Fred Hutch Cancer Center

Transfusion Medicine: Pretransfusion Testing, Blood Component Therapy and Adverse Reactions

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September 27, 2024

Land Acknowledgement

Fred Hutchinson Cancer Center acknowledges the Coast Salish peoples of this land, the land which touches the shared waters of all tribes and bands within the Duwamish, Puyallup, Suquamish, Tulalip and Muckleshoot nations.

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2. Explain the risks of transfusing uncrossmatched blood products
3. Describe the various blood components available for transfusion and their indications
4. Analyze the pathophysiologic mechanisms of the most common and severe non-infectious complications associated with transfusions

Pre-Transfusion Testing

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(5.06.01)
Which of the following tests are required prior to a red cell transfusion? (select all that apply)?

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Which of the following tests are required prior to a red cell transfusion? (select all that apply)?

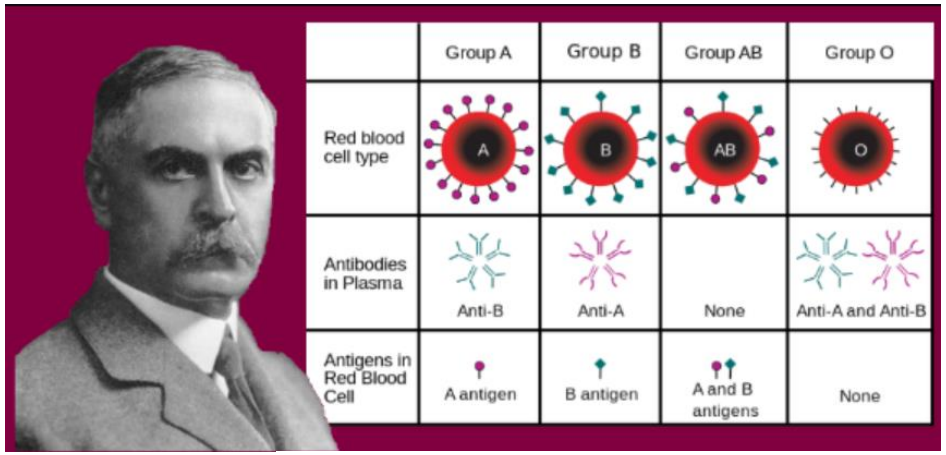
- **A) A blood type**
- **B) A second confirmatory blood type**
- **C) A crossmatch**
- D) A second confirmatory crossmatch
- **E) An antibody screen**
- F) A direct antiglobulin test

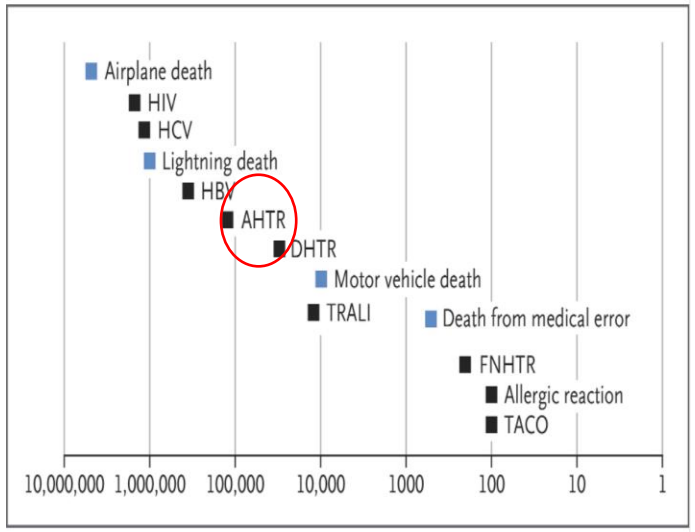
“Type and Screen”

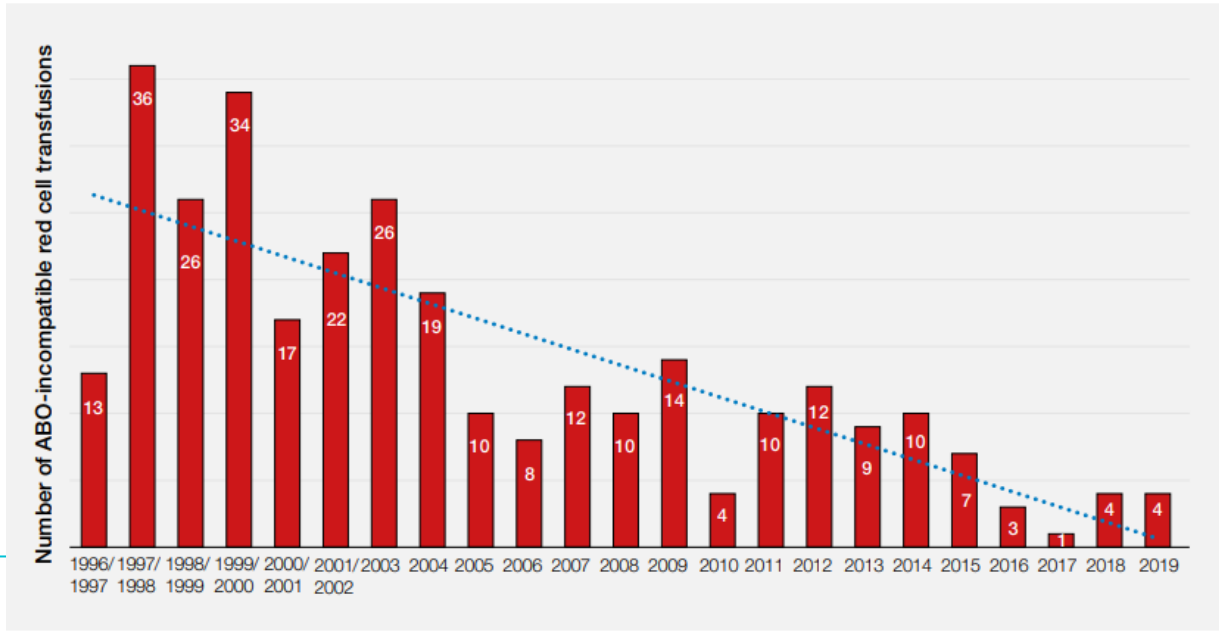
Blood Type

- ABO and RhD typing
    - Forward type: RBC Antigen (A, B, RhD)
    - Reverse type: Antibodies (anti-A, anti-B)
  - Need a second confirmatory blood type for a new patient
    - Reduced wrong blood in tube errors leading to acute hemolytic transfusion reactions

ABO incompatible Red Cell Transfusions: Never Event







Reducing the Incidence of Acute Hemolytic Transfusion Reactions

- Almost always due to error at bedside (>99%)
  - Wrong patient label on patient sample
    - 2 Samples for Type and Screen are required for every new patient
    - All samples for blood bank MUST be verified by 2 different providers
  - Wrong unit started on a patient
    - Double verification: Prior to starting a transfusion, 2 different nurses must verify the product and patient

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(5.06.02)
Type O RhD negative (O neg) red blood cells are universally safe in all patients.

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Type O RhD negative (O neg) red blood cells are universally safe in all patients.

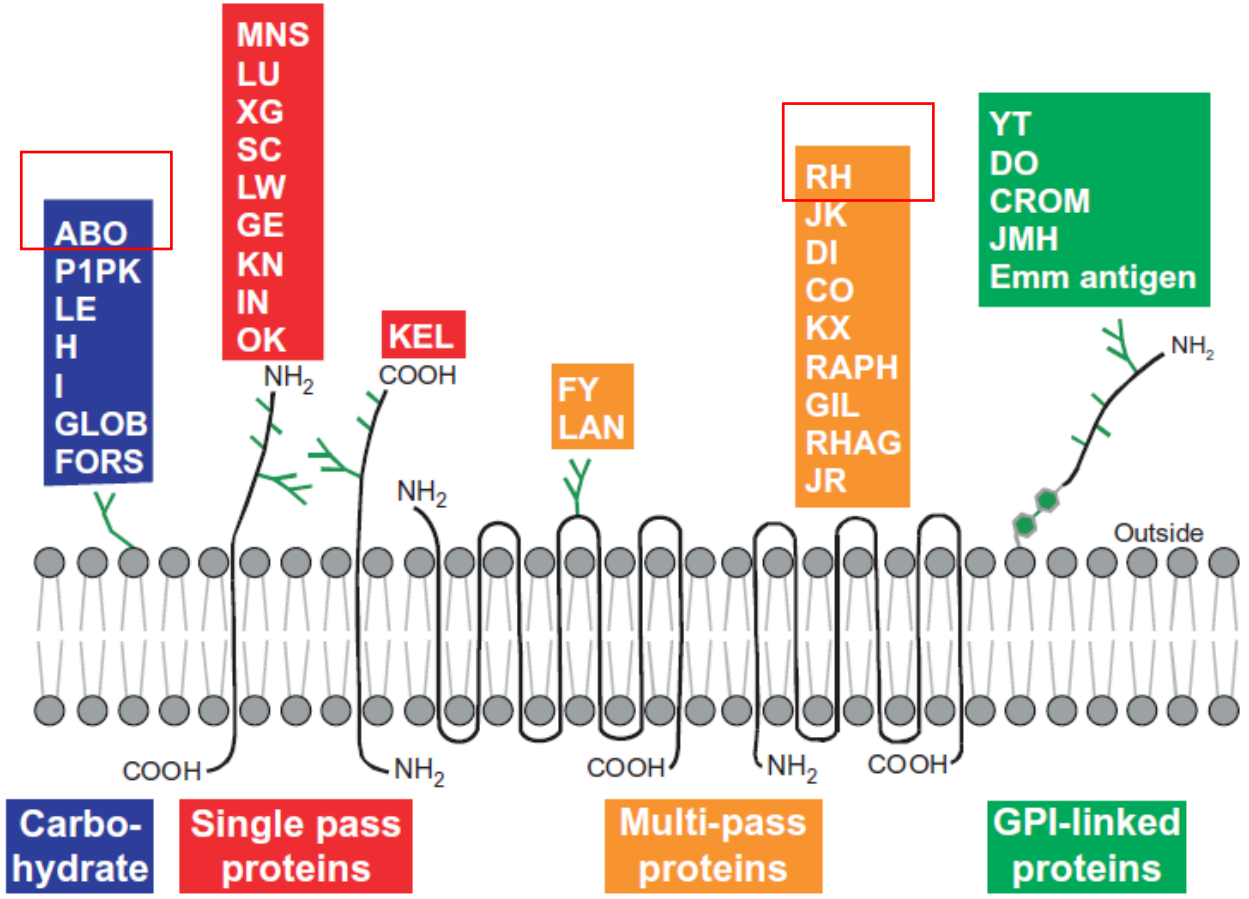
- A) True
- **B) False**

“Type and **Screen**”

Antibody Screen

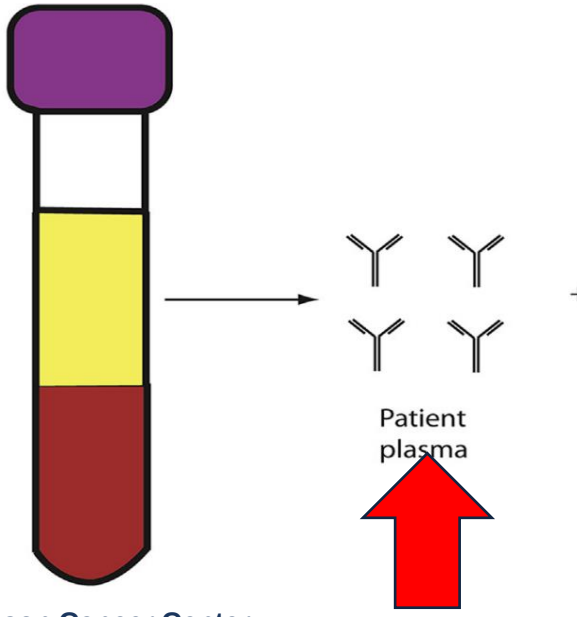
- Test patient plasma for presence of red cell *allo*antibodies
  - NOT Anti-A or anti-B
  - Anti-D, Anti-E, Anti-e, Anti-C, Anti-c, Anti-K, Anti-Jka, etc….
- Ab screen positive = universal supply of O negative RBC may not be safe

