

# Blood Bank III

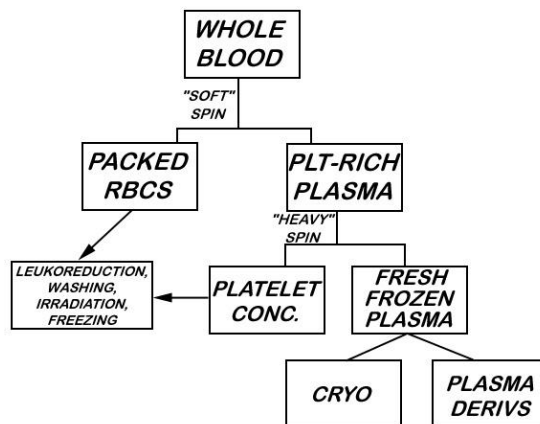
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## Blood Components and Component Therapy

### I. General

#### A. Basic concept of “component therapy”

1. More efficient use of products by giving a patient what he needs and avoiding what he doesn't need.
2. Made possible by advent of plastic bags around 1950.
3. Single unit may be made into numerous components (see figure below representing classical US method).



#### B. Anticoagulant/preservative solutions

1. Allows blood to be stored for extended periods without drastic effects on most metabolic and therapeutic qualities
2. Red cell storage defined by demonstrating 75% survival of transfused cells at 24 hours after transfusion (FDA)
3. Historic anticoagulant/preservatives
  - a. Citrate-phosphate-dextrose (CPD) and citrate-phosphate-dextrose-dextrose (CP2D)
    - 1) Allow 21 days of RBC/whole blood storage
  - b. Citrate-phosphate-dextrose-adenine (CPDA-1)
    - 1) Very similar to CPD but with 17.3 mg of adenine (no adenine in CPD)
    - 2) Allows 35 days of RBC/Whole Blood storage
4. Additive solutions (“Adenine Saline” additives)
  - a. Increases shelf life of RBCs to 42 days
  - b. Most common types
    - 1) AS-1 (Adsol®)
    - 2) AS-3 (Nutricel®)
    - 3) AS-5 (Optisol®)
  - c. Specifics vary, but all add more dextrose and adenine to increase blood shelf life.
  - d. AS-1 and AS-5 contain mannitol for RBC preservation

## Pathology Review Course

5. Preparation of additive solution RBCs:
  - a. RBCs with additive solution process:
    - 1) Blood collected in CPD or CP2D (NOT CPDA-1), spun, then mixed with 110 mL additive solution for 500 mL collections (100 mL for 450 mL bags)
    - 2) This gives a product with more volume and less plasma (HCT usually 55-65%)
6. Know storage details for various products (Table 1)

Product	Storage	Product	Storage
<b>RBCs / Whole blood</b>	35 days (CPDA-1) 42 days (Additives) 1-6 C	<b>Granulocytes</b>	24 hours; 20-24 C (no agitation)
		<b>Fresh Frozen Plasma</b>	1 year; -18 C OR 7 years, -65 C; 24 hours at 1-6 C after thaw
<b>Frozen RBCs</b>	10 years; -65 C; 24 hours after thaw	<b>CRYO</b>	1 year at -18 C 6 hours at 20-24 C after thaw (4 hours if pooled)
<b>Washed RBCs</b>	24 hours; 1-6 C		
<b>Platelets</b>	5 days; 20-24 C (gentle agitation); 4 hours if pooled		

### C. Quality control of blood products

1. Blood is a controlled product that is tightly regulated by the FDA (with more regulations from AABB & CAP).
2. Very specific, detailed requirements acceptability

Product	QC	Product	QC
<b>RBCs</b>	HCT < 80% (all), ≥ 50 g HGB in 95% (apheresis RBCs)	<b>Apheresis platelets</b>	≥ 3.0 x 10 <sup>11</sup> and pH ≥ 6.2 in 90%
<b>RBCs leukoreduced</b>	≤ 5 x 10 <sup>6</sup> WBCs in 95%, retain 85% of RBCs	<b>Apheresis platelets leukoreduced</b>	Above + < 5.0 x 10 <sup>6</sup> residual WBCs in 95%
<b>Platelets (PC)</b>	≥ 5.5 x 10 <sup>10</sup> and pH ≥ 6.2 in 90%	<b>CRYO</b>	Factor VIII ≥ 80 IU (all) Fibrinogen ≥ 150 mg (all)
<b>Platelets (PC) leukoreduced</b>	≥ 5.5 x 10 <sup>10</sup> in 75%, pH ≥ 6.2 in 90%, AND < 8.3 x 10 <sup>5</sup> WBCs in 95%	<b>Granulocyte concentrate</b>	≥ 1.0 x 10 <sup>10</sup> in 75%

## II. Blood and Components

### A. Blood components

1. **Red blood cells/additive solution red blood cells**
2. **Platelets**
  - a. Whole blood-derived platelets (WBD-PLTs)
  - b. Apheresis-derived platelets (AD-PLTs)
3. **Modified RBCs and platelets**
  - a. Leukocyte reduced products

- b. Irradiated products
- c. Frozen products
- d. Washed products
- 4. **Plasma and derivatives**
  - a. Fresh frozen plasma (FFP)
  - b. FFP alternatives (including FP24)
  - c. Cryoprecipitate (“antihemophilic factor”)
  - d. Factor concentrates
  - e. Other plasma derivatives
- 5. **Miscellaneous products**
  - a. Granulocyte concentrate
  - b. DDAVP
  - c. Recombinant activated factor VII (NovoSeven)

## **B. Whole blood**

- 1. The original blood product!
- 2. Minimal availability in most blood banks today
- 3. Specifics:

<b>Volume:</b>	450-500 mL
<b>Contents:</b>	RBCs (200-250 mL) Plasma (250-300 mL) WBCs ( $10^9$ ) Platelets Anticoagulant (63 or 70 mL)

- 4. Potential indications:
  - a. Massive blood loss (30-40% or more of blood volume)
    - 1) Trauma/emergency transfusions most commonly
    - 2) Use may lead to less exposure by providing coag factors (and maybe a few functional platelets), as well as volume
    - 3) Whole blood must be ABO identical due to plasma; tougher to use in emergencies.
  - b. Exchange transfusions in neonates (more often “reconstituted” from separate RBCs and FFP)
  - c. Autologous transfusions
- 5. Contraindications:
  - a. Anything where something more specific to the patient’s needs would be better.
- 6. Storage Time and Conditions
  - a. Length depends on anticoagulant/preservative used
  - b. 1-6°C.

## **C. Red blood cells with and without additives**

- 1. The most commonly used blood component
- 2. Prepared by centrifugation and removal of most of plasma layer of whole blood, or by apheresis collection.
  - a. May be transfused without modification after preparation or may use additive solution

3. Specifics:

<b>Volume:</b>	350 mL (incl. additive)
<b>Contents:</b>	RBCs (200-250 mL) Plasma ( $\leq$ 50 mL) WBCs ( $10^9$ ) and PLTs Anticoagulant (63 or 70 mL) Additive solution 200-250 mg iron

4. QC Requirements:

- a.  $HCT \leq 80\%$  for all RBCs
- b. Apheresis RBCs: 95% must have  $>50$  g HGB or 150 mL of RBCs

5. Indications

a. **Need for increased oxygen-carrying capacity**

1) How do you decide?

- a) Hemoglobin level is a very inaccurate indicator of the need for transfusion
- b) Anemia compensation (HGB dissociation curve shift to right, increased cardiac output, decreased blood viscosity) must be considered
- c) Cardiac factors,  $O_2$  demand often overlooked
- d) Measuring mixed venous saturation ( $S_{vO_2}$ ) and comparing to arterial levels ( $S_{aO_2}$ ) gives an estimate of current oxygen use
  - Example: 25% extraction ( $S_{aO_2}$  100%,  $S_{vO_2}$  75%) is normal; extraction may go up to 75% or more when necessary (exercise, etc)
  - Heart muscle has little reserve; extracts close to 75% normally
  - Overall oxygen extraction ratio of 0.5 (50%) or more at rest is deemed "critical."
- e) All factors (including blood volume, heart function, ability to increase cardiac output, and  $O_2$  requirements) should be addressed when considering transfusion.
  - Because of compensation, chronic anemia is less likely to need transfusion (and may be dangerous!)

2) Situations that *may* require red cell transfusion:

- a) Acute hemorrhage (over 30% of blood volume acutely)
- b) Hemolysis
- c) Marrow failure

b. **Exchange transfusions**

- 1) Sickle cell patients in crisis or presurgery
- 2) Hemolytic disease of the newborn/fetus (HDFN)

c. **Thresholds**

- 1) Using a universal threshold like HGB 10 g/dL, HCT 30% is outdated, silly, and bad practice

- 2) All specialties have published recommendations that agree with statement above
- 3) General guidelines:
  - If HGB < 6 g/dL, transfusion usually needed
  - If HGB >10 g/dL, transfusion rarely needed
  - If HGB is between 6 and 10, clinical judgment, assessment of situation, etc, is required
- 4) Based on published studies:
  - It is *reasonable* to transfuse to higher threshold after acute MI (9-10 g/dL HGB)
  - It is *reasonable* to withhold transfusion in orthopedic or ICU patients (non-acute MI) until HGB is below 7-8 g/dL
  - It is *reasonable* to assess need for further transfusion after each unit given
6. Contraindications
  - a. Acute hemorrhage < 20% of blood volume
    - 1) Crystalloids are adequate in most of these cases
  - b. Chronic nutritional anemias (folate, B<sub>12</sub>, iron)
7. Expected effect (per unit)
  - a. HCT increases 3%, HGB 1 g/dL (without acute bleeding or hemolysis).
  - b. Effect can be measured 15 minutes after transfusion.
8. ABO compatibility
  - a. ABO type of transfused RBCs must be compatible with recipient plasma ABO antibodies
  - b. Always protect the transfused cells! (See chart)

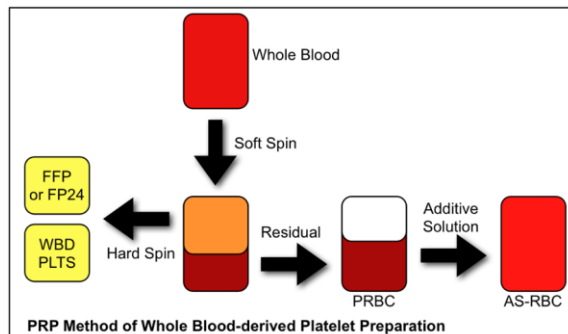
		DONOR			
		A	B	AB	O
RECIPIENT	A	✓			✓
	B		✓		✓
	AB	✓	✓	✓	✓
	O				✓

9. Storage and shipping
  - a. Same as for whole blood if CPD, CPDA-1 used
  - b. 42 days at 1-6 C if additive solutions used
  - c. Shipping temperature 1-10 C
10. Compatible fluids
  - a. Normal saline (0.9%)
  - b. ABO compatible plasma
  - c. 5% albumin
  - d. Red cells should not contact lactated Ringer's (LR), D5W, 0.45% NS, antibiotics/other drugs, or TPN
    - 1) Hypotonic solutions - red cells swell and burst; hypertonic solutions - red cell shrinkage.
    - 2) LR has enough calcium to counteract the citrate anticoagulant in blood (LR has 3 mEq Ca<sup>2+</sup>/L)

