New Developments in Fetal and Neonatal Alloimmune Thrombocytopenia

James B. Bussel, MD, Emilie L. Vander Haar, MD, Richard L. Berkowitz

PII: S0002-9378(21)00436-1

DOI: https://doi.org/10.1016/j.ajog.2021.04.211

Reference: YMOB 13795

To appear in: *American Journal of Obstetrics and Gynecology*

Received Date: 2 December 2020

Revised Date: 26 March 2021

Accepted Date: 2 April 2021

Please cite this article as: Bussel JB, Vander Haar EL, Berkowitz RL, New Developments in Fetal and Neonatal Alloimmune Thrombocytopenia, *American Journal of Obstetrics and Gynecology* (2021), doi: https://doi.org/10.1016/j.ajog.2021.04.211.

New Developments in Fetal and Neonatal Alloimmune Thrombocytopenia

James B. Bussel[1,2], MD, Emilie L. Vander Haar, MD[2,3] and Richard L Berkowitz[3,4]
[1]Professor Emeritus, Department of Pediatrics, [2]Weill Cornell Medicine, New York, New York
[3]Division of Maternal Fetal Medicine, Department of Obstetrics and Gynecology
[4]Columbia Vagelos College of Physicians and Surgeons, Columbia University Irving Medical Center, New York, New York

 # Corresponding Author:

Dr. James B. Bussel

Professor Emeritus of Pediatrics, Weill Cornell Medicine, New York, New York

Email: jbussel@med.cornell.edu, 917-291-5091

115 east 67th street, New York NY 10065

Dr. Bussel Reports the following disclosures:

Consultant: CSL-Behring (IVIG manufacturer), UCB, Argenx, Momenta/Janssens (manufacturers of FcRn inhibitors), and RallyBio (company interested in screening and prophylaxis of FNAIT)
DSMB: CSL-Behring

Dr Berkowitz reports: Momenta/Janssens (manufacturers of FcRn inhibitors), and RallyBio (company interested in screening and prophylaxis of FNAIT)

Abstract: 294 words

Manuscript: 3514 words

Abstract

Fetal and Neonatal AlloImmune Thrombocytopenia (FNAIT), the platelet equivalent of Hemolytic Disease of the Fetus and Newborn (HDFN), can have devastating effects on both the fetus and neonate. Current management of FNAIT in a subsequent affected pregnancy involves antenatal administration of IVIG and prednisone to the pregnant woman in order to prevent the development of severe fetal thrombocytopenia and secondary intracranial hemorrhage (ICH) in utero. That therapy has proven to be highly effective, but is associated with maternal side effects and is expensive. This commentary describes four advances that could substantially change the current approach to detecting and managing FNAIT in the near future. The first would be introduction of a program to screen all antepartum patients in this country for pregnancies at risk to develop FNAIT. Strategies to implement this complex process are described. A second advance is testing of cell free fetal DNA (cffDNA) obtained from maternal blood to non-invasively determine the fetal HPA-1 genotype. A third, in preliminary development, is creation of a prophylactic product that would be the platelet equivalent of Rh immune globulin (Rhogam). Finally a fourth major potential advance is development of neonatal Fc receptor (FcRn) inhibitors to replace the current medical therapy administered to pregnant women with an affected fetus. FcRn recycles plasma IgG to increase its half-life and is the means by which IgG crosses the placenta from the maternal to the fetal circulation. Blocking FcRn is an ideal way to prevent maternal IgG antibody from causing FNAIT in a fetus at risk for developing that disorder. The pertinent pathophysiology and rationale for each of these developments will be presented along with our thoughts relating to steps that must be taken and difficulties that each approach would face in order for them to be successfully implemented.

Key Words: FcRn, FNAIT, HPA-1ab, platelet, NAITgam, thrombocytopenia, intracranial hemorrhage, IVIG

Introduction:

Fetal and Neonatal AlloImmune Thrombocytopenia (FNAIT) is the platelet equivalent of Hemolytic Disease of the Fetus and Newborn (HDFN). It is a rare disease, occurring in approximately 1 in 1,000 births, but affected pregnancies can carry severe consequences including fetal/neonatal intracranial hemorrhage (ICH)[1], which may result in irreversible brain damage or death. FNAIT is caused by an incompatibility between the antigenic composition of the mother's platelets and those of the fetus, inherited from the father [2, 3]. Approximately 80 percent of FNAIT cases in Caucasians occur in a mother whose platelets express only Human Platelet Antigen 1b (HPA-1a negative) and who conceives an HPA-1a fetus. Those fetal platelets enter the maternal circulation and cause an immune reaction, leading to maternal production of HPA-1a antibodies that subsequently cross the placenta and lead to fetal thrombocytopenia(Figure 1). While HPA-1a discordance is the most common source of FNAIT, more than 30 other platelet antigen incompatibilities can cause this disorder[4], although those cases are usually less severe. The HPA-1ab polymorphism is not present in patients of Chinese or Japanese descent; platelet antigen frequencies in other ethnicities are not as well-defined[5, 6].

Since routine screening for the maternal platelet genotype is not currently performed in the United States, most women with this disorder are only discovered after having had an affected neonate. FNAIT is often suspected when, during the first day of life, an infant with unexpected signs of bruising or frank bleeding is found to have an abnormally low platelet count[7]. Fortunately, most cases of FNAIT are not complicated by clinically significant bleeding, but 10 to 20% of severely-affected newborns will have an intracranial hemorrhage, three quarters of which occur *in utero*[8]. Current management of affected

newborns with platelet counts < 30,000/uL is to initiate treatment with a random platelet transfusion, often with concomitant IVIG[9], along with radiologic evaluation for ICH.

