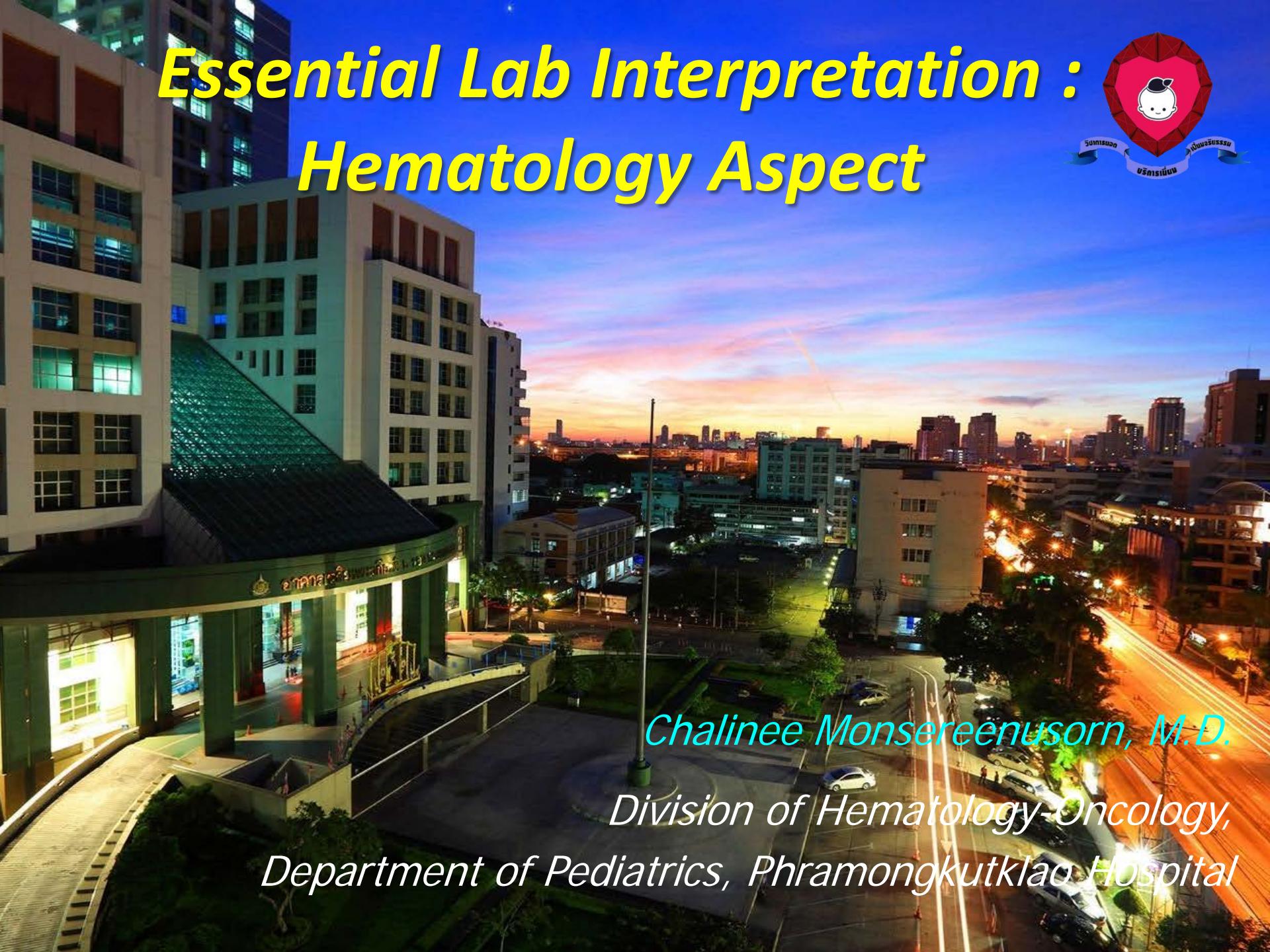
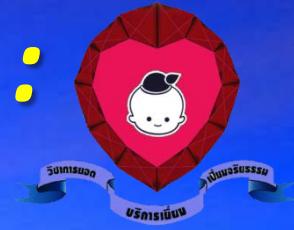


Essential Lab Interpretation : Hematology Aspect



Chalinee Monsereenusorn, M.D.

