What is this research about?

When the body attacks its own cells, the results can be devastating. **Immune thrombocytopenia** (ITP) is an autoimmune disease in which the body produces antibodies against blood platelets. These antibodies bind to platelets and tag them, causing them to be recognized as ‘foreign’ by the immune system and destroyed. Platelets work by sticking together and forming a plug that seals broken blood vessels, so the loss of too many platelets can lead to serious bleeding.

Platelets tagged with antibodies are thought to be destroyed in the spleen. Immune cells in the spleen called **macrophages** have a protein on their surface called an **Fc receptor**. Through the Fc receptor, macrophages can recognize platelets that have been tagged for destruction. The platelets are then ‘eaten’ by macrophages and destroyed. Most treatments for ITP are based on this mechanism of platelet destruction. These include prescribing intravenous immunoglobulin (IVIG), a plasma-derived drug, or in some cases removing the patient’s spleen. IVIG has anti-inflammatory activity that primarily shuts down macrophage/Fc receptor-dependent platelet destruction.

Most cases of ITP are spontaneous and without any clear cause. Although ITP is considered a single disorder, the progression and severity of the disease varies from patient to patient. Patients respond very differently to treatment and previous work by the same researchers who conducted this study hinted at possible reasons why. In most cases of ITP, the anti-platelet antibodies target one of two proteins found on the surface of blood platelets: **GPIbIIa** or the **GPIb complex**. These researchers and others have shown that when ITP is caused by antibodies against the GPIb complex, patients often do not respond to therapy targeting the Fc receptor (e.g. IVIG). These intriguing observations suggested that other mechanisms not involving the Fc receptor may lead to platelet destruction in some cases of ITP. The researchers investigated that possibility with ground-breaking results.

What did the researchers do?

The researchers generated antibodies against **GPIb alpha** (part of the GPIb complex) and GPIIbIIa. To understand how the antibodies work, the researchers compared the effects of these antibodies in plasma samples from patients with ITP. They conducted a series of **in vitro** (in test tubes) and **in vivo** (in a mouse model of ITP) experiments, investigating how the antibodies affect platelets’ ability to activate and aggregate (stick together), important first steps in sealing broken blood vessels. The researchers tested whether these antibodies cause **desialylation**, which is the removal of sugar residues from proteins. Desialylation changes the shape and structure of proteins on the surface of platelets and triggers platelet destruction in an organ other than the spleen: the liver. They tracked how platelets are cleared from the blood circulation following treatment with GPIb alpha antibodies. Finally, they tested a potential new treatment for ITP based on their findings.

What did the researchers find?

- Antibodies against GPIb alpha caused higher platelet activation than antibodies against GPIIbIIa.
- Desialylation of platelet proteins occurred mostly with antibodies against GPIb alpha, and less so with antibodies against GPIIbIIa.
These findings showed that the effects of antibodies against GPIb alpha are fundamentally different to the effects of antibodies against GPIIbIIIa. This suggests that antibodies against GPIb alpha and GPIIbIIIa lead to different mechanisms of platelet destruction. Investigating this concept, the researchers showed that:

- Platelets tagged with anti-GPIb alpha antibodies are frequently destroyed in the liver via Ashwell-Morell receptors on liver cells.
- This mechanism of platelet destruction is independent of macrophages in the spleen and the Fc receptor. The researchers also demonstrated for the first time that platelet activation and desialylation act together in a positive feedback loop that leads to destruction of platelets in the liver.
- The positive feedback means the effect of the antibodies is magnified, and may explain why relatively low levels of antibodies in ITP patients sometimes cause severely low platelet counts.
- Taking advantage of the link between desialylation and platelet destruction, the researchers found that blocking desialylation using sialidase inhibitors prevented platelet destruction.

**How can you use this research?**

This study shows for the first time that liver cells play an important and previously unknown role in platelet destruction in ITP. This could have major implications for diagnosing and successfully treating patients with this bleeding disorder. To date, there is no reliable way to predict the success or failure of any ITP treatment in the clinic. Measuring platelet desialylation may be a new way to determine if platelet destruction is happening in the liver, which could help guide treatment choices.

IVIG is often the drug of choice to treat ITP, but it is in limited supply and extremely expensive. Although IVIG is generally effective, it can be harmful. Using a test to identify patients who will not respond to IVIG could prevent unnecessary use of this drug and limit harmful side effects in patients who will not benefit from receiving this treatment in the first place. Importantly, this research also shows that platelet destruction in the liver can be blocked using sialidase inhibitors. Sialidase inhibitors are commercially available as anti-influenza drugs and may be a potential treatment for ITP patients who do not respond to other treatments. Clinical trials are needed to investigate the value of measuring platelet desialylation as a diagnostic test for this mechanism of ITP and the potential of sialidase inhibitors as an ITP treatment.

**About the research team:** This study was conducted in the laboratory of Dr. Heyu Ni, a Canadian Blood Services scientist and a professor in the departments of laboratory medicine and pathobiology, physiology and medicine at the University of Toronto. Dr. Ni is a member of the Toronto platelet immunology group at the Keenan research centre for biomedical science at St. Michael’s Hospital, and the platform director for hematology, cancer, and immunologic diseases at St. Michael's Hospital. The research team included members of Dr. Ni’s laboratories, and collaborators from Brigham and Women’s Hospital at Harvard Medical School, Boston, MA, Anhui Medical University, Hefei, China and Qilu Hospital, Shandong University, Jinan, China.

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