11. Red cell types (most covered in other parts of handout)
  - a. Red Blood Cells, Low Volume
    - 1) Prepared from a low-volume (less than 450 mL in 500 mL bag) whole blood collection
  - b. Red Blood Cells, Adenine Saline Added
  - c. Red Blood Cells, Leukocyte-reduced
  - d. Red Blood Cells, Frozen
  - e. Red Blood Cells, Deglycerolized, and Red Blood Cells, Washed
  - f. Red Blood Cells, Irradiated
  - g. Red Blood Cells, Apheresis (often “double” products)
  - h. Red Blood Cells, Rejuvenated
    - 1) RBCs may be “rejuvenated” up to 3 days after expiration in CPD or CPDA-1, or up to the expiration date in AS-1 with “Rejuvesol.”
    - 2) Restores ATP, 2,3-DPG levels to near-normal levels
    - 3) Rejuvenated product is often frozen (up to 10 years for CPD, CPDA-1 RBCs; up to 3 years for AS-1)
    - 4) Thawed (or immediately transfused) product must be washed, then transfused within 24 hours.

**D. Platelets (See Oct 2010 podcast on iTunes)**

1. **Whole blood-derived platelets (platelet concentrate, “random platelets”, WBD-PLTs)**
  - a. Prepared via centrifugation (“soft” spin then “hard” spin in the US) from a single whole blood unit.



- b. May be pooled at blood center or transfusion service
- c. Specifics

<b>Volume:</b>	40-60 mL
<b>Contents:</b>	PLTs ( $\geq 5.5 \times 10^{10}$ in 90%) Plasma (40-60 mL) WBCs ( $10^7$ ) pH $\geq 6.2$ (90%)

- d. Traditional dose
  - 1) 1 unit per 10 Kg body weight
    - a) Typically given six bags at a time in adults
  - 2) 10-15 mL/Kg in neonates

2. **Apheresis platelets (“single donor” platelets, AD-PLTs)**
  - a. Made from one donor via apheresis procedure
    - 1) Apheresis = removing whole blood from body, taking what you want, then returning the rest.
    - 2) Roughly 85% of platelets transfused in the US today are apheresis-derived
    - 3) Double or triple products can come from 1 donor (each must have  $\geq 3.0 \times 10^{11}$  PLTs)
  - b. Specifics:

<b>Volume:</b>	100-150 mL (or more)
<b>Contents:</b>	PLTs ( $\geq 3.0 \times 10^{11}$ in 90%)
	Plasma (100-150 mL)
	WBCs ( $< 5.0 \times 10^6$ )
	pH $\geq 6.2$ (90%)

- c. Why choose AD-PLTs over WBD-PLTs?
  - 1) **Limiting exposure**
    - a) For infectious disease transmission
      - 1 exposure vs. at least six for PC decreases risk of viral transmission
      - Also decreases risk of bacterial contamination
    - b) For HLA immunization?
      - Traditional thought: fewer donor exposures mean less immunization.
      - Not true! Risk more dependent on number of foreign antigens (not donors) seen; so leukoreduction is more important.
      - “TRAP” study: *NEJM*. 1997;337, No. 26, 1861-9.
  - 2) **Platelet refractoriness (see BB Practical)**
    - a) Lack of response to platelet transfusion (immune and nonimmune causes)
    - b) May be used as HLA-matched or crossmatched doses for immune refractoriness.
3. Indications for platelet transfusion:
  - a. **Thrombocytopenia**
    - 1) Transfusions may be prophylactic or therapeutic
    - 2) Prophylactic
      - a) Data supports a prophylactic threshold of  $<5,000$  (but most use 10K).
      - b) 20K threshold if patient has risk factors
        - i) Fever, sepsis, bleeding, thrombocytopeny
      - c) 50K if about to have major surgery
    - 3) Therapeutic
      - a) 50K if bleeding
      - b) 100K likely necessary for patients with intracranial and pulmonary hemorrhage

- 4) Controversy in LP, liver biopsy, endoscopy
  - a) Many use 50K prophylactic threshold
  - b) Not proven; count not predictive of bleeding
  - c) Outcome is operator-skill dependent!
- b. **Thrombocytopathy**
  - 1) Prophylactic transfusions not indicated
  - 2) Therapeutic indications:
    - a) Congenital defects with bleeding
    - b) Drugs (Plavix, ASA most common)
      - Increased bleeding in emergency surgical patients, especially cardiac surgery
    - c) External agents
      - Cardiac bypass
      - ECMO
    - d) Metabolic effects (chronic renal failure)
      - PLTs are *not* first-line defense for renal failure-related PLT dysfunction! (dialysis, DDAVP, Cryo, conjugated estrogens, etc).
4. Contraindications to platelet transfusion
  - a. **Thrombotic thrombocytopenic purpura (TTP)**
    - 1) ADAMTS13 deficiency leading to large vWF multimers and subsequent platelet microthrombi
    - 2) More platelets could lead to more thrombi
    - 3) May be overblown fear; studies show success. Can use PLTs in life-threatening situations
    - 4) Hemolytic-uremic syndrome (HUS) similar to TTP without neurologic symptoms; avoid PLTs
  - b. **Heparin-induced thrombocytopenia, type II**
    - 1) Antibody vs. heparin/platelet factor 4 complex
    - 2) These patients are at great risk for thrombosis, and platelets should be avoided if possible
  - c. **Immune/idiopathic thrombocytopenic purpura (relative)**
    - 1) Doesn't help platelet count, and patients don't usually bleed
    - 2) Use only if significant bleeding occurs
  - d. **Post-transfusion Purpura (PTP)**
    - 1) Uncommon antibody vs. transfused PLTs
    - 2) Most transfused PLTs will be antigen-positive
5. General comments about platelets
  - a. Dose
    - 1) WBD-PLTs traditionally 4-6 bags/dose
    - 2) AD-PLTs 1 bag/dose (assuming  $3.0 \times 10^{11}$  count)
    - 3) Recent study analyzing dosage: No difference in outcomes when given less than  $3.0 \times 10^{11}$  dose
    - 4) Neonates: 10-15 mL/Kg (may be concentrated)
  - b. Expected effect
    - 1) Generally, if one hour post-count increases by  $>20,000-30,000$ , response is adequate ("eyeball")