In the subsequent pregnancy of a woman whose fetus is known to be carrying the offending antigen, antepartum management with IVIG and steroids is recommended in order to increase the fetal platelet count until delivery[1, 3, 8, 10-14]. The anticipated severity of the disorder is related to whether, and if so when, the fetus suffered an ICH in the prior affected pregnancy. However, currently the only way to assess the actual degree of fetal thrombocytopenia is to directly measure the platelet count in utero by cordocentesis, which is an invasive procedure that may have serious adverse consequences. That recognition has led to a severity-based, minimally invasive medical approach for antenatal management of affected pregnancies (Figure 2). In order to be certain that the fetal platelet count has achieved sufficient levels without resorting to serial cordocentesis, all patients eventually are escalated to "maximal therapy" (IVIG 2g/kg/wk and prednisone 0.5mg/kg/day).

This level of therapy, however, is reached at different gestational ages according to the severity risk, and, in the highest risk group, the prednisone dose administered reaches 1mg/kg/day (Figure 2). Serial weekly platelet transfusions administered directly to the fetus is an invasive form of therapy complicated by increased fetal morbidity and mortality, which is now very infrequently used, and is not recommended.

HDFN and FNAIT: 2 peas in a pod?

Both HDFN AND FNAIT are caused by parental blood cell antigen incompatibilities in which the mother becomes sensitized by transplacental transmission of fetal cells into the maternal circulation. The mother makes IgG antibodies to a paternal antigen on the surface of these cells, which then cross the placenta, attack red cells or platelets , and cause fetal anemia or thrombocytopenia respectively. If severe enough, each of these effects can have fatal consequences for the fetus or newborn. Much of what we know about FNAIT has evolved based on recognizing its similarity to HDFN.

With the passage of time, however, it has become clear that there are important differences between these disorders. Amongst others these include:

- Screening for Rh incompatibility is virtually ubiquitous in the United States, and the use of Rh Immune Globulin (RhIG) has almost entirely eliminated HDFN in this country, which is one of the most outstanding medical developments of the 20th century[15]. In contrast, there is currently no screening for patients at risk of developing FNAIT, and although that disorder is less common than HDFN, its consequences can be equally devastating[7].
- FNAIT often becomes manifest in the first pregnancy and may be quite severe at that time[16], whereas in HDFN, the infant in the first pregnancy is rarely affected, and fetuses become progressively more severely affected in subsequent gestations[15, 17].
- The marked severity of thrombocytopenia in FNAIT is partly due to an inhibition of fetal megakaryocyte production by the maternal anti-platelet antibodies[18], whereas, except for cases of Kell incompatibility, the fetal

anemia in HDFN appears to be exclusively due to destruction of circulating red blood cells.

- HDFN causes anemia in utero which can be monitored non-invasively with serial MCA Doppler studies[17] whereas there are no biomarkers of fetal thrombocytopenia that can currently be assessed non-invasively prior to the occurrence of an ICH [19].
- Maternally-administered IVIG and steroids during the antepartum period is the way most cases of severely-affected FNAIT pregnancies are managed in the US, whereas in utero red cell transfusion and/or early delivery constitute standard management for severe cases of HDFN[15, 17].
- Fetal blood sampling is rarely performed in FNAIT since the medical treatment described above is virtually 100% effective in raising and then maintaining fetal platelet counts in a safe realm, whereas, women with HDFN are followed with serial MCA Doppler studies until it is determined that they need to be transfused in utero or delivered[15, 17].
- In HDFN, the fetus/newborn can suffer postnatal neurologic damage from hyperbilirubinemia caused by hemolysis, as well as developing other sequelae resulting from prolonged hypoxia, whereas in FNAIT, irreversible damage is almost always limited to cases complicated by intracranial hemorrhage[20].

In this commentary, we will describe several new developments at different stages of their evolution that we believe may, to differing degrees, potentially revolutionize how FNAIT will be diagnosed and managed in the foreseeable future.

Screening: Identifying the at-risk population

If screening and subsequent effective prophylaxis for FNAIT can be developed in a cost effective and safe manner, it would greatly reduce the incidence of that disorder. Several large population screening studies for FNAIT have been successfully performed in Europe[21-23] and have demonstrated the following important findings:

- Those studies have been performed with relatively small loss to follow-up and with platelet antigen typing having a very low (<<1%) error rate.
- 75% of women who were found to have FNAIT caused by the HPA-1a antigen became sensitized at delivery in their first pregnancy[21-23]. This is markedly different from cases detected by clinically documented neonatal thrombocytopenia, in which 60% are affected in the mother's first pregnancy[16]
- Other features of FNAIT are also different if cases accessed by screening are compared to those clinically detected in neonates by documenting thrombocytopenia. For example, the incidence of severe thrombocytopenia is 3-5 fold higher in the clinically identified cases. In other words, population screening may be identifying much more mild, asymptomatic disease.

It is important to note that approximately 28% of HPA-1a negative American women carry the DRB3*0101 HLA antigen. The latter identifies an immune response gene

without which HPA-1a negative women almost never produce high levels of anti-HPA-1a antibodies[24,25]. The association of high titer anti-HPA-1a with DRB3*0101 is one of the strongest links of a specific antibody response to an immune response gene that is currently known. It is extremely rare for an HPA-1a negative woman to become significantly sensitized to her HPA-1 positive fetus in the absence of having the DRB3*0101 antigen. Therefore, detection of that antigen is an essential component of a screening program designed to detect cases at risk for developing FNAIT.

