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GADJAH MADA

PEMROSESAN KOMPONEN DARAH

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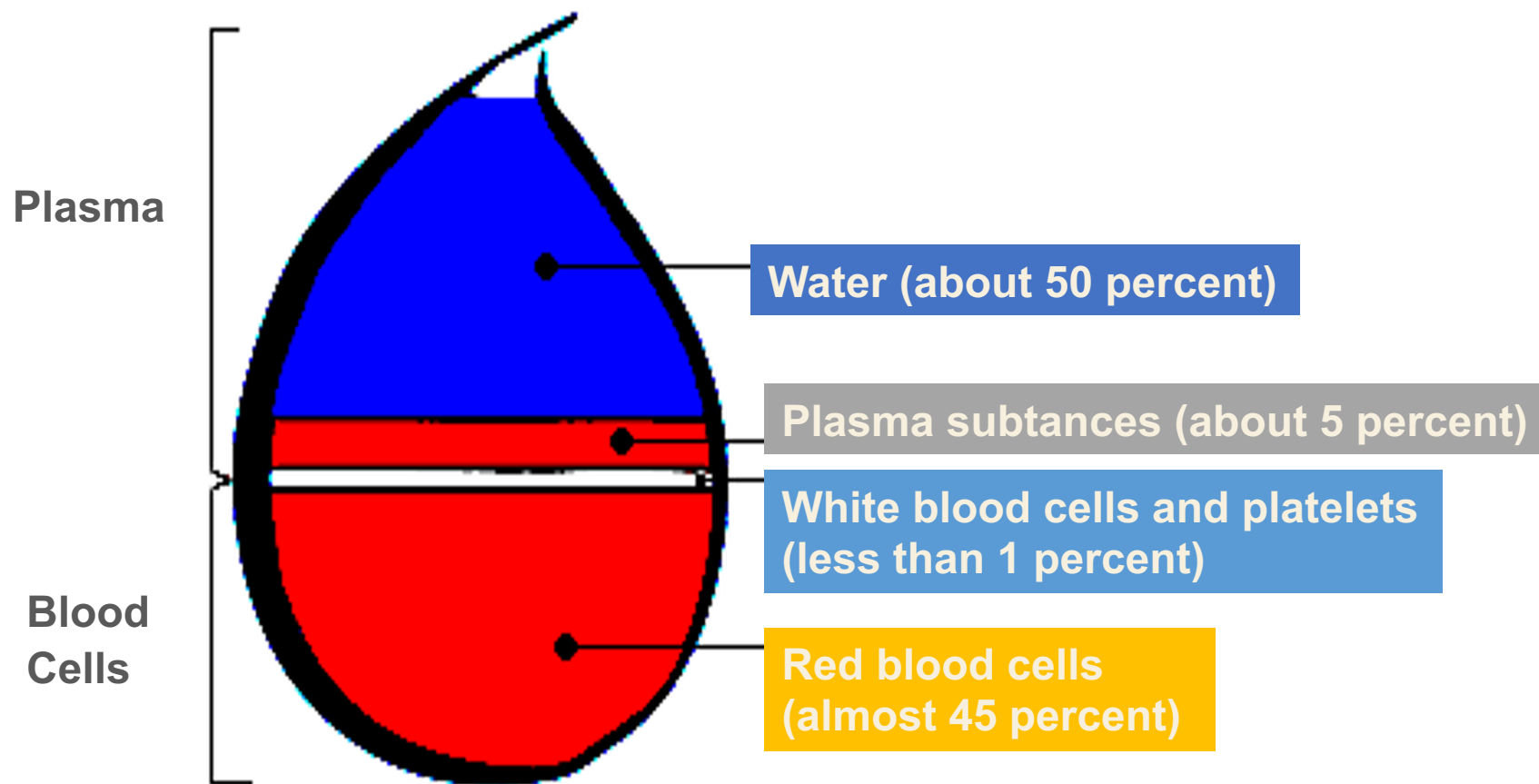
RSUP dr. Sardjito

