



# Principles of Perioperative Autologous Cell Processing

AmSECT International Conference

Reno, Nevada: April 27, 2010

Preparation / Review for IBBM PBMT Exam

**No disclosures or conflicts to report**

# Perioperative Blood Management Technologist [PBMT]

## Job Domain Analysis

*Theoretical Hierarchical Construct for K/S/A for Competency Exam*

<b>Environmental Factors</b>	<b>Equipment / Disposables</b>	<b>Patient Care Procedures</b>	<b>Critical Incidents</b>
Assertiveness, lead team when required [1.5]	Application and operation of equipment [2.5]	Suggest changes to and author clinical procedure guidelines [3.5]	Design and practice team drills for critical incidents [4.5]
Integration into surgical team and participate in care planning and quality management [1.4]	Follow manufacturer instructions-for-use and assembly [2.4]	Follow guidelines recognizing contraindications and exceptions [3.4]	Communication with team during critical incident and crisis management [4.4]
Inter-team member communication and patient privacy [1.3]	Disposable supplies and interface with hardware [2.3]	Follow guideline indications for use and record keeping [3.3]	Respond correctly to critical incidents and emergencies [4.3]
Social structure and vocabulary of quality-care surgical teams [1.2]	Principles of operation for equipment [2.2]	AABB (FDA, JCAHO) standards and guidelines [3.2]	Diagnose, troubleshoot, and report critical incidents [4.2]
Rules for sterile environment: OSHA, CDC training [1.1]	Hardware and device technical knowledge [2.1]	Body of medical knowledge: physiology, pharmacology [3.1]	Body of medical knowledge [4.1]

**Increasing complexity, proficiency and difficulty**

Riley, April, 2008

# April 2010: Examination Plan

Section	Label	Items	Percent	Col	Percent
1.1	Sterile environment	4	0.04		
1.2	Social structure	1	0.01		
1.3	Communication	0	0.00		
1.4	Team integration	0	0.00		
1.5	Leadership	0	0.00	5	0.05
2.1	Device knowledge	2	0.02		
2.2	Equipment operation	9	0.08		
2.3	Disposable supplies	8	0.07		
2.4	Manufacturer's IFUs	5	0.05		
2.5	Equipment applications	5	0.05	29	0.26
3.1	Physiology, pharmacology	23	0.21		
3.2	Standards and guidelines	4	0.04		
3.3	Indications for use	11	0.10		
3.4	Contraindications and exceptions	6	0.05		
3.5	Author CPGs	1	0.01	45	0.41
4.1	Medical knowledge	8	0.07		
4.2	Diagnose and troubleshoot	11	0.10		
4.3	Critical incident response	11	0.10		
4.4	Team crisis management	1	0.01		
4.5	Design safety drills	0	0.00	31	0.28
<b>Total</b>		110	1.00	110	1.00

# Perioperative Blood Management Technologist [PBMT] Job Domain Analysis

*Theoretical Hierarchical Construct for K/S/A for Competency Exam*

Environmental Factors	K/S/A	Label	Count	Percent
Assertiveness, lead when required [1.5]	K	Knowledge	45	0.41
Integration into surgical team and participate in care planning and quality management [1.4]	S	Skills	31	0.28
	A	Application	34	0.31
Inter-team member communication and patient privacy [1.3]	<b>Total</b>		<b>110</b>	<b>1.00</b>
	and interface with hardware [2.3]	indications for use and record keeping [3.3]	critical incidents and emergencies [4.3]	
	Principles of operation for equipment [2.2]	AABB (FDA, JCAHO) standards and guidelines [3.2]	Diagnose, troubleshoot, and report critical incidents [4.2]	
	Hardware and device technical knowledge [2.1]	Body of medical knowledge: physiology, pharmacology [3.1]	Body of medical knowledge [4.1]	
Social structure and vocabulary of quality-care surgical teams [1.2]				
Rules for sterile environment: OSHA, CDC training [1.1]				

Increasing complexity, proficiency and difficulty

Riley, April, 2008

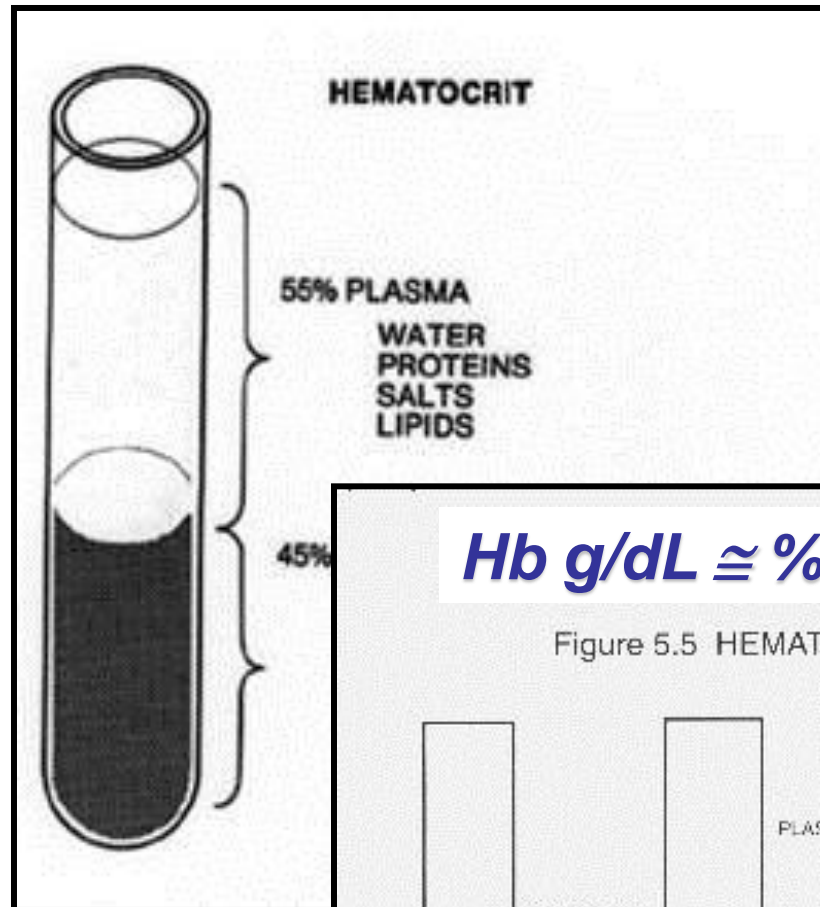
# Intraoperative Autologous Transfusion

## Principles of Cell Washing for the PBMT

### Objectives / Review Areas



- RBCs
- WBCs
- Platelets
- Buffy Coat
- Plasma
- Hematocrit
- Hemoglobin
- Hemolysis
- Hemoglobinuria
- Blood typing / Rh

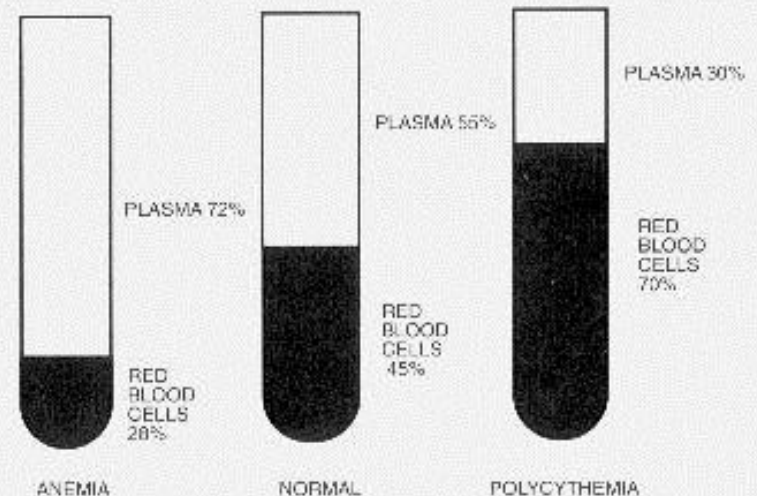


ponents

n density

$$Hb \text{ g/dL} \cong \% HCT / 3$$

Figure 5.5 HEMATOCRITS



# Effects of Hemodilution

- Reduces blood viscosity
- Reduced hematocrit decreases total vascular resistance
- Marked dropped in perfusion pressure followed by compensatory increase in cardiac output
- Patients with arterial occlusive disease may be susceptible to ischemia
- Surgical bleeding results in less RBC loss
- Transfusion triggers are important
- What is ANH (acute normovolemic hemodilution)?

