

# Persistent neonatal thrombocytopenia can be caused by IgA anti platelet antibodies present in breast milk of immune thrombocytopenic mothers

Hagit Hauschner, Nurit Rosenberg, Rafael Mendelsohn, Uri Seligsohn, Nechama Sharon



Sheba - Academic Medical Center Hospital

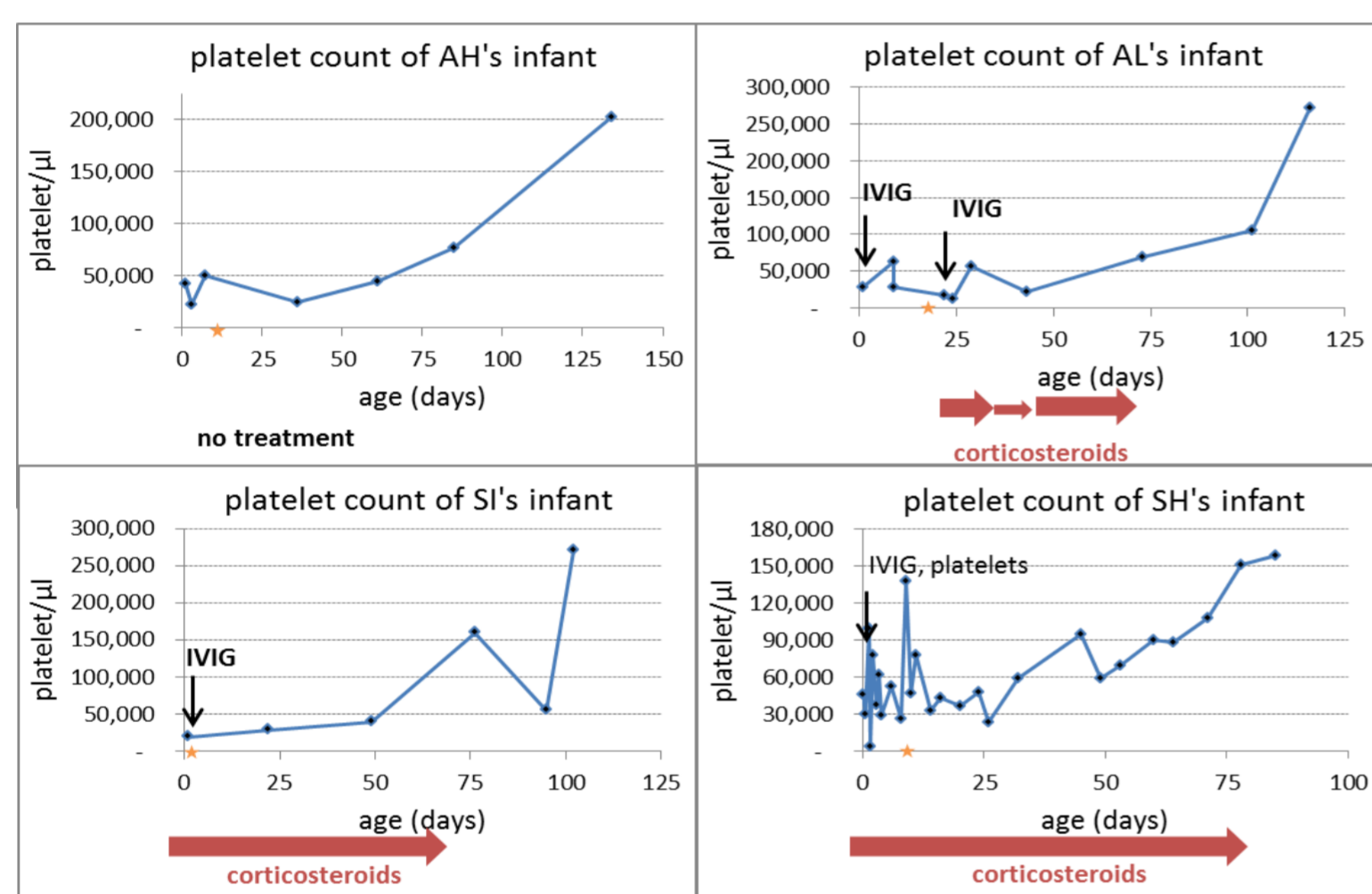
## ABSTRACT

Immune thrombocytopenia (ITP) in pregnant women can cause neonatal thrombocytopenia by transport of antiplatelet autoantibodies across the placenta. Usually, an infant's platelet count normalizes within 2 months. We observed neonatal thrombocytopenia that persisted more than 4 months and disappeared following discontinuation of breastfeeding.

