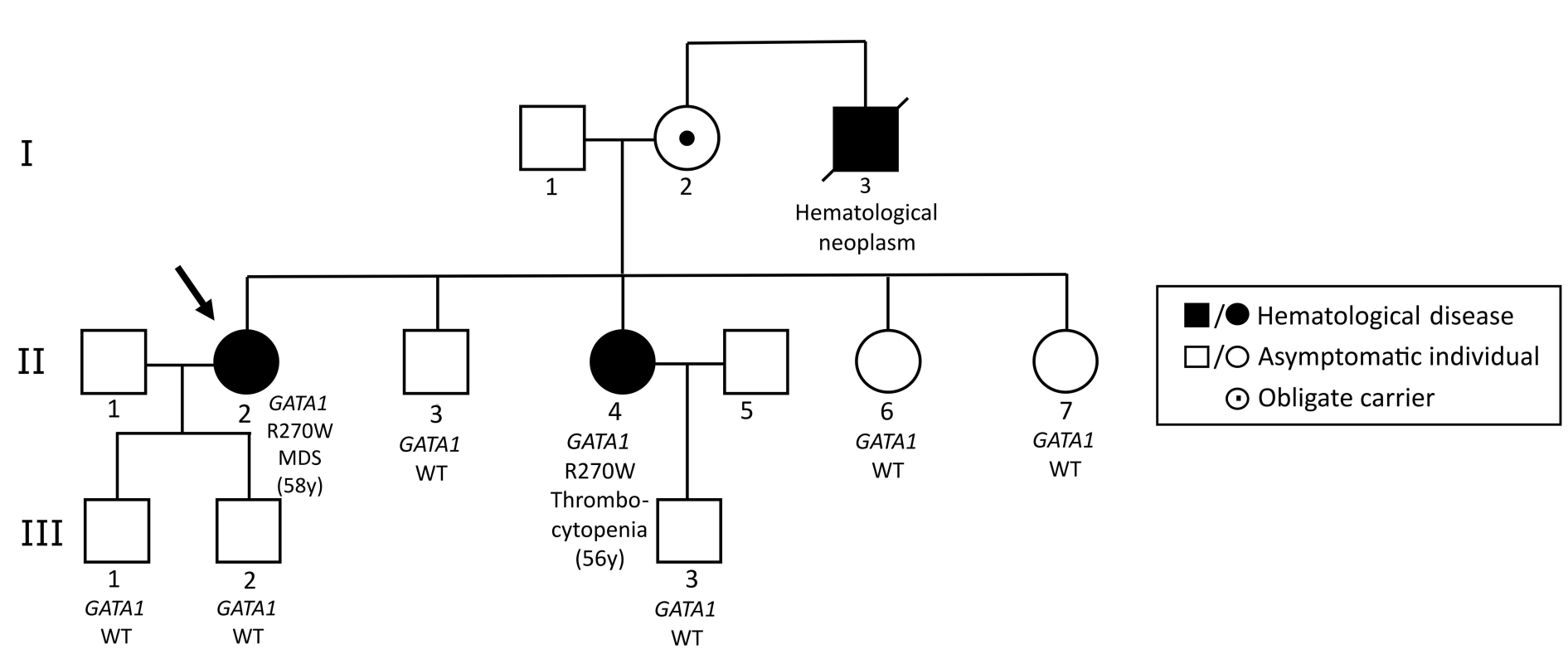
17. Centurione L, Di Baldassarre A, Zingariello M, et al. Increased and pathologic emperipolesis of neutrophils within megakaryocytes associated with marrow fibrosis in GATA-1low mice. Blood. 2004;104(12):3573-3580.

