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Foetal and neonatal alloimmune thrombocytopenia

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A B S T R A C T

Foetal or neonatal thrombocytopenia results from alloimmunisation during pregnancy. Maternal alloantibodies can be formed following exposure to paternally derived human platelet antigens (HPAs) on foetal platelets, in case of incompatible HPA type. These alloantibodies are of the immunoglobulin G subclass and can therefore enter the foetal circulation through active placental transport mediated by the neonatal Fc-receptor. After entering the foetal circulation, these alloantibodies can cause destruction of foetal platelets and potentially damage other foetal cells containing the specific antigen. Subsequent clinical presentation in foetuses or neonates can vary widely, from an asymptomatic thrombocytopenia to a broad spectrum of bleeding complications. Most frequently encountered are minor skin haemorrhages, such as hematomas or petechiae, but also more devastating haemorrhages can occur. Of these, an intracranial haemorrhage is the most feared complication because of its high risk of life-long major neurological handicaps or perinatal death.

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Background

Foetal and neonatal alloimmune thrombocytopenia (FNAIT) is the most important and most frequent cause of foetal and early neonatal thrombocytopenia in term-born infants. As in adults,

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normal foetal and neonatal platelet counts range from 150×10^9/L to 450×10^9/L. Foetal or neonatal thrombocytopenia is defined as a platelet count $<150 \times 10^9$/L, and a platelet count $<50 \times 10^9$/L is called a severe thrombocytopenia.

Pathophysiology

FNAIT is a form of alloimmunisation during pregnancy. Consecutive conditions to be fulfilled are an incompatibility between mother and foetus/child, maternal alloantibody formation, active placental transport of antibodies into the foetal circulation and destruction of foetal platelets. The incompatibility comprises human platelet antigens (HPAs). Exposure to the foreign, paternally derived HPA can occur physiologically as well as in pathological conditions. Foetal blood cells enter the maternal circulation, a phenomenon called fetomaternal haemorrhage. This can occur spontaneously and often asymptomatically in healthy pregnancies, during or after delivery, as a result of invasive procedures or after abdominal trauma. In addition, the maternal circulation is exposed to the foetal side of the placenta, in particular to the syncytiotrophoblast cells, which express the integrin β3 containing various HPA epitopes [1].

To date, 37 different HPAs have been identified and known to cause FNAIT. Usually, a distinction between high- and low-frequency antigens is made. Twelve high-frequency HPAs are clustered into six biallelic groups (HPA-1,2,3,4,5 and 15). The epitope of the antigens on platelets is located on glycoprotein structures that are expressed on the platelet membranes. These glycoproteins, also called integrins, are present in complexes: IIb/IIIa, Ib/IX, Ia/IIa and CD109. The glycoprotein that carries the most HPAs is glycoprotein IIIa (integrin β3 or CD61), including HPA-1 [2]. In the Caucasian population, HPA-1a is the most important antigen, responsible for approximately 80% of the cases of FNAIT, followed by HPA-5b, which accounts for circa 10% [3–5]. Genetic differences between ethnic populations lead to a variance in distributions of these incidences. For example, in the Asian population, anti-HPA-4b is the most frequently involved antibody, followed by anti-HPA-3a and anti-HPA-21b [6–8]. Furthermore, antibodies targeted against glycoprotein IV (also called CD36) are rarely seen in Caucasian FNAIT but are more frequently involved in FNAIT in African and Asian population [9,10].

Endothelial cells

Glycoproteins containing the epitopes of HPAs are not solely present on platelets. Glycoprotein IIIa or integrin β3, containing the most HPAs, is expressed on the membranes of endothelial cells as well as in a complex with integrin αV (αVβ3). This raises an interesting dynamic: considering the fact that the pathogenic mechanism resulting in devastating intracranial haemorrhage (ICH) in FNAIT has never been adequately understood. Generally, alloantibodies are thought to enter the foetal circulation and cause bleeding complications and thrombocytopenia due to the destruction of foetal platelets, a theory that is not exactly airtight, given that only a small proportion of even severely thrombocytopenic newborns actually suffer from bleeding complications; furthermore, severe bleedings have been described in only moderate thrombocytopenia. Additionally, research shows that mice that are completely lacking circulating platelets or fibrinogen survive in utero and do not bleed [11]. This combination of unexplained pathogenic bleeding mechanisms and the fact that the most involved antigen in FNAIT is present on endothelial cells has led to new insights. First, in vitro studies illuminated the direct interaction between anti-HPA-1a and human umbilical vein endothelial cells (HUVECs), demonstrated by a decreased HUVEC spreading as well as a decreased integrity of their monolayer in electric cell-substrate impedance sensing assays [12]. Second, a large in vivo study with murine models of FNAIT showed that ICHs in these mice occurred regardless of platelet count. Also, HPA-1a antibodies inhibited angiogenic signalling, induced endothelial cell apoptosis and decreased vessel density in affected brains as well as retinas [13]. Lastly, a recent study with human sera of women with HPA-1a alloantibodies that caused FNAIT suggested a correlation between the interaction and binding of the antibodies with αVβ3 and whether or not an ICH had occurred in these pregnancies [14].

Syncytiotrophoblast cells

In addition to endothelial cells, αVβ3 is expressed on placental tissue by syncytiotrophoblast cells [1]. Although there is no direct evidence, it has been suggested that anti-HPA-1a might be able to induce placental insufficiency through interaction with these cells. In line, an association with intrauterine growth restriction, as well as with intrauterine foetal demise and miscarriages, has been reported [15,16]. Another group has reported a significant increase of chronic villitis in untreated FNAIT compared to cases treated with IVIG [17]. Furthermore, the expression of HPA-1a on placental tissue might lead to increased and early exposure during pregnancy, which might be an explanation for the high proportion of affected first pregnancies and first-born children in FNAIT.

Incidence

FNAIT is the most important cause of thrombocytopenia in otherwise healthy term infants. Approximately half of the newborns with an early severe thrombocytopenia suffer from FNAIT [18,19].

In the absence of population-based screening, incidence and prevalence numbers have to be extracted from, ideally, large prospective and preferably non-intervention studies. Such studies can be performed either postnatally, first screening for neonatal platelet count, followed by for platelet-specific alloantibody testing, or antenatally, first screening for HPA-type, then testing for antibody formation followed by assessment of neonatal outcome.