The true at-risk population for the development of HPA-1a incompatible FNAIT in screening is approximately 1 in 200 women (0.5%). This assumes that very few DRB3*0101, HPA-1a negative women being screened will have pre-existing anti-HPA-1a antibodies. Approximately 85% of those women who are HPA-1a negative, will have an HPA-1a potentially affected fetus. In order to identify this subset, the following studies would need to be performed:

1. Maternal blood (already being sent for red blood cell typing on the first prenatal visit) would also be used to have her platelet genotype determined (figure 3). Currently maternal platelet typing has to be sent to a specialty laboratory. Whether this will continue to be necessary in the future remains to be seen. The availability of high-throughput Elisa-based testing for HPA-1a may make this screening cheaper and easier to standardize[26], but this has not undergone large scale testing.
2. In the screened subset of HPA-1a negative women, it would also be necessary to determine the DRB3*0101 status of the mother, the genotype of the fetus using

cff DNA obtained from the maternal blood sample, and the presence or absence of anti-HPA-1A antibodies in maternal serum (figure 3).

Therefore, the net effect of screening would be first to identify mothers at high risk of making anti-HPA-1a antibodies, i.e. those who are both HPA-1a negative and DRB3*0101 positive, and then to see if they have an HPA-1a positive fetus. In addition, screening will also identify a further subset in which such a patient already has produced anti-HPA-1a antibodies. The latter group would be directly referred to an MFM specialist to undergo evaluation for antenatal therapy.

Fetal Typing: Cell-Free Fetal cffDNA

Approximately 98% of Caucasian Americans are HPA-1a positive, and 75% of those individuals are homozygous for that antigen[27, 28]**.** If, however, the father of the baby (FOB) in a pregnancy conceived with an HPA-1a negative woman is a heterozygote, which occurs in approximately 25% of cases, 50% of their conceptions will be negative for the HPA-1a antigen and thus not at risk of FNAIT. Therefore, although the majority of pregnancies conceived by couples that include an HPA-1a negative woman and an HPA-1a positive male will have an HPA-1a positive fetus, approximately 15% will not. As a consequence, if the genotype of the FOB is not known to be HPA-1a1a, the genotype of the fetus must be determined. Previously, there were only three ways to perform fetal platelet antigen typing: e.g. obtaining fetal cells from chorionic villus sampling, amniocentesis[29], or fetal blood sampling. Those methods are all invasive and thus have some associated risks. Presently, fetal platelet HPA-1a antigen typing can be done reliably as early as 10 weeks of

gestation using cell-free fetal DNA (cffDNA) detection in maternal blood obtained from a venipuncture[30]. This testing is now available in the United States and Western Europe for HPA-1a1b, but not yet for other platelet antigens. This form of assessing the fetal genotype for other platelet antigens may become available in the future, and, if so, will further optimize and make safer the management of those less common causes of FNAIT.

Prophylaxis: From Rhogam to "NAITgam"

HDFN

Prophylaxis has been remarkably successful in preventing cases of HDFN. RhD immunoglobulin (e.g. anti-D, Rhogam) is thought to prevent sensitization to RhD by blocking maternal production of her own anti-D antibodies when administered at 26-28 weeks of gestation, at the time of delivery, and following episodes of maternal bleeding, invasive fetal testing procedures, or maternal trauma that may be associated with fetal-maternal hemorrhage. Rh D immunoglobulin is a hyperimmune anti-D gammaglobulin that is manufactured by collecting the plasma of donors who have markedly elevated titers of antibody to Rh factor D[31]. In the past, these donors were highly sensitized women who had had newborns affected with HDFN. Currently, many donors are Rh negative men, or women who are not capable of, or have chosen not to become pregnant. Those donors are periodically given injections of Rh-positive cells to ensure that their levels of anti-D antibodies remain high. A number of years ago, several studies explored whether polyclonal, plasma-derived anti-D serum could be replaced by a monoclonal anti-D antibody[32]. However, the many different structural variations in the D antigen were found to prevent a single monoclonal antibody from being highly effective in preventing the

239 development of maternal sensitization[33]. Therefore, anti-D immunoglobulin remains a
240 polyclonal, plasma-derived product.
241
242 "NAITgam":
243 It is certainly possible that hyperimmune gammaglobulin derived from the plasma of
244 women who had prior pregnancies affected with FNAIT could provide prophylaxis in a
245 similar fashion to that obtained with Rh immunoglobulin. One lot of such a product, called
246 "NAITgam", has in fact been produced from plasma donations of women who have high
247 circulating levels of antibody to HPA-1a[34]. There will soon be a "proof of principle" trial to
248 determine if giving that product to non-pregnant HPA-1a negative recipients who have
249 been deliberately exposed to HPA-1a1a platelets prevents the recipients from making
250 antibody to HPA-1a.
251
252 There are very many uncertainties as to whether "NAITgam" can actually prevent FNAIT
253 from occurring as effectively as anti-D immunoglobulin does for HDFN. There is no reason
254 *per se* to doubt this, but very little experimental work outside of limited animal studies[35]
255 exists to support it. On the positive side, this product probably has very little maternal
256 toxicity. However, even if "NAITgam" is shown to be highly effective in providing
257 prophylaxis in studies of non-pregnant patients, appropriate dosing and a schedule of
258 administration must be determined during actual gestations. This will require large-scale
259 trials of antepartum testing to demonstrate that an experimentally-verified strategy works,
260 and that "NAITgam" is safe for both the mother and the fetus. Since severe cases of FNAIT
261 caused by HPA-1a can not only occur in the first pregnancy but may occur as early as 18

weeks, prophylaxis for those cases would need to be started by the beginning of the second trimester[12], and likely repeated on multiple occasions throughout the pregnancy.