Yogyakarta, Indonesia





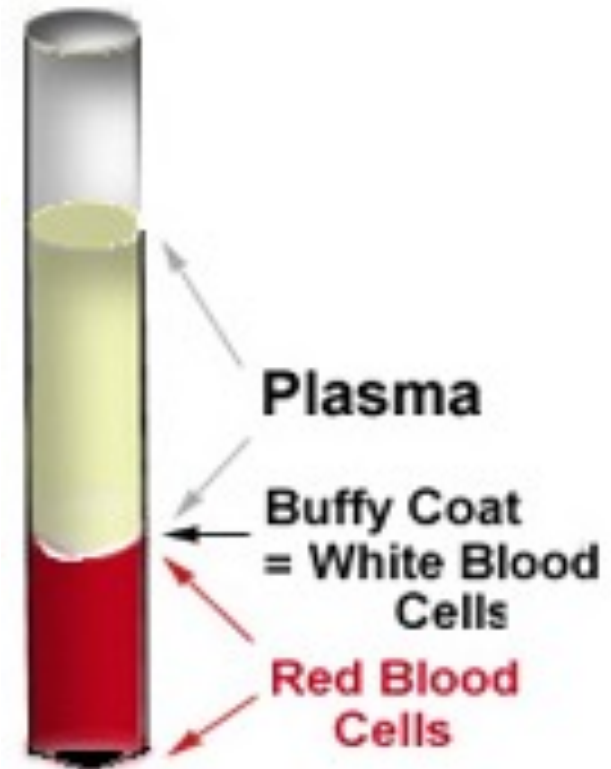
Komposisi Darah





Blood and Blood Cells

- Blood is heavier and 3 – 4 times more viscous than water.
- Most blood cells are formed in bone marrow
- Blood platelets = cell fragments





Mengapa Harus Komponen?

- Benefit several different patients
- Patients receive only the component(s) necessary
- Reduces the risk of transfusion reactions
- Storage conditions can be optimized
- Offers logistic, ethical and economic advantages



Prosedur Preparasi

- Post-donation : traditional manner
- During collection : apheresis.



Collection Basis

- Blood is collected in a primary bag that contains anticoagulant-preservatives
- Satellite bags may also be attached, depending on what components are needed
- Anticoagulant-preservatives minimize biochemical changes and increase shelf life



Bag Aystem

- A wide variety of PVC (polyvinyl chloride) plastic bag is available
- Sterile and pyrogen-free
- Storage of platelet requires a plastic with increased oxygen permeability.



SINGLE BAG

- No further processing into components is performed
 - The unit is transfused as wholeblood.





DOUBLE BAG

- An additional empty bag is attached
- Plasma can be transferred through the tubing to the transfer bag creating two components :red cells & plasma.





TRIPLE BAG

- Used to manufacture PC from PRP, or to harvest cryoprecipitate from FFP





QUADRUPLE BAG

- Similar to triple bag, but has additional bag containing red cell additive solution
- Usually used in automated system to prepare:
 - red cell concentrate
 - buffy coat
 - plasma





TOP AND BOTTOM BAG

- Plasma out of the top, red cells out of bottom leaving BC in primary bag.



Prinsip Sentrifugasi

- Blood constituents can be separated because they differ in size and density and will sediment at different rates when centrifugal force is applied
- Other factors are viscosity of the medium and flexibility of the cells which are temperature dependent
- The choice to be made is the speed and time of centrifugation to separate desired component.



Prinsip Sentrifugasi

- **First phase:** the surrounding fluid is only a mixture of plasma and anticogulant solution. Leukocyte and redcells now sediment more rapidly than platelet
- **Later phase:** leukocytes and red cells settle at lower half, PRP at upper half
- **End phase:** cell-free plasma is in the upper part, red cells at the bottom



Prinsip Sentrifugasi

To establish optimal centrifugation:

- Collect parameter that indicate the desired outcome of procedure e.g. Hematocrit, plasma volume)
- Establish a base line setting with regard to speed (rpm or g)
- Prepare a number (>10) using this setting and measure the parameters
- Adjust speed and time

	Mean density (g/ml)	Mean volume (10e-15 litre)
Plasma	1.026	
Platelet	1.058	9
Monocyte	1.062	470
Lymphocytes	1.070	230
Neutrophile	1.082	450
Red cell	1.100	87



Pemrosesan Komponen

Components of whole blood are centrifuged:

- “light spin” – short time, low RPM
- “heavy spin” – longer spin, high RPM

Procedures are in the *AABB Technical Manual*.