- 2) CCI has value in determining adequacy of response, even if count doesn't increase much
- 3) One-hour post-transfusion count is standard.
- c. Storage and shipping
  - 1) **5 days at 20-24 C** (with gentle agitation)
  - 2) Shipping range "as close as possible" to 20-24 C
  - 3)  $\leq 24$  hours without agitation during shipping
  - 4) 4 hours after transfusion service pooling (applies to WBD not pre-pooled in blood center)
- d. ABO and Rh
  - 1) PLTs do not require pretransfusion crossmatches, and ABO-incompatible platelets commonly given.
  - 2) Rh antigens are NOT present on platelets.
    - a) Platelet preparations may contain a few contaminating RBCs with Rh antigens.
    - b) Risk of making anti-D is low (0-18%)
    - b) Consider Rh prophylaxis in childbearing age women (1 vial RhIG per 2-3 weeks; effective as long as anti-D is detectable)
  - 3) Platelet ABO incompatibilities
    - a) Major = platelet ABO antigens incompatible with recipient plasma (like A plts to O recip)
      - Cleared from circulation faster; less effect
    - b) Minor = donor ABO *antibodies* incompatible with *recipient RBCs* (like O plts to A recipient)
      - A concern in children, neonates, and high-titer donors ("reverse" hemolytic reactions)
    - c) ABO-identical is best, but not always practical
- e. **Platelet sterility**
  - 1) *AABB Standards* requires centers to both *limit* and *detect* bacterial contamination of ALL platelets
  - 2) Limiting contamination
    - a) Careful skin preparation
    - b) Discarding initial 20-30 cc of blood
    - c) Exclusive use of apheresis platelets
  - 3) Detecting contamination
    - a) Culture-based methods
      - Require 24 hour wait before taking sample
      - BacT/ALERT (bioMerieux, Inc)
      - Enhanced Bacterial Detection System (eBDS)
    - b) Pre-issue testing
      - Verax PGD (Verax Biomedical)
    - c) Less sensitive methods
      - Gram stain, swirling, glucose checks
      - These methods no longer acceptable alone per AABB Standard 5.1.5.1.1

**E. Modifications to red cells and platelets**

**1. Leukocyte reduction (LR)**

a. Definitions (as of *AABB Standards*, 26<sup>th</sup> ed.)

1) In US:  $\leq 5 \times 10^6$  residual WBCs

2) In Europe:  $\leq 1 \times 10^6$  residual WBCs

3) US rules

a)  $\leq 5 \times 10^6$  white cells in 95% of tested units defines leukocyte-reduced:

- Red blood cells
- Apheresis platelets
- Whole blood

b)  $\leq 8.3 \times 10^5$  WBCs in 95% of tested units defines leukocyte-reduced:

- WBD-PLTs (NOTE:  $8.3 \times 10^5 \times 6 = 5 \times 10^6$ )

c) Each must also retain at least 85% of original component and meet all other QC standards.

b. Methods

1) Leukocyte reduction filters

a) 99.99% of white cells (“4 log” reduction)

2) Apheresis collection devices

a) Built-in leukoreduction methods.

b) Methods vary by manufacturer.

c. “Universal” LR is not mandated in the US, but vast majority of cellular components are leukoreduced

d. Types

1) “Prestorage” leukocyte reduction

a) Usually  $< 72$  hours after draw (no set guideline).

b) Inline filters at time of collection or post collection filters for red cells.

c) Apheresis LR is prestorage by definition

2) “Pretransfusion” leukocyte reduction

a) Immediately prior to transfusion (“bedside”)

- Least desirable
- Lack of available QC, poor training
- Many older studies done with this method

b) Better done in transfusion service before issuing

e. *Established* benefits:

1) **Prevention of febrile nonhemolytic transfusion reactions**

a) Benign reactions, but mimic early hemolysis

b) First type: WBCs secrete pyrogenic cytokines in bag *before* transfusion: Common with PLTs

c) Second type: Pyrogenic cytokines secreted *after* transfusion.