Whole blood

Plasma

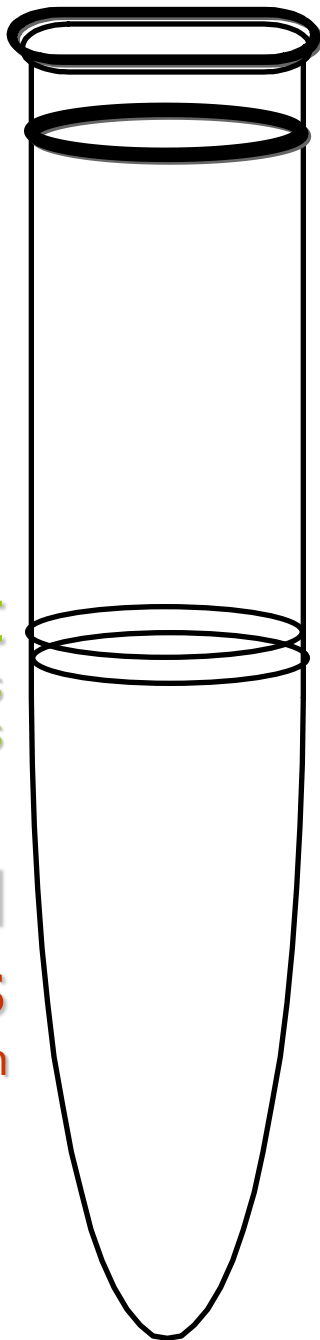
Proteins / Lipids  
Antibodies  
Electrolytes  
Water

'Buffy' coat

Platelets  
White blood cells

Red blood cells

Hemoglobin



# Blood separation technology



plasma components,  
clotting factors

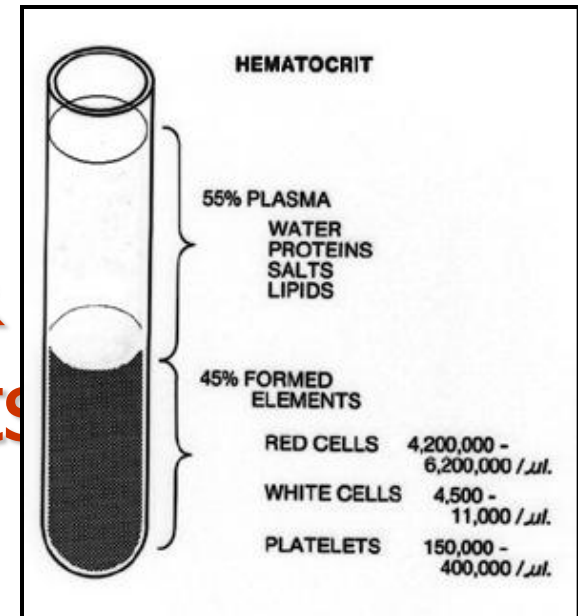


platelets & WBCs



Packed  
RBCs &  
platelets

**PRP v. PPP**





Whole blood

Plasma

Proteins / Lipids  
Antibodies  
Electrolytes  
Water

'Buffy' coat

Platelets  
White blood cells

Red blood cells

Hemoglobin

Patient

Platelet rich plasma (PRP)  
sequestration, platelet gel / glue,  
AGF™, Stem cell harvesting

collect whole blood

separate\*  
plasma, RBCs  
and 'buffy' coat

Return RBCs

YES

Keep the cells?

NO

Decant concentrated  
'buffy' coat  
rich with  
PLTs

Return PP plasma

YES

Keep the plasma?

PLT rich  
concentrate  
delivered to  
surgeon

\* Via centrifuge

# Common BCST Terminology

- adsorption column
- aggregated growth factor
- antibodies
- apheresis
- autoimmune disease
- autotransfusion
- bovine
- 'buffy' coat
- cell processing
- centrifugation
- colony stimulating factors
- cryoglobulin
- erythrocyte (RBC)
- fibrinogen
- filtration
- granulocytes (WBC)
- hemoconcentration
- hemofiltration
- leukocyte (WBC)
- lymphocyte (WBC)
- leukodepletion
- photopheresis
- platelet derived growth factor
- platelet gel or glue
- platelet poor / rich plasma
- platelet-pheresis
- rheumatoid arthritis
- thrombin
- thrombocyte (platelet)
- transforming growth factor

# Clotting Factors in plasma

## Clotting Factors

### Normal Lab Values

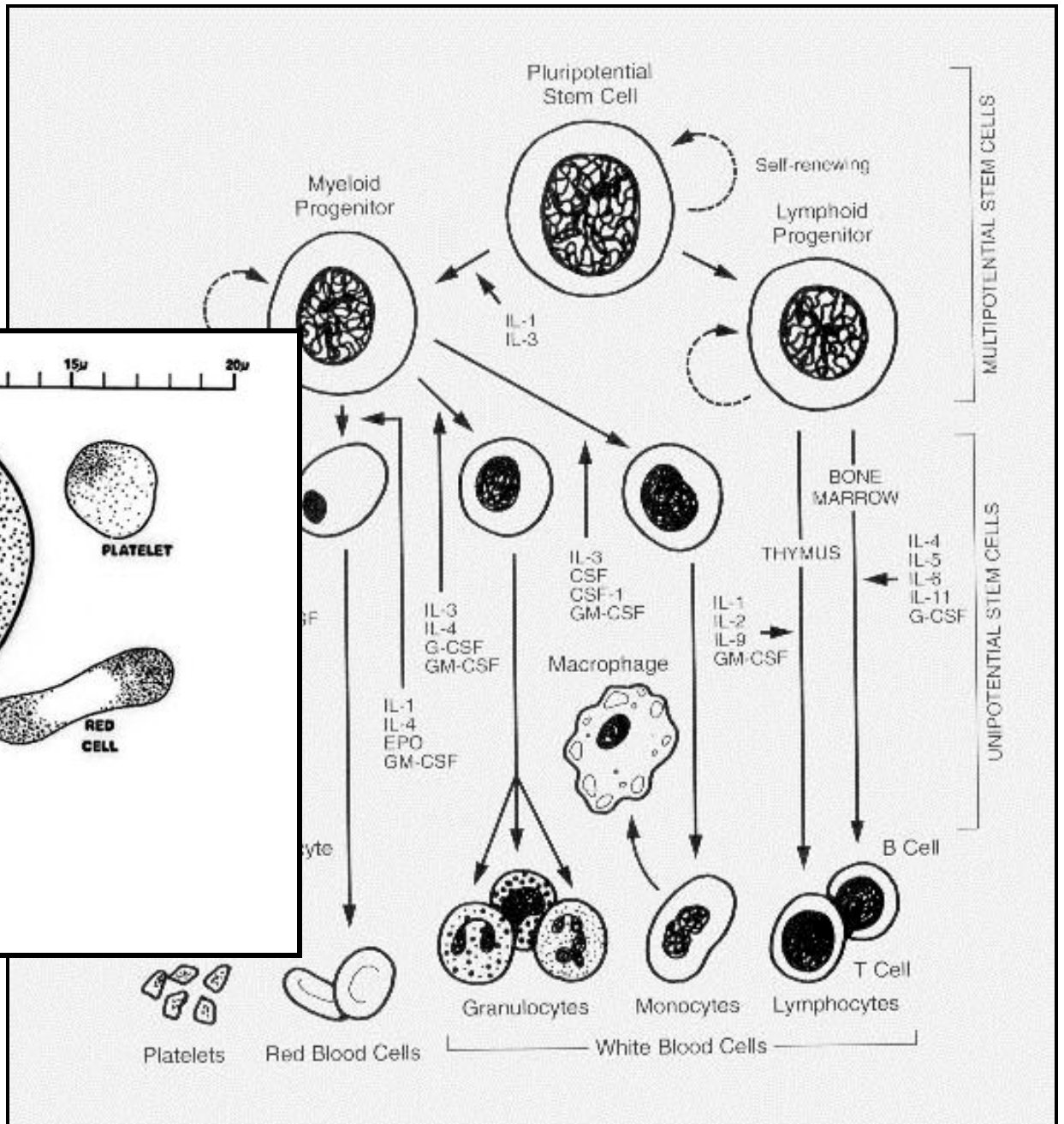
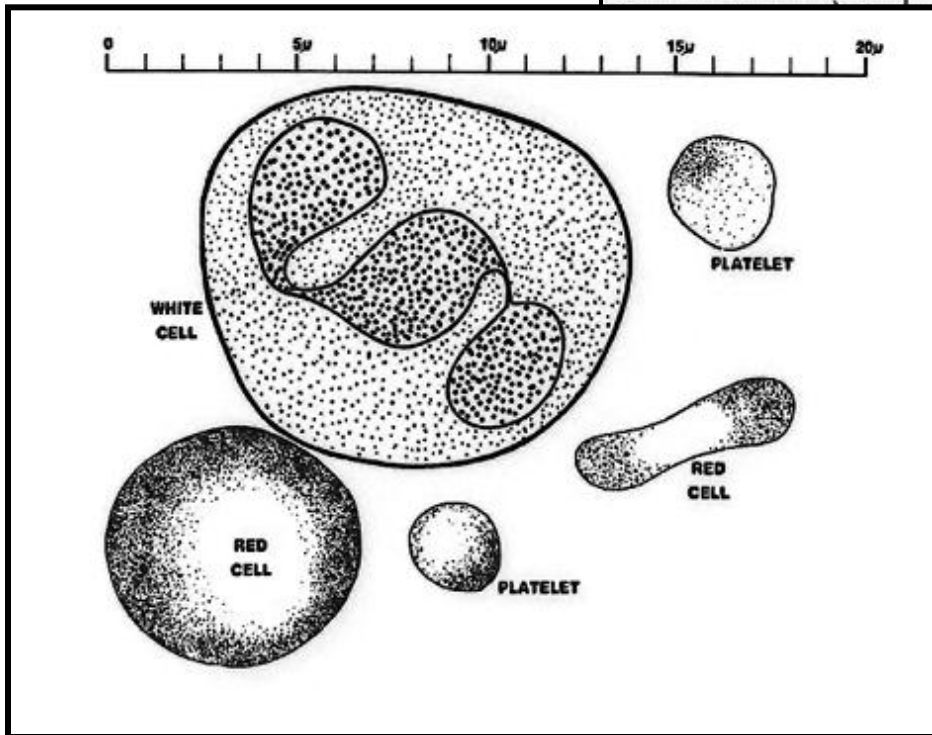
Factor I	Fibrinogen	0.15-0.35 gm/100 ml
Factor II	Prothrombin	60-140% of control
Factor III	Thromboplastin	
Factor IV	Calcium	
Factor V	Proaccelerin	
Factor VI	None	
Factor VII	Proconvertin	
Factor VIII	Antihemophilic	
Factor IX	Plasma Thromboplastin	
Factor X	Stuart Factor	
Factor XI	Plasma Thromboplastin	
Factor XII	Hageman Factor	
Factor XIII	Fibrin Stabilizing Factor	