Treatment: Inhibition of FcRn

Fc receptor (FcRn) is a unique Fc gamma receptor which binds IgG only at acidic pH[36, 37]. Free, unattached IgG is brought into an endothelial cell by pinocytosis from plasma, and passed into an endocytic vesicle called an endosome (figure 4). If the IgG is not "free" but has formed an immune complex with a platelet antigen, it may enter the endosome using a different pathway. The acidic pH of the endosome leads to dissociation of the immune complex. The free IgG is then able to bind to FcRn, which is located on the inner membrane of the endosome. The IgG-FcRn complex is then translocated to the cell surface. Because plasma has a neutral pH, the IgG dissociates from the receptor and is released back into the maternal circulation (figure 4)[38]. This recycling of IgG by FcRn results in maintenance of the normal half-life of IgG. [39, 40]The same mechanism of IgG transport occurs within the placenta where it allows IgG to cross into the fetal circulation. When maternal IgG contains anti-HPA-1a antibodies, this delivery system leads to destruction of fetal platelets containing that antigen.

Initial development of inhibitors of FcRn for clinical use was conceptualized as a therapeutic agent that should lower IgG levels, since recycling of that moiety would be significantly impaired[36]. Lowering all IgG levels would also diminish IgG autoantibody levels and, as such, would potentially have a beneficial effect on any IgG antibody-mediated disease. While not proven, it appears that anti-platelet antibodies in ITP are reduced by

FcRn inhibition to a greater extent than are the levels of free IgG. Several phase 2 and 3 clinical studies have now demonstrated the efficacy and safety of FcRn inhibition as treatment in patients with IgG-mediated autoantibody disorders such as ITP[39, 40], Pemphigus Vulgaris[41] and Myasthenia Gravis[42], with over 50% response rates in most of those trials.

Although when administered at a dose of 1g/kg/wk in the antenatal management of FNAIT, IVIG is effective in maintaining a fetal platelet count >30,000/uL in many patients, higher doses are required in more severe cases. One placental perfusion study found that only by giving IVIG at a dose of 2g/kg/wk can 90% blockade of maternal transfer of IgG into the fetal circulation be achieved[43]. However, if FcRn blockade were complete, as can safely be achieved by appropriate dosage and scheduling of FcRn inhibitors, this should totally block passage of maternal IgG across the placenta in patients affected with either HDFN or FNAIT. Therefore, the use of these inhibitors could entirely eliminate the need to employ prednisone and IVIG at any doseage, with their attendant risk of substantial adverse side effects. An additional potential benefit of using FcRn inhibitors is that by substantially reducing the half life of IgG in the maternal circulation these agents would also significantly reduce the levels of anti-fetal platelet antibodies in her bloodstream.

A potential drawback of FcRn inhibition in pregnant women, however, is that both maternal and fetal IgG levels would become greatly reduced. The mother, however, would continue to have normal IgA and IgM levels as well as unimpaired T cell function [37]. Phase 3 studies using serial infusions of FcRn inhibitors will clarify whether there is any risk of that

therapy causing increased maternal infection, but thus far there has been no apparent evidence of that occurring in the studies that have been performed to date[17,18]. The fetus is not thought to "need" IgG *in utero.* After birth, however, a hypogammaglobulinemic neonate clearly would be at significantly increased risk for developing sepsis[44]. One approach to deal with this issue would be to stop FcRn inhibition approximately 2 weeks prior to birth, and then to administer high dose IVIG to the mother several days prior to delivery in an attempt to both reconstitute her IgG reserve as well as to deliver transplacental "normal" IgG to the fetus. This approach is currently being utilized in the HDFN study described below. An alternative, or supplementary, approach might be to administer IVIG intravenously to the neonate on the first day of life perhaps via the umbilical cord.

An ongoing study of FcRn inhibition in severely affected pregnancies affected by HDFN is currently enrolling patients at several sites around the world (See clinicaltrials.gov NCT03842). In this study, beginning in the second trimester, nipocalimab, an inhibitor of FcRn, is administered weekly to pregnant women with a history of severe HDFN in prior pregnancies in order to prevent maternal anti-D from crossing the placenta. Despite its current relative rarity, HDFN was chosen over FNAIT for this proof of concept study because the availability of a non-invasively obtainable biomarker in HDFN safely allows for frequent monitoring of the effectiveness of FcRn inhibition therapy in the at-risk fetus. In other words, if MCA Doppler studies indicate that a fetus being treated with FcRn inhibition is failing therapy and developing significant anemia, rescue therapy with intrauterine red blood cell transfusions can be administered[15, 17]. No parallel non-invasive approach to fetal

monitoring currently exists for FNAIT, so the need for emergent rescue therapy could not be anticipated. If this mode of treatment proves to be as beneficial as anticipated for HDFN, it should clearly be studied for FNAIT. Success in those trials could then lead to studying other maternal anti-fetal IgG-mediated diseases such as those caused by anti-Ro/La and anti-thyroid antibodies.

Conclusion

It is exciting to report that all of the modalities mentioned above are actively being investigated, and each of them may become important to varying degrees. Testing of free fetal DNA in maternal blood to determine fetal genotype for HPA-1a is now routinely attainable in the US.. This should be utilized whenever possible right now to eliminate invasive procedures done exclusively for that purpose. DRB3*0101 is now well-accepted as a crucial component in determining the risk of sensitization to HPA-1a, and will have to be an integral part of population screening for HPA 1a. High throughput platelet antigen screening is now possible, so the technology for instituting population screening of women at risk for acquiring FNAIT certainly exists. Studies for testing the efficacy of FNAIT propylaxis are in the very early stages, but one lot of "NAITgam" has already been made and is being investigated in proof of concept studies. The likelihood of combining both screening and prophylaxis to drastically reduce the incidence of FNAIT, however, is years away. Large scale clinical trials will need to be performed to evaluate efficacy, safety and cost-benefit before screening of all pregnancies and "NAITgam" prophylaxis of women at risk for developing FNAIT can become a reality.

Blocking transfer of maternal IgG to the fetus by inhibition of FcRn in HDFN is currently being studied. At least 4 different forms of this agent are also being evaluated for a number of indications in non-pregnant patients. If the studies in severe cases of HDFN prove to be safe and successful, we believe they hold great promise for revolutionizing the management of FNAIT.