- Seen more commonly with RBC transfusions
- Recipient antibodies against transfused WBC antigens or immune complexes of donor WBCs and coating antibodies binding to macrophages

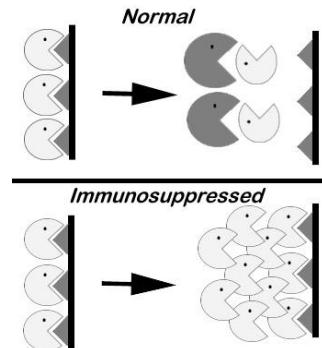
- d) Pretransfusion reduction works fine for second type, prestorage necessary to prevent first.
- 2) **Prevention of HLA immunization**
  - a) HLA antibody formation requires antigens be presented by transfused lymphocytes
  - b) LR works well to prevent this interaction
    - “TRAP” Study: NEJM 1997,337;26:1861-9.
  - b) Use for multiple transfusion recipients
  - c) UV-B treatment also works; not available in US.
- 3) **Prevention of CMV transmission**
  - a) Virus carried only in WBCs (monocytes).
  - b) *Blood*. 1995;86,3598-3603; “The Bowden Study” – landmark LR study
    - Filtered products equivalent to CMV seronegative in preventing seroconversion.
    - Neither method is perfect; very early infection may escape testing and/or leukoreduction
  - c) Generally, CMV- and LR products are considered equivalent (“CMV-safe”)
- f. *Potential* benefits:
  - 1) **Prevention of immunosuppressive effects of transfusion**
    - a) Controversial, not universally accepted
    - b) Transfusion probably immunosuppresses recipient (“immunomodulation”).
      - Many (but not all) studies show increased post-op infections and increased cancer recurrence in transfused patients
    - c) Donor WBCs are thought to be the cause of the immunosuppression
  - 2) **Reduction of bacterial/parasitic contamination**
    - a) Some studies suggest reduction in organisms like *Yersinia* or *Leishmania* with leukoreduction.
    - b) Probably not reliable enough to depend on!
  - 3) **Reduction of reperfusion injury post cardiac bypass**
  - 4) **Reduction in the risk of prion disease**
    - a) One of the reasons for universal LR in Europe
    - b) Prion-specific filters developed
- g. Contraindications:
  - 1) Prevention of graft vs. host disease.
    - a) Irradiation is only proven method (cases of TA-GVHD with leukoreduced blood reported)
  - 2) Transfusion of previously frozen products (FFP, cryo, etc)
  - 3) Transfusion of granulocyte concentrate
- 2. **Washing**
  - a. 1-2 L of saline removes about 99% of plasma.
  - b. Generally takes one to several hours (automated)

- c. Shelf life
    - 1) Red cells: 24 hours post-wash
    - 2) Platelets: 4 hours post-wash
  - d. Why bother?
    - 1) **Removal of plasma proteins for hypersensitivity (RBCs and platelets)**
      - a) Classic example: IgA deficiency
        - A few IgA deficient patients develop anti-IgA; exposure leads to anaphylaxis.
        - Requires intense washing (3L or so)
        - IgA-deficient donors are alternative
      - b) Removal of unwanted antibodies
        - ABO antibodies (neonatal transfusions)
        - T-activation (polyagglutination)
    - 2) **Neonatal alloimmune thrombocytopenia (NAT, NAIT)**
      - a) Severe thrombocytopenia usually due to maternal anti-HPA-1A (80%); similar to HDFN in concept
        - Exposure through pregnancy or transfusion
        - Antibody crosses placenta and attacks baby's platelets
        - Can occur in first pregnancy (rapid antibody formation)
        - 10-30% get intracranial hemorrhage
      - b) Washed, irradiated maternal platelets are blood bank treatment of choice after birth (lack offending antigen and antibody); IVIg also used
      - c) Maternal IVIG used before birth (+/- intrauterine washed/irradiated maternal platelet transfusion)
    - 3) **Removal of unwanted electrolytes (RBCs and platelets)**
      - a) Especially in neonatal transfusions
      - b) Large-volume or irradiated products
3. **Freezing**
- a. Cryopreservative agents protect component while freezing and thawing.
    - 1) Most common: Glycerol at 40% concentration
    - 2) Glycerol must be removed (deglycerolization) before transfusion; basically a washing process
    - 3) DMSO for platelets, but recovery is very poor (1/3).
  - b. Why bother?
    - 1) **Storage of rare, autologous, or O-negative units**
    - 2) **Plasma hypersensitivities (as with washed)**
    - 3) **Repeated febrile reactions (as with washed)**
  - c. Storage
    - 1) Before freezing:
      - a) CPD, CPDA-1, or CP2D: Maximum 6 day shelf life before glycerolization

- b) Additive solutions (AS-1, AS-3, AS-5): Up to full 42 day shelf life before glycerolization
- 2) Red Cells
  - a) 10 years at -65 C (40% glycerol)
  - b) 24 hours at 1-6 C after thawing/deglycerolizing
- 3) Platelets
  - a) At least two years at -80 C (not FDA licensed)

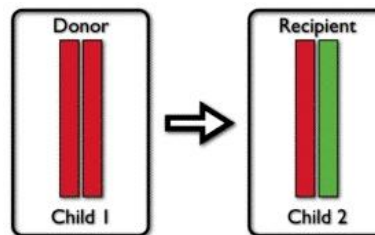
4. **Irradiation**

- a. Irradiation is effective in deactivating lymphocytes without significantly damaging anything else.
- b. Prevent transfusion-associated graft vs. host disease
- c. TA-GVHD sequence/requirements:
  - 1) Viable, active lymphocytes are transfused.
    - a) Minimum number needed unknown.
  - 2) Donor and recipient are not HLA-identical.
  - 3) Recipient is unable to respond to neutralize the effect of the transfused WBCs.
- d. The normal response:
  - 1) Transfused lymphocytes (CD4 and CD8) mount an immune response against foreign HLA host tissues.
  - 2) Normally, host lymphs (CD8) counterattack and neutralize the response (see top of figure below).



