Persistent neonatal thrombocytopenia can be caused by IgA antiplatelet antibodies in breast milk of immune thrombocytopenic mothers

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Key Points

- Persistent thrombocytopenia was observed in breastfed neonates of ITP women.
- Breast milk of ITP women may contain immunoglobulin A antiplatelet antibodies, which target αIIbβ3 integrin.
- Milk samples from 3 groups of women: ITP group, 7 women who had ITP during pregnancy; R-ITP group, 6 women who recovered from ITP before pregnancy; and 9 healthy controls. We found increased levels of antiplatelet antibodies of the immunoglobulin A type in the milk of ITP patients compared with the other 2 groups. Similar increase was demonstrated for antibodies binding to αIIbβ3 expressed in cultured cells. Thus, transfer of antiplatelet antibodies from ITP mothers by breastfeeding can be associated with persistent neonatal thrombocytopenia.

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Introduction

Immune thrombocytopenia (ITP) is present in 4.1% of thrombocytopenic pregnant women.1 In ITP patients, immunoglobulin G (IgG) autoantibodies can be formed, targeting platelet αIIbβ3 or glycoprotein Ib-IX. Maternal IgG can be actively transported across the placenta resulting in neonatal thrombocytopenia. Following decline in neonatal platelet count during the 7 days after delivery, the count gradually increases corresponding to the half-life of the passively transported maternal antiplatelet IgG. Usually, normalization of the infant’s platelet count occurs within 2 months.2

We recently observed a neonate of a mother with ITP whose platelet count was persistently low for 4 months and recovered upon discontinuation of breastfeeding. The objective of our study was to discern the mechanism responsible for the persistently low platelet count in breastfed newborns and examine additional thrombocytopenic neonates whose mothers had ITP during pregnancy. Another group of patients were women who had a history of ITP yet had normal platelet counts during the current pregnancy and no neonatal ITP.

Study design

Index patient

A.H. (age 36 years) had severe ITP during pregnancy and was treated with corticosteroids and intravenous gamma globulin (IVGG) infusions. At term, her platelet count was 16,000/μL and her newborn’s count was 42,000/μL. The infant’s counts reached a nadir of 25,000/μL at 1 month. At 3 months, breastfeeding was discontinued, and within 1 month the platelet count recovered. A milk sample was taken 1 month after delivery.

Groups of patients

Milk samples were obtained from 6 additional ITP patients whose neonates had thrombocytopenia (Table 1). Maternal ITP was diagnosed by exclusion of other causes of thrombocytopenia, presence of purpura, and no antiphospholipid antibodies.

Six other patients (designated R-ITP) had a history of ITP but recovered before their current pregnancy. Normal platelet counts were observed in these women and their neonates. Milk was also obtained from 9 healthy women. The study was approved by the institutional review boards of Laniado Hospital and Tel Aviv Sourasky Medical Center and conducted in accordance with the Declaration of Helsinki.

The online version of this article contains a data supplement.
Detection of antiplatelet antibodies by using washed platelets

Platelets from 3 healthy donors were prepared by centrifugation and washing and were then mixed together. The washed platelets were incubated with Ig extracted from the milk by using Ig Adem Kit (Odemtech, Pessac, France). Details are provided in supplemental Data available at the Blood Web site.

Antiplatelet antibodies were detected by anti-human total Ig (IgA/IgM/IgG)–fluorescein isothiocyanate (Dako, Glostrup, Denmark), or anti-human IgA–fluorescein isothiocyanate (Millipore, Temecula, CA), or anti-human IgG–fluorescein isothiocyanate (Millipore). The samples were tested by flow cytometry. Samples were considered positive when the results were greater than mean ratios of control samples plus 2 standard deviations.

Results and discussion

Representative patient

A.H. had ITP diagnosed during pregnancy, and her infant had thrombocytopenia that persisted for 4 months. We examined A.H.’s milk sample for antiplatelet antibodies by using an antibody that recognizes IgG, IgA, and IgM. Figure 1A demonstrates a high level of antiplatelet antibodies in the milk-Ig sample from A.H. compared with a milk-Ig sample from a control. Next, we tested whether the antibodies were of IgG, as in the sera of most patients with chronic ITP, or IgA, the predominant type in breast milk. The milk sample from A.H. contained antiplatelet antibodies that were solely IgA (Figure 1B-C). To identify the specific antigen against which the antibodies reacted, we tested the Ig samples with cultured cells expressing integrin αIIbβ3 or compared the results to mock cells. Figure 1D shows that anti-αIIbβ3 antibodies were abundant in A.H.’s milk-Ig sample.

Presence of antiplatelet antibodies in the milk of the 3 groups of patients

Table 1 lists clinical and immunologic data for 7 ITP and 6 R-ITP lactating patients.
Two of ITP women (A.H. and A.L.) exhibited a high frequency of positive results for IgA/IgM/IgG compared with controls ($P < .05$) but low frequency of IgA platelet antibodies in their milk compared with ITP women ($P < .05$) (Figure 1E). The milk-Ig was against $\alpha_{\text{IIb}}$ in only 1 R-ITP woman (M.A.). In controls, 1 of 9 samples was positive for anti-$\alpha_{\text{IIb}}$ antibody, resulting in a significant difference between the ITP group and controls ($P < .01$).

Our study provides the first evidence for the presence of antplatelet antibodies in breast milk of ITP patients associated with neonatal persistent thrombocytopenia. These antibodies were of the IgA type and were recognized by epitopes on $\alpha_{\text{IIb}}$. IgA antibodies can be absorbed along the infant’s gastrointestinal tract and gain access to the circulation. Anti-$\alpha_{\text{IIb}}$ antibody was also found in 1 control sample, which might be explained by the broad spectrum of IgA antibodies found in colostrum samples of healthy mothers, and was suggested to be important for the anti-idiotypic network.

The primary mechanism for platelet destruction in ITP is thought to be autoimmune-dependent phagocytosis, especially in cases of anti-$\alpha_{\text{IIb}}$ antibodies. IgA, which interacts with FcRRII expressed on macrophages, can initiate antibody-dependent cell-mediated cytotoxicity. Conversely, an infant’s immune system is immature during the first few months after birth. Therefore, the mechanism for the breastfed neonate’s thrombocytopenia is an issue for further investigation.

The hypothesis that milk from ITP women can cause thrombocytopenia in the infant was discussed in a series of articles and letters. Kelemen et al. suggested that the colostrum of ITP patients contributed to the lowering of circulating platelet counts in the first postnatal days. This was supported by high levels of Ig found in colostrum-fed infants on postpartum day 5 compared with formula-fed infants.9 Hence, lactation was discouraged. In contrast, Meschengieser and Lazzari11 described an ITP patient who gave birth to a thrombocytopenic preterm infant who was breastfed from day 5 without apparent adverse effects on his platelet count. In our view, breastfeeding should not be discouraged, but when low platelet counts persist, discontinuing breastfeeding is a viable solution.

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Authorship

Contribution: H.H. designed and performed the study, interpreted the findings, and wrote the manuscript; N.R. and U.S. designed the study, interpreted the data, and wrote the manuscript; R.M., Y. Shiff, and S.A. identified patients; A.S. and Y. Schachter contributed to writing the article; and N.S. made the initial observation, collected the data, and wrote the article.

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References


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