Postnatal - When combining results from postnatal screening studies in all newborns, the incidence of severe FNAIT (platelet count $< 50 \times 10^9$/L) was 0.04%, corresponding to 1 in 2500 newborns, which led to an ICH in 25% of these cases [20].

Antenatal – Incidence numbers based on antenatal studies are most adequately estimated by a systematic review that combined a total of 176,084 pregnant women from ten screening studies [21]. The authors found that 2.1% (1 in 50) of pregnant women were HPA-1a negative and therefore at risk for FNAIT. In these women, 9.7% developed anti-HPA-1a alloantibodies (1 in 500). Further, 31% (1 in 1500) of alloimmunised pregnancies developed severe FNAIT, of which approximately 10% (1 in 15,000) led to an ICH or intrauterine foetal demise. While this review combines the largest screening studies performed thus far, only four of the ten studies did not perform an antenatal intervention. This leaves a total of 52,994 women observed, in which only a single ICH occurred. Therefore, no conclusions on natural history of the disease can be made and the above-mentioned estimates are likely an underestimation of the true incidence of FNAIT caused by HPA-1a. The suggestion of underestimation was also put forth by an Irish study, which reported that merely 7% of expected cases were clinically detected [22].

Clinical characteristics

FNAIT has a broad spectrum of clinical presentations. First, an asymptomatic thrombocytopenia might be detected as a chance finding without other signs of FNAIT. In these cases, FNAIT is usually only suspected after exclusion of other causes of foetal and early neonatal thrombocytopenia (Table 1). Second, mild bleeding symptoms might be present in newborns. These children might experience haematomas, petechiae or small visceral bleeding. Also, transient haematuria or bloody stools might be seen [4]. Lastly, FNAIT can present with severe bleeding symptoms, essentially major internal organ haemorrhages. These can virtually occur in every organ (lung, bowel, kidney, etc.), but an ICH is the most feared bleeding because of its associated risk of lifelong disability and mortality [23,24]. Analysis of the short-term outcome in 43 cases of ICH showed that more than one-third cases (35%) resulted in perinatal death within four days of life [24]. Another cohort of 21 consecutive cases of ICH at a single tertiary centre showed an even higher mortality rate of 48% [25]. Long-term follow-up in this study showed that 60% of surviving children had severe neurological developmental impairment, defined as a cerebral palsy, bilateral deafness or blindness or a severe motor and/or cognitive developmental delay (<−2SD) [25]. Over 80% of ICHs are estimated to occur before birth, two-thirds before 34 weeks' gestation and 54% even before 28 weeks' gestation [24,26]. In contrast to severe disease caused by red

Table 1
Causes of foetal and early neonatal thrombocytopenia.

Increased destruction
Immune thrombocytopenia
Maternal autoimmune (ITP, SLE)
Foetal/Neonatal Alloimmune Thrombocytopenia (FNAIT)
Severe fetal haemolytic disease due to red cell alloimmunisation
Alloimmune drug-induced (penicillin, anti-epileptica, quinidine and indomethacin)
Peripheral consumption
Hypersplenism
Kasabach–Merritt
Disseminated intravascular coagulation (DIC)
Thrombosis (e.g. aortic and renal vein)
Decreased production
Genetic disorders (TAR syndrome, trisomy 13,18,21, triploidy, Turner's syndrome, amegakaryocytosis, Wiskott–Aldrich, May–Hegglin, Bernard–Soulier and Alport syndrome)
Bacterial infection (GBS, *E.Coli*, *Listeria and* Syphilis)
Viral infection (CMV, parvo, rubella, HIV an HSV)
Parasite infection (toxoplasmosis)
Asphyxia
Placental insufficiency (pre-eclampsia, IUGR, diabetes, premature birth)

ITP idiopathic thrombocytopenia; SLE, systemic lupus erythematosus; GBS, group-B Streptococcus; TAR, thrombocytopenia-absent radii syndrome; CMV, cytomegalovirus; HSV, Herpes Simplex Virus; HIV, human immunodeficiency virus; IUGR, intrauterine growth restriction.

blood cell alloimmunisation, 23% of ICHs occurred in primigravida women and 63% affected the first-born child.

Diagnosis

In the absence of routine antenatal screening, suspicion of FNAIT usually arises in case of a clinically affected newborn. Therefore, in the majority of the cases, the diagnostic work-up is performed post-natally. However, antenatal suspicion and subsequent diagnostic work-up may be performed as well. The reason can be antenatal ultrasound detection of foetal abnormalities, especially in the brain (Fig. 1) or because a sister of the pregnant women had a pregnancy complicated by FNAIT.

First, when FNAIT is suspected, diagnostic work-up should include HPA-typing of mother, father and child [27]. This way, possible HPA incompatibilities can be established. Second, an antibody screening should be performed to identify maternal platelet-specific alloantibodies, preferably using the MAIPA assay [28]. Also, maternal serum can be tested for auto-antibodies [27]. FNAIT can be confirmed in case of a maternal-neonatal or maternal-paternal HPA-incompatibility combined with the detection of alloantibodies for this specific HPA.

Obstetric management

In current practice, preventive measures are virtually only available for subsequent pregnancies in women with a previously affected child. A rare exception concerns cases in which diagnostic work-up for FNAIT was performed following a sister with an affected child. Pregnancies at risk for FNAIT are best managed in a tertiary centre with both obstetric and neonatal expertise in this disease. First, paternal genotype should be taken into account to assess the risk of an incompatible pregnancy. In case of paternal homozygosity, every next pregnancy for this couple will be incompatible by definition. In case of paternal heterozygosity, however, there is a 50% chance that the foetus is compatible with the mother and the pregnancy is not at risk to be complicated by FNAIT. In these cases, foetal genotype has to be determined to assess the need for monitoring and potential preventive treatment. For HPA-1a, the predominantly involved alloantibody, foetal status can be determined using non-invasive testing in maternal plasma, using cell-free foetal DNA [29]. Despite promising research, in current practice, no

