

Case Report

# Neonatal Intracranial Hemorrhage with a Dramatic Outcome Due to Maternal Anti CD36 Antibodies

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**Abstract:** Fetal/neonatal allo-immune thrombocytopenia (FNAIT) results from maternal immunization against fetal platelet-specific antigens (HPA) inherited from the father. Most cases involve HPA located on glycoproteins (GP) IbIX, IaIIa and IIbIIIa. Iso-immunizations can also occur in the absence of expression of membrane proteins, such as GPIIb or GPIIIa in Glanzmann patients. CD36 (also called glycoprotein GPIV) deficiency is observed in 3 to 5% of Asian and African populations. We report here the case of a 41-year-old Canadian woman originated from Africa, who delivered a male dead new-born at 39 weeks of gestation. A massive intracranial haemorrhage was identified as being the obvious cause of death. No platelet antibody against GPIbIX, IaIIa, and IIbIIIa was identified by the gold-standard Monoclonal Antibody-specific Immobilization of Platelet Antigens (MAIPA) assay. Surprisingly, anti CD36 iso-antibodies were identified in the maternal serum with a new bead-based multiplex assay. The *CD36* gene was sequenced for both parents, and a mutation was identified on Exon 10 of the mother's *CD36* gene, which was absent for the father: NM\_000072.3:c.975T>G inducing a STOP codon at position 325 of the mature protein. The absence of CD36 expression on the mother's platelets was confirmed by flow cytometry.

**Keywords:** neonatal allo-immune thrombocytopenia; intracranial hemorrhage; CD36

## 1. Introduction

Fetal/neonatal allo-immune thrombocytopenia (FNAIT) results from the maternal immunization against fetal platelet-specific antigens (HPA) inherited from the father. The major risk of FNAIT is intracranial hemorrhage (ICH), a majority occurring in utero, leading to neurological sequelae in 20%, or to death in 10%, of reported cases. Most cases involve HPA located on glycoproteins (GP) IbIX, IaIIa and IIbIIIa. Iso-immunizations can also occur in the absence of expression of membrane proteins, such as GPIIb or GPIIIa in Glanzmann patients. CD36 (also called GPIV) deficiency is observed in 3 to 5% of Asian and African populations [1]. This transmembrane glycoprotein of 88-kDa is usually expressed by capillary and mammary endothelium, macrophages, erythroblasts, adipocytes, platelets and monocytes.

## 2. Case Presentation

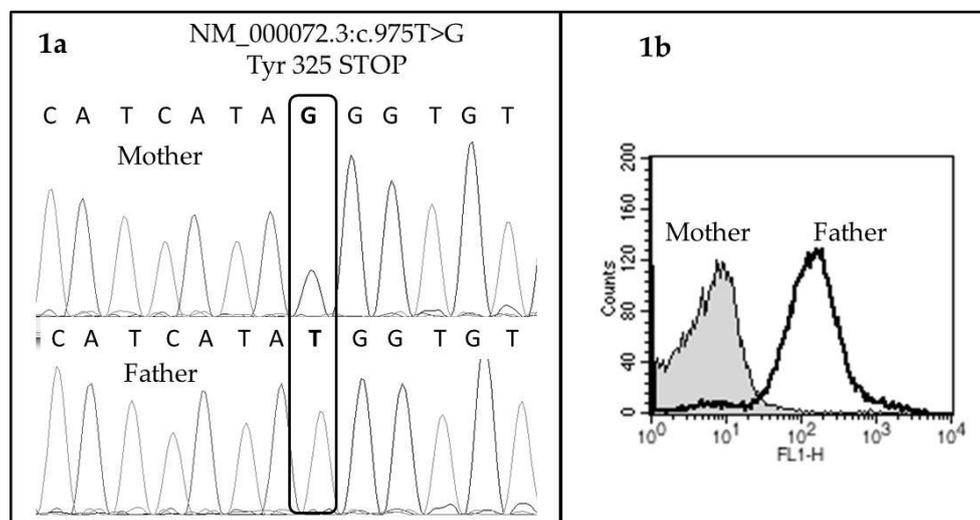
We report here the case of a 41 year old Canadian woman originated from Africa, who delivered a male dead newborn at 39 weeks of gestation. This was her fourth pregnancy and first newborn, with a history of three voluntary abortions. Unfortunately, the platelet count of the newborn was not defined, but intracranial haemorrhage was identified as being the obvious cause of death.