**The aim of our study was to discern whether breast milk of ITP mothers contained antiplatelet antibodies causing persistent thrombocytopenia.**

We collected 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  $\alpha\text{IIb}\beta\text{3}$  expressed in cultured cells. Thus, transfer of antiplatelet antibodies from ITP mothers by breastfeeding can be associated with persistent neonatal thrombocytopenia.



4 infants of mothers with maternal ITP that were breastfed developed persistent thrombocytopenia which ceased after stopping breastfeeding and switching to formula.

## METHODS

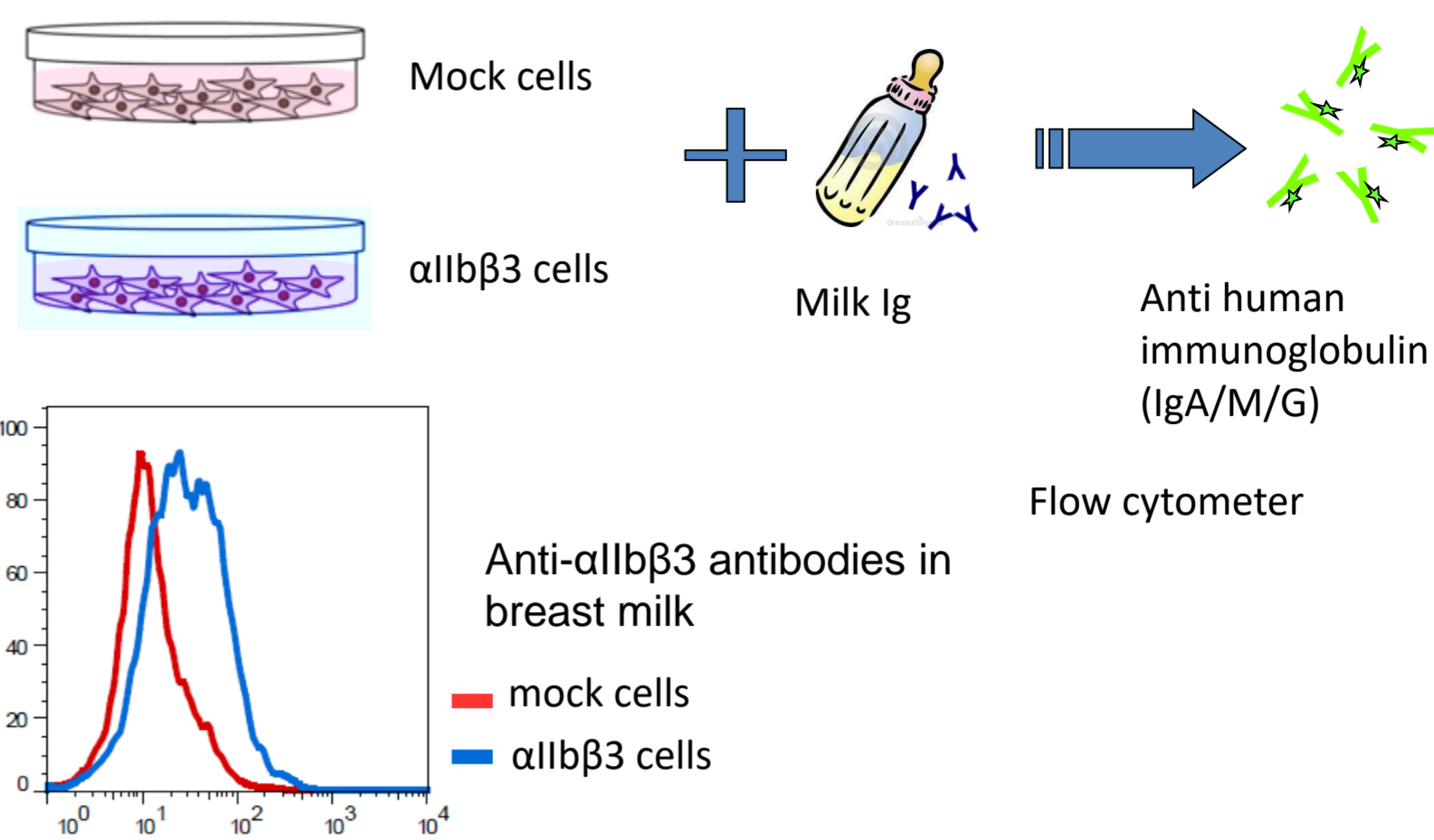
We evaluated breast milk from 22 women.