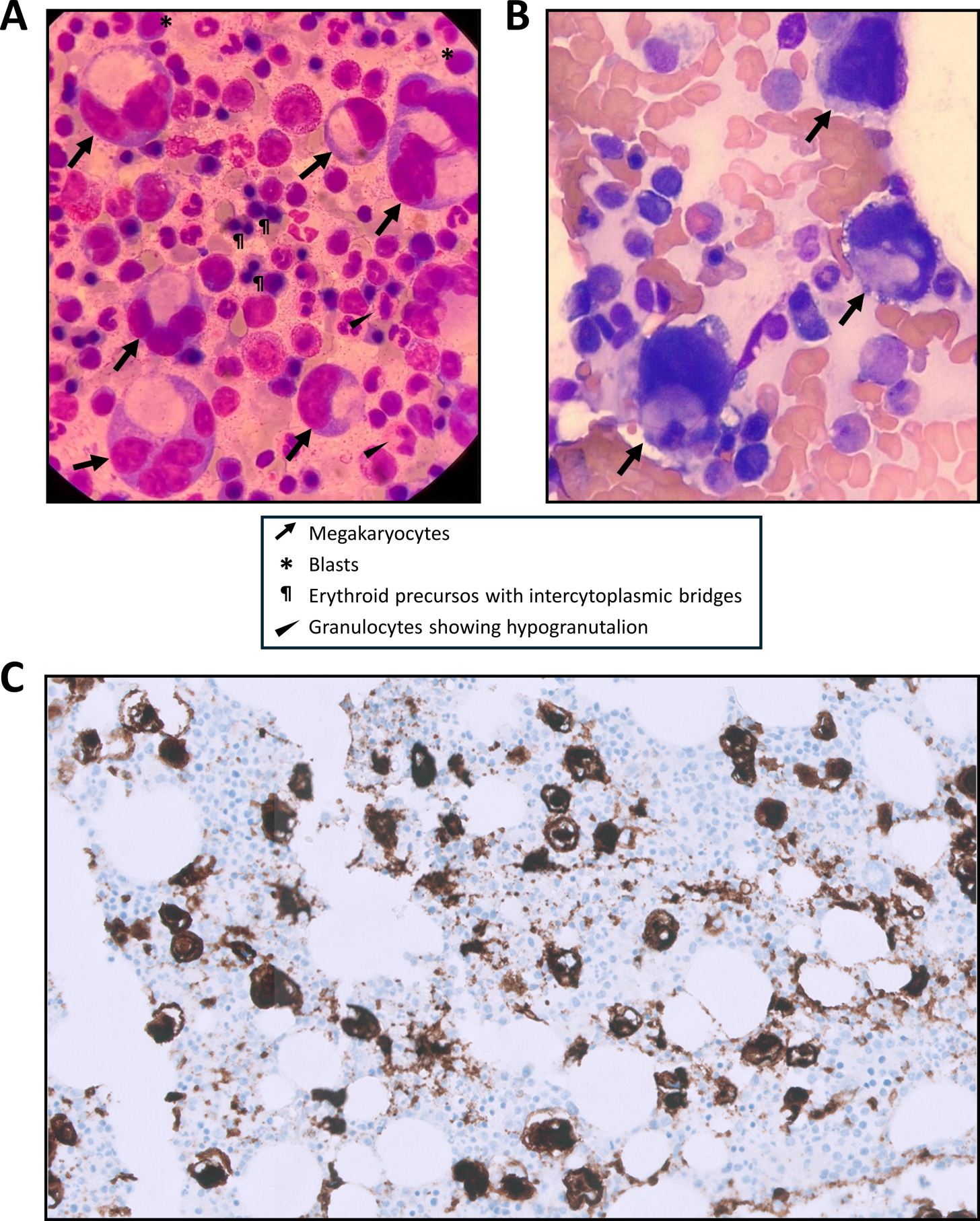
Figure legends

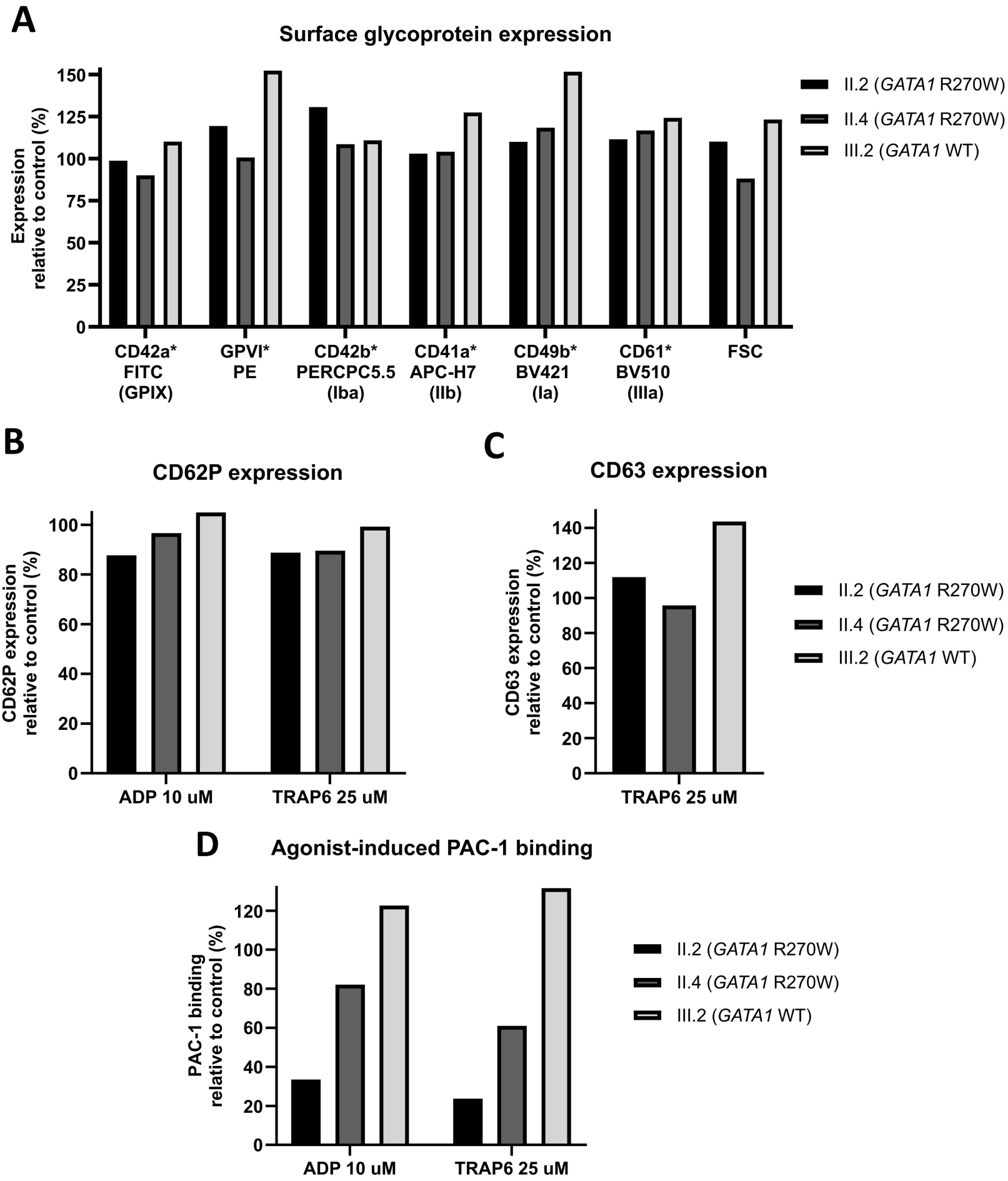
Figure 1.
Pedigree of the family carrying the germline *GATA1* R270W variant.
Squares represent males and circles denotes females. Filled symbols indicate individuals with hematological abnormalities, while slashes mark deceased individuals. The proband (II.2), is indicated by an arrow. The BM study of II.2 revealed multilineage dysplasia characterized by 70% hypoplastic megakaryocytes displaying unilobed or bilobed nuclei and cytoplasmic inclusions, dysgranulopoiesis with more than 20% of granulocytic precursors showing hypogranulation and pseudo-Pelger morphology, and erythroid precursors with intercytoplasmic bridges. The blast percentage was 6%, consistent with a diagnosis of myelodysplastic syndrome with excess blasts (MDS-EB). Cytogenetic analysis revealed a normal female karyotype (46,XX,inc[7]), and fluorescence in situ hybridization (FISH) studies were negative for deletions in 5q and 7q. The sister (II.4) presents with isolated mild thrombocytopenia over a 10-year period. Genotypes and hematologic status are indicated where available. WT: wild-type.

