Understanding the Complement Cascade and Its Role in Cold Agglutinin Disease
Instructions

This information is provided as an educational resource for healthcare providers. It is not intended to be a substitute for review of the underlying reference materials and scientific literature. Healthcare providers should make all treatment decisions based on their medical judgement and the clinical profile of the individual patient. The following document cannot be copied, modified, used or distributed without the express written consent of Bioverativ.
Pathogenesis of Cold Agglutinin Disease

- Cold agglutinin–associated lymphoproliferative bone marrow disorder
- A distinct type of LPD

LPD, lymphoproliferative disorder.
Pathogenesis of Cold Agglutinin Disease

Hemolysis is entirely classical complement pathway dependent

C, complement; CA, cold agglutinin.
Pathogenesis: Therapeutic Implications

- Distinct clonal B-cell bone marrow disorder\(^1\)
  - Therapeutic implications

- Hemolysis is entirely classical complement pathway dependent\(^2\)
  - Therapeutic implications

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C, complement; Ig, immunoglobulin; INH, inhibitor.

The Complement System: A Family of Circulating Proteins in the Blood Responsible for Immune Surveillance

C, complement protein; MBL, mannan-binding lectin.

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The C1 Complex Is the Activation Mechanism of the Classical Complement Pathway¹

**C1 complex activates the classical complement pathway:**

1. The pattern recognition molecule C1q binds items such as immune complexes, apoptotic cells, etc.
2. C1q undergoes a conformational change following binding to a target, resulting in C1r autoactivation.
3. Activated C1r cleaves and activates C1s.
4. Active C1s cleaves C4 and C2, forming the CP/LP C3 convertase.

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C, complement protein; CP, classical pathway; LP, lectin pathway; MBL, mannan-binding lectin.

The Classical Pathway Activates Upon Binding to Antibodies: A Bridge Between the Innate and Adaptive Immune Systems

C, complement protein; IgG, immunoglobulin G.

Cold Agglutinin Disease

**Overview:**
- Autoimmune hemolytic anemia
- Prevalence: ~1/60,000–1/100,000; incidence: ~1/300,000–1/1,000,000
- ~5000 patients in the United States
- Patients are often elderly and present with anemia, fatigue, hemoglobinuria, and acrocyanosis

**Etiology:**
- Primary (idiopathic)
- Secondary: caused by an underlying condition (e.g., infection, malignancy)
- Majority of patients have detectable clonal B cells responsible for the production of an autoantibody that binds to RBC <37°C (cold)

**Clinical symptoms:**
- Frequent need for transfusions
- Chronic anemia with related symptoms (decreased quality of life)
- Agglutination-associated symptoms
- Thromboembolic complications

C, complement protein; CA, cold agglutinin; RBC, red blood cell.

Patients With Cold Agglutinin Disease Are Hypocomplementemic for Upstream Classical Pathway Substrates

Compared with healthy individuals, patients with cold agglutinin disease have lower levels of C4 and C2 but not C5, suggesting that the classical pathway is consumed in these patients, but the terminal pathway is not activated (ie, C5 cleavage)\(^1\)

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Understanding the Role of Complement in Mediating RBC Destruction in Cold Agglutinin Disease

Patient Sample

Flow cytometry

Normal RBC

RBC coated with antibodies

+ Complement

Lysis

Complement-coated RBC

FL1-A, fluorescence intensity; FSC-A, forward scatter; RBC, red blood cell.

Cold Agglutinin Titers Correlate With the Amount of Complement Deposited on the RBC

\[ r = 0.76 \quad P < 0.0001 \]

C3 fragment-positive RBC (%)

Cold agglutinin titer (1:X)

N = 40 samples

C, complement protein; RBC, red blood cell.

C3b Is an “Eat Me” Signal to Cells of the Immune System

Macrophage

C3b-coated cells

Phagocytosis
C3b is deposited on the target cell’s membrane, promoting its phagocytosis\(^1\)

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C, complement protein.

An In Vitro Proxy for Extravascular Hemolysis

Normal RBC + RBC coated with complement + Complement ± inhibitor + Green dye + Green dye

Phagocytosis

RBC, red blood cell.
C1s Inhibition Prevents Phagocytosis of RBCs Exposed to Cold Agglutinin Disease Autoantibodies

C1s Inhibition Prevents Complement Dependent Hemolysis Driven by Cold Agglutinin Disease Autoantibodies

Thus, an upstream classical pathway inhibitor would prevent both extravascular and intravascular hemolysis\(^1\)

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C, complement protein; IgG, immunoglobulin G; SE, standard error.

C1s Inhibitors Prevent Anaphylatoxin Production Generated by Cold Agglutinin Disease Autoantibodies

Summary

- Cold agglutinin disease is an autoimmune hemolytic anemia\(^1\)
- Cold agglutinins bind to the RBC and activate the classical complement pathway, leading to opsonin (C3b) deposition on the RBC surface and, in some cases, direct cellular lysis\(^2,3\)
- Opsonin-coated RBCs travel to the liver where they are phagocytosed\(^2,3\)
- Classical pathway inhibitors that target upstream of C3 and prevent opsonin deposition rescue cells from being phagocytosed or lysed by complement, specifically demonstrating the role of the classical pathway in mediating cold agglutinin–driven complement activation\(^4\)
- Classical pathway inhibitors, therefore, could potentially be efficacious in the treatment of cold agglutinin disease\(^4\)

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