I	Fibrinogen
II	Prothrombin
III	Platelet factor 3 (thromboplastin)
IV	Calcium
V	Labile factor ( proaccelerin)
VI	Not assigned
VII	Stable factor ( proconvertin)
VIII	Antihemophilic factor A (AHF)
IX	Antihemophilic factor B (Christmas factor)
X	Stuart-Prower Factor
XI	Antihemophilic factor C (PTA)
XII	Hageman factor
XIII	Fibrin-stabilizing factor (FSF)

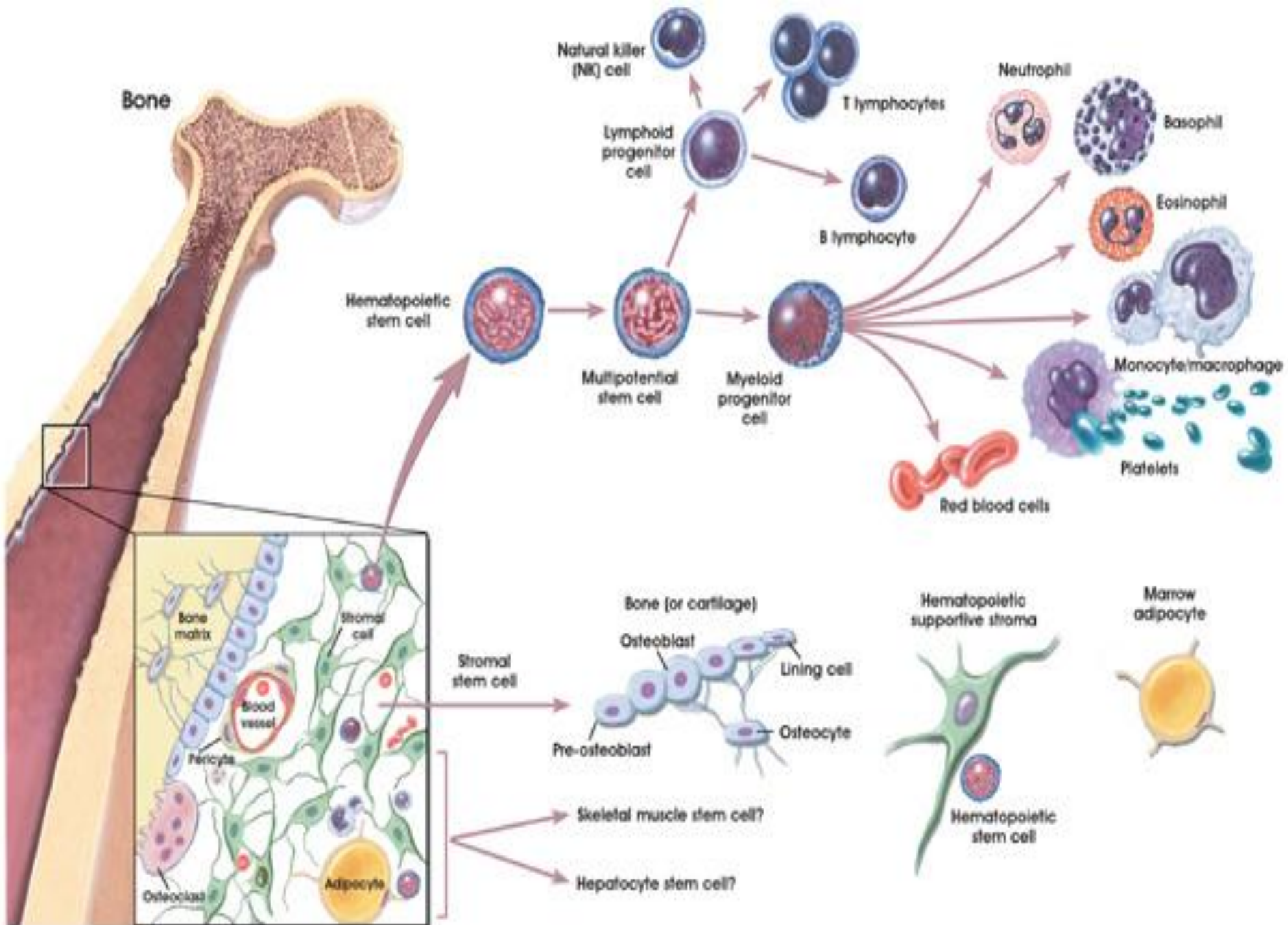
*\* Factors V and VIII are not true serine proteases, but are commonly referred to as such.*

Dailey pp 61;  
Brodie pp 41

# Stem Cells



Dailey pp 4;  
Austin pp 11



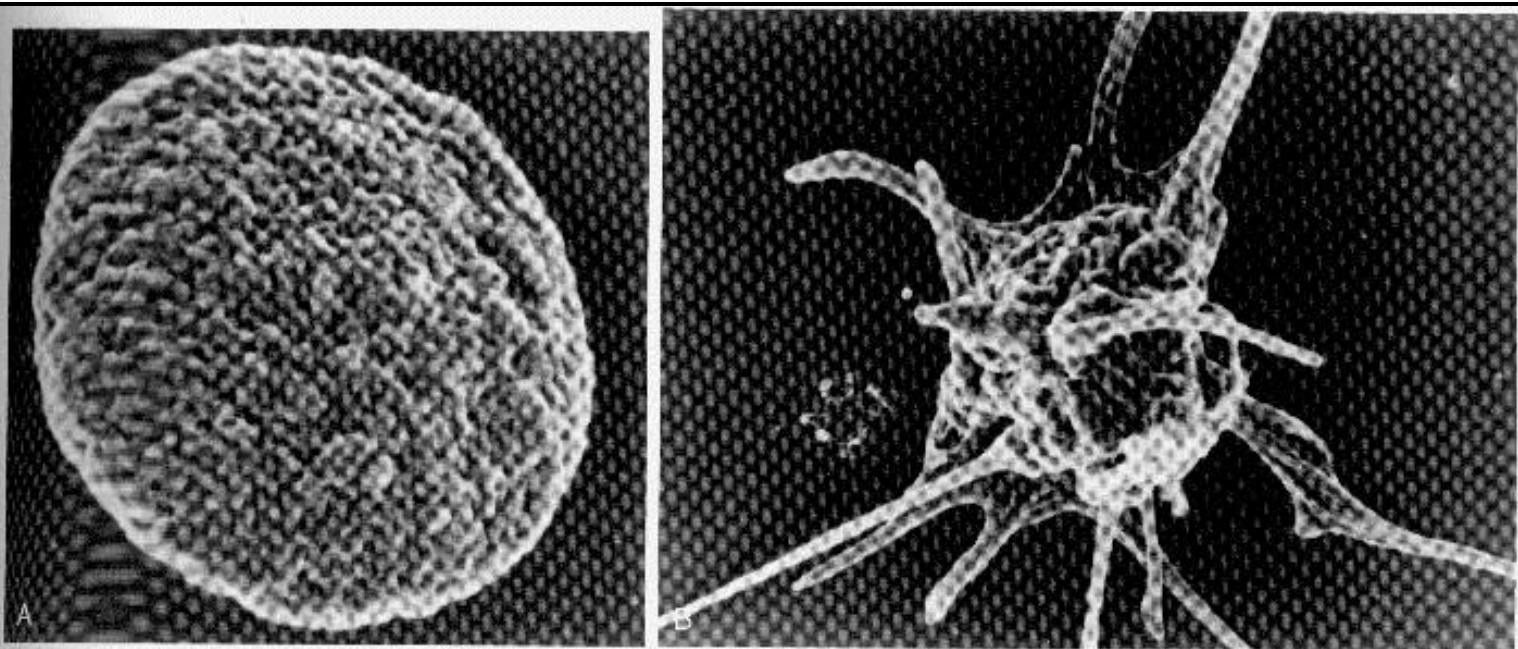
# Potential Uses for Stem Cells

Figure 11: Potential Uses for Stem Cells

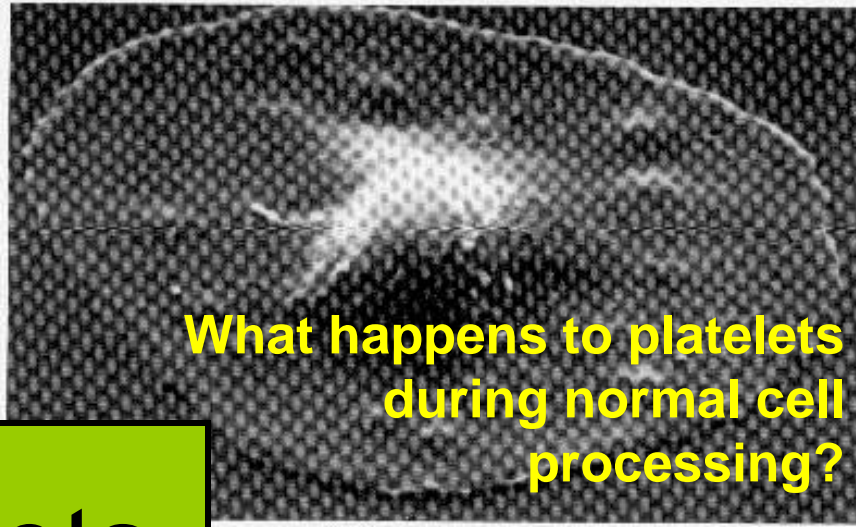
- Growing nerve cells to repair spinal injuries and restore function to paralyzed limbs.
- Growing heart muscle cells to replace useless scar tissue after a heart attack.
- Making brain cells that would secrete dopamine for the treatment and control of Parkinson's disease.
- Growing cells that make insulin, creating a lifelong treatment for diabetes.
- Growing bone marrow to replace blood-forming organs damaged by disease or radiation.
- Making blood cells genetically altered to resist specific disease, such as HIV, to replace diseased blood cells.