What could FNAIT look like in 2030 ? Ideally if screening and effective prophylaxis for this disease in HPA 1a positive women become standard of care, severe FNAIT would be almost entirely eliminated, as has occurred with Rho D incompatibility in HDFN. In those few cases of FNAIT that were subsequently identified,which occurred because of lack of screening, failure of prophylaxis, or secondary to other platelet antigens, antenatal management with an inhibitor of FcRn to block transplacental transfer of disease-causing maternal IgG would become routine. In turn, that safe and highly effective form of therapy would make treatment with IVIG and prednisone a thing of the past. It remains to be seen, however, whether these aspirations can be achieved.

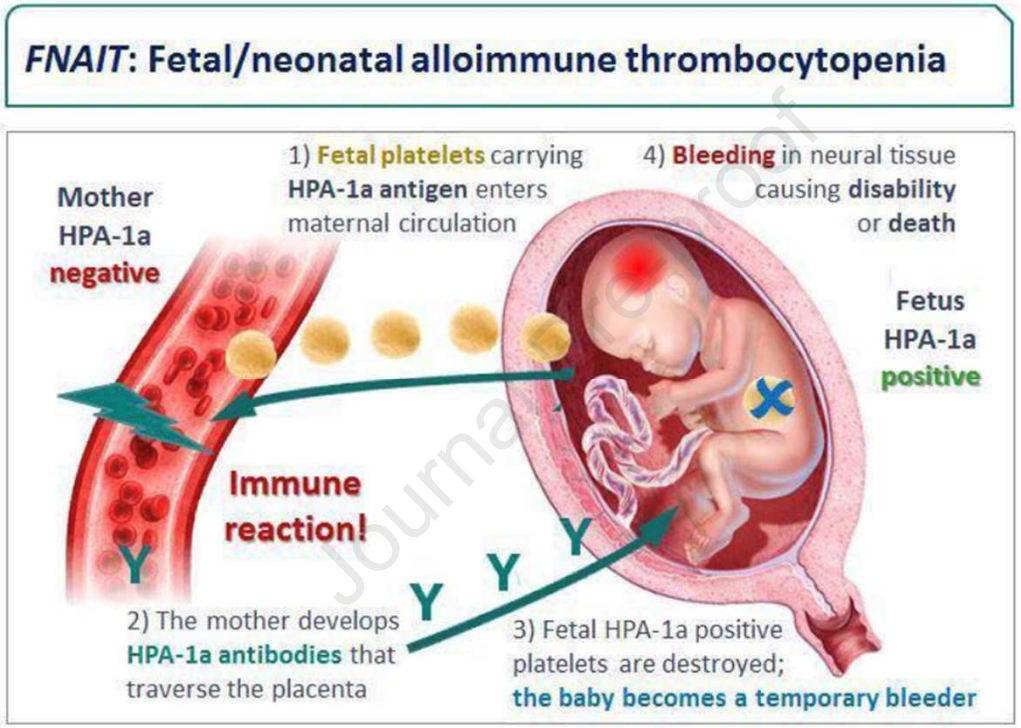
References:

1. Pacheco LD, Berkowitz RL, Moise KJ, Jr., Bussel JB, McFarland JG, Saade GR. Fetal and neonatal alloimmune thrombocytopenia: a management algorithm based on risk stratification. Obstet Gynecol 2011;118:1157-63.
2. Mueller-Eckhardt C, Kiefel V, Grubert A, et al. 348 cases of suspected neonatal alloimmune thrombocytopenia. Lancet 1989;1:363-6.
3. Lieberman L, Greinacher A, Murphy MF, et al. Fetal and neonatal alloimmune thrombocytopenia: recommendations for evidence-based practice, an international approach. Br J Haematol 2019;185:549-62.
4. Peterson JA, Gitter M, Bougie DW, et al. Low-frequency human platelet antigens as triggers for neonatal alloimmune thrombocytopenia. Transfusion 2014;54:1286-93.
5. Ohto H. [Neonatal alloimmune thrombocytopenia]. Nihon Rinsho 1997;55:2310-4.
6. Xue M, Liu YC, Wei P. [Genetic polymorphism of human platelet antigens 1-18 in Chinese Nanjing Han population]. Zhongguo Shi Yan Xue Ye Xue Za Zhi 2012;20:1235-9.
7. Bussel JB, Zacharoulis S, Kramer K, McFarland JG, Pauliny J, Kaplan C. Clinical and diagnostic comparison of neonatal alloimmune thrombocytopenia to non-immune cases of thrombocytopenia. Pediatr Blood Cancer 2005;45:176-83.
8. Lakkaraja M, Berkowitz RL, Vinograd CA, et al. Omission of fetal sampling in treatment of subsequent pregnancies in fetal-neonatal alloimmune thrombocytopenia. Am J Obstet Gynecol 2016;215:471 e1-9.
9. Kiefel V, Bassler D, Kroll H, et al. Antigen-positive platelet transfusion in neonatal alloimmune thrombocytopenia (NAIT). Blood 2006;107:3761-3.
10. Bussel JB, Berkowitz RL, McFarland JG, Lynch L, Chitkara U. Antenatal treatment of neonatal alloimmune thrombocytopenia. N Engl J Med 1988;319:1374-8.
11. Bussel JB, Berkowitz RL, Lynch L, et al. Antenatal management of alloimmune thrombocytopenia with intravenous gamma-globulin: a randomized trial of the addition of low-dose steroid to intravenous gamma-globulin. Am J Obstet Gynecol 1996;174:1414-23.
12. Bussel JB, Berkowitz RL, Hung C, et al. Intracranial hemorrhage in alloimmune thrombocytopenia: stratified management to prevent recurrence in the subsequent affected fetus. Am J Obstet Gynecol 2010;203:135 e1-14.
13. Berkowitz RL, Kolb EA, McFarland JG, et al. Parallel randomized trials of risk-based therapy for fetal alloimmune thrombocytopenia. Obstet Gynecol 2006;107:91-6.
14. Arnold DM, Smith JW, Kelton JG. Diagnosis and management of neonatal alloimmune thrombocytopenia. Transfus Med Rev 2008;22:255-67.
15. McBain RD, Crowther CA, Middleton P. Anti-D administration in pregnancy for preventing Rhesus alloimmunisation. Cochrane Database Syst Rev 2015:CD000020.
16. Jin JC, Lakkaraja MM, Ferd P, et al. Maternal sensitization occurs before delivery in severe cases of fetal alloimmune thrombocytopenia. Am J Hematol 2019;94:E213-E15.
17. Moise KJ, Jr., Argoti PS. Management and prevention of red cell alloimmunization in pregnancy: a systematic review. Obstet Gynecol 2012;120:1132-9.
18. Liu ZJ, Bussel JB, Lakkaraja M, et al. Suppression of in vitro megakaryopoiesis by maternal sera containing anti-HPA-1a antibodies. Blood 2015;126:1234-6.