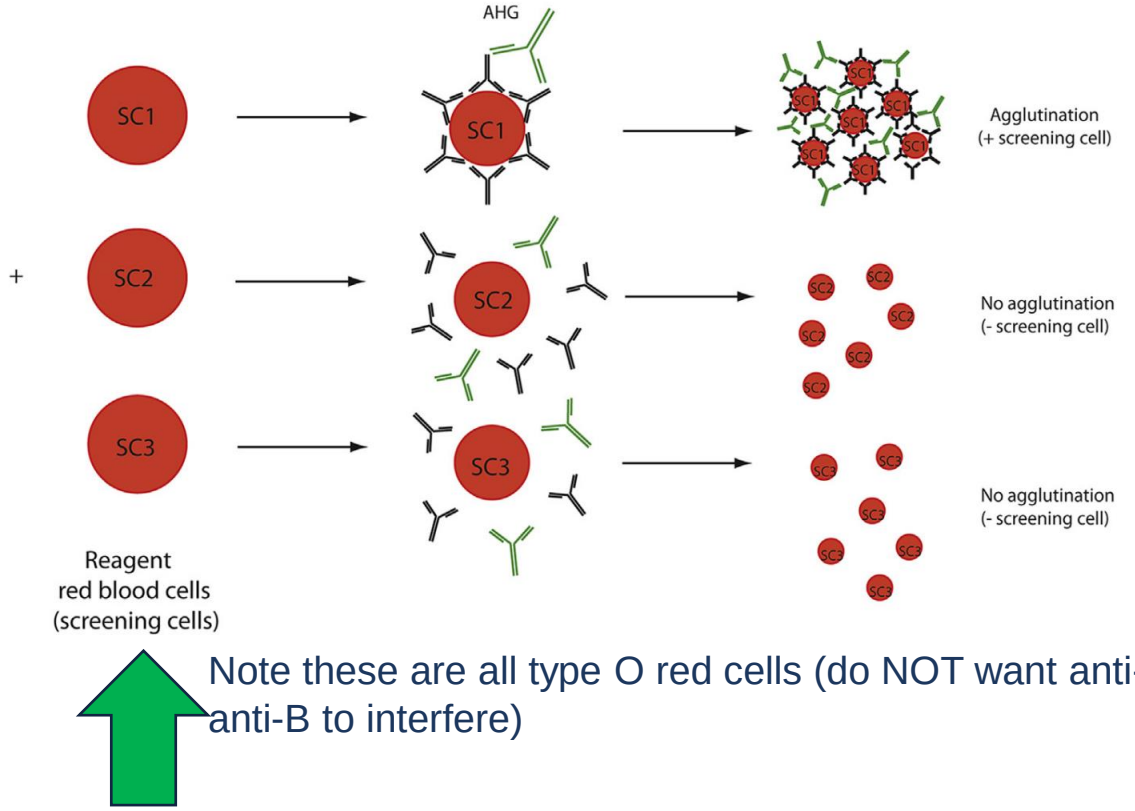
Blood Groups



Why is an antibody screen needed?

- Screen: Presence of unexpected antibodies *(anti-K, anti-E, anti-e, etc)*
  - *Not naturally occurring*
  - *Expected result: negative screen*



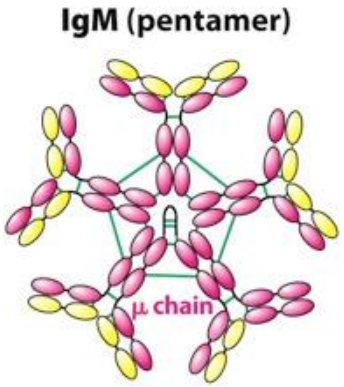


Note these are all type O red cells (do NOT want anti-A, anti-B to interfere)

Immunoglobulin Class: ABO and alloantibodies antibodies

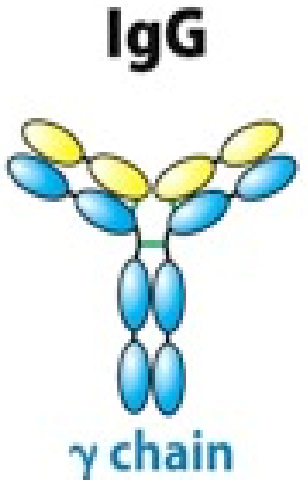
ABO Antibodies (Reverse type)

- Naturally occurring (expected antibodies)
- Cold reacting, No incubation needed (5-10 minutes)
- Strong activators of complement pathway
- Intravascular hemolysis (hemoglobinuria, hemoglobinemia)



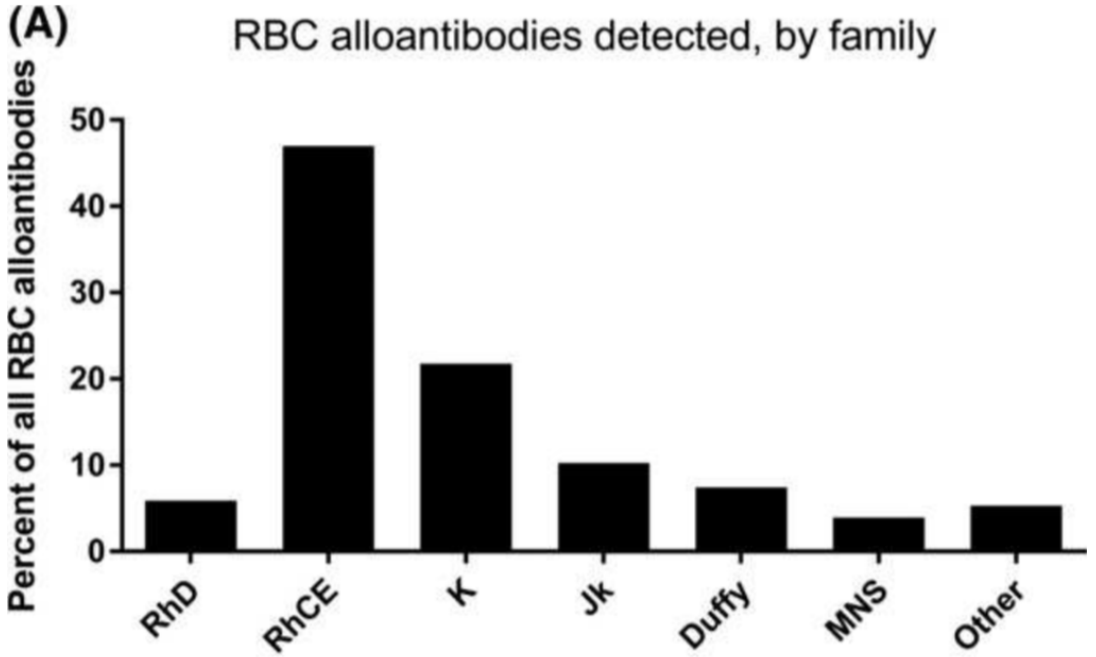
Alloantibodies (antibody screen)

- After alloimmunizing event (unexpected antibodies)
- Warm reacting, Incubation needed at 37C with AHG (30-60 minutes)
- Weaker activators of complement pathway
- Extravascular hemolysis (hyperbilirubinemia, slowly declining Hg)



RBC antigen alloimmunization

- Rate of RBC antigen allo-immunization in the US: 2-6%
- SCD, SLE, MDS, other diseases, are associated with higher rates of RBC allo-immunization



How to transfuse patient with alloantibodies?

- Example: Patient is A positive with a positive antibody screen
  - Antibody Identification: anti-Fy(a)
- Transfuse: Type O or A (Rh positive or Rh negative) RBC that is negative for Fy(a) antigen
  - Units are not routinely tested to determine if positive/negative for blood group antigens outside of ABO (Fy, Jk, K, etc)
  - Need specific units that are tested to be Fy(a) negative
  - Universal type O RBC units available in hospitals for emergency release are generally NOT tested for these antigens

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(5.06.03)
In patients with a negative antibody screen is negative, a crossmatch is not required for red cell transfusions.

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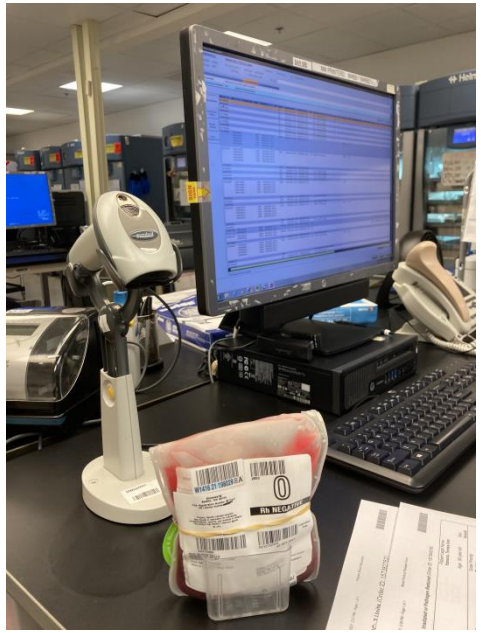
In patients with a negative antibody screen, a crossmatch is not required for red cell transfusions.

- A) True
- **B) False**

All Products With > 2mLs of RBCs are Required to Be Crossmatched (RBC, Whole Blood, Granulocytes)

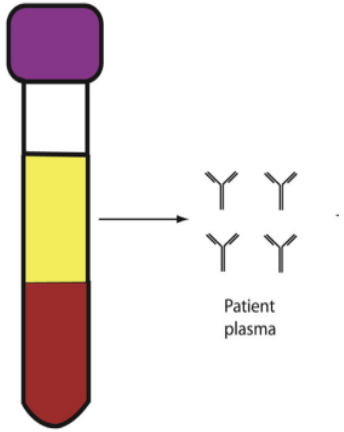
Electronic Crossmatch

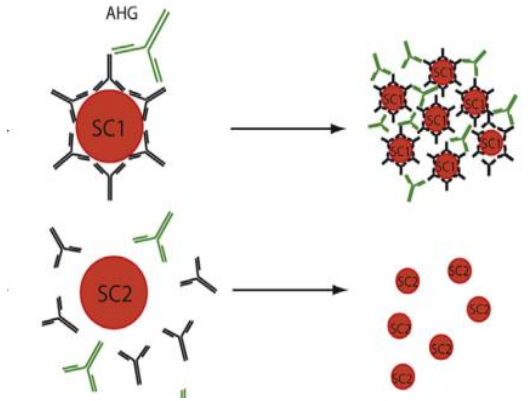
- Negative antibody screen
- ABO compatibility
- 5 minutes



Serologic Crossmatch

- Positive antibody screen
- Minor antigen compatibility
- 45-60 minutes

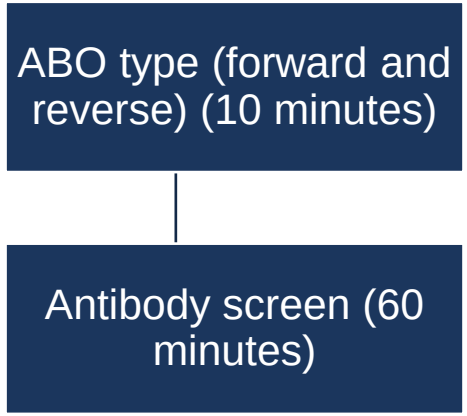




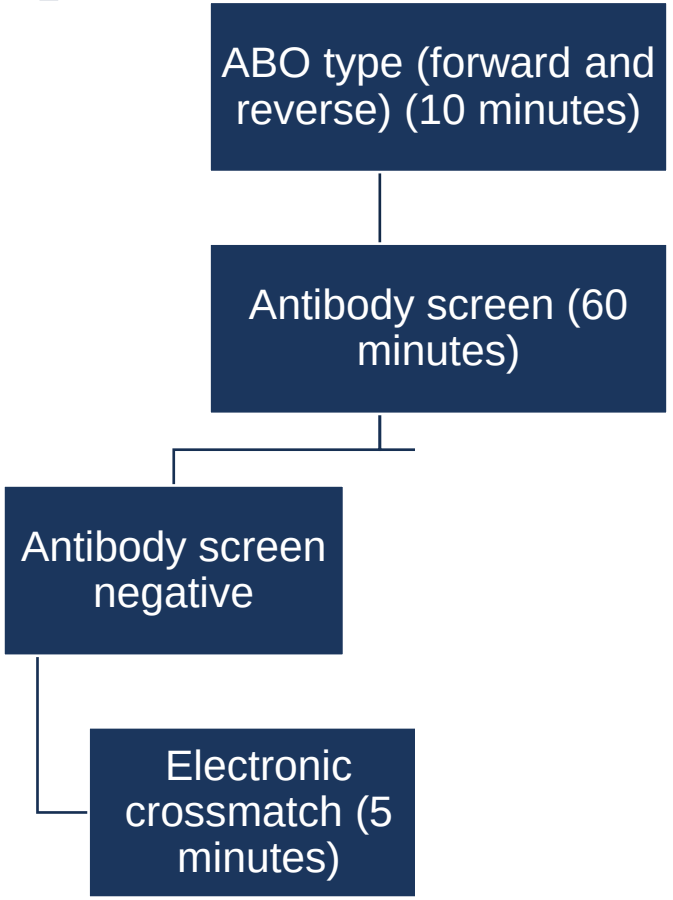
Turn around time for completion of testing and proving crossmatched products:

ABO type (forward and reverse) (10 minutes)

Turn around time for completion of testing and proving crossmatched products:



Turn around time for completion of testing and proving crossmatched products:

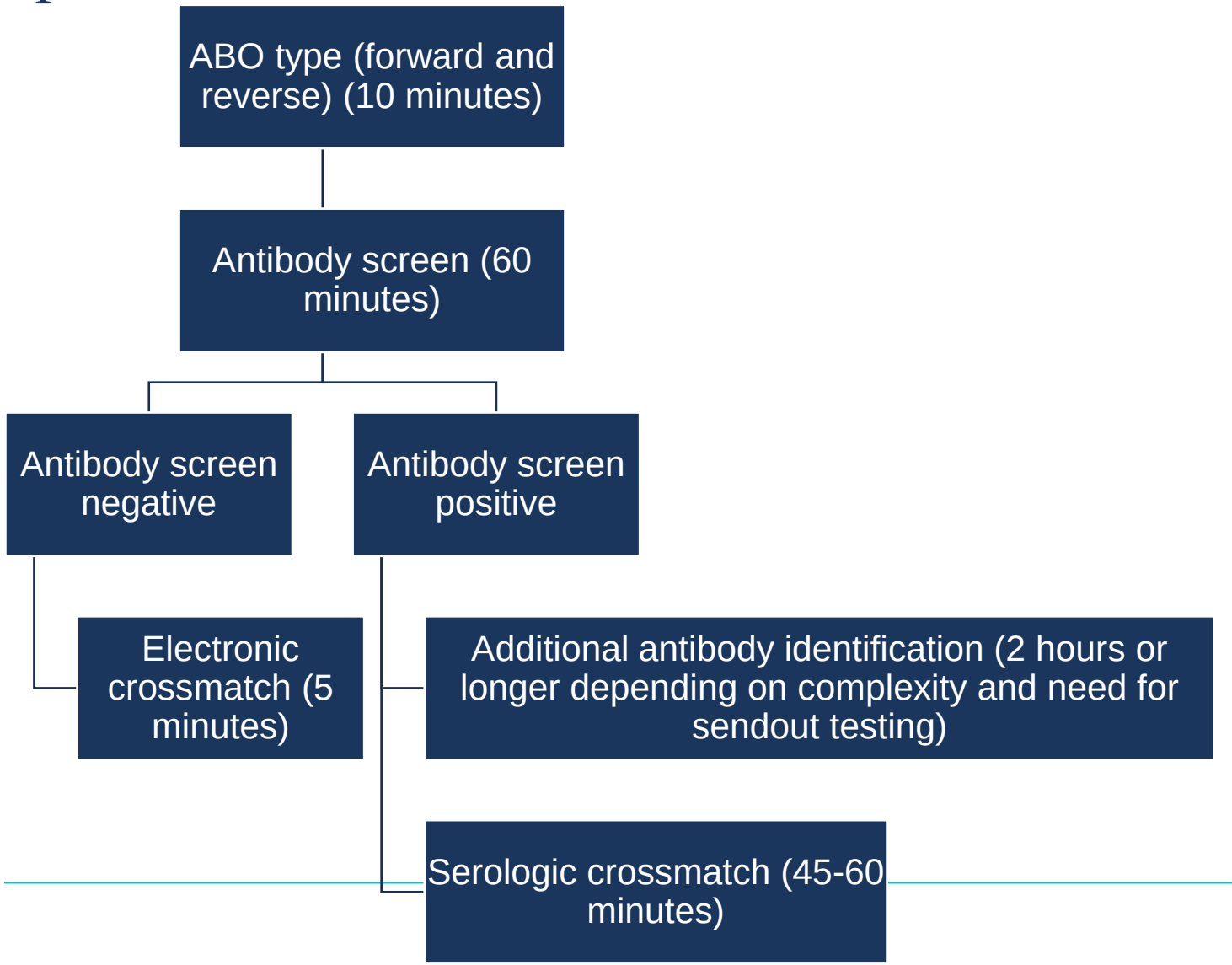


Time to crossmatch compatible blood products:
1-2 hours

If patient has active T&S → electronic CXM RBC have same turn around time as uncrossmatched products

Turn around time for completion of testing and proving crossmatched products:

Time to crossmatch compatible blood products:
1-2 hours



If patient has active T&S → electronic CXM RBC have same turn around time as uncrossmatched products

Time to crossmatch compatible blood products:
3-4 hours or longer (up to days in complex cases)

BLOOD PRODUCTS

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(5.06.04) Which of the following blood products has the most limited supply?

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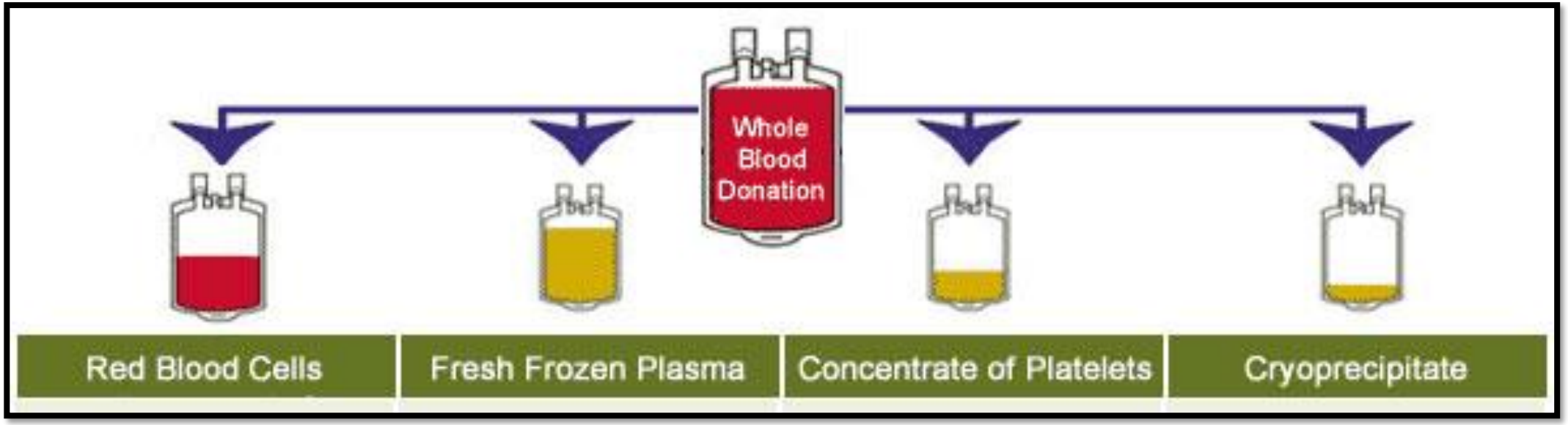
Which of the following blood products has the most limited supply?

- A) Whole Blood
- B) Red Blood Cells
- **C) Platelets**
- D) Plasma
- E) Cryoprecipitate

Whole blood donation



Whole Blood

The Evolution of Blood Transfusion in the Trauma Patient: Whole Blood Has Come Full Circle

Jonathan A. Black, MD[1] Virginia S. Pierce, MD[1] Jeffrey D. Kerby, MD, PhD[1] John B. Holcomb, MD[1]

- Characteristics
  - Low Titer Type O Whole Blood
  - Titer of anti-A and anti-B to make sure below certain threshold (can give to patients who are A and B)
  - 35 days shelf life
- Indication:
  - Trauma resuscitation (patient's blood type is unknown)
  - Pediatric surgery, ECMO priming

Semin Thromb Hemost 2020;46:215–220.

One Red Blood Cell Unit

- Content:
  - 200 mL Red blood cells (Hct of unit is about 55-65%)
  - 100 mL storage solution (anticoagulant and additive)
  - 30 mL plasma
- Testing:
  - ABO/Rh crossmatched compatible
- Effect: Increase hemoglobin 1g/dL (or 3% increment)
- Storage: 42 day shelf life, 2-6C (refrigerator)

Transfusion Thresholds

Clinical Review & Education

JAMA | Special Communication

Red Blood Cell Transfusion
2023 AABB International Guidelines

JAMA. 2023;330(19):1892-1902. doi:10.1001/jama.2023.12914
Published online October 12, 2023.

Recommendations for Adults

Recommendation 1

For hospitalized adult patients who are hemodynamically stable, the international panel recommends a restrictive RBC transfusion strategy in which the transfusion is considered when the hemoglobin concentration is less than 7 g/dL (strong recommendation, moderate certainty evidence).

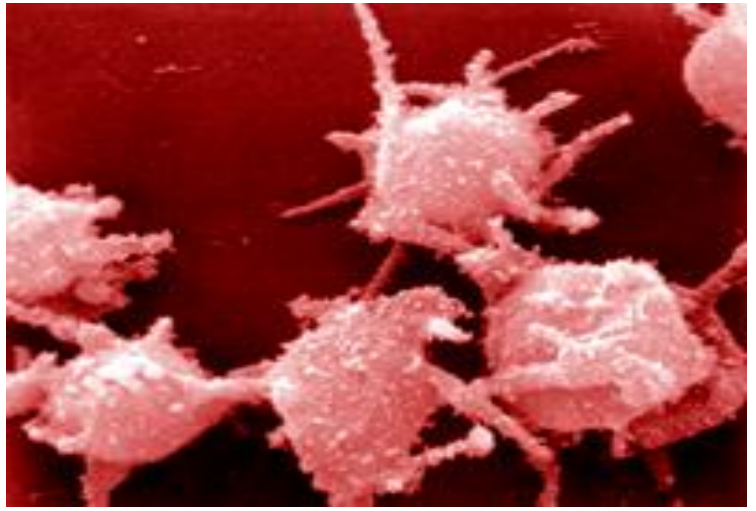
Recommendation 2

For hospitalized adult patients, the panel suggests a restrictive RBC transfusion strategy in which transfusion is considered when the hemoglobin concentration is less than 7 g/dL in those with hematologic and oncologic disorders (conditional recommendation, low certainty evidence).

CONCLUSIONS AND RELEVANCE It is good practice to consider overall clinical context and alternative therapies to transfusion when making transfusion decisions about an individual patient.