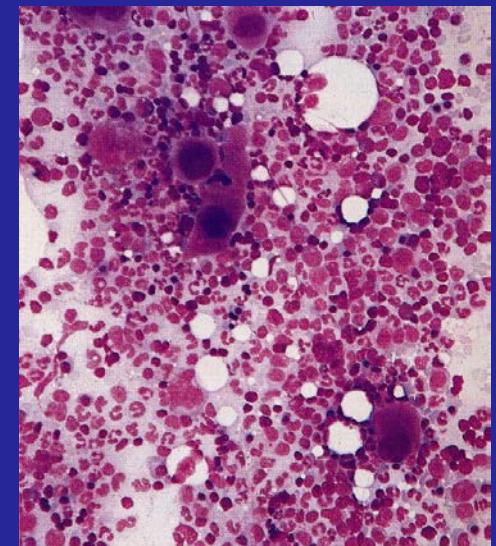
Division of Hematology-Oncology,

Department of Pediatrics, Phramongkutklao Hospital

Outlines

- Basic knowledge for laboratory in Benign Hematology :

- ✓ CBC and PBS
- ✓ Osmotic fragility test
- ✓ Platelet function testing
- ✓ Coagulation studies



CBC interpretation

CBC (WITH PLATELET COUNT)(30101)(7.1.1.1)

WBC = 3.9 (4.4 - 10.8)*1000/uL

RBC = 4.9 (3.9 - 5.2)*1000000/uL

Hematocrit = 38.2 (35.7 - 45.1)%

Neutrophil = 45 (39.2 - 70.8)%

Lymphocyte = 36 (20 - 48.4)%

Monocyte = 12 (3 - 9.9)%

Eosinophil = 7 (.5 - 5.5)%

Basophil = 0 (.1 - 1.9)%

MCV = 78 (80.4 - 95.2)fL

MCH = 26.9 (26.5 - 32.2)pg

MCHC = 34.6 (32.3 - 34.5)g/dL

RDW = 13.7 (12.2 - 14.8)%

Platelet count = 244 (184.1 - 422)*1000/uL

MPV = 9.4 (6.5 - 9.5)fL

Hemoglobin = 13.2 (12.1 - 14.7)g/dL

- RBC : Hb, Hct, MCV, MCH, MCHC, RDW
- WBC : Total and differential count
- Platelet : Total count, MPV



RBC indices

- MCV = mean corpuscular volume = size
- MCH = mean corpuscular hemoglobin = staining
- MCHC = mean corpuscular hemoglobin concentration
If $> 35 \text{ g/dl}$ spherocytosis
- Red blood cell distribution width = RDW
detect variation in red cell size = detect anisocytosis



Classification of anemia according to erythrocyte size

Microcytic **MCV < 80 fL**

Iron deficiency
Thalassemia
Sideroblastic anemia

Normocytic **MCV 81-90 fL**

Acute blood loss
Renal anemia
Spherocytosis
G6PD deficiency

Macrocytic **MCV > 95 fL**

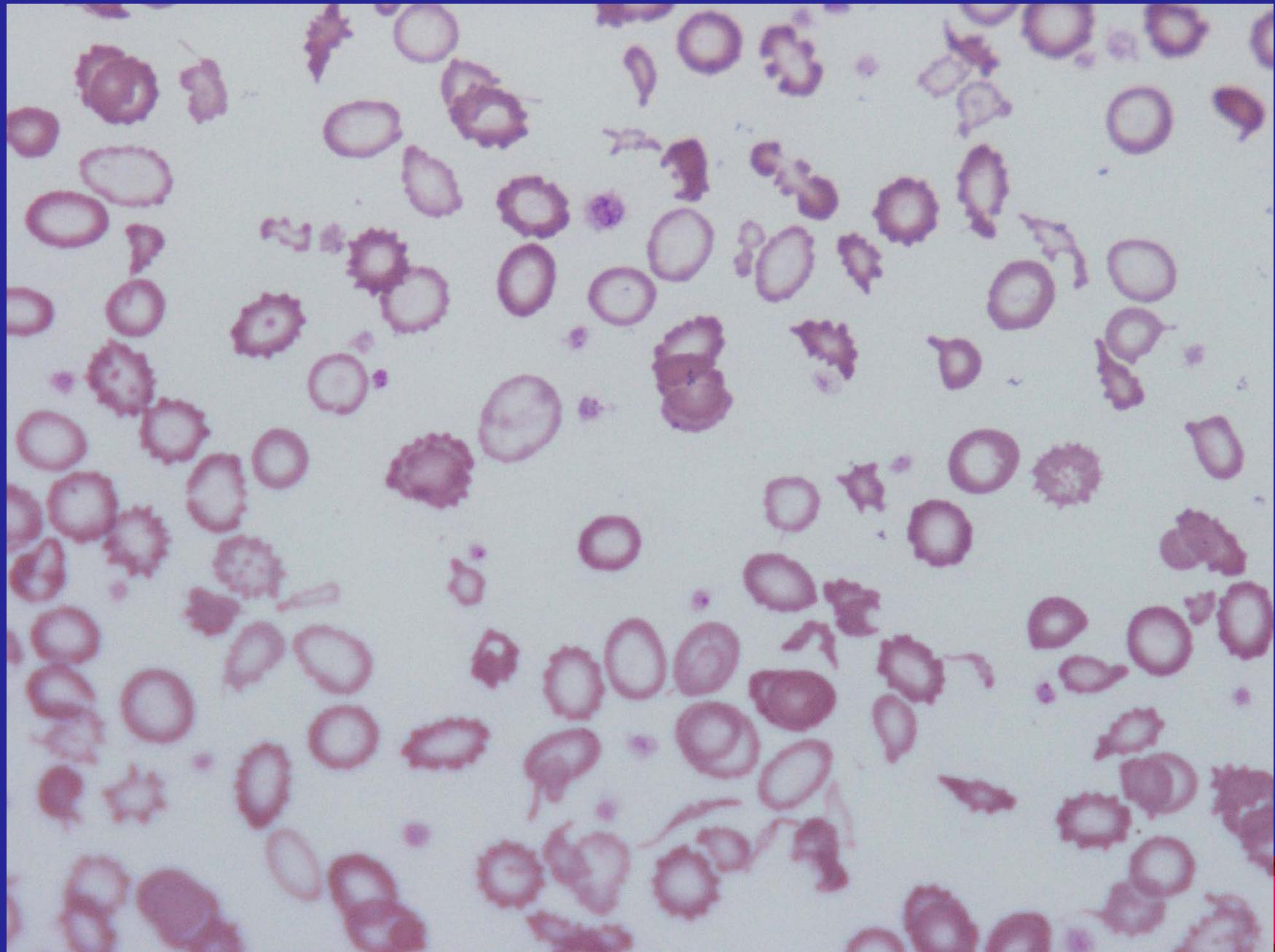
Vit B₁₂ deficiency
Folic acid deficiency
Chemotherapy
Myelodysplasia
Bone marrow failure syndrome
Hemolysis