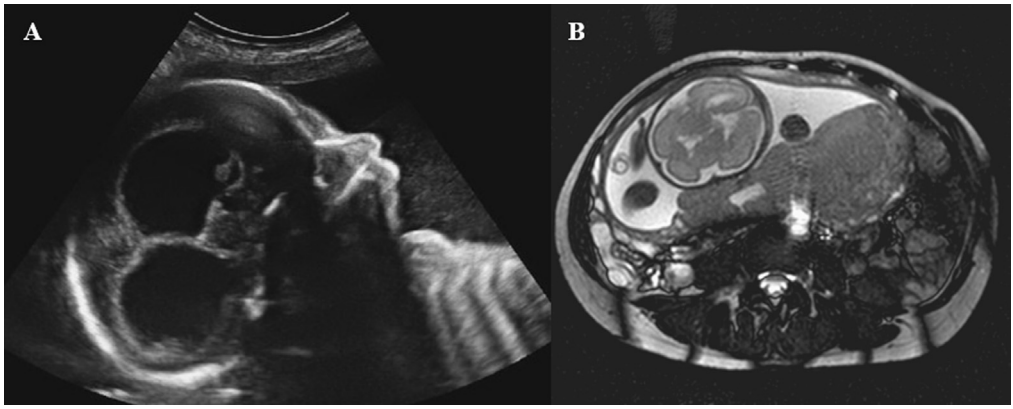

Fig. 1. Antenatal detection of FNAIT. **A.** Ultrasound image of foetal post-haemorrhagic ventriculomegaly, at 28 weeks' gestation, because of foetal alloimmune thrombocytopenia caused by maternal antibodies against HPA-5b. **B.** Prenatal MR image of a 28-week-old foetus with a parenchymal bleeding in the left parietal region with extension into the cortex. The mother was known to be at risk for FNAIT, with HPA-1a alloantibodies.

such non-invasive tests for other HPAs are available yet [30]. In these cases, an amniocentesis is advised to assess the foetal genotype.

Once incompatibility between mother and foetus is confirmed, close ultrasound monitoring, especially of the foetal brain, should be performed every 2–4 weeks. At this stage, clinicians should ideally be able to evaluate and monitor foetal disease severity as well as predict the occurrence of severe bleeding. Unfortunately, unlike in haemolytic disease of the foetus and newborn (HDFN), the red cell counterpart of FNAIT, there are no antenatal non-invasive diagnostic tests available to assess disease severity before severe complications occur. The only possibility is to assess foetal platelet count by foetal blood sampling (FBS), which means puncturing the umbilical cord. Besides the fact that this procedure is risky, in particular when platelets are low, platelet counts are not linearly correlated to disease severity. Because of this lack of reliable non-invasive diagnostic tools to guide obstetric management and treatment, research has focused on assessing factors to select pregnancies at high risk.

Antibody level - In some centres, antibody levels and titres are monitored by titration and quantification. While high titres do seem to be associated with severe FNAIT, this is not a consistent relationship, and there are cases of severe haemorrhages with barely detectable antibody levels [31]. Therefore, monitoring antibody titres, if performed at all, is currently mostly in research setting and rarely influences obstetric treatment.

*HLA-DRB3*0101* - The HLA-DRB3*0101 genotype is positively correlated with the occurrence of alloimmunisation in HPA-1a incompatible pregnancies [32,33]. However, besides this correlation to immunisation, no additional link to disease severity, which would enable identifying immunized pregnancies at high risk, has been made.

Glycosylation – Another proposed predictive laboratory factor is the glycosylation pattern of the Fc-part alloantibodies. Antibodies vary in glycosylation pattern, which influences the affinity and amount of binding to Fc-receptors [34,35]. In FNAIT, a decreased fucosylation and increased galactosylation are reported to correlate to neonatal platelet counts and disease severity [36].

Endothelial function – Most recently, as discussed previously, binding and interaction with endothelial cells have been proposed to be correlated to the occurrence of ICH [14].

Next to these laboratory parameters, various clinical characteristics have been evaluated as well [37]. So far, the only clinical parameter directly correlated to disease severity is the occurrence of an ICH in a previously affected pregnancy. Estimated recurrence rate of an ICH, without antenatal treatment, is as high as 79% [37,38]. Therefore, the only parameter that can currently guide the antenatal treatment regime is the occurrence of an ICH in a sibling.

Antenatal treatment

In current practice, without tools to assess which alloimmunised pregnancies are at truly high risk for bleeding complications, preventive antenatal treatment is initiated in all pregnancies with known platelet-specific alloantibodies and an antigen-positive foetus. The preventive toolkit in these pregnancies consists of invasive and non-invasive treatment options. An overview of the antenatal management for pregnancies complicated in the Dutch national referral centre is displayed in Fig. 2.

Foetal blood sampling – intrauterine platelet transfusion

The first prenatal strategy was adapted from the very successful and still routinely applied treatment of foetal anaemia. In 1984, Daffos was the first to perform ultrasound-guided FBS followed by an intrauterine platelet transfusion [39]. This strategy allowed both the assessment of foetal platelet count and the ability for direct treatment if necessary. Compared to serial intrauterine transfusions as treatment for foetal anaemia in HDFN, there are two major differences to its application in FNAIT. First, half-life of platelets is a few days, considerably shorter than that of red blood cells [40]. This results in the need for at least weekly foetal platelet transfusions. And even after a week, pre-transfusion platelet counts are often well-below 50×10^9/L, indicating that even weekly transfusions will not be enough to