SORVALL RC12BP

0

577:07

TEMPERATURE 1
21

H-12000

SORVALL
77003 04

4700

SUSAN
AT ROUNT



Plasma Extractor

(blood press) is used to apply pressure to a centrifuged unit of blood in order to transfer part of it to transfer bag.





Automatic Blood Separator

Use light sensor to detect cells in primary bag and tubing.





Basic Blood Components

- Red Blood Cells
- Platelets
- Fresh Frozen Plasma (FFP)
- Cryoprecipitated Anti-hemophilic Factor
- Granulocytes.



Basic Blood Components

RBCs

- 1-6° C (stored); 1-10° C (shipped)
 - 21, 35, or 42 days depending on preservative or additive
 - Hematocrit should be $\leq 80\%$
 - One unit increases hematocrit 3%
- Once the unit is "opened" it has a 24 hour expiration date!





Red Blood Cells

- Prepared by removing part of the plasma from centrifuged whole blood
- Enough plasma is removed to achieve hematocrit to 0.65 - 0.70.



Red Blood Cells

- RBCs (frozen)
 - $\leq -65^{\circ}\text{C}$ for 10 years
- RBCs (deglycerolized or washed)
 - Good at $1-6^{\circ}\text{C}$ for 24 hours
- RBCs (irradiated)
 - $1-6^{\circ}\text{C}$ for 28 days



Leukoreduced-Red Blood Cells

Leukocyte-Reduced RBCs are for:

- Patients who receive a lot of transfusions to prevent antibody production toward WBC antigens
- Patients transfused outside of a hospital
- Patients who have reacted to leukocytes in the past.



Leukocyte Reduction Filters (maintains closed system)



Courtesy LifeSouth Community Blood Centers, Gainesville, Fla.



http://www.pall.com/39378_39479.asp

Final unit must have less than
 5×10^6 WBCs



Cryopreserved-RBCs

Frozen RBCs

- Glycerol is added to cryoprotect the unit
- Glycerol prevents cell lysis (dehydration, intracellular ice)
- **Why?** Freezing RBCs preserves rare units or extends to life of autologous units.



Courtesy LifeSouth Community Blood Centers, Gainesville, Fla.



Cryopreserved-RBCs

Deglycerolized RBCs

- RBCs that have had the glycerin removed
- Thawed at 37°C
- A blood cell processor washes the cells with varying concentrations of saline
- Considered “open”, expires in 24 hrs.





Washed-RBCs

- Not effective in reducing WBCs
- For patients (with anti-IgA) that may react with plasma proteins containing IgA
- Reactions may be allergic, febrile, or anaphylactic.



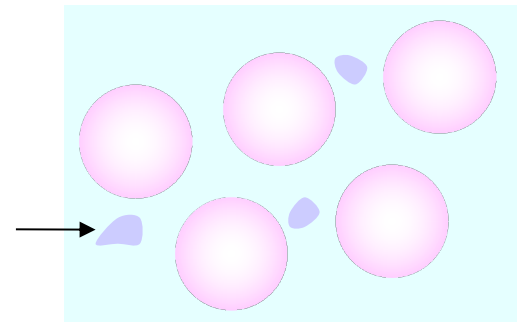
Irradiated-RBCs

- Prevents T-cell proliferation that may cause transfusion-associated graft versus host disease (GVHD)
- GVHD is fatal in 90% of those affected
- Used for:
 - Donor units from a blood relative
 - HLA-matched donor unit
 - Intrauterine transfusion
 - Immunodeficiency
 - Premature newborns
 - Chemotherapy and irradiation
 - Patients who received marrow or stem cells.