- e. Lack of host neutralization (bottom of figure above) may lead to TA-GVHD.
  - 1) Almost uniformly fatal, so thankfully rare
  - 2) Patients present with:
    - a) Fever 7-10 days post-transfusion
    - b) Face/trunk rash that spreads to extremities
    - c) Mucositis, nausea/vomiting, watery diarrhea
    - d) Hepatitis
    - e) **Pancytopenia and subsequent marrow aplasia**
- f. Radiation dose
  - 1) 2500 cGy (“rad”) dose required targeted to center of bag, with at least 1500 cGy in all parts of the bag
- g. Indications for irradiation
  - 1) **Immunosuppression**
    - a) Congenital T-cell deficiencies (DiGeorge’s, SCID, Wiskott-Aldrich)
    - b) Stem cell or marrow transplant recipients

- c) Patients taking chemo agents that attack T-cells (Fludarabine, other purine analogs)
- d) Aplastic anemia patients
- e) Patients with solid tumors getting intensive chemotherapy/radiation
- 2) **Intrauterine transfusions, premature neonatal transfusions, and neonatal exchange transfusions**
- 3) **Hematologic malignancies (esp Hodgkin's disease)**
- 4) **Granulocyte transfusions**
- 5) **Receiving blood from a first-degree relative donor or receiving HLA-matched units**
  - a) HLA-heterozygous recipient from an HLA-homozygous donor
    - Child 2 gets blood from child 1 in picture below (child 1 is HLA homozygous, while child 2 shares one haplotype with child 1)
    - Child 2 does not see child 1 as foreign, but child 1 does see child 2 as having a foreign HLA type (because of the non-shared HLA)
  - b) Recipient doesn't recognize transfused cells as foreign, so no counterattack.



- h. Patients probably NOT at risk (but often get irradiated products anyway).
  - 1) Solid organ transplant recipients
  - 2) Term neonates
  - 3) AIDS patients (CD8 cells that defend from foreign WBCs preserve function until late in disease).
  - 4) Patients receiving previously frozen blood products (FFP, cryoprecipitate)
    - a) Note that frozen/thawed/deglycerolized RBCs MUST be irradiated if patient at risk; since they were cryopreserved, viable lymphs remain.
- i. Don't use irradiation for:
  - 1) Preventing CMV transmission
  - 2) Peripheral progenitor cell infusions
- j. Maximum storage: 28 days after irradiation or regular expiration date, whichever comes first
  - 1) K<sup>+</sup> and free hemoglobin increase in plasma

**F. Plasma Group (See Mar 2010 Podcast on iTunes)**

**1. Fresh frozen plasma (FFP)**

- a. Widely used but not well-studied prospectively

- b. Whole blood-derived (see earlier diagram) or from apheresis (AD-FFP can have much higher volume)
- c. Specifics for WBD-FFP:

**Volume:** 200-250 mL  
**Contents:** All coag factors  
- 400 mg fibrinogen  
- 1 IU/mL of all others  
Almost no viable WBCs  
NOTE: No QC testing

- d. General notes about coagulation and FFP transfusion
  - 1) 30-40% of coag factors required for hemostasis.
  - 2) “Adequate” factor levels don’t necessarily give normal coag tests (mild elevations common)
  - 3) Most FFP transfusions are given for elevated PT/INR levels (Factor VII most responsible)
    - a) Factor VII *in-vivo* half-life is only about 4 hours
    - b) Correction for FVII deficit is only temporary
  - 4) Prophylactic FFP use with mild lab elevations is usually a mistake, even before procedures
    - a) No evidence of bleeding prevention or lab value correction when these patients are transfused
  - 5) No universal threshold exists, but many use INR of 2.0 as indicator of serious factor deficiency
- e. Indications
  - 1) **Bleeding patients with coagulopathy due to multiple factor deficiencies**
    - a) Hepatic failure
      - Decreased production of all hepatic factors (including pro- and anticoagulants)
      - PT doesn’t predict bleeding; avoid prophylaxis
    - b) Dilution from massive transfusion
    - c) Consumptive processes (DIC)
  - 2) **Bleeding patients requiring urgent reversal of vitamin K deficiency from warfarin effect**
    - a) Warfarin affects factors II, VII, IX, X
    - b) IV or SQ vitamin K takes hours (between 6 and 12) to replenish these factors (oral.
    - c) For non-bleeding patients, correct without FFP administration (hold dose and give vitamin K)
    - d) In bleeding patients, Prothrombin Complex Concentrate (PCC) may be better choice than FFP (but will still need small amount of FFP since US PPC preps have minimal FVII)
    - e) In general, need at least 10-20 ml/Kg of FFP to attain hemostasis in these patients (usually more)
    - f) *Chest* 2008;133:160S-198S; get this article!

- 3) **Trauma transfusion**
  - a) Recent literature suggesting that trauma patients given plasma in close to 1:1 ratio with RBCs have better survival (first reported in military)
  - b) Much current argument pro and con
  - c) Many trauma centers have established “trauma/massive transfusion protocols” that attempt to make 1:1 ratio automatic
  - d) Concerns about plasma transfusion complications (TRALI) and AB plasma wastage
  - e) Not proven in randomized, prospective studies
- 4) **Dilutional coagulopathy**
  - a) Transfusion of multiple coag factor poor products (RBCs and crystalloids in massive transfusion) dilutes coag factors.
  - b) Usually not apparent until after at least 10-15 or more units of RBCs (with accompanying fluid) in a 24 hour time span
  - c) May be less of an issue with massive transfusion protocols and 1:1 ratios mentioned above
- 5) **Transfusion or plasma exchange for TTP/HUS**
  - a) Acquired or congenital ADAMTS13 deficiency; large vWF multimers lead to platelet thrombi
  - b) FFP has normal amounts of ADAMTS13
  - c) Plasma exchange standard treatment
  - d) Treatment until PLT count is at least 100K with near-normal LDH
- 6) **Other factor-specific coagulopathies without an available factor concentrate (V, X, XI in US)**
- 7) **C1-esterase inhibitor deficiency**
- f. **Contraindications:**
  - 1) Volume expansion
    - a) Albumin, crystalloids are safer.
  - 2) Heparin reversal
    - a) Antithrombin, which potentiates heparin!
    - b) Use protamine sulfate or just stop the heparin.
  - 3) Factor deficiencies with available concentrates
  - 4) Prophylactic or pre-procedure treatment of mild elevations of PT/PTT; see above
  - 5) “Nutrition,” “Wound healing,” or “well-being”
- g. **Preparation/storage**
  - 1) **Pre-storage**
    - a) Centrifuged, separated, and placed at -18 C within 8 hours (kept for up to one year)
    - b) May also be kept at -65 C for up to 7 years
  - 2) **Pre-transfusion**
    - a) Thawed at 30-37 C.
    - b) Stored at 1-6 C for 24 hours