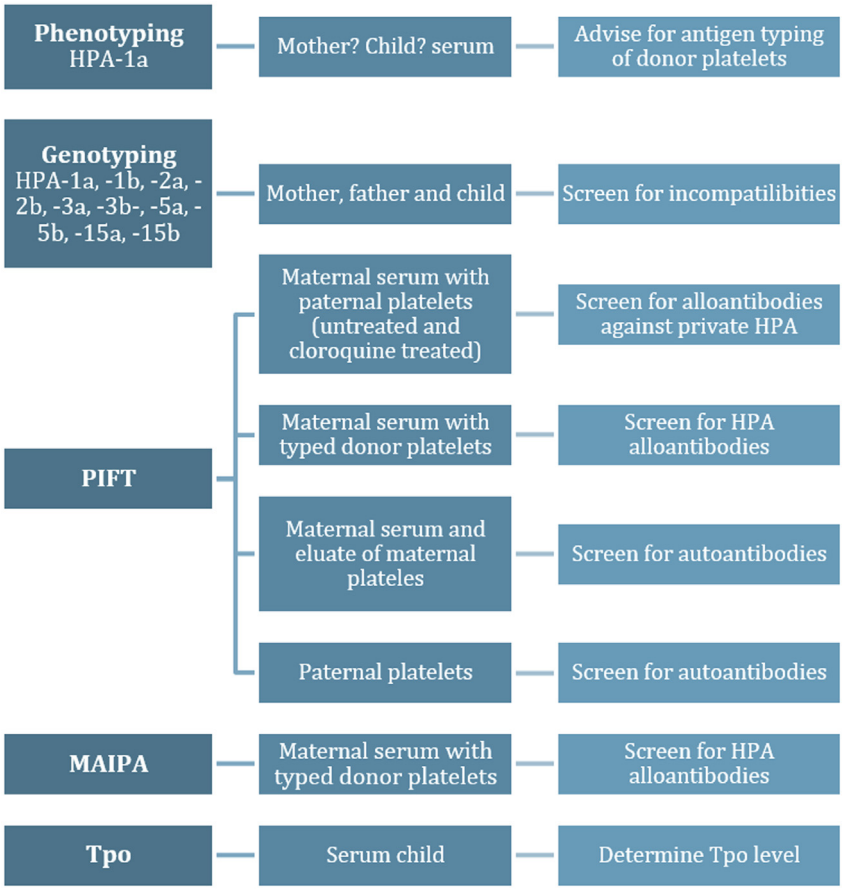

Fig. 2. Overview of obstetric management in FNAIT.

maintain safe platelet counts [41,42]. Second, cordocentesis in a thrombocytopenic foetus introduces a high risk of complications. These complications include a risk of bleeding, including exsanguination, due to this thrombocytopenic status. Also, foetal bradycardia is more often noted, which might possibly be attributed to the higher plasma volume transfused [41]. This combined with the required interval of transfusions leads to a cumulative estimated complication risk as high as 11% per pregnancy [43].

Intravenous immunoglobulins

Endeavouring to replace the risky treatment strategy with a safer non-invasive alternative, Bussel was the first, in 1988, to report the effect of maternal IVIG in a pregnancy complicated by FNAIT [40]. The treatment, as well as the dose of 1 g/kg maternal body weight, was adapted from the treatment of idiopathic thrombocytopenic purpura, caused by platelet autoantibodies. Antenatal treatment with IVIG rapidly gained ground and is currently standard practice. A recent systematic review, analysing 315 pregnancies treated with IVIG only, reported a success rate of 99% in preventing ICH from occurring [43]. Although most centres have completely replaced invasive treatment by IVIG administration (with or without corticosteroids), differences on its optimal regime with regards to dose and start of treatment still remain. The most commonly used dose is 1 g/kg per week. However, whether this could be reduced or increased in certain subgroups remains unclear. Side-effects are clearly dose-dependent, and IVIG is a relatively expensive drug, produced from human multi-donor plasma. Kamphuis and colleagues showed that a lower dose of 0.5 g/kg per week was not inferior to the 1 g/kg per week in standard risk pregnancies (i.e. previous sibling without ICH) [44]. Also, a higher dose of 2 g/kg per week has been suggested in pregnancies with a sibling with an early antenatal ICH [45,46]. Additionally, as the only evident risk factor for developing an ICH thus far is the occurrence of an ICH in a sibling, pregnancies are usually stratified into two groups with two different IVIG treatment strategies regarding the dose as well as the gestational age at the start. The start of IVIG is mainly based on the estimated onset of ICHs. The largest study describing ICHs reported the gestational age of onset to be less than 28 weeks in over half of the cases [24]. This would support starting IVIG earlier than 28 weeks (commonly used in Europe), e.g. 24 or even 20 weeks, the latter being most common in the United States of America.

The working mechanism of IVIG remains unsolved, although there are several theories [47]. One theory states that the presence of IVIG might dilute and lower the HPA-alloantibody levels in maternal serum and would therefore result in a lower titre and level of antibodies. Another theory proposes that IVIG might compete with HPA-alloantibodies on the neonatal Fc-receptor on the placenta, leading to a lower amount of antibodies transported into the foetal circulation. Third and fourth, this concept of competition might occur in the foetal circulation and spleen as well as leading to fewer antibodies binding to foetal platelets or fewer platelets destroyed in the spleen [47–49].

Despite IVIG being common practice in most specialised foetal therapy centres, the use of IVIG in pregnancies at risk for FNAIT is still off-label. This might be because of uncertainty on the possible, long-term immunostimulative or immunosuppressive effects of IVIG to the maturing foetal immune system. One cohort study assessed neurodevelopmental outcome in 37 children exposed to IVIG during foetal life and reported no clinically apparent adverse effects in early childhood [48].

Corticosteroids

Another non-invasive treatment is the administration of corticosteroids. These can be administered either as separate therapy or, more often, in addition to IVIG. When comparing IVIG to corticosteroids, both applied as singular treatment, corticosteroids are less efficient [50–52]. As an addition to IVIG, corticosteroids are thought to reduce possible headache complaints, as side effect from IVIG, and support its efficiency. This strategy of adding steroids to IVIG treatment was also first described by Bussel and colleagues [53]. They started with dexamethasone 1.5–5 mg/kg, but because of limited beneficial effects and significant side effects such as oligohydramnios, this therapy was stopped [54]. Dexamethasone was replaced by prednisone, which seemed to have less side effects at a dose of 0.5 mg/kg/day. Its benefit, however, is debated. Only one study reported a significant increase in platelet count when adding prednisone to IVIG. It has to be remarked, however, that in

order to find this significant increase they used the following self-instated, non-predefined outcome measure: platelet count >25 $\times$ 10^9/L at second sampling, or an increase by > 10 $\times$ 10^9/L or a platelet count >40 $\times$ 10^9/L that was not decreased by > 10 $\times$ 10^9/L [50]. All other studies comparing IVIG treatment to a treatment with IVIG and steroids, including a (underpowered) randomised controlled trial, did not find any significant differences in platelet count, ICH or mortality [45,46,51,53–55].