Source: James Thomson, assistant professor of anatomy at the University of Wisconsin Medical School, and John Gearhart, a professor of GYN/OB and physiology at Johns Hopkins University School of Medicine



Speiss pp 65



**What happens to platelets  
during normal cell  
processing?**

# Platelets

# RBC antigens and antibodies

Figure 4.1 ABO BLOOD GROUPING COMPATIBILITY

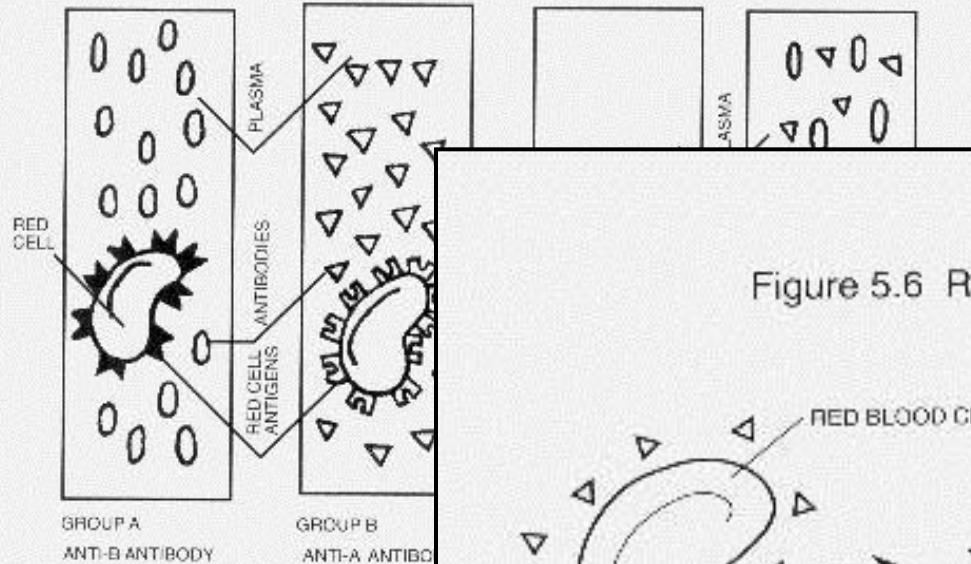
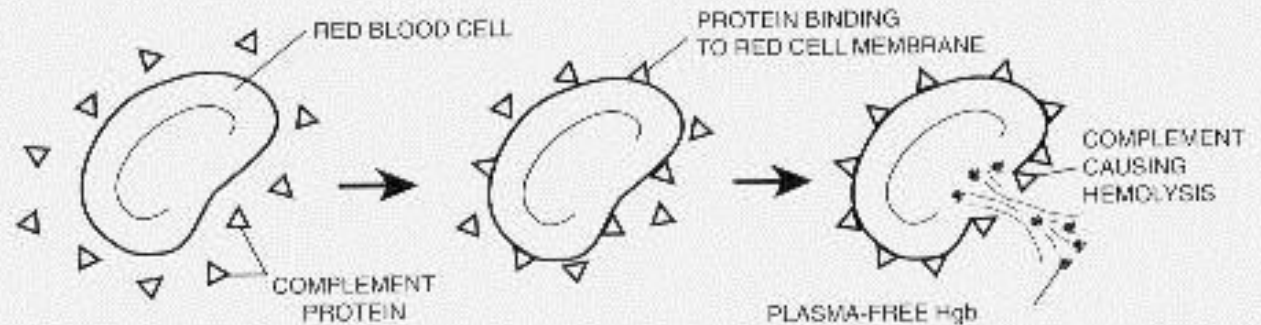


Figure 5.6 RED BLOOD CELL HEMOLYSIS



Dailey, pp 26, 38

One cause of red cell hemolysis is the opsonization of complement protein on the red cell membrane.



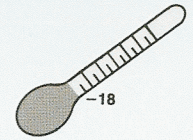
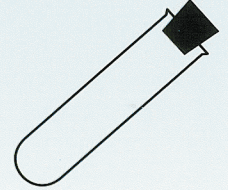
# Blood Typing: RBCs

- Group A has anti-B antibodies
- Group B has anti-A antibodies
- Group AB has both antigens and no antibodies
- Group O has no antigens and both antibodies



## TYPES OF COMPONENTS

	Volume/Unit
Plasma (FFP)	180-250 mL
Plasma Cryoprecipitate Reduced	180-250 mL
Cryoprecipitate	10-20 mL



## COMPATIBILITY

### ABO

- Plasma components should be ABO compatible.

Patient Group	Compatible Donor Groups for Plasma*
A	A, AB
B	B, AB
AB	AB
O	A, B, AB, O



\*Cryoprecipitate may be given without regard to ABO type in adults.

### Rh

- Plasma and cryoprecipitate may be transfused without regard to Rh type.

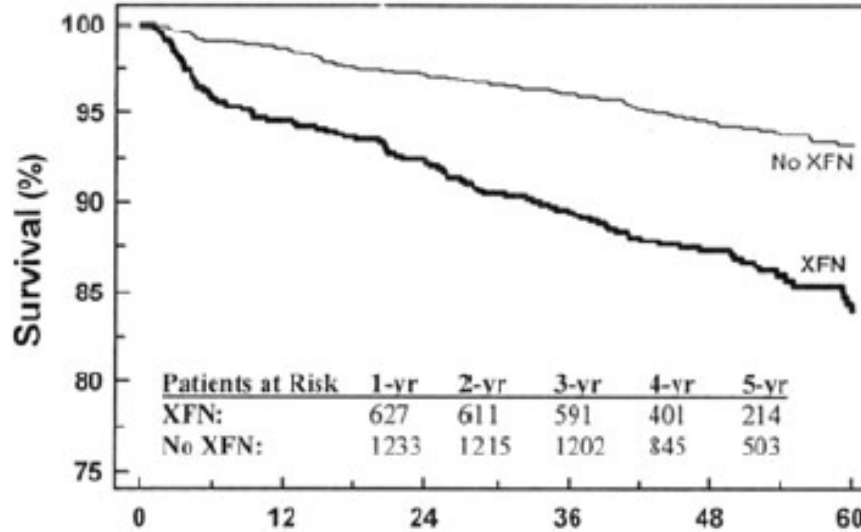
# Risks Allogeneic Transfusion

**TABLE 1. Risks of transfusion**

**Risk**

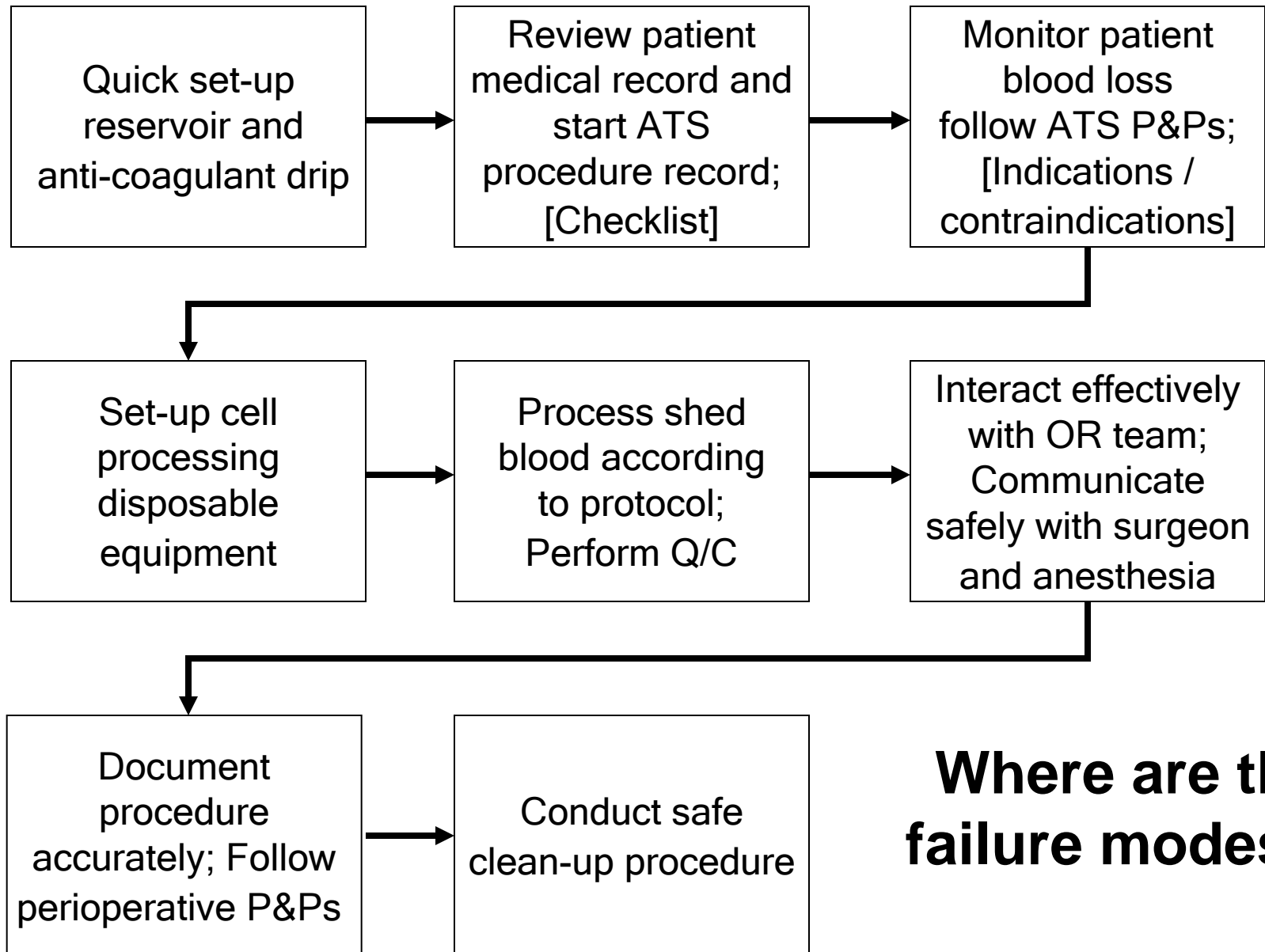
1. Hepatitis B
2. Hepatitis C
3. HIV
4. HTLV
5. TTV
6. West Nile virus
7. Cytomegalovirus conversion
8. Epstein-Barr virus
9. TRALI
10. ABO-Rh mismatch  
Occurrence  
Mortality
11. Delayed hemolytic reaction
12. Alloimmunization (PLTs and WBCs)
13. Alloimmunization (RBCs)
14. Allergic reactions
15. Febrile reaction
16. GVHD
17. Volume overload
18. Depressed erythropoiesis

\* Some of the reported risks of transfusion are either as risk per number of units transfused or as risk per number of units transfused.



