**Phenotypic characterization of a patient with Glanzmann Thrombasthenia caused by a novel homozygous mutation in the ITGA2B gene**.

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**Background/introduction:**

Glanzmann Thrombasthenia (GT) is a rare congenital bleeding disorder caused by a defect in platelet receptor αIIbβ3. It is characterized by impaired platelet aggregation that classically causes mild to severe mucocutaneous bleedings. We present here the case of a 52-year old woman who was diagnosed with GT at birth due to umbilical cord bleeding. In 2009, the patient presented with hemorrhage in the upper gastro-intestinal tract, which led to hemorrhagic shock. Platelet transfusion turned out to be inefficient, as the patient had developed anti-platelet antibodies (anti-HLA class II, anti-GPIbIX and anti-αIIbβ3). Hemostasis was restored using recombinant FVIIa. Since then, the patient exhibited recurrent but moderate epistaxis.

**Aims:**

To better characterize the patient hemostatic dysfunction and identify the underlying IIb3 mutation.

**Methods/Materials**

Initial laboratory work-up consisted in standard routine tests including whole blood cell count and coagulation screens (prothrombin time, PT; activated partial thromboplastin time, aPTT; thrombin time, TT and fibrinogen levels). Primary hemostasis was evaluated by measuring closure time using PFA-100® and by light-transmission aggregometry in platelet-rich plasma (PRP). Platelet surface IIb3 levels were assessed in whole blood by flow cytometry analysis using PerCP-coupled anti-CD61 antibody. Clot retraction was evaluated in whole blood drawn on a tube containing a clot activator (silica particles) and incubated for 24h at 37°C. In addition, thrombus retraction was assessed in PRP in which erythrocytes (2%, V/V) were added to visualize the platelet clot. Either calcium (20 mM) or thrombin (1U/ml) was added to the PRP and tests tubes were incubated at 37°C. Thrombus formation and retraction were evaluated every 30 minutes up to 4h. *ITGA2B* and *ITGB3* genes were analyzed using high resolution melting and direct exon sequencing.

**Results**

Laboratory investigations displayed the classical phenotypic presentation of GT. Blood cell count and in particular platelet count was within normal range (150-350 x103/mm3). Routine coagulation tests (PT, aPTT, TT) showed no abnormality. PFA-100 closure time was markedly prolonged using both collagen-epinephrine (>300 sec) and collagen-ADP cartridges (>292 sec). As expected, PRP aggregation assay showed absence of platelet response to ADP (5 µM), collagen (2 µg/ml), epinephrine (5 µM) and arachidonic acid (1 mM), while ristocetin-induced agglutination remained normal. The expression of IIb3 integrin was reduced to 14% of normal as compared to an age-matched control population. Clot retraction was severely impaired in both recalcified PRP and whole blood. Thrombus retraction was virtually absent 20 min and up to 4 hours upon recalcification and remained strongly inhibited even 4 hours after addition of 1 U/ml thrombin, as compared to PRP from a healthy donor. Genetic analysis revealed a previously unidentified mutation in exon 18 of the ITGA2B gene. The missense mutation (c.1722A>C) led to the substitution of the Asp591 to an Ala residue of the IIb subunit. Intriguingly, our patient was homozygous for the mutation although no notion of consanguinity appeared in her family history.

**Summary/Conclusions**

We identified a novel mutation in the IIb subunit resulting in Glanzmann Thrombasthenia.

Category : “Clinical and Laboratory”