19. KJAER M, BERTRAND G, BAKCHOUL T, et al. Maternal HPA-1a antibody level and its role in predicting the severity of Fetal/Neonatal Alloimmune Thrombocytopenia: a systematic review. Vox Sang 2019;114:79-94.
20. WARD MJ, PAULINY J, LIPPER EG, BUSSEL JB. Long-term effects of fetal and neonatal alloimmune thrombocytopenia and its antenatal treatment on the medical and developmental outcomes of affected children. Am J Perinatol 2006;23:487-92.
21. WILLIAMSON LM, HACKETT G, RENNIE J, et al. The natural history of fetomaternal alloimmunization to the platelet-specific antigen HPA-1a (PlA1, Zwa) as determined by antenatal screening. Blood 1998;92:2280-7.
22. TURNER ML, BESSOS H, FAGGE T, et al. Prospective epidemiologic study of the outcome and cost-effectiveness of antenatal screening to detect neonatal alloimmune thrombocytopenia due to anti-HPA-1a. Transfusion 2005;45:1945-56.
23. KILLIE MK, KJELDSEN-KRAGH J, HUSEBEKK A, SKOGEN B, OLSEN JA, KRISTIANSEN IS. Cost-effectiveness of antenatal screening for neonatal alloimmune thrombocytopenia. BJOG 2007;114:588-95.
24. KJAER M, GEISEN C, AKKOK CA, et al. Strategies to develop a prophylaxis for the prevention of HPA-1a immunization and fetal and neonatal alloimmune thrombocytopenia. Transfus Apher Sci 2020;59:102712.
25. KJELDSEN-KRAGH J, FERGUSSON DA, KJAER M, et al. Fetal/neonatal alloimmune thrombocytopenia: a systematic review of impact of HLA-DRB3*01:01 on fetal/neonatal outcome. Blood Adv 2020;4:3368-77.
26. WINKELHORST D, PORCELIJN L, MUIZELAAR E, OLDERT G, HUISKES E, VAN DER SCHOOT CE. Fast and low-cost direct ELISA for high-throughput serological HPA-1a typing. Transfusion 2019;59:2989-96.
27. FONTAO-WENDEL R, WENDEL S, ODONE V, CARNEIRO JD, SILVA L, ISFER E. A case report of neonatal alloimmune thrombocytopenic purpura: the importance of correct diagnosis for future pregnancies. Sao Paulo Med J 2005;123:198-200.
28. KJELDSEN-KRAGH J. New elegant methods for maternal and fetal HPA-1a typing. Transfusion 2018;58:2253-54.
29. MCFARLAND JG, ASTER RH, BUSSEL JB, GIANOPOULOS JG, DERBES RS, NEWMAN PJ. Prenatal diagnosis of neonatal alloimmune thrombocytopenia using allele-specific oligonucleotide probes. Blood 1991;78:2276-82.
30. VAN DER SCHOOT CE, THURIK F, SCHEFFER PG, et al. Prenatal Fetal DNA Testing for Predicting HDFN, FNAIT, and RhIG Candidacy. Blood 2013;122:SCI-51-SCI-51.
31. BRINC D, LE-TIEN H, CROW AR, SIRAGAM V, FREEDMAN J, LAZARUS AH. Immunoglobulin G-mediated regulation of the murine immune response to transfused red blood cells occurs in the absence of active immune suppression: implications for the mechanism of action of anti-D in the prevention of haemolytic disease of the fetus and newborn? Immunology 2008;124:141-6.
32. KUMPEL BM. Monoclonal anti-D development programme. Transpl Immunol 2002;10:199-204.
33. FLEGEL WA. The genetics of the Rhesus blood group system. Blood Transfus 2007;5:50-7.
34. WENG YJ, HUSEBEKK A, SKOGEN B, KJAER M, LIN LT, BURNOUF T. Anti-Human Platelet Antigen-1a Immunoglobulin G Preparation Intended to Prevent Fetal and Neonatal Alloimmune Thrombocytopenia. PLoS One 2016;11:e0162973.