Platelets

- Important in maintaining hemostasis
- Help stop bleeding and form a platelet plug (primary hemostasis)
- People who need platelets:
 - Cancer patients
 - Bone marrow recipients
 - Postoperative bleeding.





Wholeblood-derived Platelets

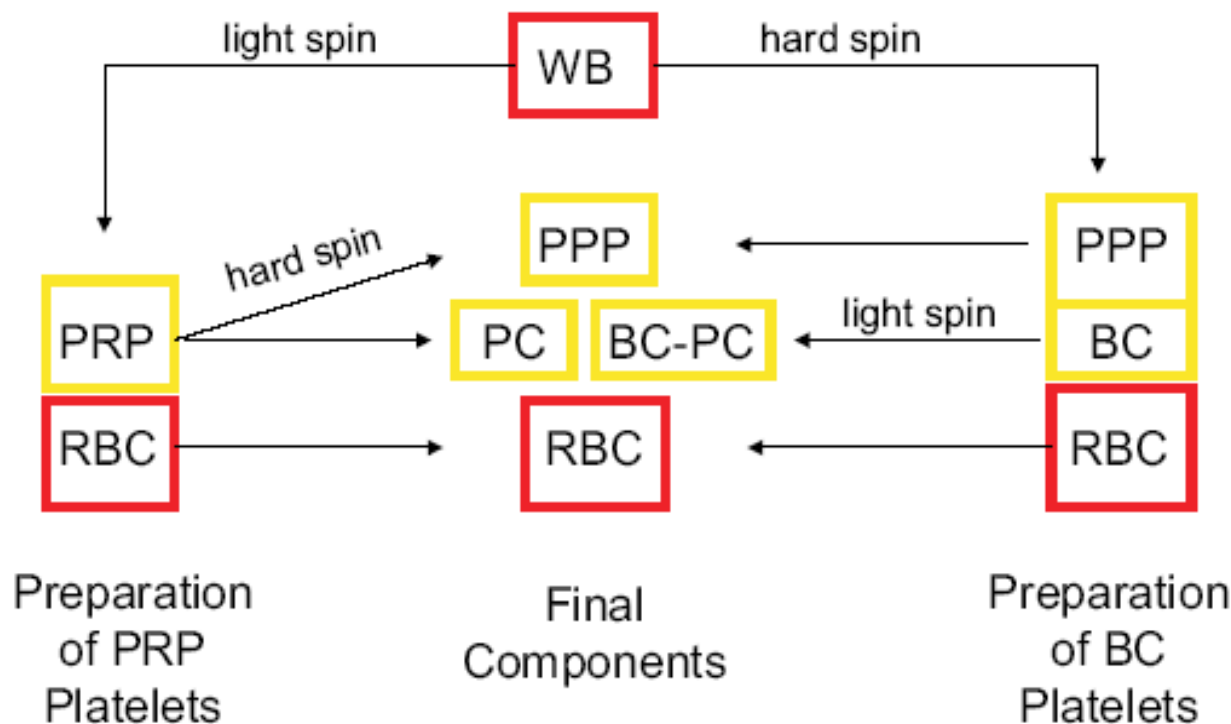


Figure 13-1. Preparation of whole-blood-derived platelets. WB = whole blood; PRP = platelet-rich plasma; PPP = platelet-poor plasma; PC = platelet concentrate; BC = buffy coat; RBC = red blood cell.