Mode and timing of delivery

The final part of antenatal management comprises the mode and timing of delivery. In the Norwegian screening study, the 170 pregnancies with anti-HPA-1a were all managed by performing a planned, near-term caesarean section, with platelets for immediate transfusion directly available [33]. Fifty-seven neonates with severe FNAIT were born, of which 3 suffered severe complications (ICH or intrauterine death). These numbers were compared to 15 previously published prospective studies combined (10 severe complications in 51 alloimmunised pregnancies), which led to the conclusion that a near-term caesarean resulted in a lower number of severe complications. A few remarks, however, can be made. First, it was not described whether a routine ultrasound of the neonatal brain was performed to detect ICH. Second, a remarkable 21.5% of the neonates suffered prematurity issues and were treated at the neonatal intensive care unit. Third, the design of the 15 prospective studies, that were used as historic controls, was highly heterogenic. Most studies with a high proportion of severe complications identified their cases after birth, based on diagnostic work-up in thrombocytopenic neonates instead of antenatal screening, which introduces a selection bias. The rationale for management of FNAIT by near-term elective caesarean section is based on three perceived advantages. Early delivery reduces exposure time to the 'hostile environment', in which pathogenic antibodies can cross the placenta. Immediate measuring of platelet counts and if needed treatment may reduce the chance of postnatal ICH. And lastly, the concept that vaginal birth may be more traumatic, and that in case of low platelets the birth process may lead to ICH. A major drawback to this management plan is the fact that most ICHs occur before 36 weeks' and even 28 weeks' gestation. In addition, there has been one cohort study performed that did not find an association between vaginal delivery and ICH [56]. So in our view, especially in women with a previous vaginal delivery without a sibling who suffered from ICH, a planned induction of labour can be considered a safe strategy. Although no evidence exists to advise on a mode of delivery in case of an in utero ICH, most centres will be performing a near-term caesarean section in these cases. For all deliveries, it is recommended to avoid potential traumatic events, such as scalp electrodes, scalp blood samplings or assisted vaginal delivery. Directly after delivery, cord blood platelet measurement should be performed.

Neonatal management

Neonatal management is aimed at reducing bleeding tendency by increasing platelet counts [57]. Initial neonatal evaluation should always include clinical assessment, platelet count and cranial ultrasound assessment. The combination of clinical and laboratory parameters determines the need for treatment. First choice of treatment is a platelet transfusion, administered as quickly as possible. Ideally, the transfused product contains platelets that lack the involved HPA (HPA-compatible or HPA-matched transfusion). In case of a subsequent pregnancy or antenatal diagnosis of FNAIT, the responsible alloantibody is known and appropriate platelets can be ordered in time. In newly detected cases, however, a confirmation of the diagnosis by laboratory assays usually takes some time and the responsible alloantibody can be unknown. In these cases, ideally HPA-1bb/5aa platelets are transfused, which are antigen negative for 90% of the FNAIT cases [58]. An alternative can be a platelet transfusion with random-donor platelets. Kiefel and colleagues showed in a small cohort that multiple random platelet transfusions can be sufficient in increasing platelet counts [59]. In line, a recent cohort study found no increased need for additional transfusion of random compared to compatible platelet transfusions [60].

Immunoprophylaxis

In HDFN, the red cell counterpart of FNAIT, the implementation of anti-D prophylaxis has led to a great decrease of mortality and morbidity caused by RhD immunisation [61]. Historically, RhD, like HPA-1a in FNAIT, was the most frequently involved antigen of severe HDFN [62,63]. The possibility of immunoprophylaxis for HPA-1a immunisation in FNAIT as a prophylactic equivalent to anti-D is debated for years and is an important focus for some research groups. In vivo animal studies have reported that antibody mediated immune suppression, induced with anti-D prophylaxis, can also occur in FNAIT mouse models [64]. In these murine studies, β3 integrin-deficient (β3−/−) mice are used to mimic HPA-1a negativity. After injection with HPA-1a positive platelets in β3−/− female mice, the administration of human anti-HPA-1a strongly reduced the β3 antibody response. Besides a drop in β3-antibody level of 90%, there were fewer miscarriages, fewer stillborn pups, fewer pups with ICH and significantly higher platelet counts in the pups. For human follow-up, a recombinant anti-HPA-1a antibody (B2G1Δnab) was produced that was able to block binding of maternal polyclonal HPA-1a antibodies to platelets [65]. In vivo studies were performed and B2G1Δnab was tested successfully in healthy human volunteers, where it effectively cleared HPA-1a positive platelets [66]. An alternative approach is producing an anti-HPA drug using a similar way of production as for Rh-D prophylaxis. Pooled plasma from many donors is used to extract anti-HPA IgG, with which a multi-centre international study is currently being planned (www.profnait.eu).

Although the effectiveness of immunoprophylaxis after delivery is proposed, no consensus exists regarding the optimal timing. The concept of prophylaxis after delivery emerged from the largest prospective, Norwegian, screening study, that reported that most alloimmunisations in HPA-1a negative primigravid women occurred at or soon after delivery [33]. Although skewed toward more severe cases, retrospective data report a higher number of affected newborns after first pregnancies. The previously mentioned analysis of 43 ICHs caused by FNAIT, for instance, illuminated that two-thirds of these severe bleedings occurred in the first-born child [24]. In these cases, a prophylaxis after delivery would not have prevented the bleedings, which is eminently the target of the intervention. For a program of antenatal prophylaxis, such as now common in Rh-D negative pregnancies, it is vital that good safety data become available because unlike the D-antigen on red cells, the HPA-1 antigen is also present on other cells than platelets.

Prenatal screening

The debate on whether or not implementation of prenatal screening in order to timely detect and prevent first occurrence of FNAIT would be (cost-effective and beneficial has been going on for decades [58,67–69]. The massively reduced mortality and morbidity caused by (RhD-as well as non-RhD-mediated) HDFN, after introduction of population-based screening programs, is a great example [70]. Although pathophysiologic mechanisms are principally comparable, there are major differences as well. For example, severe cases of ICH in FNAIT occur in first-born children in over half of the children, underlying the need for a screening and possible prophylaxis program to be early antenatal instead of late antenatal or postnatal [24,25]. Also, monitoring of immunised pregnancies to identify pregnancies at high risk for complications is different. Whereas, in red blood cell immunisation, laboratory assays (antibody titres, quantitation and ADCC-test) as well as ultrasound markers (Doppler measurement of the middle cerebral artery) are available, no parameters can determine the risk of bleeding in immunised pregnancies in FNAIT. This lack of risk-assessment and monitoring possibilities together with a relatively unknown knowledge on incidence natural history of the disease are important focus points for research. With more answers, it might be possible to solve the ongoing debate and answer questions on feasibility and effectiveness of population-based HPA-screening in pregnancy for FNAIT. Given the high burden and costs of surviving children with brain damage due to ICH, the relatively straightforward screening (adding HPA-typing to blood group typing), the limited proportion of HPA-1a negative women actually producing antibodies and the highly effective IVIG treatment, we expect that routine screening will be a cost-effective strategy.