**Fig. 1. The Kaplan-Meier mortality curves for those patients transfused perioperatively and for those not transfused. Those patients not transfused had approximately 2.5-fold better survival than those that received one or more units of RBCs during their CABG hospital stay. These data were after propensity analysis weighting confounding events and risks. Reprinted with permission from Engoren et al.<sup>38</sup>**

# Simulation flow for basic PBMT defined competencies



**Where are the failure modes?**

# Blood Management Techniques During Phases of Operative Period

## Perioperative Blood Management

### Pre-Op

- Hematologic analysis
- Plan for hemorrhage
- Pharmacology
- Pre-donation**
- Pheresis
- Exchange transfusion
- Genetic therapy
- Transfusion

### Anesthetic

- Pharmacology
- BCST, Cellular therapies
- Plasma sequestration
- RBC sequestration**
- Hypotension
- Transfusion
- Non-blood vol expansion
- Artificial blood
- Hematologic monitoring

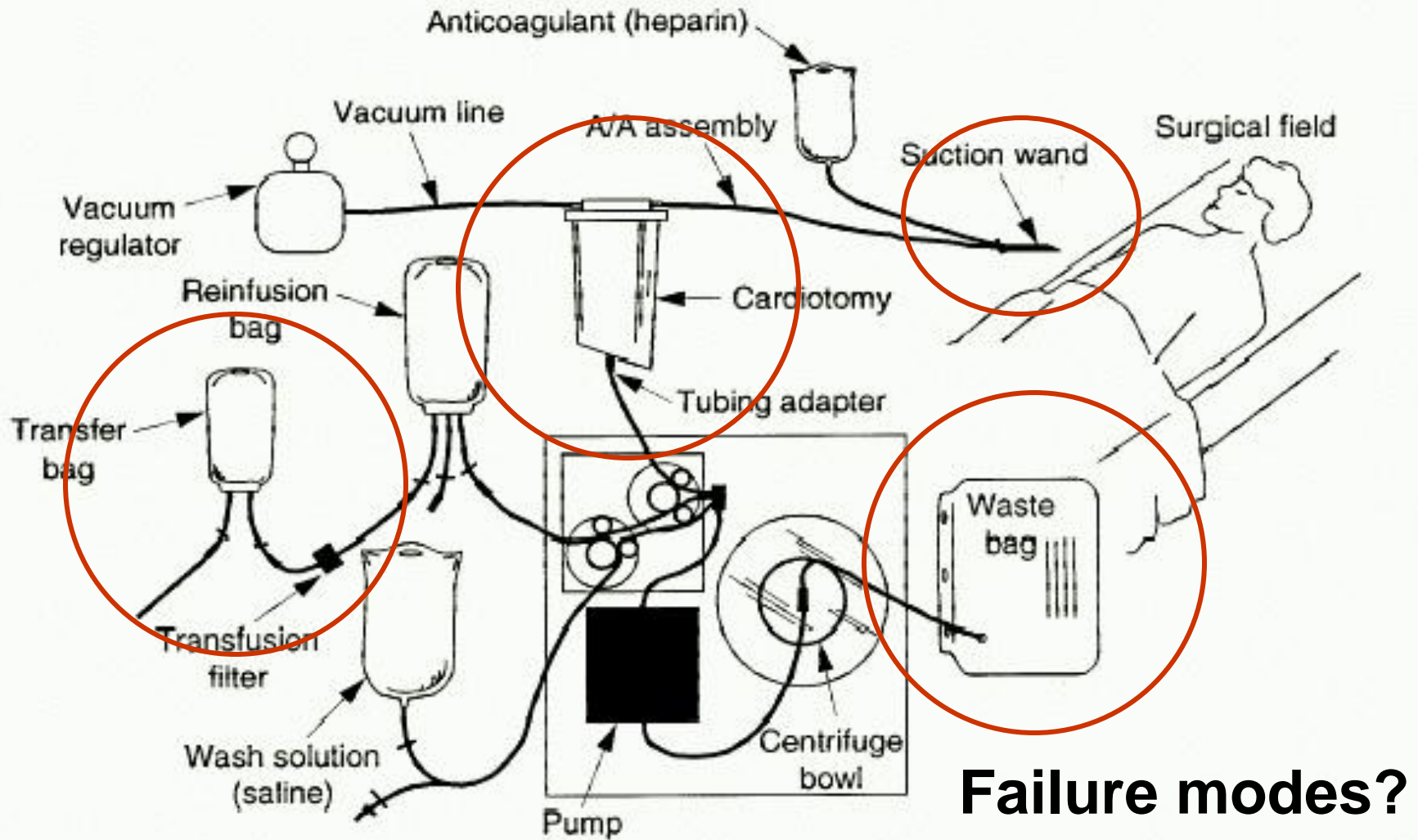
### Operative

- Meticulous hemostasis
- ATS: Cell processing**
- Tissue glue
- Platelet gel**
- Ultrafiltration**
- Surface treatments
- Transfusion
- Rapid infusion
- Artificial blood
- Hematologic monitoring

### Post-Op

- ATS: Cell processing**
- Cell washing**
- Transfusion
- Total leukocyte depletion
- Ultrafiltration
- Hematologic monitoring

# Safe IAT Circuit



# Hemoconcentration

- Hemoconcentrators
  - Dialysis
  - Ultrafiltration
- Centrifuge
  - Single bowl
  - Continuous processing
- Filtration
  - Micro-aggregate filtration
  - Leukocyte-depleting filters



# Anticoagulation for ATS

- ACD, CPD
  - 15 ml/100 ml shed blood
  - 1:7 ratio
  - $[Ca^{+2}]$
  - Thrombocytes
- Heparin solution
  - (30,000 IU/L)
  - 1:7 ratio
  - Antithrombin



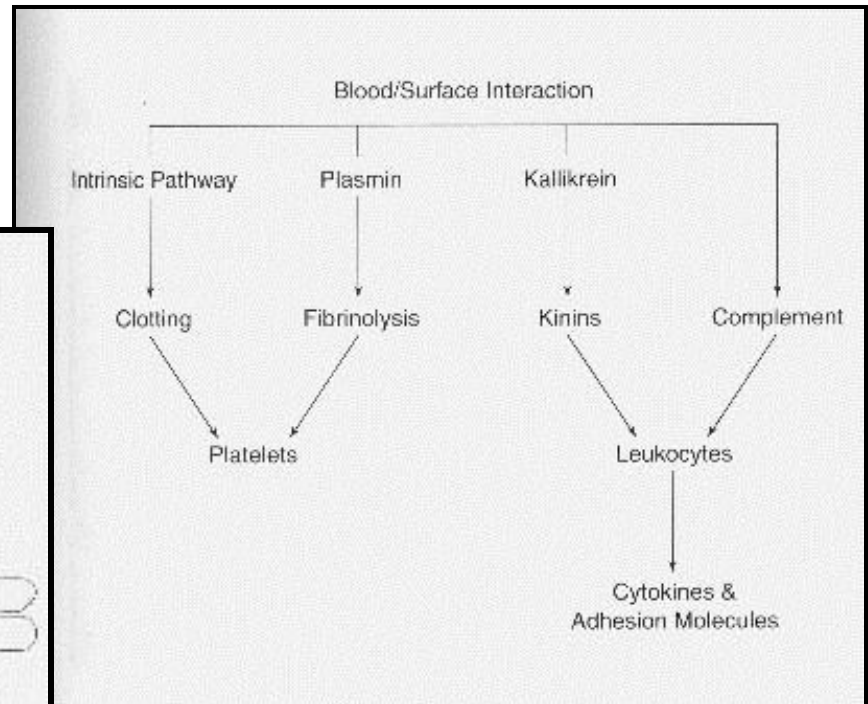
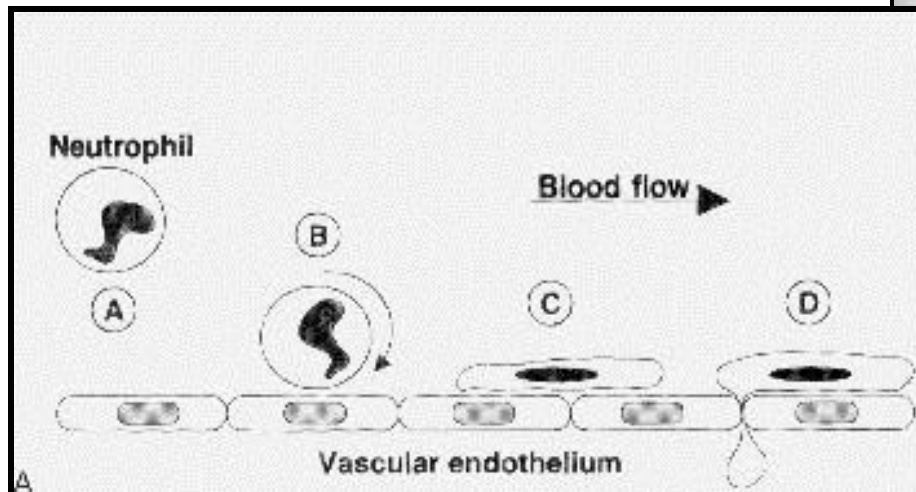
# Organizations