### 2.1. Laboratory Investigations

Laboratory investigations were performed by the Platelet Immunology laboratory of Hema-Quebec (Montreal, Canada). HPA genotyping of the parents did not reveal any incompatibility in HPA-1 to -6, -9, and -15. No platelet antibody against GPIbIX, IaIIa and IIbIIIa was identified by the gold-standard MAIPA assay (Monoclonal Antibody-specific Immobilization of Platelet Antigens [2]). A cross-match of maternal serum against the father's platelets was also negative for these GP using the same MAIPA assay. Additional investigations were performed with a new bead-based multiplex assay (Luminex, PakLx, Immucor, Waukesha, WI, USA). Surprisingly, anti CD36 iso-antibodies were identified in the maternal serum.

### 2.2. CD36 Deficiency on Maternal Platelets and Identification of the Mutation

Biological samples were sent to a second reference laboratory for confirmation (BloodCenter of Brittany EFS, Platelet Immunology Laboratory, Rennes, France). Maternal anti CD36 iso-antibodies were confirmed with the same Luminex technology. Unsurprisingly, those antibodies were undetectable in MAIPA (FA6-152, 10.5 and TR9 anti-CD36 mouse monoclonal antibodies). This method has already been described as poorly sensitive for detecting anti CD36 iso-antibodies, as human anti CD36 antibodies recognize the same epitopes blocked by mouse monoclonal antibodies [3]. To understand the origin of those maternal anti CD36 iso-antibodies, the entire coding sequence of CD36 gene was sequenced for both parents, as previously described [4]. We identified a mutation on Exon 10 of the mother's CD36 gene, which was absent for the father: NM\_000072.3:c.975T>G inducing a STOP codon at position 325 of the mature protein (Figure 1a). This mutation has already been described as the most frequent in African people [5] and is already recorded as rs3211938 in the international database. Using flow cytometry, we noticed the absence of CD36 expression on the mother's platelets (Figure 1b), in contrast to the father's platelets with a normal expression of CD36.



**Figure 1.** Platelet CD36 deficiency in a context of maternal iso-immunization. Molecular basis of the mutation by Sanger sequencing of Exon 10 CD36 gene (a), and flow cytometry of platelets from the mother and the father using FITC-labeled anti CD36 mouse monoclonal antibody clone 10.5 (b).

### 3. How to Manage a Future Pregnancy?

As the parents wish to have another baby, their obstetrician asked for consultation on how to manage a pregnancy and delivery. In order to prevent fetal/neonatal thrombocytopenia, a non-invasive antenatal therapy based on intravenous immunoglobulin (IvIg) is recommended, even if the literature reports only a few cases of treated women in a context of CD36 deficiency [6]. In a context of fetomaternal anti-HPA allo-immunization, this therapeutic option is the most effective

in the prevention of fetal and neonatal bleeding [7]. At delivery, the platelet count of the newborn must be measured to evaluate antenatal therapy effectiveness. In case of severe thrombocytopenia, the newborn will be transfused with the platelets of a particular phenotype. In the case reported here, CD36 negative platelets can be frozen (e.g., mother's platelets collected before pregnancy), and thawed if the newborn needs to be transfused.

#### 4. Conclusions

To conclude, we report here a rare case of maternal iso-immunization against CD36 with a dramatic outcome, but clearly elucidated which procedures allow for the safe management of a future pregnancy.

**Conflicts of Interest:** The authors declare no conflict of interest.

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