Platelets

- Testing:
  - Do not have to be crossmatched
  - Plasma compatible
- Storage:
  - room temperature (20-24C with gentle agitation)
    - Highest risk of infectious contamination
  - Shelf life: 5 days (up to 7 with delayed sampling)



Activated Platelets

Platelet Transfusion Thresholds

Supporting evidence (Quality)	Strength of Recommendation	Platelet count (K/uL)	Indication
		<150	Accepted definition of thrombocytopenia
Low/None	Not graded	**100**	Surgery on the brain or the posterior eye- BSCH
High		**80**	NO linear relationship between platelet count and bleeding from 6-80K/uL; Uhl et al. 2017
Moderate	Weak	**70**	Neuraxial anesthesia
Low/None	Weak	**50**	Lumbar puncture, major non neuraxial surgery - AABB
Low/None	Not graded	**50**	Therapeutic enteroscopy - ASGE
Low/None	Not graded	**50**	Liver, renal, transbronchial biopsy - JPAC
Low/None	Weak	**20**	Central line placement - AABB
Low/None	Not graded	**20**	Diagnostic enteroscopy - ASGE
Low/None	Not graded	**20**	Bronchoscopy with lavage - Brit Thoracic Soc.
Moderate	Strong	**20**	Unstable, febrile, bleeding patients
Moderate	Strong	**10**	Ppx. for spontaneous bleeding – AABB; BM biopsy
High		**5**	Spontaneous bleeding; Uhl et al. 2017

Adapted from Nagrebetsky, Brit J Anaesthesia, 2019

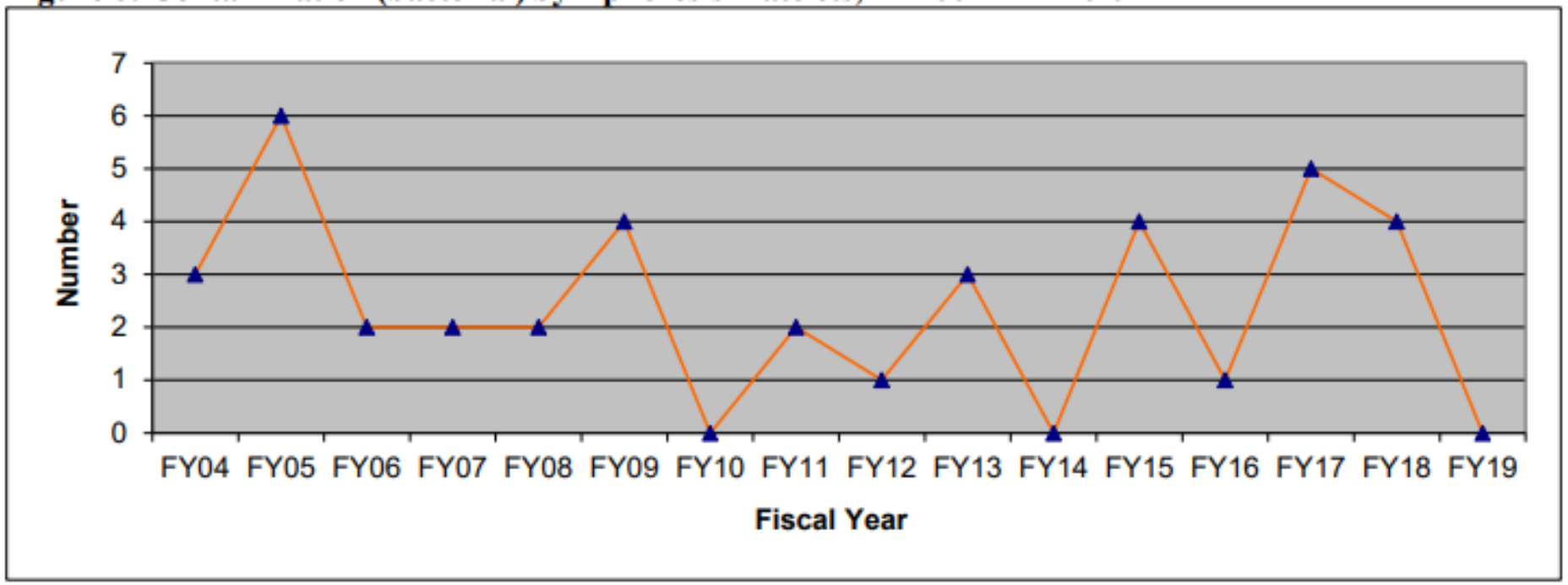
Type of Platelet Products

- Source:
  - Apheresis (single donor) vs. Pooled (whole blood from 4-6 donors)
- Storage solution
  - Plasma vs. Platelet additive solution (PAS)
- Storage temperature
  - Room temperature
- Bacterial testing
  - Bacterial culture vs. Pathogen inactivated (psoralen treatment)

Transfus Med Rev 2020 Oct;34(4):207-208

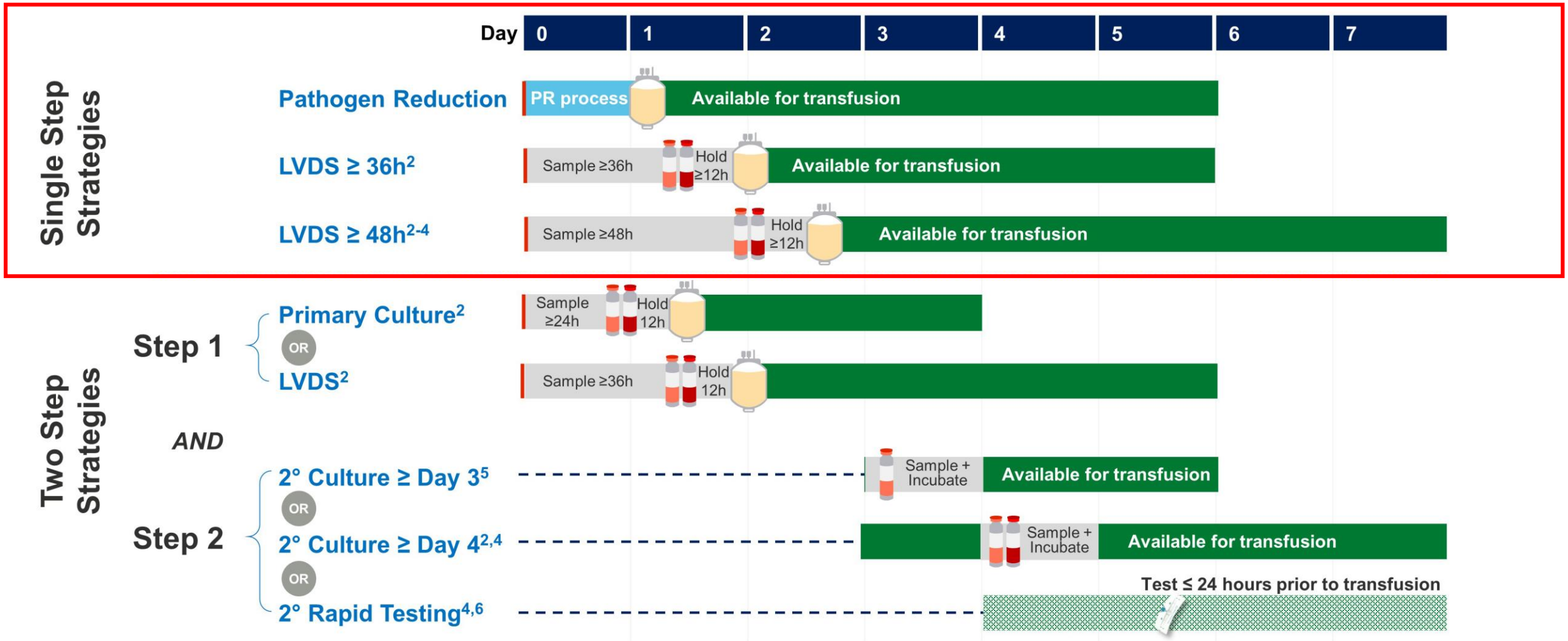
FDA Reported Fatalities from Bacterial Contamination

Figure 6: Contamination (bacterial) by Apheresis Platelets, FY2004 – FY2019



- ~4 deaths per year due to bacterial contamination of apheresis platelets
- ~1:500,000 platelet transfusions may result in fatality due to contamination
- ~1:1500 to 1:3000 non-fatal septic transfusion reaction rate with passive surveillance

FDA Guidance on Bacterial Safety September 2019[1]



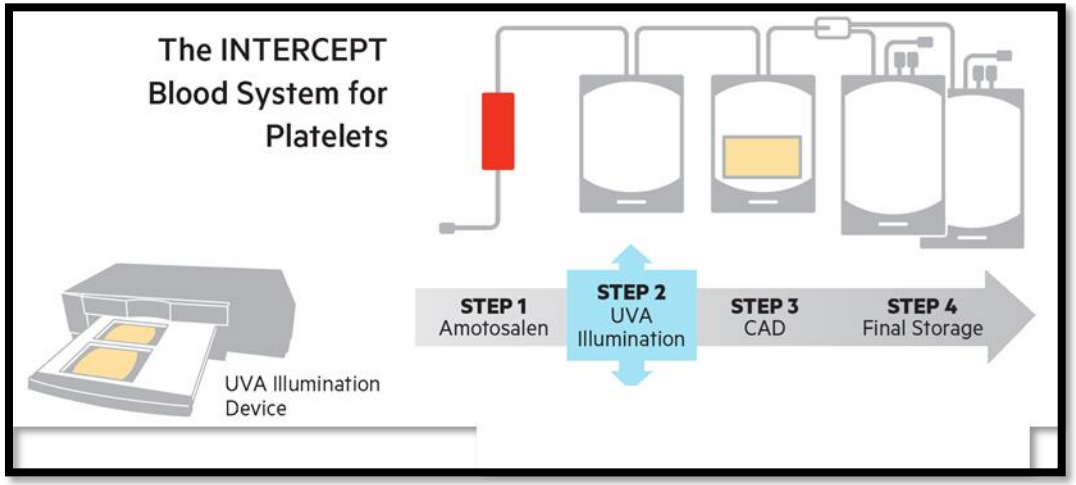
1. Bacterial Risk Control Strategies for Blood Collection Establishments and Transfusion Services to Enhance the Safety and Availability of Platelets for Transfusion: Guidance for Industry. US FDA; Sept 2019. 2. Aerobic and Anaerobic, 3. At the time of the finalization of this guidance, the instructions for use of the culture-based device currently labeled as a "safety measure" require a primary culture and secondary test to extend dating of platelets. Therefore, the LVDS no sooner than 48 hours strategy for a 7-day dating period cannot be implemented until appropriately labeled devices are available. 4. Platelets may only be stored beyond day 5 and up to day 7 if each component is tested using a bacterial detection device cleared by FDA and labeled for use as a "safety measure" according to its instructions for use, and if the platelet storage container has been cleared or approved for 7-day storage. 5. Aerobic. 6. Rapid testing practices vary and should be performed according to bacterial testing device instructions for use. Institutions may test daily to ensure availability of units (non-reactive test valid 24 hours prior to transfusion) or may choose to quarantine unit then test within 24 hours of transfusion. (Harm SK, et al. Transfusion. 2018 Apr;58(4):938-942. Ruby KN, et al. Transfusion. 2018 Jul;58(7):1665-1669).

cerus

Pathogen Inactivation – Amotosalen treatment

- Only FDA approved method for pathogen inactivation (PI)
- Can be used for Platelets and Plasma
- Platelet shelf life is 5 days
- PI can replace irradiation to prevent TA- GVHD
- Questionable hemostatic equivalence between treated and non-treated platelets

Spring Trial Transfusion 2005 Dec;45(12):1864-75.

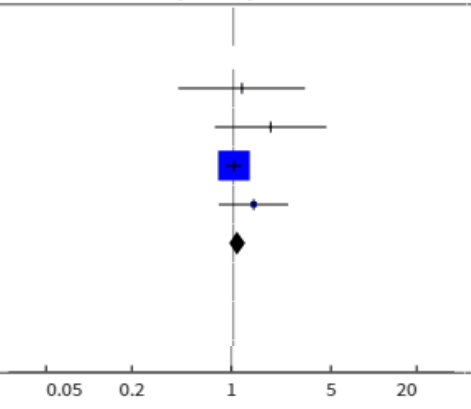


- Enveloped viruses
- Non-enveloped viruses
- Gram-negative bacteria
- Gram-positive bacteria
- Spirochetes
- Protozoa
- Leukocytes

Efficacy of Psoralen Treated Platelets – No increase in significant bleeding events

Analysis 1.4. Comparison 1 Pathogen-reduced platelets versus standard platelets for the prevention of bleeding, Outcome 4 Number of participants with 'clinically significant' bleeding (WHO grade ≥ 2 or equivalent) - follow-up more than 7 days.