- FDA
- OSHA
- CDC
- JC
- CAP
- CMS
- AABB
- ASA
- AmSECT
- Hand hygiene
- Body fluid precautions
- Blood labeling
- Storage time
- Body fluid exposure
- Sharps
- Contaminated waste
- PPE
- Guidelines for PABCT
- GLP
- POCT



# Collection System / Vacuum

- Filtered vs. non-filtered
- Safe vacuum levels / Suction tips
- Blood-gas interface
  - SIRS
- Activated WBCs



# Tonicity

- Osmolarity
  - Ions (osmotic force)
  - Proteins (oncotic force)
- Hypotonic
  - Cells placed in a hypotonic solution swell
- Isotonic
- Hypertonic
  - Cells placed in a hypertonic solution shrink
- Hemolysis

# Wash solutions

- Saline
- PlasmaLyte-A
- NormoSol-R
- Lactated Ringers – contains calcium ions
- D<sub>5</sub>W – do not use as a wash solution
- Anticoagulant compatibility
- IV compatibility
- Type of shed blood (procedure-specific)

# Blood and plasma volume

- Body weight: pounds to kg
- Estimated blood volume: % kg
- Estimated plasma volume:  $(1.0 - f_{\text{Hct}})$
- Red cell mass (L)
  - Patient
  - ATS reservoir
- ANH volumes
- PRP (plasma pheresis fraction)

# Pharmacology

- Anticoagulation
  - Anti-platelet drugs
- Antibiotics
  - Plasma-bound
- IV wash solutions – FDA indications
- Electrolytes / supplement
- Procoagulants
  - Topical hemostatic agents
- Allogeneic blood products

## Theory and practice of Latham Bowl ATS

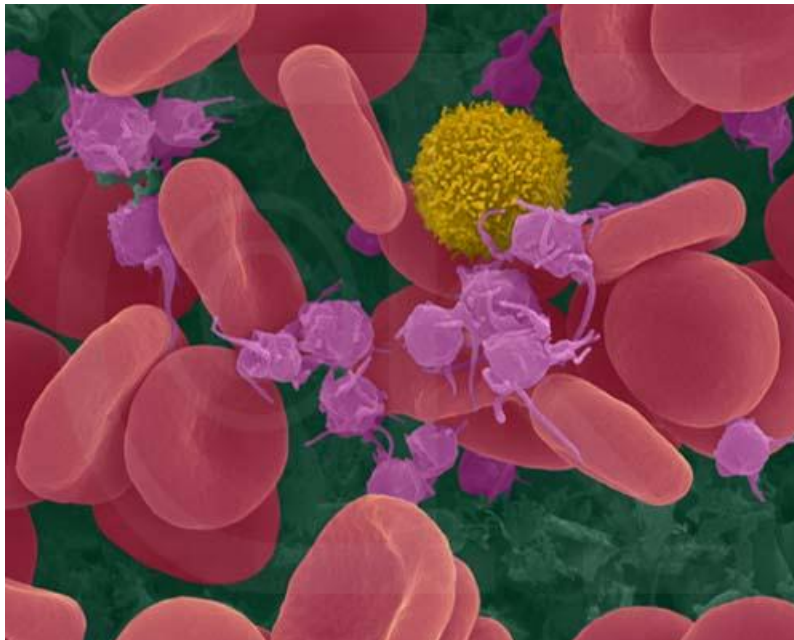


Reeder G. Autotransfusion theory or operation: a review of the physics and hematology. *Transfusion*. 2004;44:35S-39S

Reeder G. Autotransfusion theory or operation: a review of the physics and hematology. *Transfusion*. 2004;44:35S-39S

**TABLE 1. Density range for the constituents of blood**

Blood component	Component density range
Plasma	1.025-1.029
Thrombocytes	1.060-1.067
WBCs	1.065-1.090
RBCs	1.085-1.097



**Megakaryocytes are similar in density to the lower density RBCs, so some platelets are found in the top of the RBC pack**

# Centrifugation

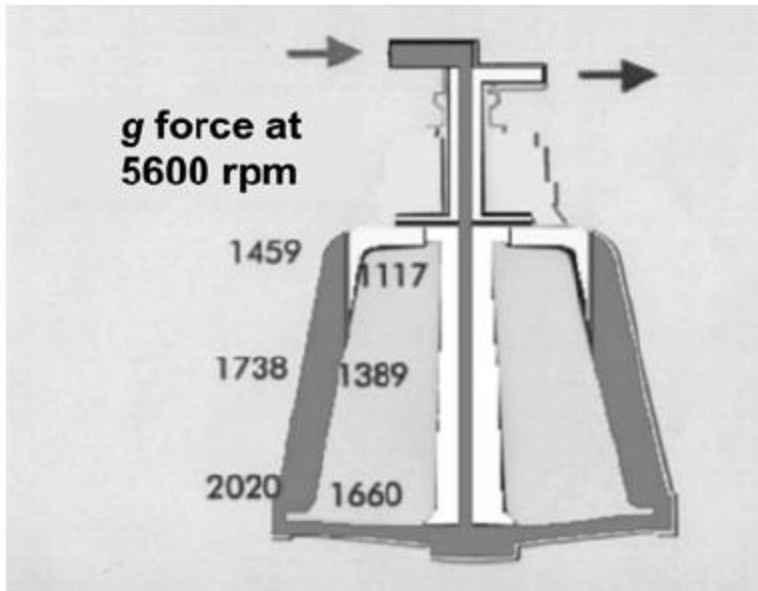
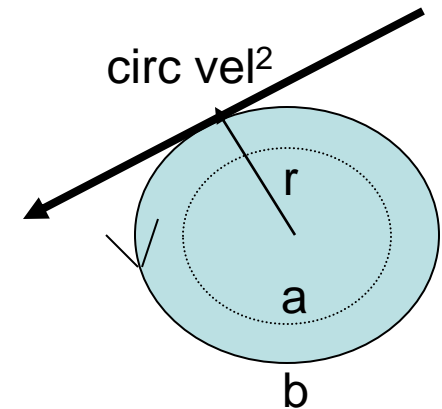


Fig. 2. Bowl g forces at 5600 rpm.

$$F = m \left[ \frac{v^2}{r} \right]$$

Newton



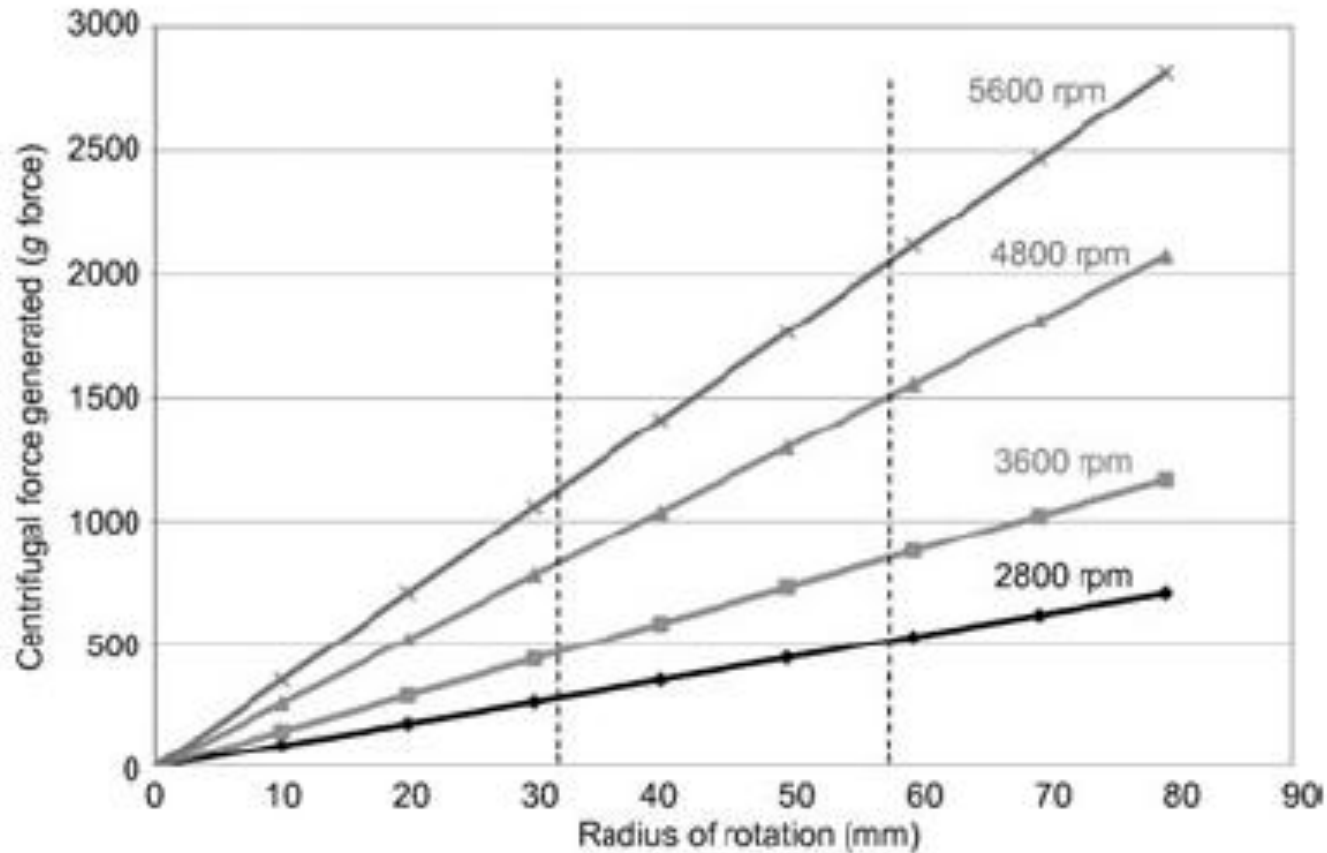
**Contaminants vary, always consider the source of the shed blood**