- g. Dosage
  - 1) Most often given two bags at a time in adults, but this dose is inadequate.
  - 2) 10-20 mL/Kg is more appropriate dosage (that would be about 3-7 bags in a 70 Kg adult!)
  - 3) 10-15 mL/Kg is an appropriate dose in neonates.
- h. Effect
  - 1) Standard dose increases factor levels by about 20-30% in a 70 Kg person.
  - 2) Transient due to short half-lives (FVII)
  - 3) Greatly elevated PT/PTT more affected than mild
- i. ABO and Rh
  - 1) Donor antibodies compatible with recipient RBCs.
  - 2) Give without regard to Rh.

		<i>DONOR</i>			
		A	B	AB	O
RECIPIENT	A	✓		✓	
	B		✓	✓	
	AB			✓	
	O	✓	✓	✓	✓

2. Plasma variants
  - a. **Plasma frozen within 24 hours of phlebotomy (FP24)**
    - 1) Not frozen in 8 hours like FFP, but 24 hours
    - 2) Factor V levels essentially equal to those in FFP, while factor VIII levels decline by about 20-25% in comparison to FFP
    - 3) Stored and managed just like FFP (including “thawed plasma” conversion below)
    - 4) Except for DIC patients, can be used identically to FFP (low FV and/or FVIII is uncommon).
  - b. **Thawed plasma**
    - 1) FFP/FP24, once thawed, is only good for 24 hours.
    - 2) Thawed FFP/FP24 may be relabeled as “thawed plasma” and kept at 1-6 C for up to 5 days.
      - a) This process is not recognized by the FDA
    - 3) Indications are essentially identical to FFP.
  - c. **Plasma, cryoprecipitate reduced (“cryo-reduced plasma”, “cryosupernatant”)**
    - 1) Residual plasma that remains after cryoprecipitate removed from FFP (see below).
    - 2) Less vWF, so may be useful in TTP plasma exchanges if regular FFP doesn’t work (literature shows mixed results on this).
    - 3) Storage and transfusion just like FFP.
  - d. **Liquid plasma/plasma**
    - 1) Plasma separated from whole blood up to 5 days after expiration

- 2) Liquid plasma: stored at 1-6 C, not frozen
  - a) Can be transfused up to 5 days after whole blood expiration date, but rarely used
- 3) Plasma: stored at -18 C or below; rarely used

e. **Source plasma**

- 1) Apheresis collection, usually paid donors
- 2) Used for manufacture, not transfusion
- 3) Licensed product

f. **Recovered plasma**

- 1) Plasma from volunteer whole blood donation
- 2) Unused units of frozen plasma may be relabeled as recovered and sold for further manufacture
- 3) Unlicensed product

3. **Cryoprecipitate**

- a. Also has seen increased use in recent years.
- b. Specifics:

<b>Volume:</b>	15 mL
<b>Contents:</b>	≥ 150 mg fibrinogen
	≥ 80 IU Factor VIII
	80-120 IU vWF
	40-60 IU Factor XIII
	<u>Fibronectin</u>

c. ABO and Rh

- 1) ABO compatible preferred
- 2) Give without Rh concern

d. Indications

1) **Fibrinogen deficiency (congenital or acquired)**

- a) General threshold: 100 mg/dl for adequate hemostasis post-surgery.
- b) Calculation in BB Practical section
- c) Many use 10-20 bags per dose in adults, more if fibrinogen is less than 50 mg/dl.
- d) 10 bags deliver about 2500 mg of fibrinogen in about 150 ml of volume
  - > 1 liter FFP needed for same amount!

2) **Treatment of uremic thrombocytopeny**

- a) Acquired adhesion defect (probably) which may respond to vWF supplementation
- b) Generally seen with creatinine levels > 3 mg/dL
- c) Second line of defense (after DDAVP, dialysis)
- d) Also: Conjug. estrogens, inc. HCT to ~30%
- e) *Am J Med.* 1994;96:168-79 describes treatment of uremic thrombocytopeny.

3) **Factor XIII deficiency**

4) **Topical “glue”**

- a) Mixed with bovine thrombin and applied directly to raw surfaces
- b) Currently available fibrin sealants (treated, virus-free) have made this less common.