Summary

FNAIT is the most important cause of early neonatal thrombocytopenia. Maternal alloantibodies targeting foetal platelets as well as foetal endothelial and placental tissue might lead to a wide spectrum of clinical consequences. Of these, an ICH is the most feared bleeding complication with a high risk of associated morbidity and mortality. Fortunately, in subsequent pregnancies, with known immunisation, severe bleeding complications can be effectively prevented with antenatal weekly intravenous immunoglobulin infusions. Because of still insufficient knowledge on natural history and the absence of safe diagnostic tools to assess pregnancies at high risk, no population-based screening in order to prevent bleeding complication from occurring in first pregnancies as well is implemented yet. Future research might not only provide evidence on possible feasibility and efficacy of screening but also help guiding and configuring potential prenatal HPA-screening in pregnancy.

Practice points

- Most cases of FNAIT are caused by maternal alloantibodies targeted against foetal HPA-1a, which is not only present on foetal platelets but on foetal endothelial cells and syncytiotrophoblast cells as well.
- Clinical consequences of FNAIT vary from asymptomatic thrombocytopenia to (minor) skin bleeding or severe ICH. New follow-up data of children suffering from ICH illustrates that 60% develop severe neurodevelopmental impairment, and merely 20% survive without any neurological or psychological disorder.
- Antenatal management with weekly intravenous immunoglobulin infusions in pregnancies at risk for FNAIT is highly effective in preventing severe bleeding complications without the complication risks associated with invasive foetal procedures.

Research agenda

- The effect of HPA-1a antibodies on cells containing integrin with the HPA-1a epitope as well, such as endothelial cells and syncytiotrophoblast cells.
- Incidence and natural history of FNAIT, evaluated with large, prospective, observational and ideally non-intervention cohort studies.
- Development of a risk assessment tool to enable monitoring of immunised pregnancies and identifying pregnancies at high risk for bleeding complications.

Conflicts of interest

The authors have no conflicts of interest.

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References

[1] Campbell S, Swann HR, Seif MW, Kimber SJ, Aplin JD. Cell adhesion molecules on the oocyte and preimplantation human embryo. Hum Reprod 1995;10:1571–8.

[2] Newman PJ, Derbes RS, Aster RH. The human platelet alloantigens, PlA1 and PlA2, are associated with a leucine33/proline33 amino acid polymorphism in membrane glycoprotein IIIa, and are distinguishable by DNA typing. J Clin Invest 1989;83:1778–81.
[3] Merieux Y, Debost M, Bernaud J, Raffin A, Meyer F, Rigal D. Human platelet antigen frequencies of platelet donors in the French population determined by polymerase chain reaction with sequence-specific primers. Pathol Biol (Paris) 1997;45:697–700.
[4] Mueller-Eckhardt C, Kiefel V, Grubert A, Kroll H, Weisheit M, Schmidt S, et al. 348 cases of suspected neonatal alloimmune thrombocytopenia. Lancet (London, England) 1989;1:363–6.
[5] Davoren A, Curtis BR, Aster RH, McFarland JG. Human platelet antigen-specific alloantibodies implicated in 1162 cases of neonatal alloimmune thrombocytopenia. Transfusion 2004;44:1220–5.
[6] Kunishima S, Hayakawa A, Fujita K, Saito H. Transient macrothrombocytopenia associated with maternal-neonatal HPA-21bw incompatibility. Thromb Res 2013;131:e286–8.
[7] Ohto H. Neonatal alloimmune thrombocytopenia. Nihon Rinsho 1997;55:2310–4.
[8] Ohto H, Miura S, Ariga H, Ishii T, Fujimori K, Morita S. The natural history of maternal immunization against foetal platelet alloantigens. Transfus Med 2004;14:399–408.
[9] Tomiyama Y, Take H, Ikeda H, Mitani T, Furubayashi T, Mizutani H, et al. Identification of the platelet-specific alloantigen, Naka, on platelet membrane glycoprotein IV. Blood 1990;75:684–7.
[10] Curtis BR, Aster RH. Incidence of the Nak(a)-negative platelet phenotype in African Americans is similar to that of Asians. Transfusion 1996;36:331–4.
[11] Shivdasani RA, Rosenblatt MF, Zucker-Franklin D, Jackson CW, Hunt P, Saris CJ, et al. Transcription factor NF-E2 is required for platelet formation independent of the actions of thrombopoietin/MGDF in megakaryocyte development. Cell 1995;81:695–704.
[12] van Gils JM, Stutterheim J, van Duijn TJ, Zwaginga JJ, Porcelijn L, de Haas M, et al. HPA-1a alloantibodies reduce endothelial cell spreading and monolayer integrity. Mol Immunol 2009;46:406–15.
[13] Yougbare I, Lang S, Yang H, Chen P, Zhao X, Tai WS, et al. Maternal anti-platelet beta3 integrins impair angiogenesis and cause intracranial hemorrhage. J Clin Invest 2015;125:1545–56.
[14] Santoso S, Wihadmadyatami H, Bakchoul T, Werth S, Al-Fakhri N, Bein G, et al. Antiendothelial alphavbeta3 antibodies are a major cause of intracranial bleeding in fetal/neonatal alloimmune thrombocytopenia. Arterioscler Thromb Vasc Biol 2016;36:1517–24.
[15] Murphy MF, Hambley H, Nicolaides K, Waters AH. Severe fetomaternal alloimmune thrombocytopenia presenting with fetal hydrocephalus. Prenat Diagn 1996;16:1152–5.
[16] Tiller H, Killie MK, Husebekk A, Skogen B, Ni H, Kjeldsen-Kragh J, et al. Platelet antibodies and fetal growth: maternal antibodies against fetal platelet antigen 1a are strongly associated with reduced birthweight in boys. Acta Obstet Gynecol Scand 2012;91:79–86.
[17] Althaus J, Weir EG, Askin F, Kickler TS, Blakemore K. Chronic villitis in untreated neonatal alloimmune thrombocytopenia: an etiology for severe early intrauterine growth restriction and the effect of intravenous immunoglobulin therapy. Am J Obstet Gynecol 2005;193:1100–4.
[18] Burrows RF, Kelton JG. Fetal thrombocytopenia and its relation to maternal thrombocytopenia. N Engl J Med 1993;329:1463–6.
[19] Dreyfus M, Kaplan C, Verdy E, Schlegel N, Durand-Zaleski I, Tchernia G. Frequency of immune thrombocytopenia in newborns: a prospective study. Immune Thrombocytopenia Working Group. Blood 1997;89:4402–6.
[20] Kamphuis MM, Paridaans NP, Porcelijn L, Lopriore E, Oepkes D. Incidence and consequences of neonatal alloimmune thrombocytopenia: a systematic review. Pediatrics 2014;133:715–21.
[21] Kamphuis MM, Paridaans N, Porcelijn L, De Haas M, Van Der Schoot CE, Brand A, et al. Screening in pregnancy for fetal or neonatal alloimmune thrombocytopenia: systematic review. BJOG An Int J Obstet Gynaecol 2010;117:1335–43.
[22] Davoren A, McParland P, Barnes CA, Murphy WG. Neonatal alloimmune thrombocytopenia in the Irish population: a discrepancy between observed and expected cases. J Clin Pathol 2002;55:289–92.
[23] Winkelhorst D, Kamphuis MM, de Kloet LC, Zwaginga JJ, Oepkes D, Lopriore E. Severe bleeding complications other than intracranial hemorrhage in neonatal alloimmune thrombocytopenia: a case series and review of the literature. Transfusion 2016;56(5):1230–5.
[24] Tiller H, Kamphuis MM, Flodmark O, Papadogiannakis N, David AL, Sainio S, et al. Fetal intracranial haemorrhages caused by fetal and neonatal alloimmune thrombocytopenia: an observational cohort study of 43 cases from an international multicentre registry. BMJ open 2013;3.
[25] Winkelhorst D, Kamphuis MM, Steggerda SJ, Rijken M, Oepkes D, Lopriore E, et al. Perinatal outcome and long-term neurodevelopment after intracranial haemorrhage due to fetal and neonatal alloimmune thrombocytopenia. Fetal Diagn Ther 2018:1–8.
[26] Spencer JA, Burrows RF. Feto-maternal alloimmune thrombocytopenia: a literature review and statistical analysis. Aust N Z J Obstet Gynaecol 2001;41:45–55.
[27] Porcelijn L, van Beers W, Gratama JW, van't Veer M, De Smet A, Sintnicolaas K. External quality assessment of platelet serology and human platelet antigen genotyping: a 10-year review. Transfusion 2008;48:1699–706.
[28] Kiefel V, Santoso S, Weisheit M, Mueller-Eckhardt C. Monoclonal antibody–specific immobilization of platelet antigens (MAIPA): a new tool for the identification of platelet-reactive antibodies. Blood 1987;70:1722–6.
[29] Scheffer PG, Ait Soussan A, Verhagen OJ, Page-Christiaens GC, Oepkes D, de Haas M, et al. Noninvasive fetal genotyping of human platelet antigen-1a. BJOG An Int J Obstet Gynaecol 2011;118:1392–5.
[30] Wienzek-Lischka S, Krautwurst A, Frohner V, Hackstein H, Gattenlohner S, Brauninger A, et al. Noninvasive fetal genotyping of human platelet antigen-1a using targeted massively parallel sequencing. Transfusion 2015;55:1538–44.
[31] Bessos H, Turner M, Urbaniak SJ. Is there a relationship between anti-HPA-1a concentration and severity of neonatal alloimmune thrombocytopenia? Immunohematology Am Red Cross 2005;21:102–9.
[32] L'Abbe D, Tremblay L, Filion M, Busque L, Goldman M, Decary F, et al. Alloimmunization to platelet antigen HPA-1a (PIA1) is strongly associated with both HLA-DRB3*0101 and HLA-DQB1*0201. Hum Immunol 1992;34:107–14.