We collected 4 samples during the first month of breastfeeding.

7 women with active ITP had thrombocytopenia during the pregnancy. And their neonates also had thrombocytopenia. 6 woman had a history of R ITP, but either had normal or mildly low platelet counts and their neonates had normal platelet counts. The 9 healthy controls were woman who had a normal healthy pregnancy.

## Detection of anti- $\alpha\text{IIb}\beta\text{3}$ antibodies by using cultured cells

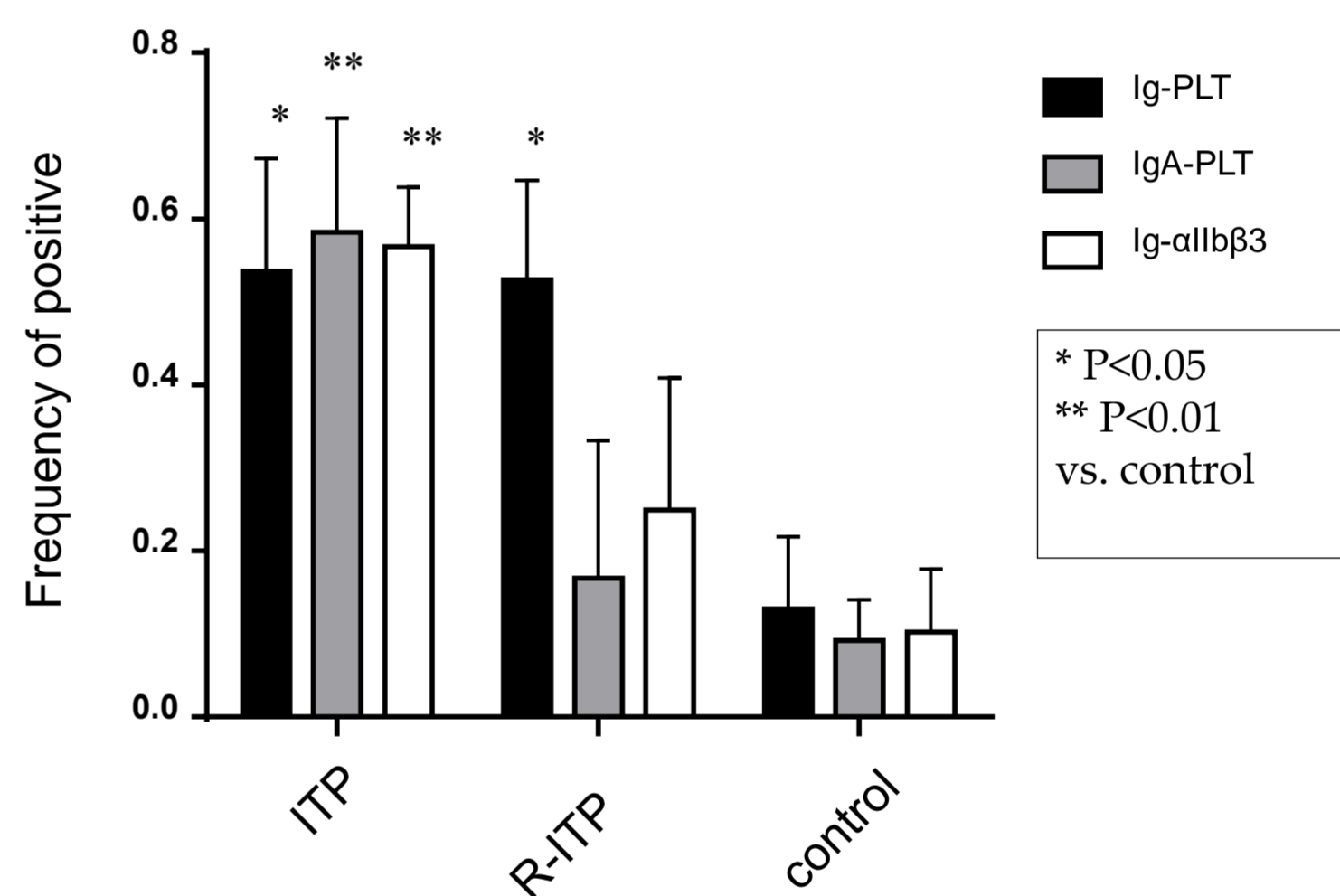
Anti- $\alpha\text{IIb}\beta\text{3}$  antibodies were detected by using baby hamster kidney cells expressing Anti- $\alpha\text{IIb}\beta\text{3}$  or only the vectors (mock). Expression of Anti- $\alpha\text{IIb}\beta\text{3}$  on the cell surface was validated by using monoclonal antibodies against Anti- $\alpha\text{IIb}\beta\text{3}$  (supplemental Data). The cell suspension in phosphate-buffered saline was incubated with milk-Ig samples for 30 minutes. Anti- $\alpha\text{IIb}\beta\text{3}$  antibodies were detected by adding total anti-human Ig phycoerythrin (Millipore) to the cells and using flow cytometry. The ratio of Anti- $\alpha\text{IIb}\beta\text{3}$  antibody binding was calculated by dividing the MFI of Anti- $\alpha\text{IIb}\beta\text{3}$  -expressing cells by the MFI of mock cells incubated with the same milk-Ig sample. Samples were considered positive when the results were greater than mean ratios of control samples plus 2 standard deviations.