(Herman & Benson, 2005)



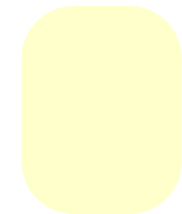
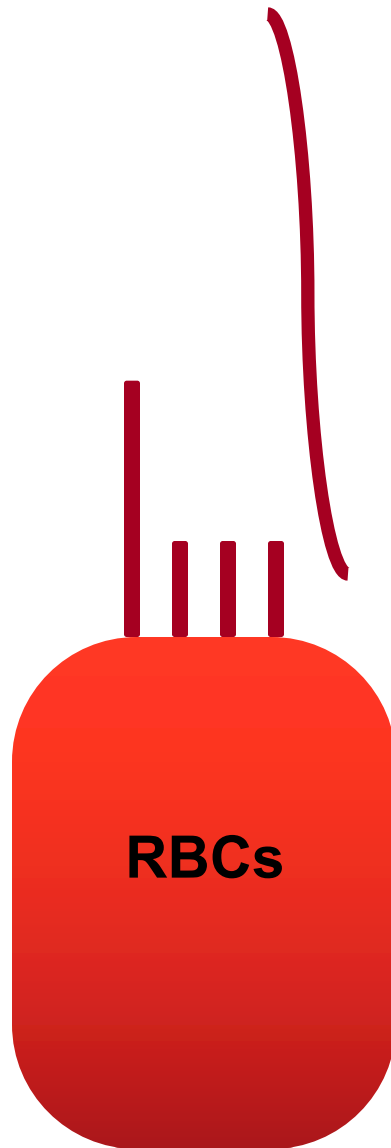
How Platelet are Processed?

Requires 2 spins:

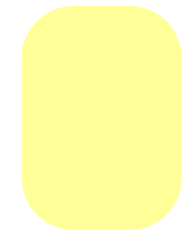
- **Soft** – separates RBCs and WBCs from plasma and platelets
- **Heavy**
 - platelets in **platelet rich plasma** (PRP) will be forced to the bottom of a satellite bag
 - 40-60 mL of plasma is expelled into another satellite bag, while the remaining bag contains **platelet concentrate**.



Preparation of PCs



Plasma



Platelet
concentrate



Platelets

- Storage Temperature
 - 20-24°C for 5 days (constant agitation)
- Each unit should contain at least 5.5×10^{10} platelets
- Each unit should elevate the platelet count by 5000 μL in a 165 lb person.



Other Type of Platelets

- **Pooled platelets**
 - Used to reach therapeutic dose
 - An “open system” occurs when pooling platelets, resulting in an expiration of 4 hours
- **Plateletpheresis** – therapeutic dose (from one donor) without having to pool platelets
 - HLA matched – for those with HLA antibodies
 - Leukocyte reduced - used to prevent febrile non-hemolytic reactions and HLA alloimmunization

Other type of Platelets





Pooling of Buffycoat





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FFP

- Plasma that is frozen within 8 hours of donation
 - -18°C or older for 1 year
- Method of freezing
 - Should bring the core temperature down to -30C or below within 60 minutes
 - Should be presented in a regular configuration to maximize exposure to the freezing process
 - If a liquid environment is used, container cannot be penetrated by the solvent

FFP

FFP is thawed before transfusion

- 30-37°C waterbath for 30-45 minutes
- Stored 1-6°C and transfused within 24 hours
- It should not be refrozen



Cryoprecipitate

- Cryoprecipitated antihemophilic factor (AHF) or “Cryo” is the precipitated protein portion that results after thawing FFP
- Contains:
 - von Willebrand’s factor (plt. adhesion)
 - Fibrinogen
 - 150 mg in each unit
 - Factor VIII
 - About 80 IU in each unit
 - Fibrinonectin



Cryoprecipitate

- Same **storage** as FFP (cannot be *re-frozen* as FFP once it is separated); -18 for 1 year
- **If thawed, store at room temp 4 hrs**
- The leftover plasma is called **cryoprecipitate reduced** or **plasma cryo**
 - Good for thrombocytopenic purpura (TTP)
- CRYO is used for
 - Factor VIII deficiency (Hemophilia A)
 - von Willebrand's Disease
 - Congenital or acquired fibrinogen defects (i.e., dysfibrinogenemia).



Granulocytes

- Neutrophils are the most numerous, involved in phagocytosis of bacteria/fungi
- Although rare, it is useful for infants with bacteremia
- Prepared by hemapheresis
- $\geq 1.0 \times 10^{10}$
- Maintained at room temp for 24 hours.



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