**TABLE 2. Contaminants found in shed wound or pump blood<sup>1,2</sup>**

Fibrin(ogen) split products and D-dimers
Activated fibrinolytic products—plasmin
Activated complement—C3a and C5a
Proteolytic enzymes
Marker enzymes, including creatine phosphokinase-myocardial fraction (CPKMB) from mediastinal drainage
Stroma, cell fragments, and internal cellular contents
Activated WBCs
Free Hb
Bacteria and endotoxins
Fats
Anticoagulants



# RPM Determine G Forces



**Fig. 1. Relationship between rpm and developed g forces in centrifugal cell processing bowls.**

# Operational Settings

TABLE 5. Operational settings (mL/min)

Machine type	Mode								
	"Wakeup"			"Standard"			"Maximum"		
	Fill	Wash	Empty	Fill	Wash	Empty	Fill	Wash	Empty
AT-1000	300	300	300	500	500	500	1000	1000	1000
Sequestra	300	300	300	500	500	500	1000	1000	1000
BRAT-2	400	800	600	500	500	500	1300	1300	1300
Compact A	400	450	500	500	500	500	1000	1000	1000
Cell Saver 4	500	600	500	500	500	500	1000	1000	1000
Cell Saver 5	200-600	200-600	500	500	500	500	1000	1000	1000

**Higher rotation rates apply higher G forces, different cell processing algorithms employ different RPMs to optimize RBC / buffy coat separation**

# Before Wash

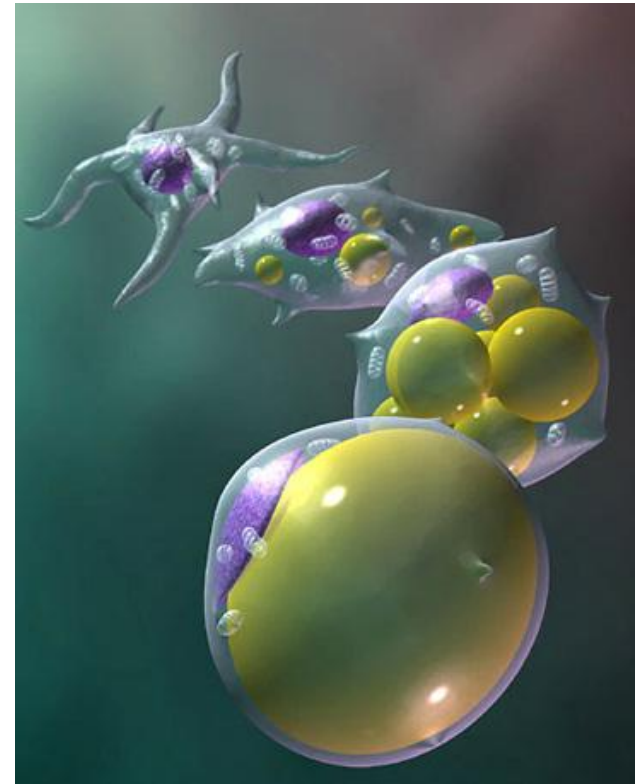
**TABLE 3. Coagulation factors in shed blood**

Coagulation factor	Venous blood	Shed blood
Factor VIII (%)	156.7	17
Factor V (%)	113.5	0
Antithrombin III (%)	97.3	46
Fibrinogen (mg/dL)	246	0
Plasminogen (%)	84.3	54.0
Protein C (%)	89.7	68.7
Protein S (%)	88.7	73.7

**TABLE 4. Biochemical debris in shed blood**

Biochemical agent	Venous blood	Shed blood
D-dimer ( $\mu\text{g/mL}$ )	0.7	1024
Fibrin(ogen) degradation products ( $\mu\text{g/mL}$ )	4.0	5120
Tissue plasminogen activator (ng/mL)	16	38.5
Complement C3a (ng/mL)	428	14784

**Fat and debris removal are issues**



# Quality Indicators of Cell



PERGAMON

Transfusion and Apheresis Science 27 (2002) 153–157

TRANSFUSION  
AND APHERESIS  
SCIENCE

[www.elsevier.com/locate/transci](http://www.elsevier.com/locate/transci)

## Intraoperative blood salvage in cancer surgery: safe and effective?

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### Abstract

To support blood supply in the growing field of cancer surgery and to avoid transfusion induced immunomodulation caused by the allogeneic barrier and by blood storage lesions we use intraoperative blood salvage with blood irradiation. This method is safe as it provides efficient elimination of contaminating cancer cells, and as it does not compromise the quality of RBC. According to our experience with more than 700 procedures the combination of blood salvage with blood irradiation also is very effective in saving blood resources. With this autologous, fresh, washed RBC a blood product of excellent quality is available for optimal hemotherapy in cancer patients.

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Keywords: cell salvaging, contraindications, neoplasm, in vitro, blood salvage, intraoperative technique, autotransfusion

TABLE 6. P

System	Hct
A	47.5
B	41.3
C	45.2
D	52.2
E	33.2
F	45.6

\* All data are per

AAE  
5.3,  
reco  
ana  
auto

profes-

1995.

# Fill

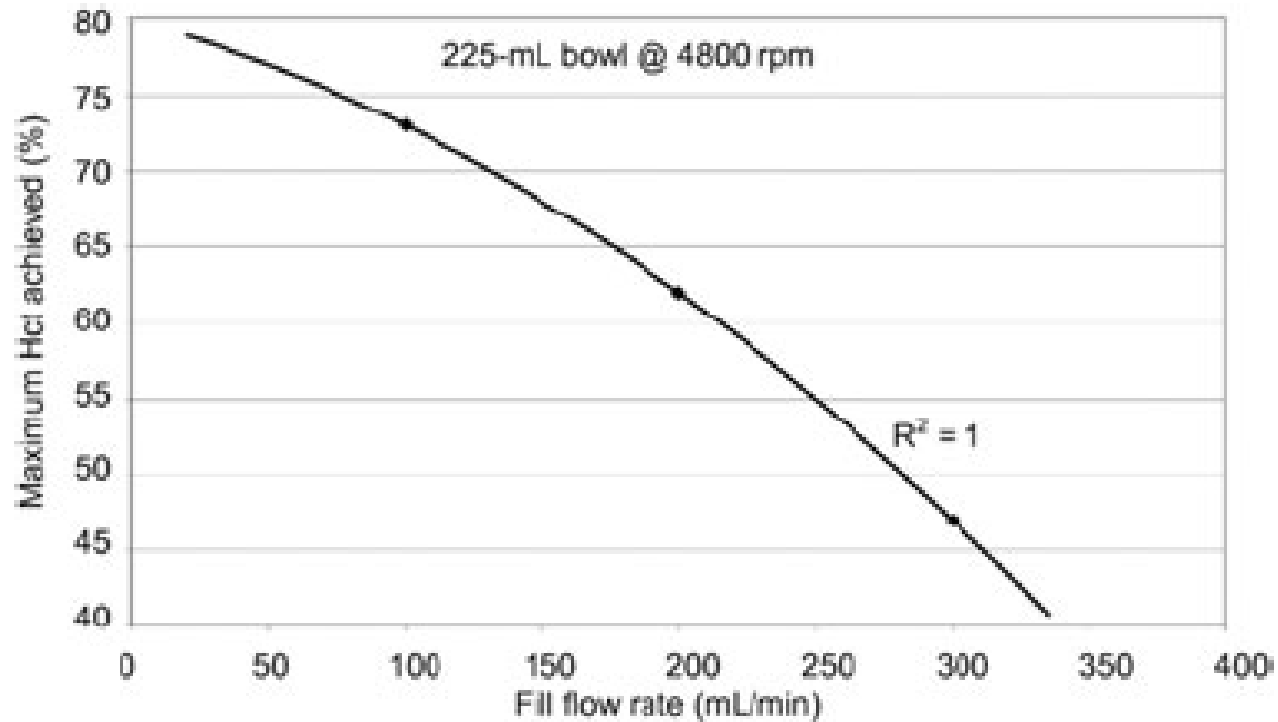


Fig. 4. Relationship between fill speed and achieved Hct.