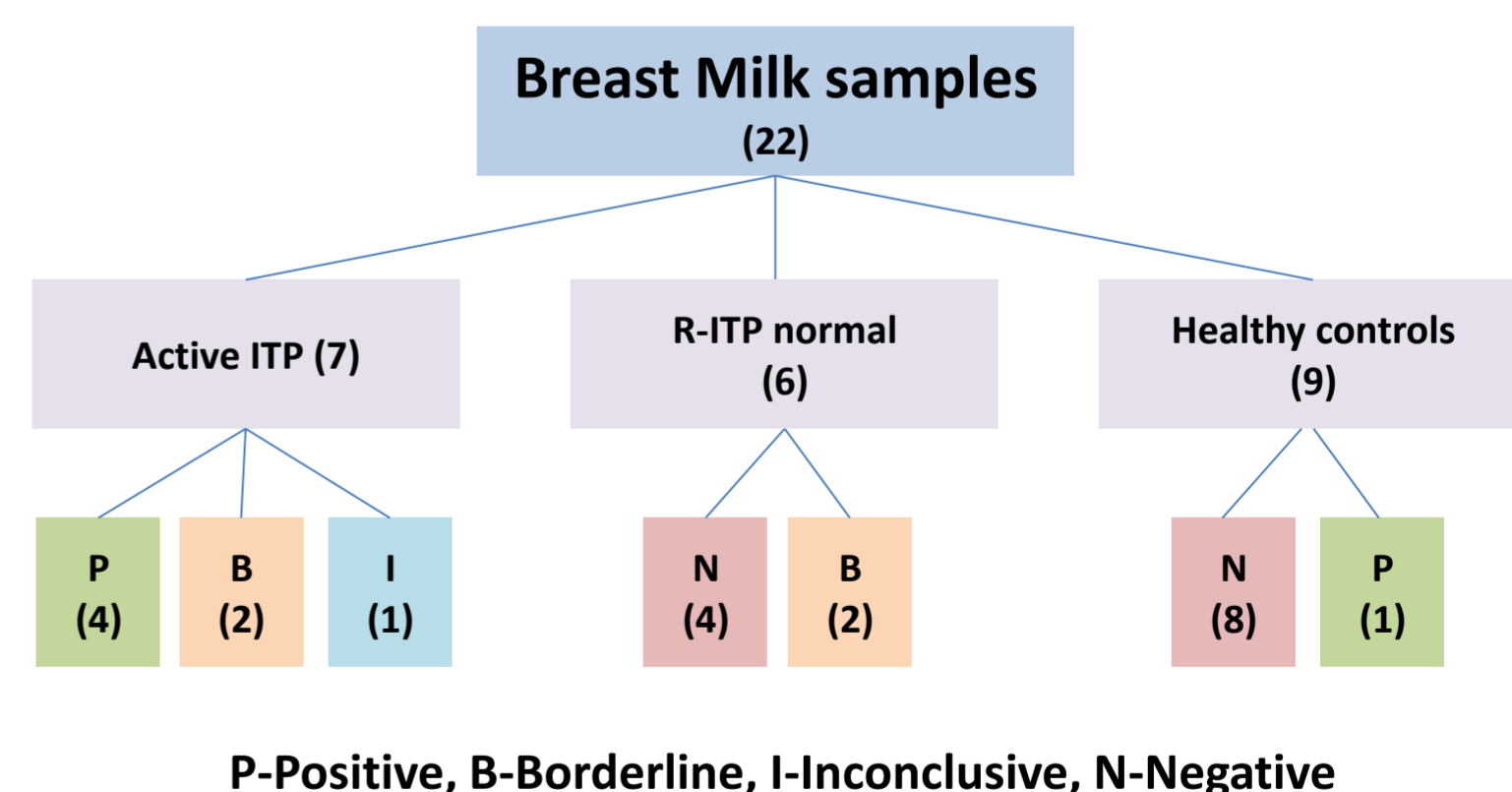
## RESULTS

We evaluated breast milk from 22 women; 7 women with active ITP during pregnancy revealed Anti- $\alpha\text{IIb}\beta\text{3}$  antibodies 4 of them were positive, 2 borderline and 1 inconclusive.

6 R-ITP, most of them were negative for Anti- $\alpha\text{IIb}\beta\text{3}$  antibodies. Control group (9) – 8 were negative for Anti- $\alpha\text{IIb}\beta\text{3}$  antibodies and 1 positive. These results demonstrate a statistically significant increased level of IgA in the milk of Active ITP patients (group 1) in comparison with the 2 other groups



The frequency of positive assay for antibodies detection in ITP-thrombocytopenic women compare to R-ITP or control subjects was analyzed by Graphpad software using 2-ways ANOVA with Tukey's multiple comparisons test.



P-Positive, B-Borderline, I-Inconclusive, N-Negative

## CONCLUSION

The majority of samples of breastmilk from women with Active ITP contained IgA anti platelet antibodies. The presence of these antibodies and the association with persistent thrombocytopenia in the neonates suggest a potential transfer of anti platelets antibodies through breastmilk.

In our view, breastfeeding should not be discouraged, but when low platelet counts persistent and the baby needs treatment such steroids, discontinuing breastfeeding is a viable solution.