Hb Typing and RBC parameters Correlation

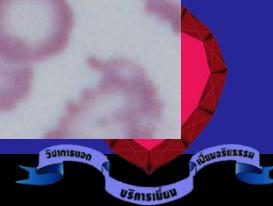
10 months old Thai boy

- Anemia
- Liver and spleen just palpable

CBC	
RBC($10^6/\text{mcL}$)	3.37 (4.1-6.1)
Hct (%)	19
Hb (g/dL)	5
MCV (fl)	45 (80-100)
MCH (pg)	18 (26-34)
MCHC (g/dL)	29.7 (31-37)
RDW (%)	28 (11.9-14.8)
Father	--SEA/ $\alpha\alpha$
Mother	β^0/β



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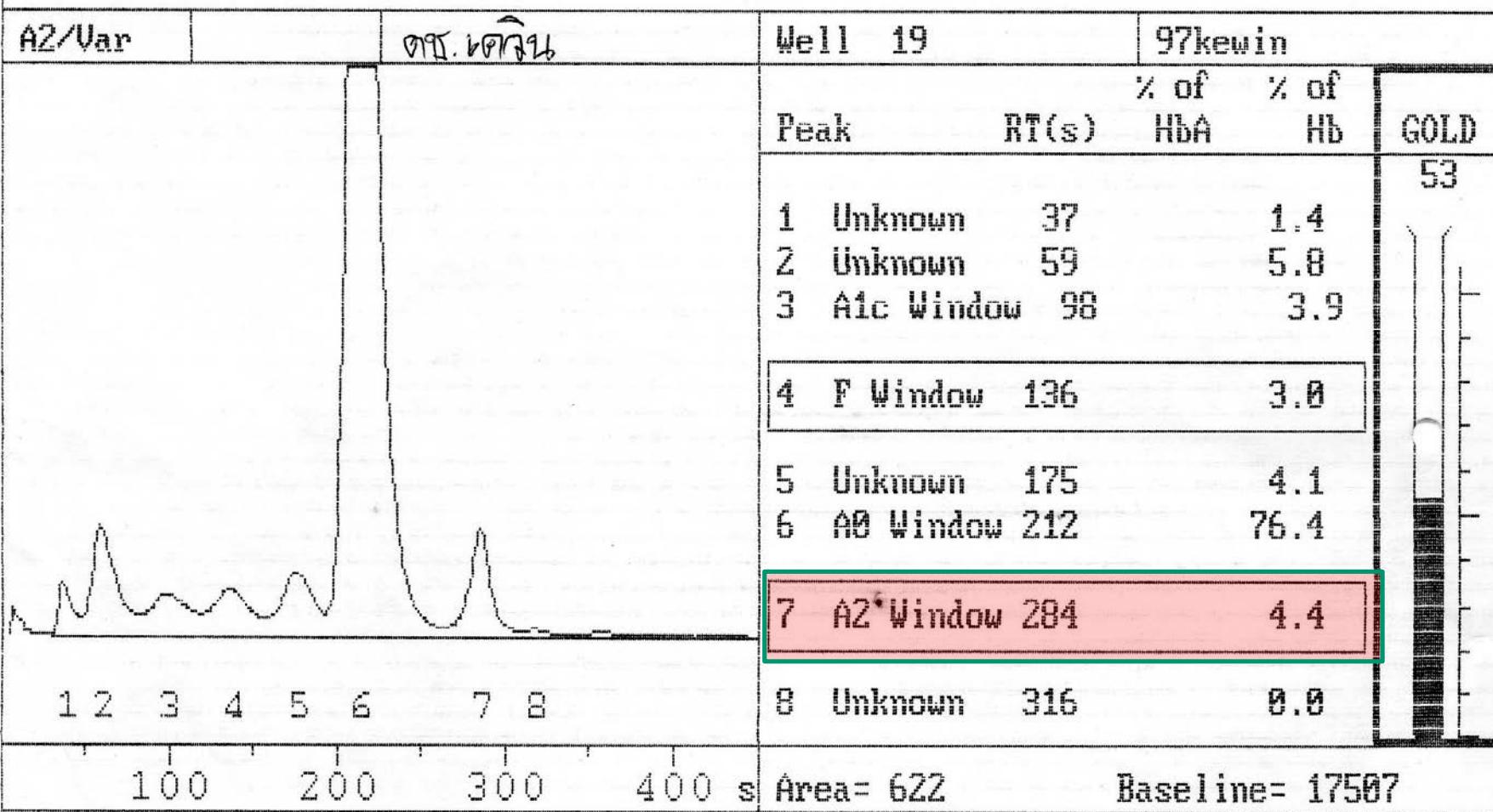
Hb-GOLD