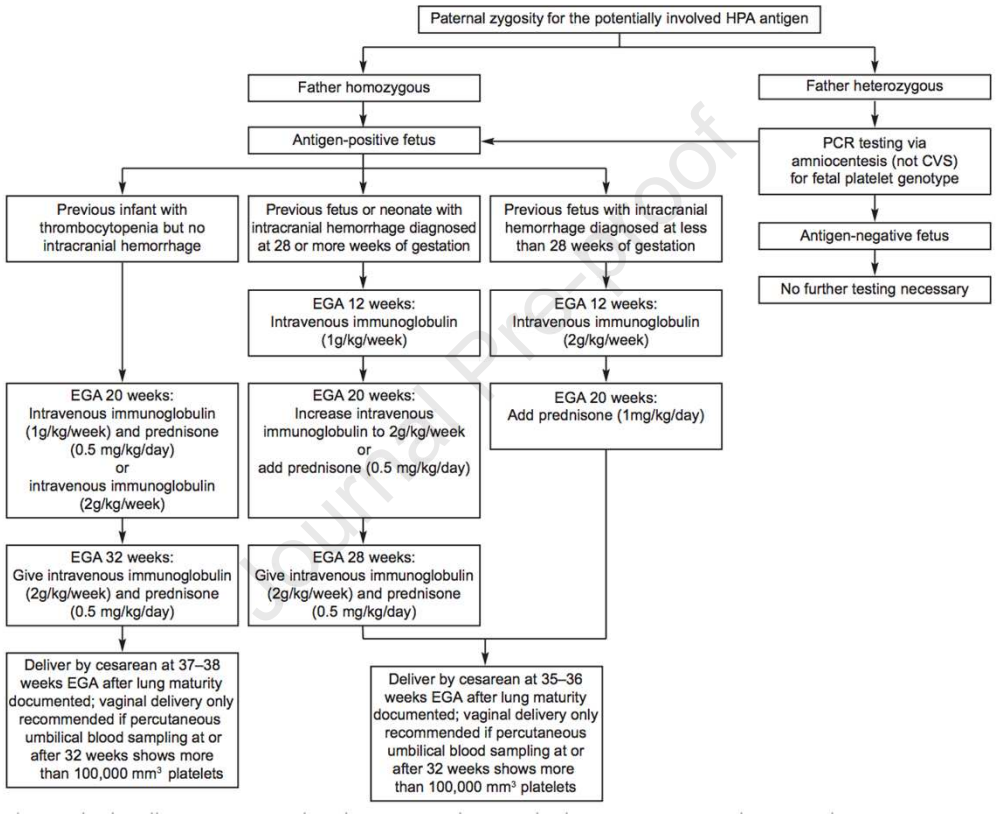
35. TILLER H, KILLIE MK, CHEN P, et al. Toward a prophylaxis against fetal and neonatal alloimmune thrombocytopenia: induction of antibody-mediated immune suppression and prevention of severe clinical complications in a murine model. Transfusion 2012;52:1446-57.
36. PYZIK M, SAND KMK, HUBBARD JJ, ANDERSEN JT, SANDLIE I, BLUMBERG RS. The Neonatal Fc Receptor (FcRn): A Misnomer? Front Immunol 2019;10:1540.
37. PATEL DD, BUSSEL JB. Neonatal Fc receptor in human immunity: Function and role in therapeutic intervention. J Allergy Clin Immunol 2020;146:467-78.
38. DAMAS OM, DESHPANDE AR, AVALOS DJ, ABREU MT. Treating Inflammatory Bowel Disease in Pregnancy: The Issues We Face Today. J Crohns Colitis 2015;9:928-36.
39. NEWLAND AC, SANCHEZ-GONZALEZ B, REJTO L, et al. Phase 2 study of efgartigimod, a novel FcRn antagonist, in adult patients with primary immune thrombocytopenia. Am J Hematol 2020;95:178-87.
40. ROBAK T, KAZMIERCZAK M, JARQUE I, et al. Phase 2 multiple-dose study of an FcRn inhibitor, rozanolixizumab, in patients with primary immune thrombocytopenia. Blood Adv 2020;4:4136-46.
41. IZUMI K, BIEBER K, LUDWIG RJ. Current Clinical Trials in Pemphigus and Pemphigoid. Front Immunol 2019;10:978.
42. MENON D, BARNETT C, BRIL V. Novel Treatments in Myasthenia Gravis. Front Neurol 2020;11:538.
43. URBANIAK SJ, DUNCAN JI, ARMSTRONG-FISHER SS, ABRAMOVICH DR, PAGE KR. Transfer of anti-D antibodies across the isolated perfused human placental lobule and inhibition by high-dose intravenous immunoglobulin: a possible mechanism of action. Br J Haematol 1997;96:186-93.
44. EL-BOHY M, POOWUTTIKUL P, SECORD E. Humoral Immune Deficiencies of Childhood. Pediatr Clin North Am 2019;66:897-903.

Figure 1: Depiction of maternal/fetal platelet antigen incompatibility in fetal/neonatal alloimmune thrombocytopenia



FNAIT is caused by an incompatibility between the antigenic composition of the mother's platelets (HPA-1b1b) and those of the fetus (HPA-1a), inherited from the father. Fetal platelets enter the maternal circulation and cause an immune reaction, leading to maternal production of HPA-1a antibodies that subsequently cross the placenta and lead to fetal thrombocytopenia. This leads to a temporary bleeding state, which can cause intracranial hemorrhage. Figure reproduced with permission from NAITbabies.org.