- 5) **Treatment of von Willebrand's disease**
  - a) Second-line therapy; should be used only if factor VIII concentrates are not available.
    - Some factor VIII concentrates (eg, "Humate-P") contain vWF.
  - b) Cryo may be used for severe forms.
    - Dose 1 bag per 10 Kg body weight q 8 hr
  - c) DDAVP can be used for milder forms.
- 6) **Treatment of hemophilia A**
  - a) Use only if emergency and no factor VIII concentrate available.
  - b) Calculation in BB Practical section for exams
  - c) General targets:
    - For hemarthrosis, GI hemorrhage, trauma without bleeding: 50% F VIII level
    - For surgery, trauma with bleeding, intracranial hemorrhage: 100% F VIII level
  - e. Manufacture
    - 1) Made from a single unit of FFP.
    - 2) Thaw FFP at 1-6 C, spin and remove liquid, re-freeze slushy precipitate within 24 hours.
  - f. Storage and preparation for transfusion
    - 1) **-18 C for 1 year**
    - 2) After thawing (at 30-37 C, like FFP), store up to 6 hours at 20-24 C (unlike FFP)
    - 3) If units are pooled, transfuse within **4** hours.
    - 4) No compatibility testing required, though ABO-compatible is preferred.
    - 5) Can give without regard to Rh status
4. **Factor concentrates**
  - a. **Factor VIII concentrate**
    - 1) Used for moderate to severe hemophilia A
    - 2) Virus inactivated or recombinant
    - 3) Dosage: discussed in BB Practical
    - 4) Target levels: as above
    - 5) May contain vWF and be used in vWD.
  - b. **Factor IX concentrate**
    - 1) Used for hemophilia B
    - 2) Virus inactivated or recombinant
    - 3) NOT the same as Factor IX Complex Concentrate
  - c. **Prothrombin Complex Concentrate (PCC)**
    - 1) Or, "Factor IX Complex Concentrate"
    - 2) Approved only for bleeding hemophilia B patients, but USED in warfarin overdose correction
    - 3) Not "activated" as in the past; much less thrombosis risk
5. **Albumin and plasma protein fraction**
  - a. Volume expanders
  - b. Virus inactivated

- c. Ridiculously expensive!
- d. Differ only in composition
  - 1) Albumin: 96% albumin, 4% globulins/others
  - 2) PPF: 83% albumin, 17% globulins/others.

### **G. Granulocyte concentrate**

- 1. Increasing use due to use of donor stimulation.
- 2. Specifics:

<b>Volume:</b>	200-300 mL
<b>Contents:</b>	WBCs ( $\geq 1.0 \times 10^{10}$ )
	RBCs (20-50 mL)
	PLTs ( $1 \times 10^{11}$ )
	Plasma and anticoagulant

- 3. Indications
  - a. Consider in premature neonates with sepsis or infections, transplant patients with infections, patients with chronic granulomatous disease
  - b. Aside from above, a clinical situation including:
    - 1) Fever for 24-48 hours,
    - 2) Proven bacterial or fungal infection
    - 3) No response to antibiotic therapy
    - 4) Neutropenia ( $<500/uL$ )
    - 5) *Reversible* bone marrow hypoplasia
- 4. Not currently indicated for:
  - a. Prophylactic use
  - b. Patients with no hope of marrow recovery
- 5. Centers are commonly stimulating volunteer donors with G-CSF (+/- dexamethasone) for much greater yield; this practice is not FDA-approved
- 6. Cans and Can'ts!
  - a. ***Can (and should) irradiate*** to prevent TA-GVHD.
    - 1) Irradiation harms lymphs but not PMNs.
  - b. ***Can't filter*** to prevent CMV transmission.
    - 1) This seems obvious, doesn't it?
    - 2) Use CMV-negative donors for "CMV-safe"
- 7. Storage conditions
  - a. 24 hours from collection at 20-24 C, *without* agitation
- 8. Caution
  - a. Must be ABO and Rh compatible (lots of RBCs)
  - b. Crossmatch required before transfusion, also

### **H. DDAVP**

- 1. Synthetic ADH used for treatment of diabetes insipidus.
- 2. As a side effect, causes release of vWF from endothelial cells; seems to functionally increase factor VIII, as well.
- 3. Potential indications:
  - a. Uremic thrombocytopeny
    - 1) 0.3  $\mu g/Kg$  IV
    - 2) Should be considered before platelets or CRYO.
  - b. Mild hemophilia A

- c. von Willebrand's disease
    - 1) Works in types without marked deficiency
    - 2) Don't use in type IIB (clotting) or III (ineffective)
  - d. Hepatic failure (for improved platelet function)
4. Effect diminishes/vanishes with repeat doses ("tachyphylaxis").

### **I. Recombinant activated factor VII (NovoSeven)**

1. Non-human-plasma-derived product that is currently FDA-approved for use in:
  - a. Hemophiliacs (A or B) with inhibitors (bleeding prevention and bleeding treatment)
  - b. Patients with congenital factor VII deficiency (bleeding prevention and bleeding treatment)
2. Widespread "off-label" use has occurred, as NovoSeven gained traction as a "magical" hemostatic agent!
3. Estimated 1-2% risk of thrombosis is concerning
  - a. *JAMA*. 2006;295:293-298 (O'Connell, et al) article reported that most serious thromboembolic complications from NovoSeven followed off-label use.
  - b. Thrombotic stroke, acute MI, and pulmonary emboli; NOTE that these were not definitely *caused* by NovoSeven, just *associated* with its use.
4. Off-label use may be most appropriate for:
  - a. Treat/prevent surgical bleeding in trauma patients
  - b. Reversal of anticoagulant therapy (warfarin and factor Xa inhibitors)
  - c. Treat/prevent surgical bleeding in advanced liver failure patients.
  - d. Perioperative blood loss prevention (after failed clotting factor replacement therapy) in cardiac surgery, neurosurgery, OB/GYN surgery, and urologic surgery
5. Many other off-label uses are being studied, including use in hemorrhagic strokes and use in platelet-related bleeding.
6. Not indicated for routine pre-procedure prophylaxis.
7. Typical doses: 20 to 40 mcg/Kg in non-emergencies, 41 to 90 mcg/Kg otherwise
  - a. Roughly 2-hour half-life, repeat dose often