[33] Kjeldsen-Kragh J, Killie MK, Tomter G, Golebiowska E, Randen I, Hauge R, et al. A screening and intervention program aimed to reduce mortality and serious morbidity associated with severe neonatal alloimmune thrombocytopenia. Blood 2007;110:833–9.
[34] Nimmerjahn F, Ravetch JV. Anti-inflammatory actions of intravenous immunoglobulin. Annu Rev Immunol 2008;26:513–33.
[35] Kapur R, Kustiawan I, Vestrheim A, Koeleman CA, Visser R, Einarsdottir HK, et al. A prominent lack of IgG1-Fc fucosylation of platelet alloantibodies in pregnancy. Blood 2014;123:471–80.
[36] Sonneveld ME, Natunen S, Sainio S, Koeleman CA, Holst S, Dekkers G, et al. Glycosylation pattern of anti-platelet IgG is stable during pregnancy and predicts clinical outcome in alloimmune thrombocytopenia. Br J Haematol 2016;174(2):310–20.
[37] Delbos F, Bertrand G, Croisille L, Ansart-Pirenne H, Bierling P, Kaplan C. Fetal and neonatal alloimmune thrombocytopenia: predictive factors of intracranial hemorrhage. Transfusion 2016;56:59–66.
[38] Radder CM, Brand A, Kanhai HH. Will it ever be possible to balance the risk of intracranial haemorrhage in fetal or neonatal alloimmune thrombocytopenia against the risk of treatment strategies to prevent it? Vox Sang 2003;84:318–25.
[39] Daffos F, Forestier F, Muller JY, Reznikoff-Etievant M, Habibi B, Capella-Pavlovsky M, et al. Prenatal treatment of alloimmune thrombocytopenia. Lancet (London, England) 1984;2:632.
[40] Bussel JB, Berkowitz RL, McFarland JG, Lynch L, Chitkara U. Antenatal treatment of neonatal alloimmune thrombocytopenia. N Engl J Med 1988;319:1374–8.
[41] Sainio S, Teramo K, Kekomaki R. Prenatal treatment of severe fetomaternal alloimmune thrombocytopenia. Transfus Med 1999;9:321–30.
[42] Knight M, Pierce M, Allen D, Kurinczuk JJ, Spark P, Roberts DJ, et al. The incidence and outcomes of fetomaternal alloimmune thrombocytopenia: a UK national study using three data sources. Br J Haematol 2011;152:460–8.
[43] Winkelhorst D, Murphy MF, Greinacher A, Shehata N, Bakchoul T, Massey E, et al. Antenatal management in fetal and neonatal alloimmune thrombocytopenia: a systematic review. Blood 2017;129:1538–47.
[44] Kamphuis M, Paridaans N, Winkelhorst D, Wikman A, Tiblad E, Lopriore E, et al. Lower-dose intravenous immunoglobulins for the treatment of fetal and neonatal alloimmune thrombocytopenia: a cohort study. Transfusion 2016;56:2308–13.
[45] Berkowitz RL, Lesser ML, McFarland JG, Wissert M, Primiani A, Hung C, et al. Antepartum treatment without early cordocentesis for standard-risk alloimmune thrombocytopenia: a randomized controlled trial. Obstet Gynecol 2007;110:249–55.
[46] Bussel JB, Berkowitz RL, Hung C, Kolb EA, Wissert M, Primiani A, et al. Intracranial hemorrhage in alloimmune thrombocytopenia: stratified management to prevent recurrence in the subsequent affected fetus. Am J Obstet Gynecol 2010;203:135.e1–135.e14.
[47] Radder CM, Kanhai HH, Brand A. On the mechanism of high dose maternal intravenous immunoglobulin (IVIG) in alloimmune thrombocytopenia. In: Radder CMt, editor. Management of fetal alloimmune thrombocytopenia. Amsterdam: Print Partners Ipskamp; 2004. p. 69–81.
[48] Radder CM, de Haan MJ, Brand A, Stoelhorst GM, Veen S, Kanhai HH. Follow up of children after antenatal treatment for alloimmune thrombocytopenia. Early Hum Dev 2004;80:65–76.
[49] Ni H, Chen P, Spring CM, Sayeh E, Semple JW, Lazarus AH, et al. A novel murine model of fetal and neonatal alloimmune thrombocytopenia: response to intravenous IgG therapy. Blood 2006;107:2976–83.
[50] Berkowitz RL, Kolb EA, McFarland JG, Wissert M, Primani A, Lesser M, et al. Parallel randomized trials of risk-based therapy for fetal alloimmune thrombocytopenia. Obstet Gynecol 2006;107:91–6.
[51] Bertrand G, Drame M, Martageix C, Kaplan C. Prediction of the fetal status in noninvasive management of alloimmune thrombocytopenia. Blood 2011;117:3209–13.
[52] Kaplan C, Daffos F, Forestier F, Cox WL, Lyon-Caen D, Dupuy-Montbrun MC, et al. Management of alloimmune thrombocytopenia: antenatal diagnosis and in utero transfusion of maternal platelets. Blood 1988;72:340–3.
[53] Bussel JB, Berkowitz RL, Lynch L, Lesser ML, Paidas MJ, Huang CL, 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.
[54] Wenstrom KD, Weiner CP, Williamson RA. Antenatal treatment of fetal alloimmune thrombocytopenia. Obstet Gynecol 1992;80:433–5.
[55] Lynch L, Bussel JB, McFarland JG, Chitkara U, Berkowitz RL. Antenatal treatment of alloimmune thrombocytopenia. Obstet Gynecol 1992;80:67–71.
[56] van den Akker ESA, Oepkes D, Brand A, Kanhai HHH. Vaginal delivery for fetuses at risk of alloimmune thrombocytopenia? BJOG An Int J Obstet Gynaecol 2006;113:781–3.
[57] te Pas AB, Lopriore E, van den Akker ES, Oepkes D, Kanhai HH, Brand A, et al. Postnatal management of fetal and neonatal alloimmune thrombocytopenia: the role of matched platelet transfusion and IVIG. Eur J Pediatr 2007;166:1057–63.
[58] Murphy MF, Williamson LM. Antenatal screening for fetomaternal alloimmune thrombocytopenia: an evaluation using the criteria of the UK National Screening Committee. Br J Haematol 2000;111:726–32.
[59] Kiefel V, Bassler D, Kroll H, Paes B, Giers G, Ditomasso J, et al. Antigen-positive platelet transfusion in neonatal alloimmune thrombocytopenia (NAIT). Blood 2006;107:3761–3.
[60] Winkelhorst D, Oostweegel M, Porcelijn L, Middelburg RA, Zwaginga JJ, Oepkes D, et al. Treatment and outcomes of fetal/neonatal alloimmune thrombocytopenia: a nationwide cohort study in newly detected cases. Br J Haematol 2018 [Epub ahead of print].
[61] Bowman J. Thirty-five years of Rh prophylaxis. Transfusion 2003;43:1661–6.
[62] de Haas M, Thurik FF, Koelewijn JM, van der Schoot CE. Haemolytic disease of the fetus and newborn. Vox Sang 2015;109:99–113.
[63] Zwiers C, Lindenburg ITM, Klumper FJ, de Haas M, Oepkes D, Van Kamp IL. Complications of intrauterine intravascular blood transfusion: lessons learned after 1678 procedures. Ultrasound Obstet Gynecol–Off J Int Soc Ultrasound Obstet Gynecol 2017;50:180–6.