Study or subgroup	PCT Plts n/N	Standard Plts n/N	Risk Ratio M-H, Fixed, 95% CI	Weight	Risk Ratio M-H, Fixed, 95% CI
1.4.1 Intercept plts vs standard plts - multiple platelet transfusion studies					
Janetzko 2005	6/22	5/21		2.2%	1.15[0.41,3.19]
Kerkhoffs 2010	11/85	7/99		2.79%	1.83[0.74,4.51]
McCullough 2004	186/318	190/327		80.68%	1.01[0.88,1.15]
Rebulla 2016	24/109	17/107		7.39%	1.39[0.79,2.43]
Subtotal (95% CI)	**534**	**554**		**93.06%**	**1.06[0.94,1.21]**
Total events: 227 (PCT Plts), 219 (Standard Plts)					
Heterogeneity: $Tau^2=0$; $Chi^2=2.96$, df=3(P=0.4); $I^2=0\%$					



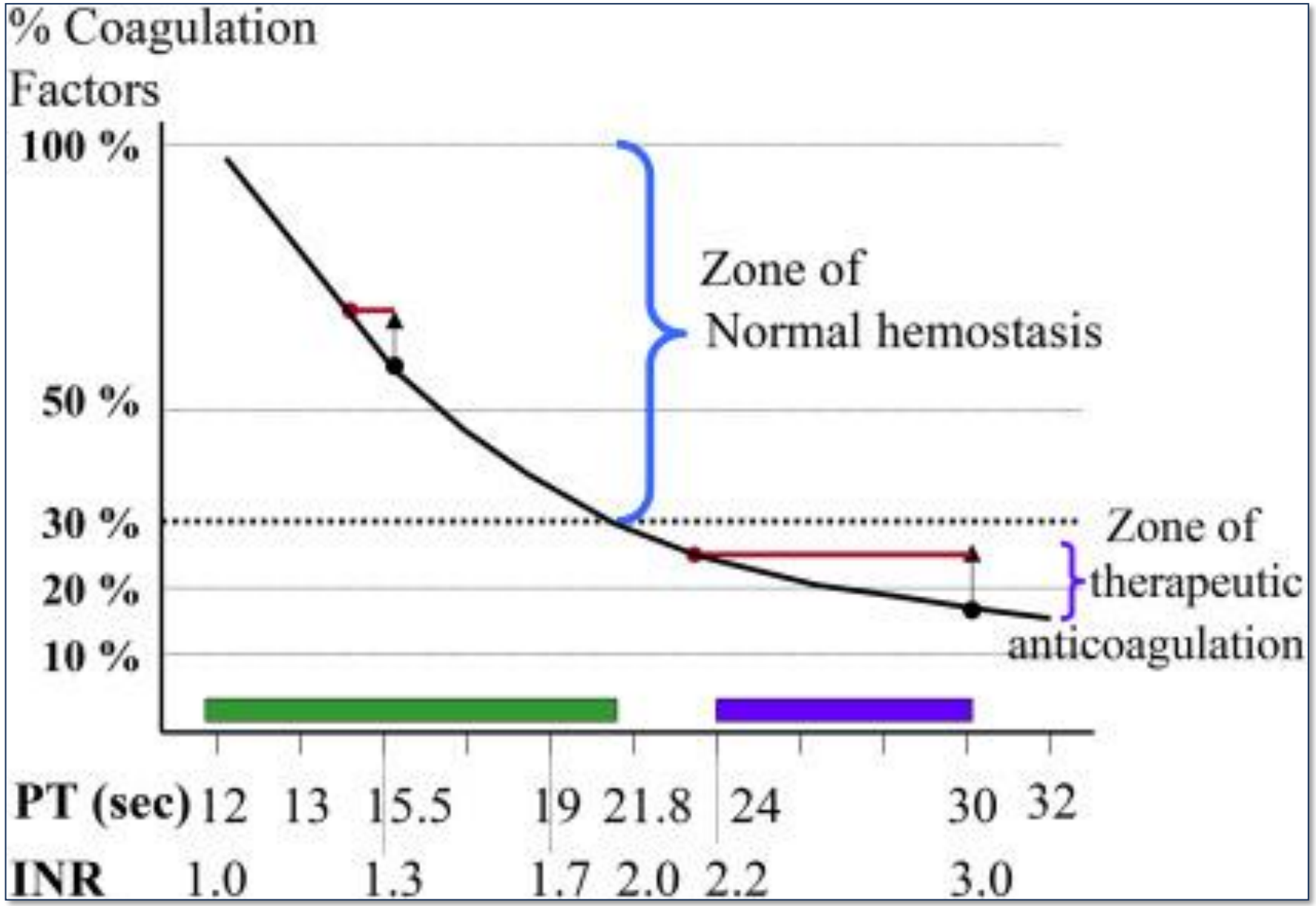
- 2017 meta-analysis of Intercept psoralen-treated platelets vs standard platelets
  - Majority Adult Hem/Onc patients
  - Moderate-quality evidence psoralen-treated platelet transfusion does not affect the risk of clinically significant or severe bleeding

Estcourt et al. Cochrane Database of Systematic Reviews 2017

Plasma

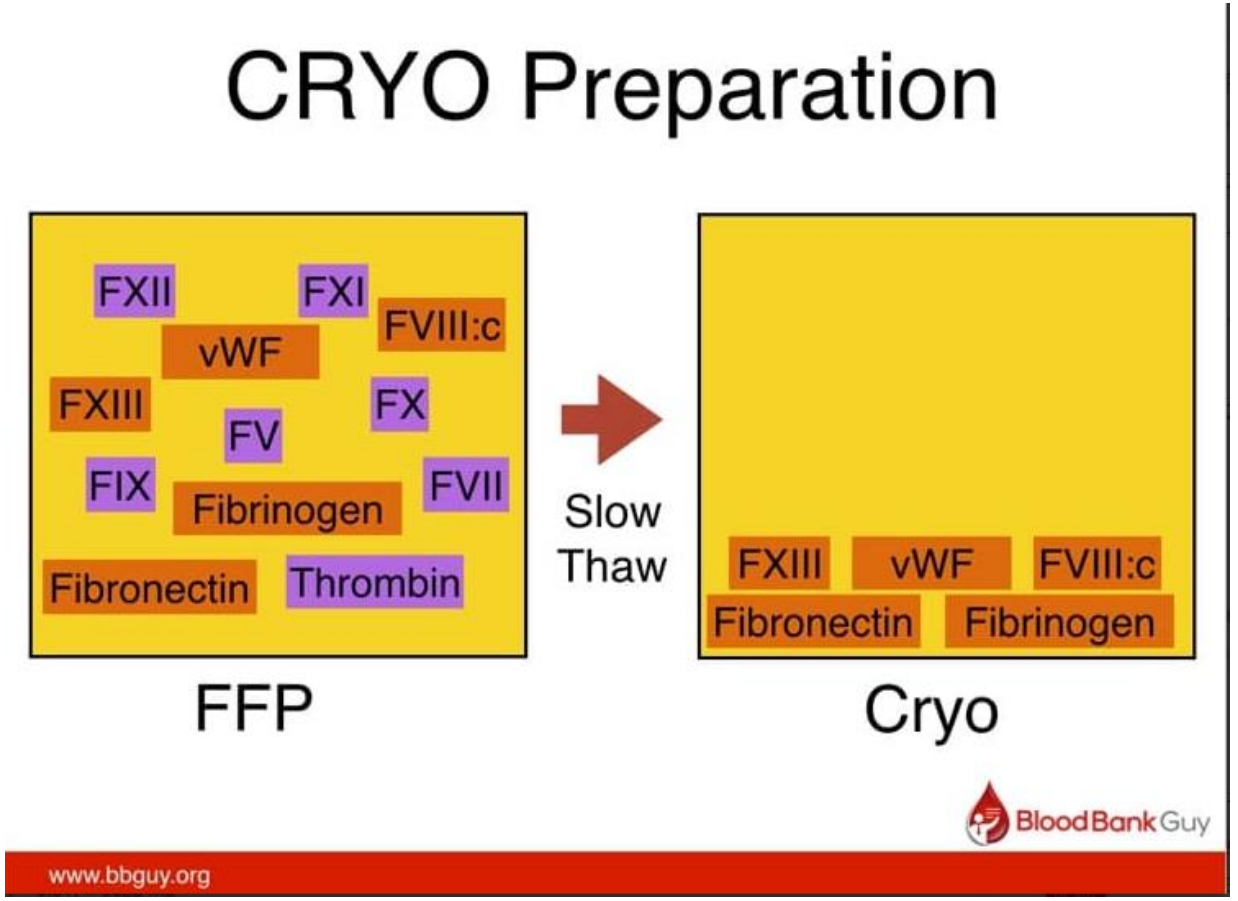
- All coagulation factors including fibrinogen
- 200-250ml volume/unit
- Dose: 10-15 mL/kg

Indication	Grade of recommendation
Liver disease with active bleeding	1C+
Warfarin reversal (If PCC unavailable)	1C+
DIC with active bleeding	1C+
Massive transfusions	1C+
Clotting factor deficiency in the absence of recombinant products	1C+
Plasmapheresis in TTP	1A+
Bleeding ppx/INR decrease prior to surgery/procedure*	2C
Volume expander	2C


% Coagulation Factors
100 %
50 %
30 %
20 %
10 %
Zone of Normal hemostasis
Zone of therapeutic anticoagulation
PT (sec) 12 13 15.5 19 21.8 24 30 32
INR 1.0 1.3 1.7 2.0 2.2 3.0

Cryoprecipitate

- Prepared by slowing thawing Plasma in the cold
  - Insoluable precipitate: Factors VIII, XIII, vWF, fibronectin, **fibrinogen**

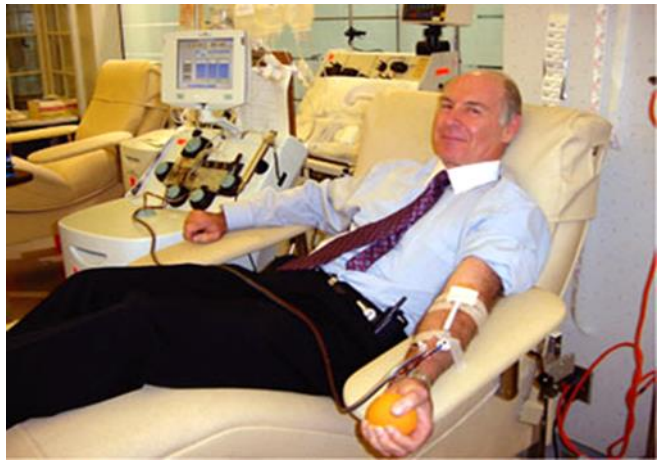


Cryoprecipitate

- Dosing
  - Dose: 1 unit/10 kilo will increase fibrinogen ~100 mg/dL
  - One Cryoprecipiate pool= 5- 6 donor units
  - **Two Cryoprecipiate pools = ~ 1 adult patient dose**
- Indicated:
  - Hypofibrinogenemia (< 100 - 150 mg)
  - Disseminated Intravascular Coagulation (DIC)
  - Obstetrical bleeding

Granulocytes

- Unit:
  - > 1 x 10^{10} granulocytes
  - Shelf-life 24 hours
- Indicated
  - To treat severe and refractory fungal infections in neutropenic patients
  - Controversial efficacy due to underpowered studies
- Not indicated
  - To prevent infections in neutropenic patients



- ✓ A donor receives stimulation with steroids and/or G-CSF
- ✓ Granulocytes are collected through apheresis procedure (~ platelets)

Estcourt LJ,. Cochrane Database of Systematic Reviews 2016, Issue 4.

Adverse Reactions

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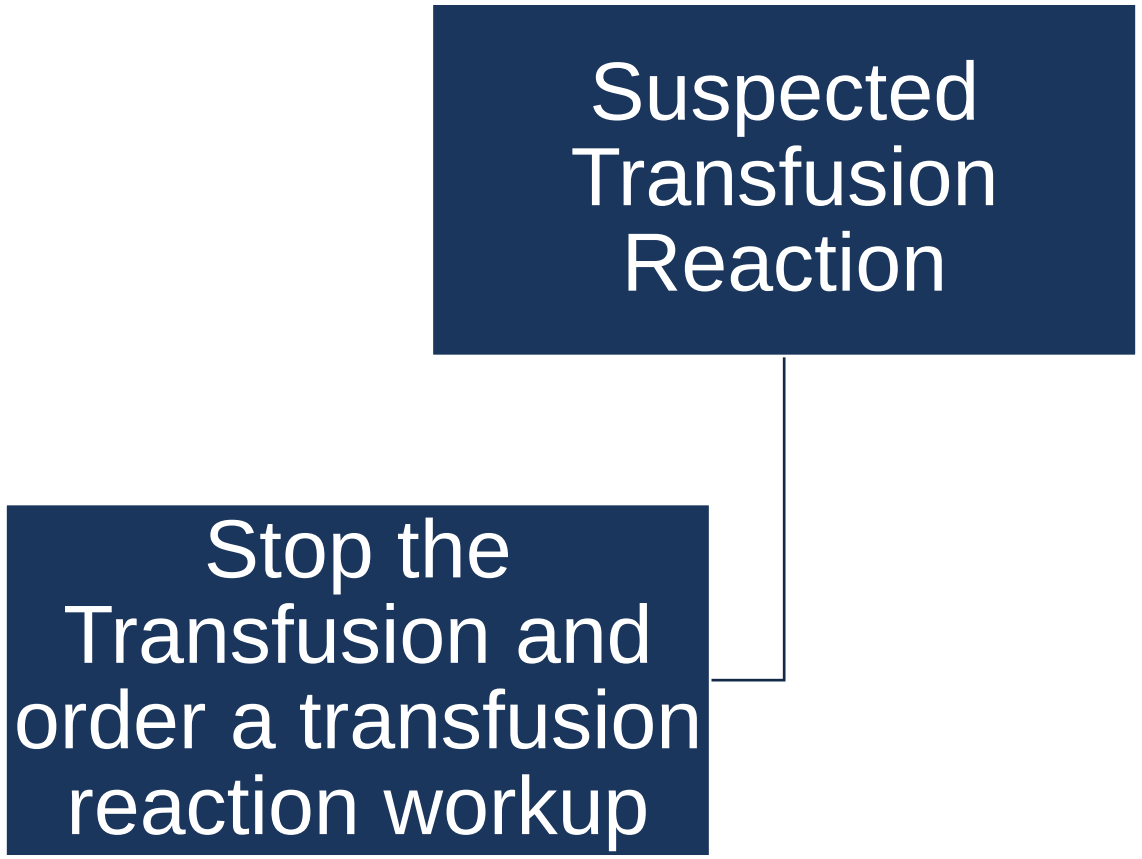
(5.06.05)
Which of the following is done in a transfusion reaction investigation? (select all that apply)

ⓘ Start presenting to display the poll results on this slide.