**Watch for spilling of RBCs**

# WASH

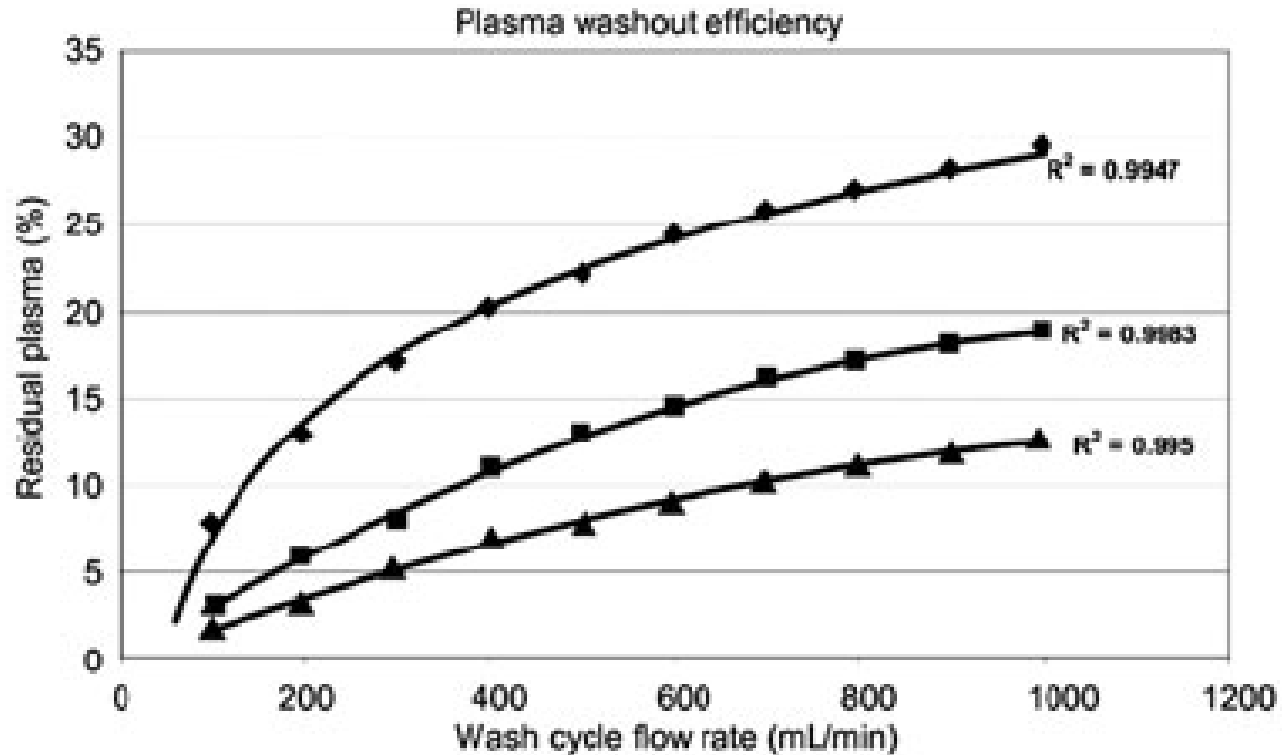


Fig. 5. Effect of flow rate and saline volume on plasma removal. (◆) 750-mL wash; (■) 100-mL wash; (▲) 1500-mL wash.

**Examine the exudate**

# Notes on Contra

**TABLE 1. General indications for CS**

Specialty	Surgical procedure	Comments
Cardiac	Valve replacement Redo bypass grafting	
Orthopedics	Major spine Bilateral knee	

- Procedure-specific should contain and contraindications
- MDs may veto contraindications in order, confirm in order
- See Waters JT Transfusion. 2004; 44S

**TABLE 2. Proposed contraindications to CS**

Pharmacologic agents
Clotting agents (Avitene, Surgicel, Gelfoam, etc.)
Irrigating solutions (Betadine, antibiotics meant for topical use)
Methylmethacrylate
Contaminants
Urine
Bone chips
Fat
Bowel contents
Infection
Amniotic fluid
Malignancy
Hematologic disorders
Sickle cell disease
Thalassemia
Miscellaneous
Carbon monoxide (electrocautery smoke)
Catecholamines (pheochromocytoma)
Oxymetazoline (Afrin)

# E-B Practice Guidelines

## ■ SPECIAL ARTICLES

Anesthesiology 2006; 105:198-208

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### *Practice Guidelines for Perioperative Blood Transfusion and Adjuvant Therapies*

“When appropriate, intraoperative or postoperative blood recovery and other means to decrease blood loss (e.g., deliberate hypotension) may be beneficial. Acute normovolemic hemodilution, although rarely used, may also be considered.”

making decisions about health care. These recommendations may be adopted, modified, or rejected according to clinical needs and constraints.

Practice guidelines are not intended as standards or absolute requirements. The use of practice guidelines cannot guarantee any specific outcome. Practice guidelines are subject to revision as warranted by the evolution of medical knowledge, technology, and practice. They provide basic recommendations that are supported by analysis of the current literature and by a synthesis of expert opinion, open forum commentary, and clinical feasibility data.

#### *and Adjuvant Therapies*

*Blood transfusion* refers to the perioperative administration of blood and blood components (e.g., autologous blood, allogeneic whole blood, red blood cells, fresh frozen plasma [FFP], platelets, and cryoprecipitate). Adjuvant therapies refer to drugs and techniques to reduce or prevent blood loss and the need for transfusion of allogeneic blood.

#### *B. Purpose of the Guidelines*

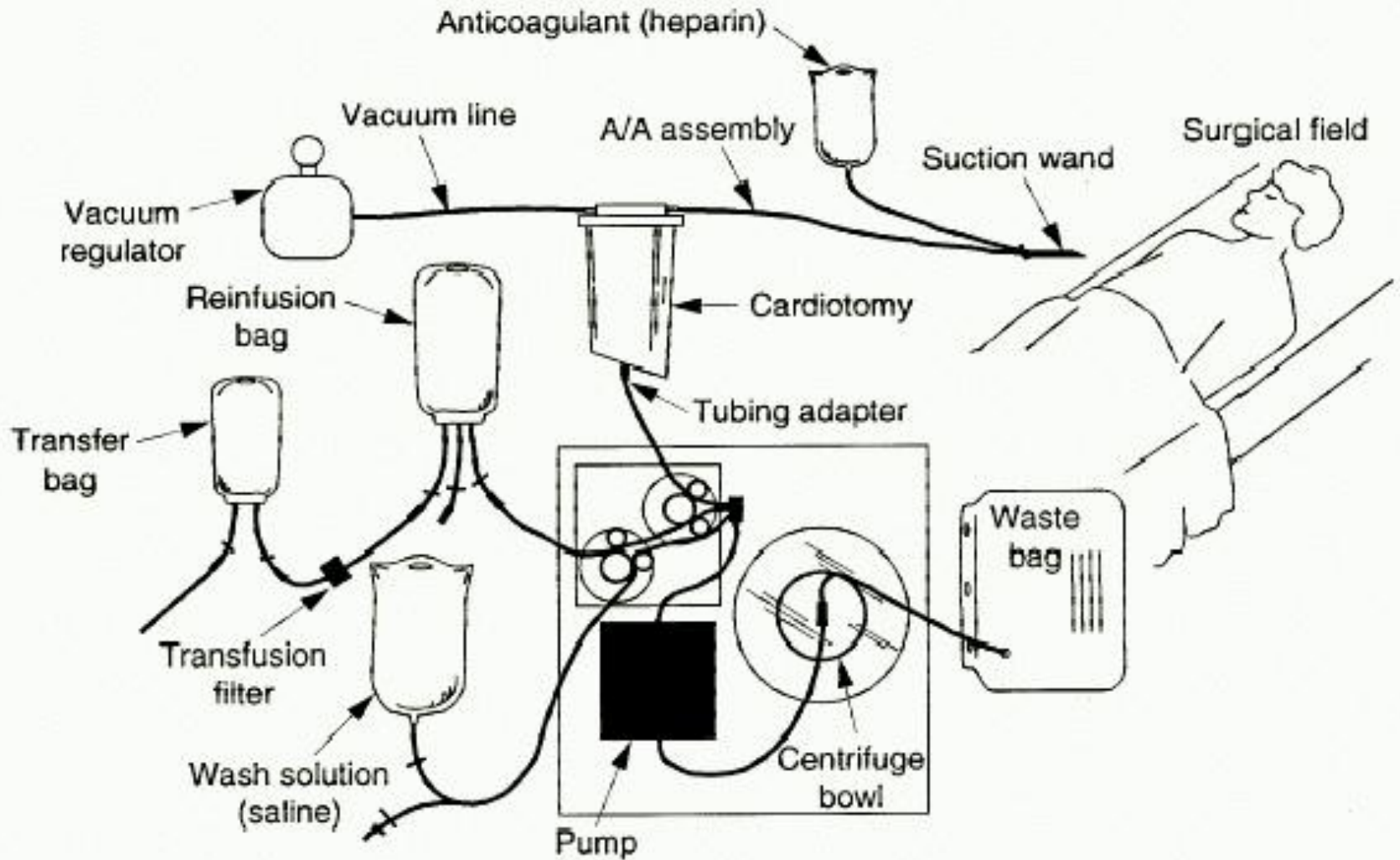
The purposes of these Guidelines are to improve the perioperative management of blood transfusion and ad-



# Quality Monitoring

- Process steps QC
- Final Product Quality Monitoring
  - Hct, [Pr], wash exudate clarity
- Process Improvement
  - Capture opportunities for improvement
  - Capture failure modes
  - Impound non-functioning equipment
- Qualifying FDA-cleared devices for a specific use [AABB]

# Safe IAT Circuit



# Critical Incidents



PERGAMON

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## Current status of bacterial contamination of autologous blood for transfusion

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### Abstract

Autologous transfusion, although not without risk, does decrease the risk of transmitted diseases via homologous transfusion. However, strict quality control is required for autologous transfusion. In Japan, a recent enactment requires that written informed consent be obtained prior to blood transfusion, which therefore requires that clinicians provide sufficient explanation of the risks involved with this procedure. To the best of our knowledge, this is the first study to comprehensively evaluate the manner in which the safety of autologous blood transfusion can be compromised

...affecting quality] [Article in German]. *Anaesthesiol Intensivmed Notfallmed Schmerzther.* 2004;39(9):569-575.

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# Critical Incidents

5. Medication errors
  1. Wrong anticoagulant drug
  2. Wrong anticoagulant drug dose
  3. Wrong anticoagulant drip solution
6. Allergic reactions
  1. Anaphylactic reaction (3)
7. Equipment failure
  1. Cell washing devices
  2. Platelet concentration devices
  3. Rapid infusion devices
  4. Blood warming devices
8. Circuit disposable **component failure**
  1. Shed blood reservoir
  2. Cell washing bowl or chamber
9. Circuit blood line separation
  1. **Blood spray**
  2. **Blood loss**

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# Critical Incidents

10. **Special patient management requirements**
  1. **Partial cell washing bowl volume (2, 4)**
  2. **Massive red blood cell and platelet loss (5)**
  3. **Massive plasma protein and clotting factor loss**
  4. **Pediatric patients (6)**
  5. **Jehovah Witness (7)**
  6. **Cancer patient (8)**
  7. **Cesarean patient (9, 10)**
  8. **Liver transplant patient**

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Questions?