[64] Tiller H, Killie MK, Chen P, Eksteen M, Husebekk A, Skogen B, 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.
[65] Ghevaert C, Wilcox DA, Fang J, Armour KL, Clark MR, Ouwehand WH, et al. Developing recombinant HPA-1a-specific antibodies with abrogated Fcgamma receptor binding for the treatment of fetomaternal alloimmune thrombocytopenia. J Clin Invest 2008;118:2929–38.
[66] Ghevaert C, Herbert N, Hawkins L, Grehan N, Cookson P, Garner SF, et al. Recombinant HPA-1a antibody therapy for treatment of fetomaternal alloimmune thrombocytopenia: proof of principle in human volunteers. Blood 2013;122:313–20.
[67] Gafni A, Blanchette VS. Screening for neonatal alloimmune thrombocytopenia: an economic perspective. Curr Stud Hematol Blood Transfus 1988:140–7.
[68] Husebekk A, Killie MK, Kjeldsen-Kragh J, Skogen B. Is it time to implement HPA-1 screening in pregnancy? Curr Opin Hematol 2009;16:497–502.
[69] Skogen B, Killie MK, Kjeldsen-Kragh J, Ahlen MT, Tiller H, Stuge TB, et al. Reconsidering fetal and neonatal alloimmune thrombocytopenia with a focus on screening and prevention. Expert Rev Hematol 2010;3:559–66.
[70] Koelewijn JM, Vrijkotte TG, van der Schoot CE, Bonsel GJ, de Haas M. Effect of screening for red cell antibodies, other than anti-D, to detect hemolytic disease of the fetus and newborn: a population study in The Netherlands. Transfusion 2008;48:941–52.