Which of the following is done in a transfusion reaction investigation? (select all that apply)

- **A) Stop the transfusion**
- **B) Clerical Check**
- **C) Return the unit to the blood bank**
- **D) Draw a post transfusion sample**
- E) Restart the unit once workup is negative

Transfusion Reaction Workflow



Transfusion Reaction Workup: Nursing Workflow

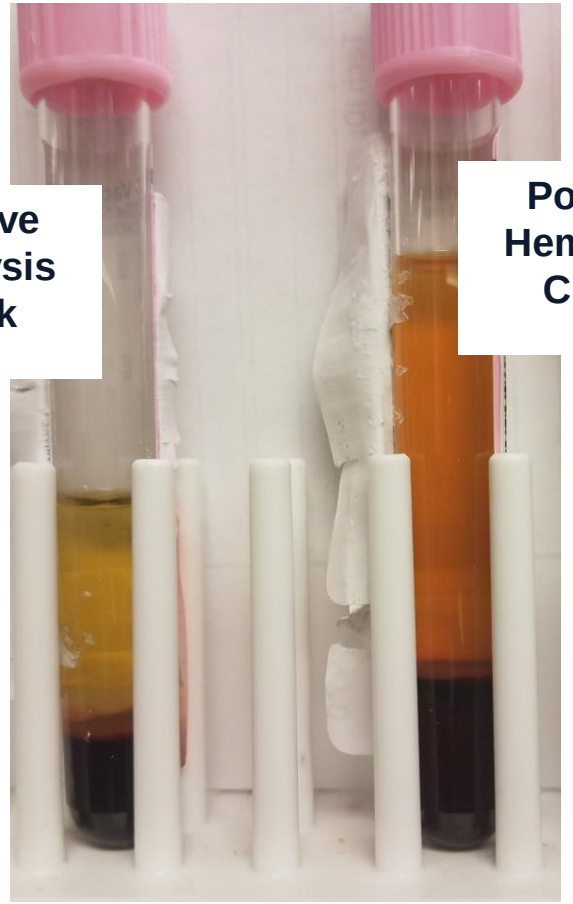
1. STOP transfusion but keep the IV open with IVF.
2. Notify Clinical Team
3. Monitor vital signs
4. Clerical Check- right unit to right patient?
5. Notify the Blood Bank
6. Send a post-transfusion sample and remainder of unit/tubing
7. Document.

Transfusion Reaction Workup: Blood Bank Workflow

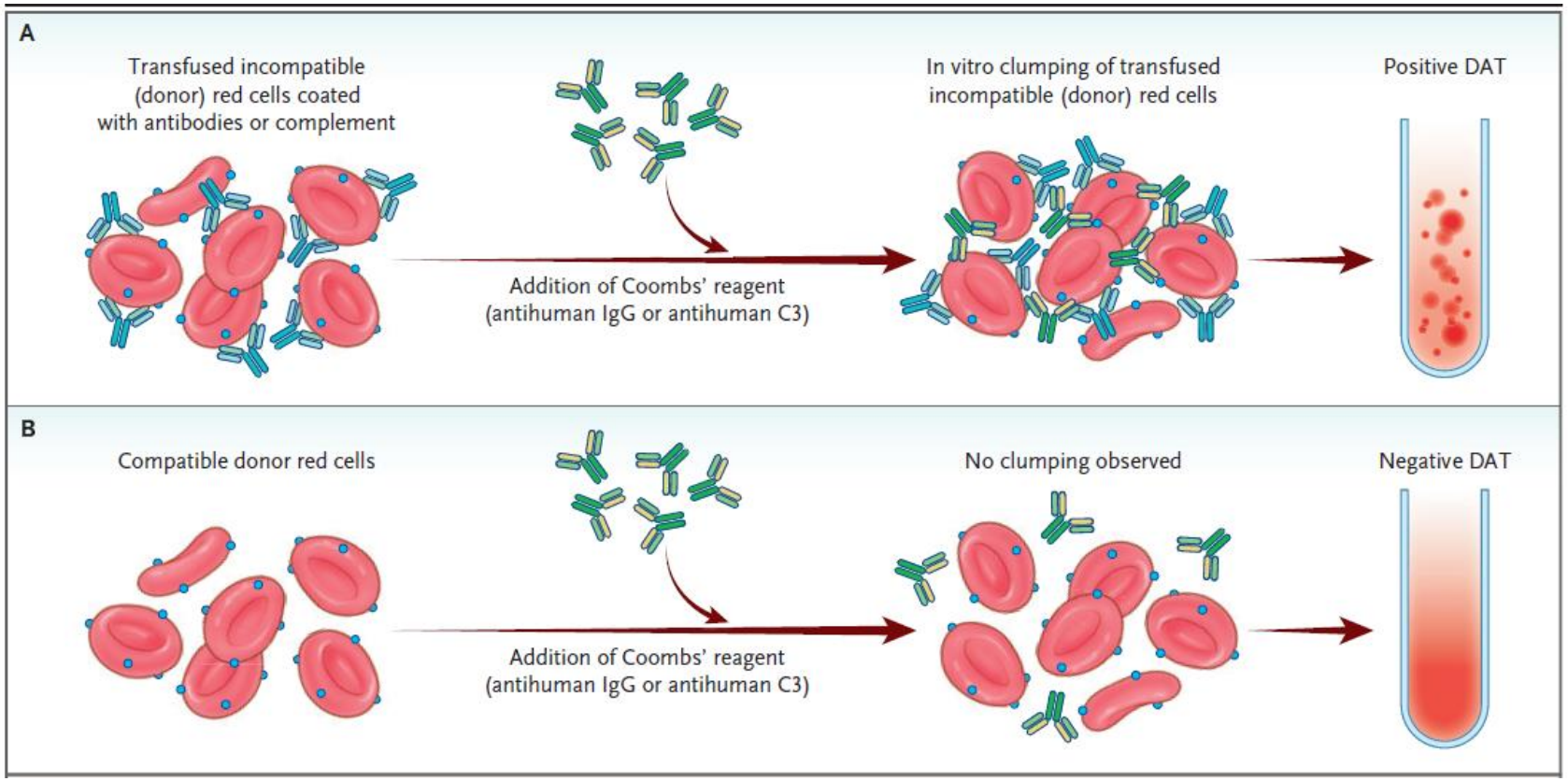
Clerical Check (Right unit for right patient?)

Post-Transfusion Sample

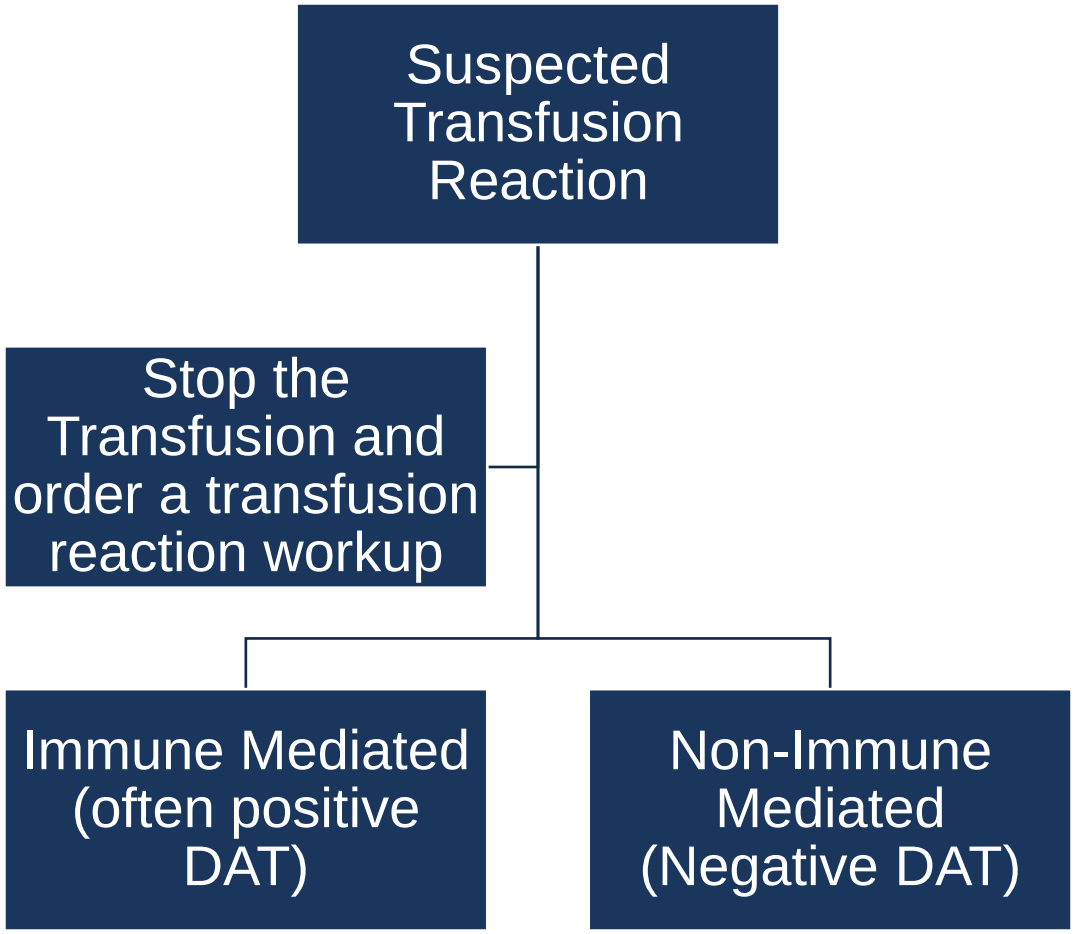
- Hemolysis check (hemoglobinemia, red plasma → intravascular hemolysis)
- Repeat ABO type (right sample sent for right patient?)
- Direct antiglobulin test (Coomb's test)



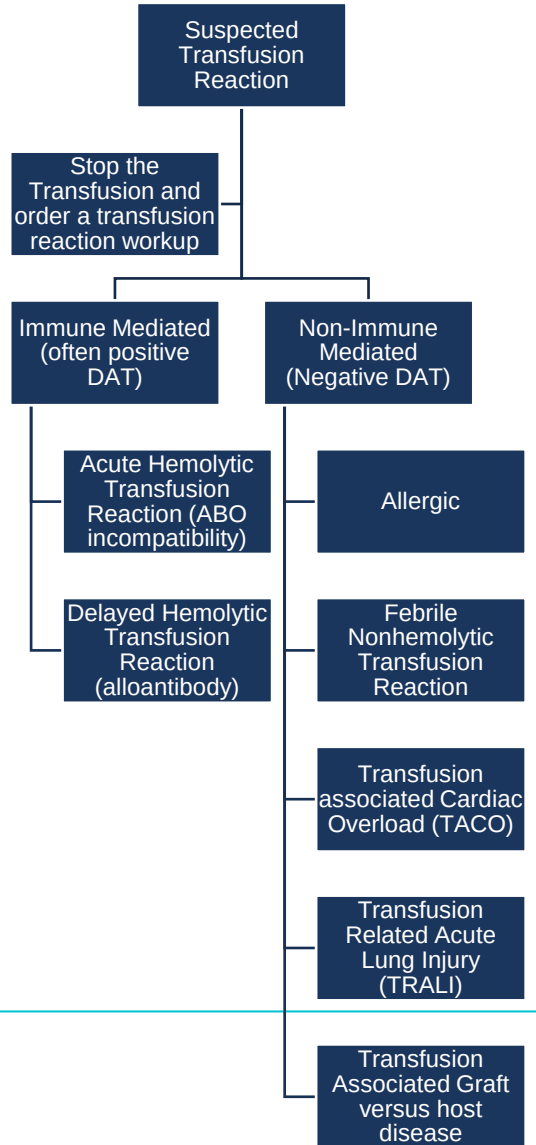
DAT (Direct Antiglobulin Test)