Figure 2.
Bone marrow morphology in carriers of *GATA1* R270W variant and MDS-EB and the male patient with the somatic *GATA1* R270W variant.
(A) Bone marrow smear from the female proband (Figure 1, II.2) carrier of the germline *GATA1* R270W showing multilineage dysplasia with marked megakaryocytic abnormalities, including small, hypolobulated megakaryocytes with basophilic cytoplasm and cytoplasmic projections (*arrows*), blasts (*asterisks*), erythroid precursors with intercytoplasmic bridges (¶), and granulocytes with hypogranulation (*arrowheads*) (May–Grünwald–Giemsa stain, ×400). **(B)** Bone marrow smear from the unrelated male patient with the same *GATA1* variant identified somatically, showing similar megakaryocytic dysplasia, including micromegakaryocytes with pseudopod-like projections (*arrows*) (×400). (C) Immunohistochemistry for CD34 in the male patient confirms CD34 positivity in immature megakaryocytes (CD34 stain, ×10).

Figure 3.
Flow cytometry analysis of platelet glycoproteins, PAC-1 binding, and granule secretion.
(A) Surface expression of platelet glycoproteins (GPIb/IX, GPIIb/IIIa, GPIa/IIa, GPVI). (B) P-selectin (CD62P) expression after agonist stimulation with ADP (10 µM) or TRAP-6 (25 µM). (C) CD63 expression after stimulation with TRAP-6 (25 µM). (D) PAC-1 binding after platelet activation with ADP (10 µM) or TRAP-6 (25 µM).
Results are expressed relative to parallel healthy control platelets for both carriers of the novel *GATA1* R270W variant (Figure 1, II.2 and II.4) and for one WT relative (Figure 1, III.2). Bars represent the patient/control ratio of fluorescence intensity (FI).


I
1
2
3
Hematological neoplasm
II
1
2
GATA1 R270W MDS (58y)
3
GATA1 WT
4
GATA1 R270W Thrombo-cytopenia (56y)
5
6
GATA1 WT
7
GATA1 WT
III
1
GATA1 WT
2
GATA1 WT
3
GATA1 WT
Hematological disease
Asymptomatic individual
Obligate carrier


A
B
Megakaryocytes
Blasts
Erythroid precursos with intercytoplasmic bridges
Granulocytes showing hypogranutalion
C


A
Surface glycoprotein expression
Expression relative to control (%)
150
125
100
75
50
25
0
CD42a* FITC (GPIX)
GPVI* PE
CD42b* PERCPC5.5 (Iba)
CD41a* APC-H7 (IIb)
CD49b* BV421 (Ia)
CD61* BV510 (IIIa)
FSC
II.2 (GATA1 R270W)
II.4 (GATA1 R270W)
III.2 (GATA1 WT)
B
CD62P expression
CD62P expression relative to control (%)
100
80
60
40
20
0
ADP 10 uM
TRAP6 25 uM
C
CD63 expression
CD63 expression relative to control (%)
140
120
100
80
60
40
20
0
TRAP6 25 uM
II.2 (GATA1 R270W)
II.4 (GATA1 R270W)
III.2 (GATA1 WT)
D
Agonist-induced PAC-1 binding
PAC-1 binding relative to control (%)
120
100
80
60
40
20
0
ADP 10 uM
TRAP6 25 uM
II.2 (GATA1 R270W)
II.4 (GATA1 R270W)
III.2 (GATA1 WT)