Figure 2: Algorithm for management of FNAIT in women with a subsequent affected pregnancy



A severity-based, minimally invasive medical approach for antenatal management of affected pregnancies. Reproduced from Pacheco et al. *Fetal and neonatal alloimmune thrombocytopenia: a management algorithm based on risk stratification*. Obstet Gynecol 2011;118:1157-63

Theoretical Screening Process for FNAIT prophylaxis: estimated affected pregnancies per 1,000,000 deliveries

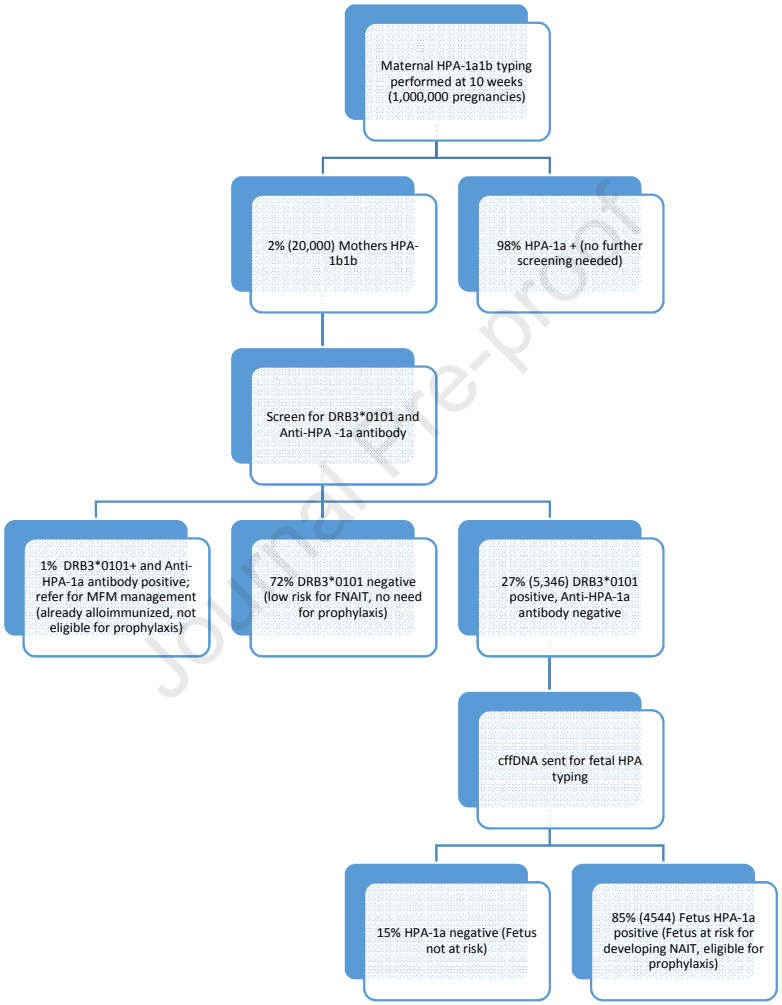
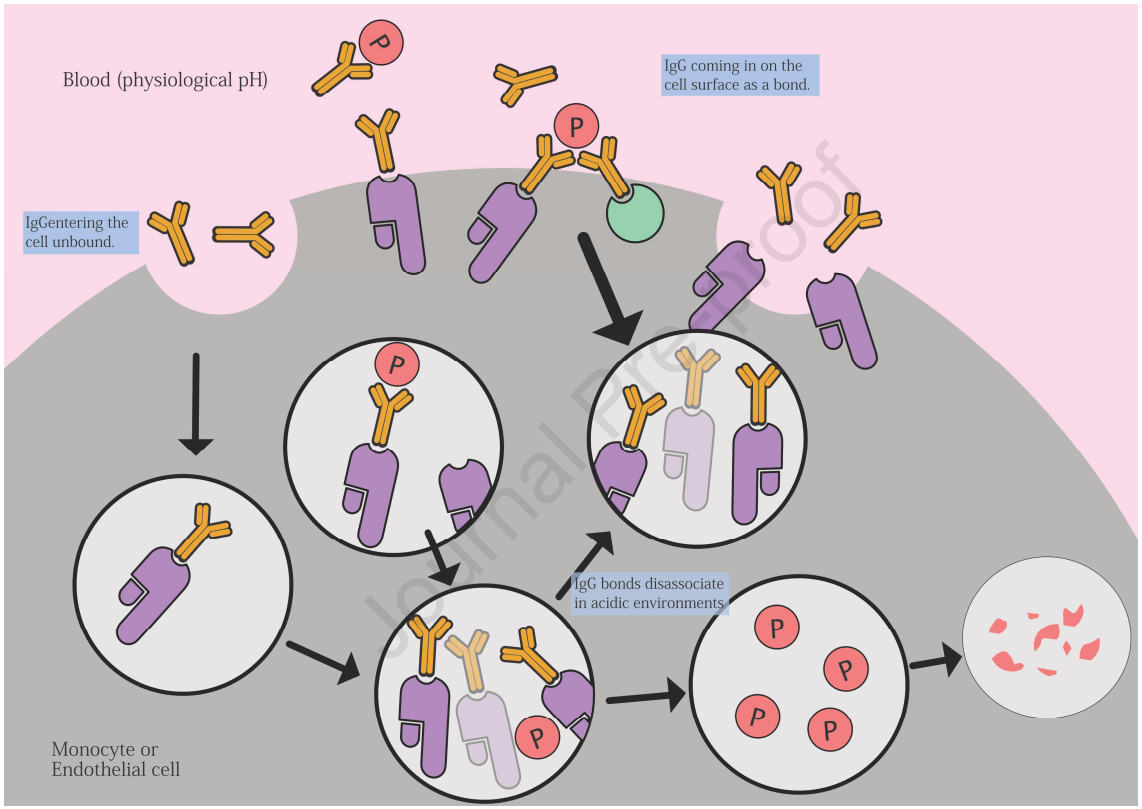


Figure 4: Neonatal Fc Receptor with free IgG and bound IgG entering cell for recycling



The neonatal Fc receptor (FcRn) is a unique Fc gamma receptor which binds IgG only at acidic pH. Free, unattached IgG is brought into an endothelial cell by pinocytosis from plasma, and passed into an endocytic vesicle called an endosome. We believe that IgG bound to blood cells enters the acidified endosome via attachment to other external FcRs and then, like IgG that enters by pinocytosis, binds to FcRn on the inner surface of the acidified endosome. The FcRn then traffics the IgG to the cell surface where it detachs from FcRn since it is in neutral (extracellular) Ph. Artwork by Iris Az.