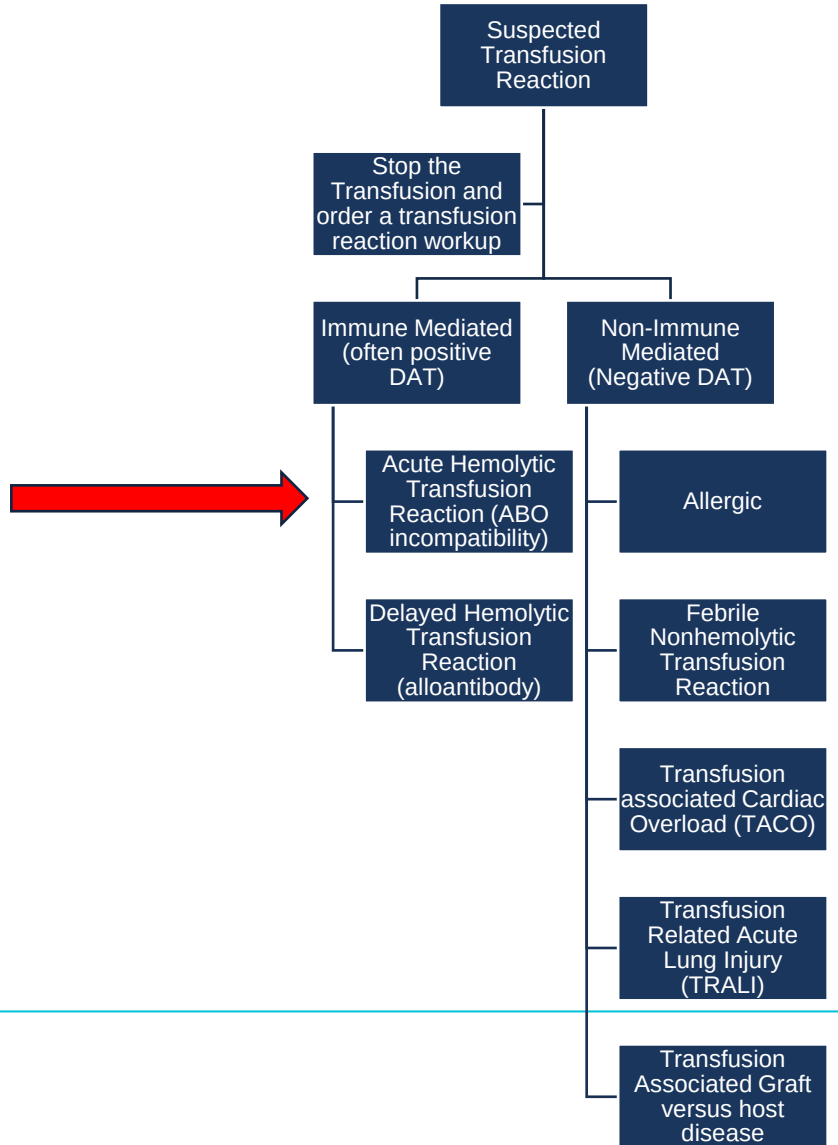
Transfusion Reaction Workflow



Transfusion Reaction Workflow



Transfusion Reaction Workflow



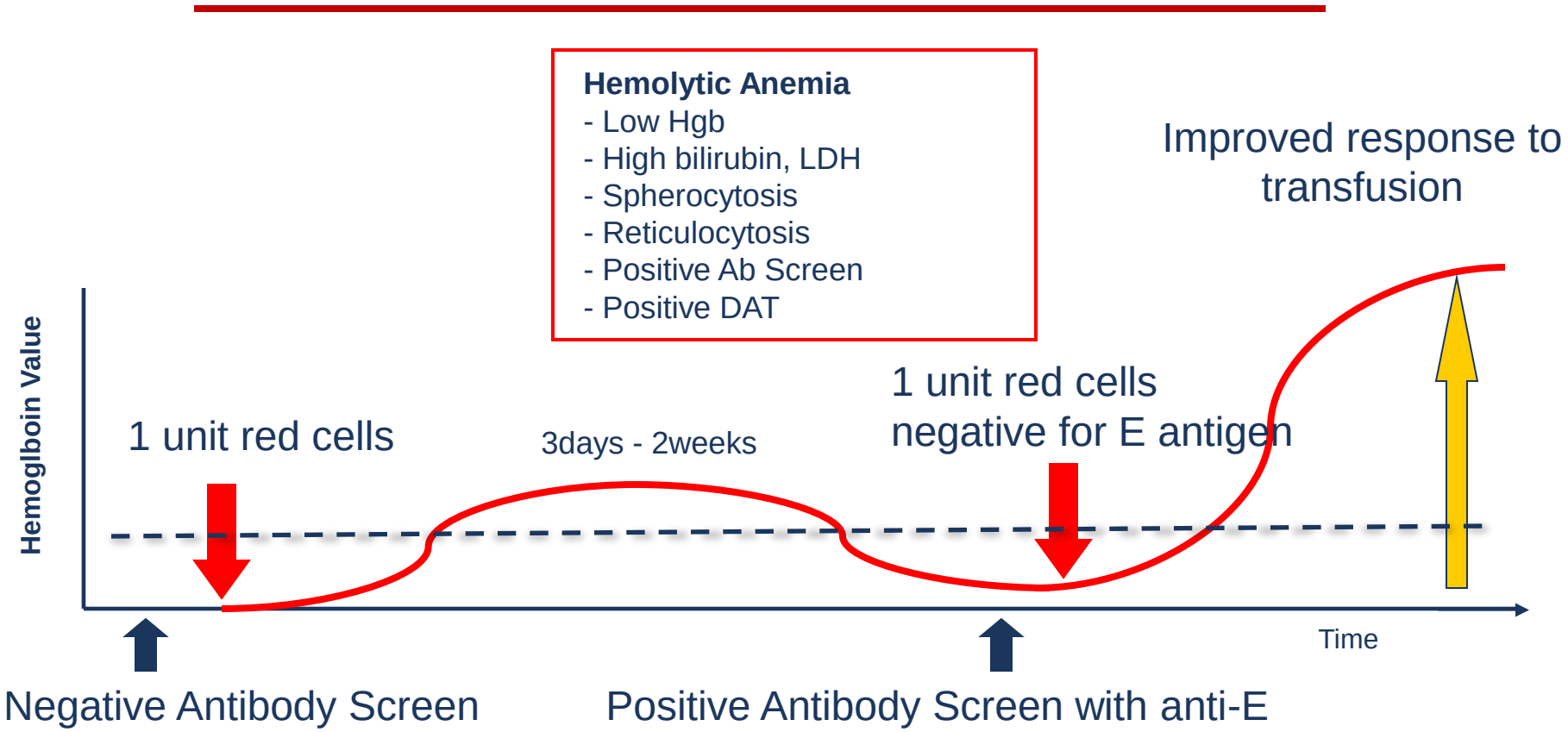
Acute Hemolytic Transfusion Reactions

- ABO incompatibility – complement fixing IgMs cause intravascular hemolysis
  - Not every patient with have the same severity of reaction to an incompatible transfusions
  - Symptoms range, however, as it can be fatal in some patients who receive as little as 30 mL, and other patients receive larger-volume transfusion with no or minor symptoms.
- *Clinical Findings:* Pain at site of infusion, sense of impending doom, chest pain, flank pain, back pain, hypotension or hypertension, hemoglobinemia, hemoglobinuria, ARF, DIC
- Treatment: Supportive care, hydration, fluids, compatible transfusions

Delayed Hemolytic Reactions

- IgG alloantibody mediated extravascular by macrophages in spleen/liver-generally late or amnestic responses
- ***Clinical Findings:*** Decrease in hemoglobin after recent transfusion (24 hours-28 days), jaundice
- *Treatment:* generally mild, supportive transfusions as needed with antigen negative units

Delayed Hemolytic Transfusion Reaction (DHTR)



Allergic Transfusion Reactions

Urticarial reactions

- Unknown cause, though to be IgE-mediated or independent
- Symptoms: Mild, flushing, pruritis, urticaria
- Management:
  - Treat with anti-histamines
  - Ok to re-start transfusion at slower rate if symptoms subside or stabilize

Anaphylactic reactions

- IgE mediated histamine release in patient
- Symptoms: Severe, Hypotension, dyspnea, airway edema, anaphylaxis
- Management:
  - Requires emergent care
  - Epinephrine, corticosteroids, antihistamines, pressors and intubation if necessary

Preventing Allergic Transfusion Reactions

- If only a mild reaction
  - Thought to be from donor product
  - Most do not recur with additional transfusions
- Data on use of pre-medication does not prevent ATRs in general even in those with history of prior reactions

Preventing Allergic Transfusion Reactions

Things that are known to decrease risk of allergic reactions

- Less plasma in the product (Plasma > Platelets > RBC)
  - Volume Reduction (remove 2/3 of plasma)
  - Platelets in additive solution (PAS) (replaces 2/3 of plasma)
  - Washing RBC + Platelets (remove > 99% of plasma)
    - A lot of resources, expiration < 24 hours

Things that have not been proven to decrease risk of allergic reactions

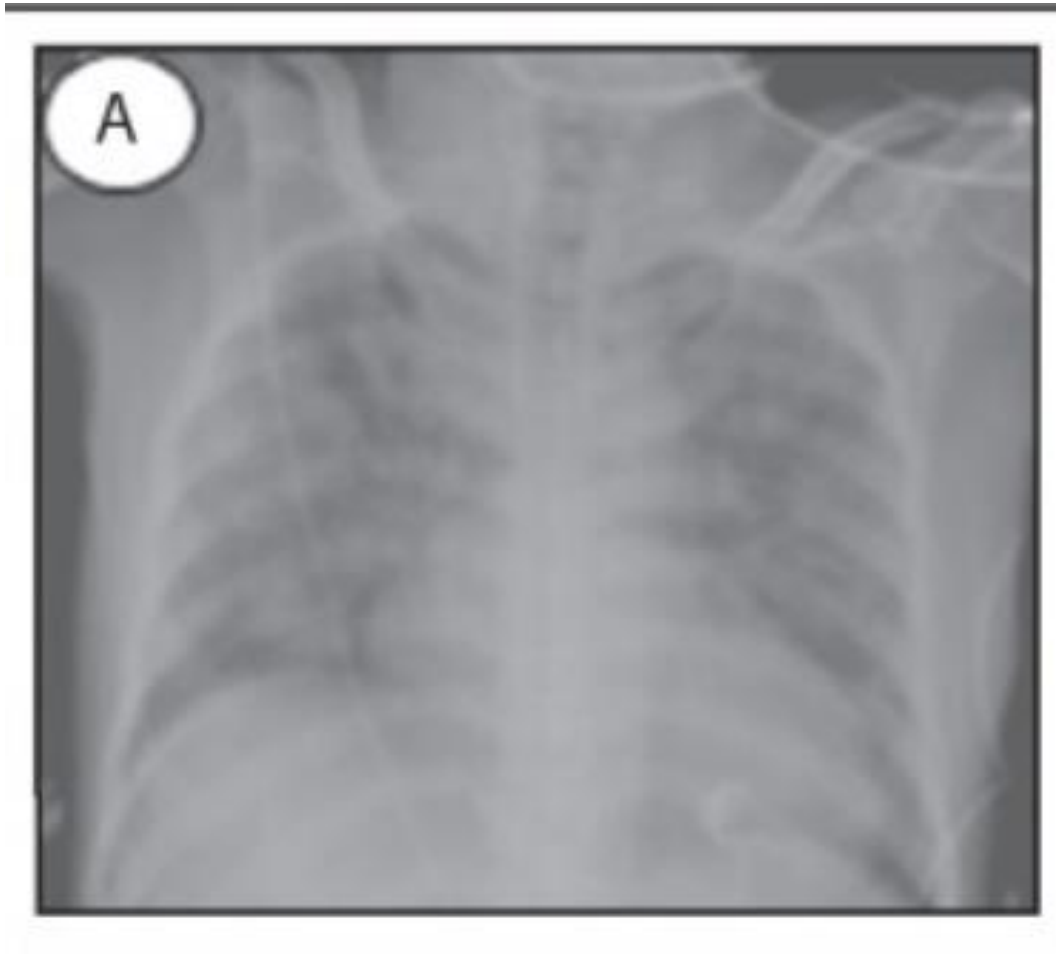
- Premedication with anti-histamines, steroids

Febrile (Non-Hemolytic) Transfusion Reaction

- Relatively common
- Cause
  - Cytokines from donor leukocytes
- Signs & Symptoms
  - Fever (2ºF), chills within a few hours of transfusion
  - N/V, hypotension
- Treatment
  - Antipyretics
  - Ok to give next transfusion if no hemolysis
- Prevention
  - Leukoreduction
  - Filtration of blood products to remove WBC
    - Decrease risk of FNTHR, HLA alloimmunization, CMV transmission

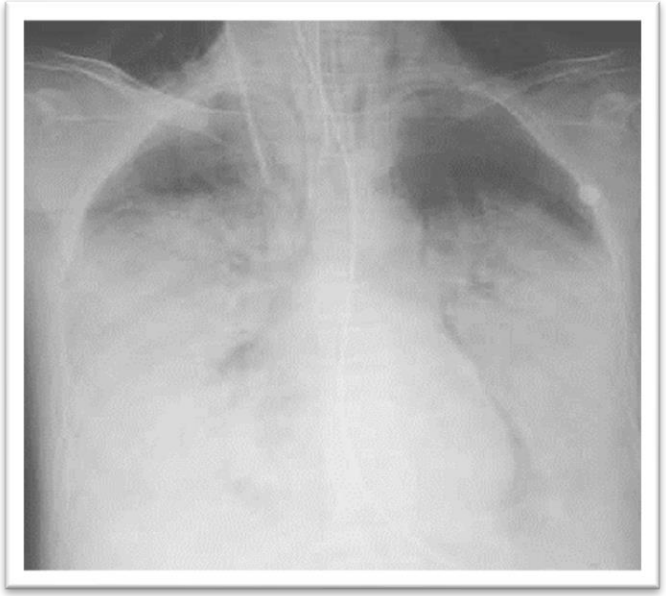
Transfusion Associated Circulatory Overload (TACO)

- Volume overload caused by transfusion
- Risk factors: elderly, heart/kidney failure, large volume/multiple transfusions
- Clinical Findings: Respiratory symptoms (Shortness of breath, rales, crackles) and chest x-ray findings (lower lobe infiltrates)
- Treatment: Diuretic
- Prevention:
  - Slower transfusions
  - Volume reduction
  - Diuretics with transfusions

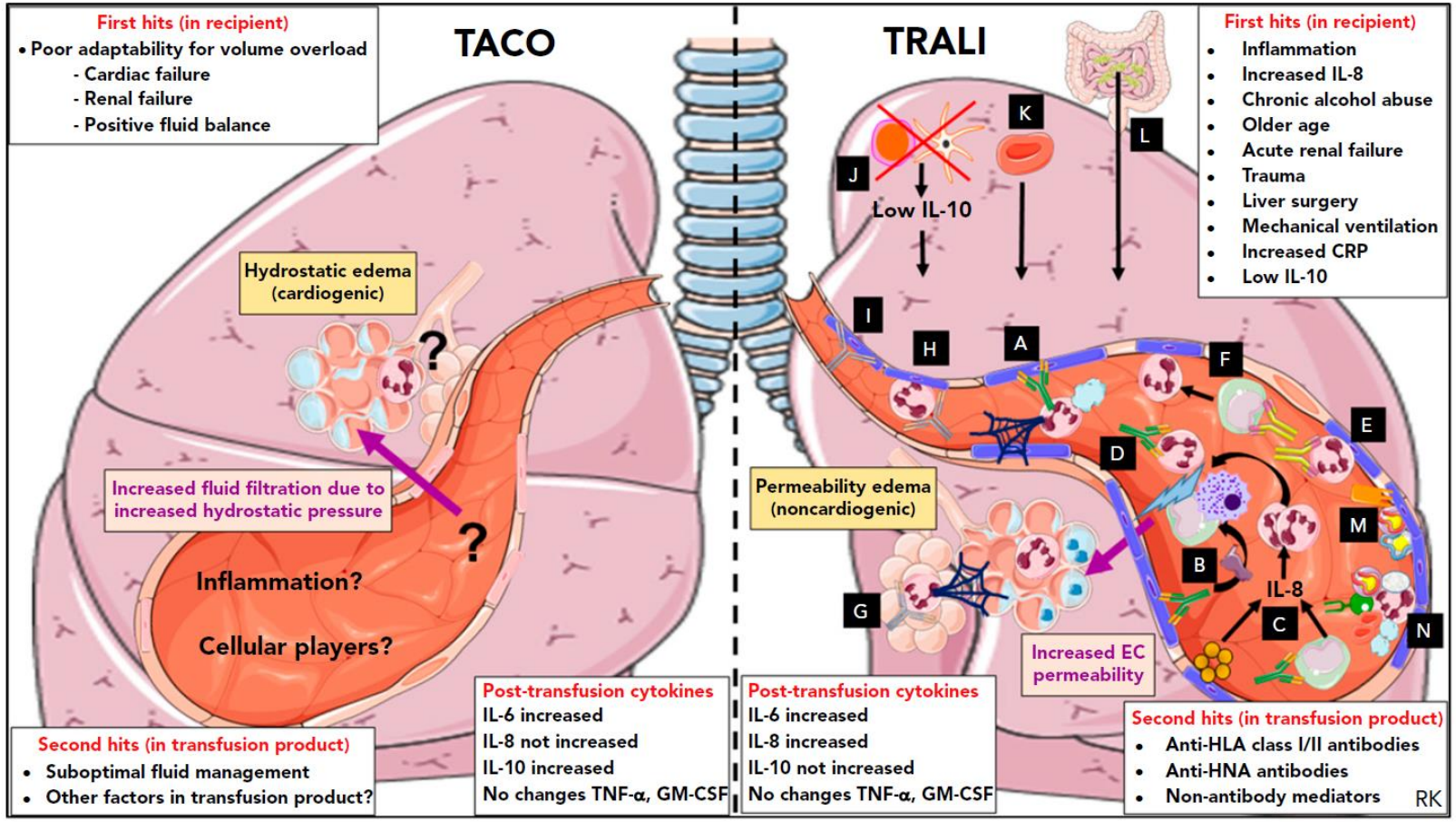


Transfusion Related Acute Lung Injury (TRALI)

- Acute respiratory distress syndrome (ARDS) within 6h of transfusion
- Cause: not well understood
  - donor HLA and HNA antibodies
  - Lipid activations of neutrophils in donor plasma Clinical findings: respiratory symptoms, chest x-ray findings (bilateral pulmonary edema)
- Treatment:
  - supportive care, improve after 72 hours, 5-10% mortality
- Prevention:
  - Use of plasma from male donors only (lower incidence of HLA/HNA antibodies)

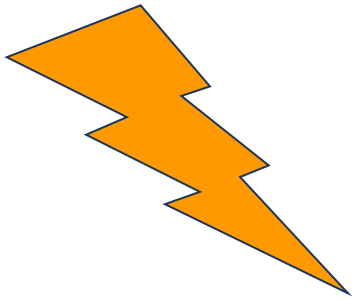


Blood Component Risks: TACO vs. TRALI



Transfusion Associated Graft Versus Host Disease (TA-GVHD)

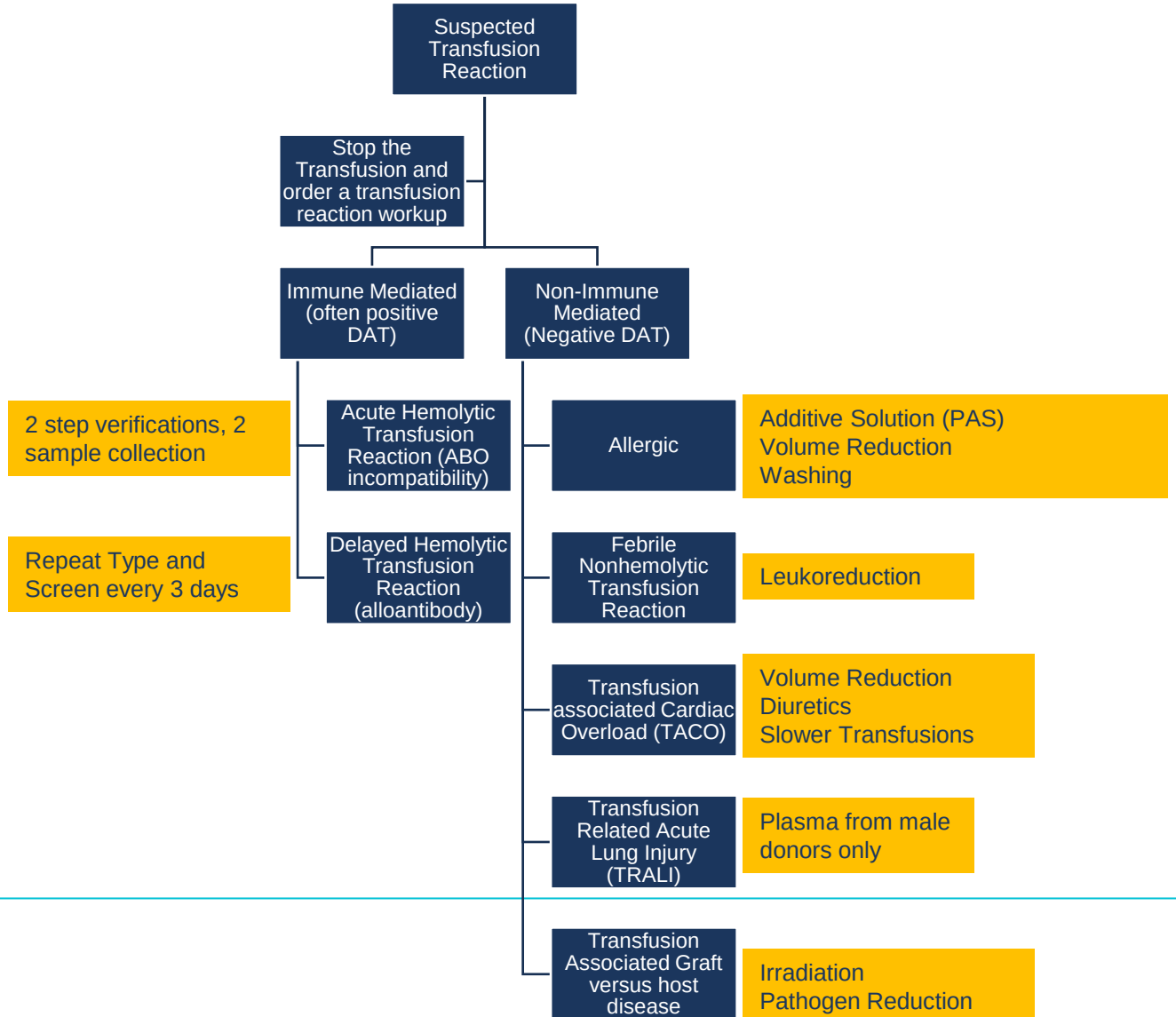
- Engraftment and proliferation of donor lymphocytes in transfusion recipient.
- Donor cells are able to attack and proliferate
  1. Hematopoietic cells → refractory pancytopenia
  2. Other organ systems: Fever, enterocolitis, rash, hepatitis
- Risk Factors: Patient with decreased T cell response to attack donor lymphocytes
- Prevention: Irradiation or Pathogen Reduction
  - Inactivates donor lymphocytes to prevention proliferation



Transfusion Associated Graft Versus Host Disease (TA-GVHD)

- Risk Factors: Patient with decreased T cell response to attack donor lymphocytes
  - Hematologic malignant
  - Bone marrow transplant
  - High dose chemotherapy
  - Premature infants
  - Immunodeficiencies
  - Patients and donors with shared HLA antigens
    - Directed donations from blood relatives
- Most people are not at risk!

Transfusion Reaction Workflow



Team Bios

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Clinical interests include: Pediatric hematology, transfusion support for neonatal patients, immune hemolytic anemias

Current research projects: Evaluation the utility of DAT in cord blood testing

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Medical Director, Division Head Transfusion Services

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Current research projects: Impact of alloimmunization in transplant, indications for red blood cell transfusions, quality of life

Fred Hutch Cancer Center

Thank you