26.7°C. Off J

C

PRAMONGKUT

15:53 17-10-2013



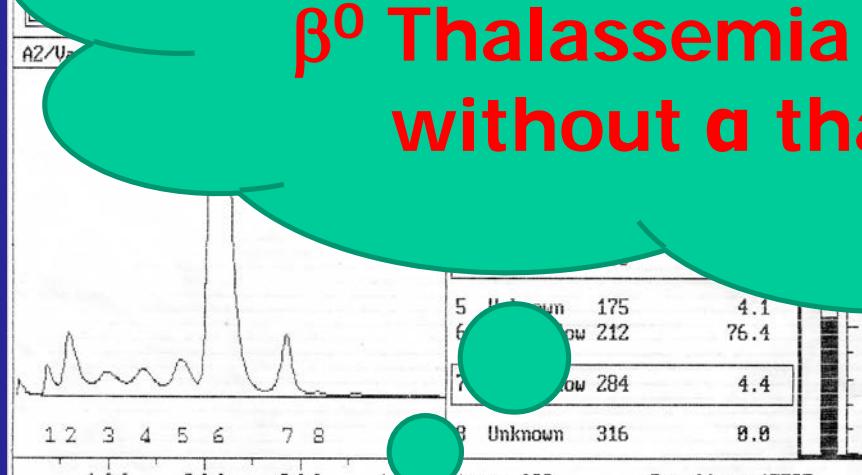
10 months old Thai boy

- Anemia
- Liver and spleen palpable

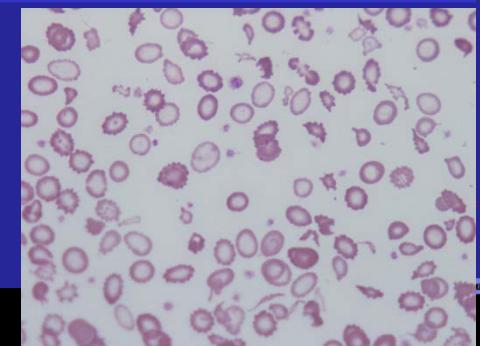
A₂A (Hb A₂ > 3.5%)

β⁰/β

β⁰ Thalassemia trait with or without α thalassemia



CBC	
RBC(10 ⁶ /mcL)	3.37 (4.1-6.1)
WBC(10 ³ /mcL)	11.9-14.8)
Platelet(10 ³ /mcL)	--SEA/αα
Mother	β ⁰ /β



What should we do next?

- Inclusion body: Negative
- PCR:
 - SEA/ $-{\alpha}^{3.7}$ with β^0/β^+

Hb H disease with β^0 Thalassemia trait with iron deficiency anemia



RBC Membrane Defect

A 5-year-old boy presents with poor appetite. Physical examination reveals pallor, moderate icteric sclerae and splenomegaly.

Automated CBC:

Hb 8.4 g/dL

Hct 26 %

MCV 82 fL (> 78fL)

MCH 34 pg (>25 pg)

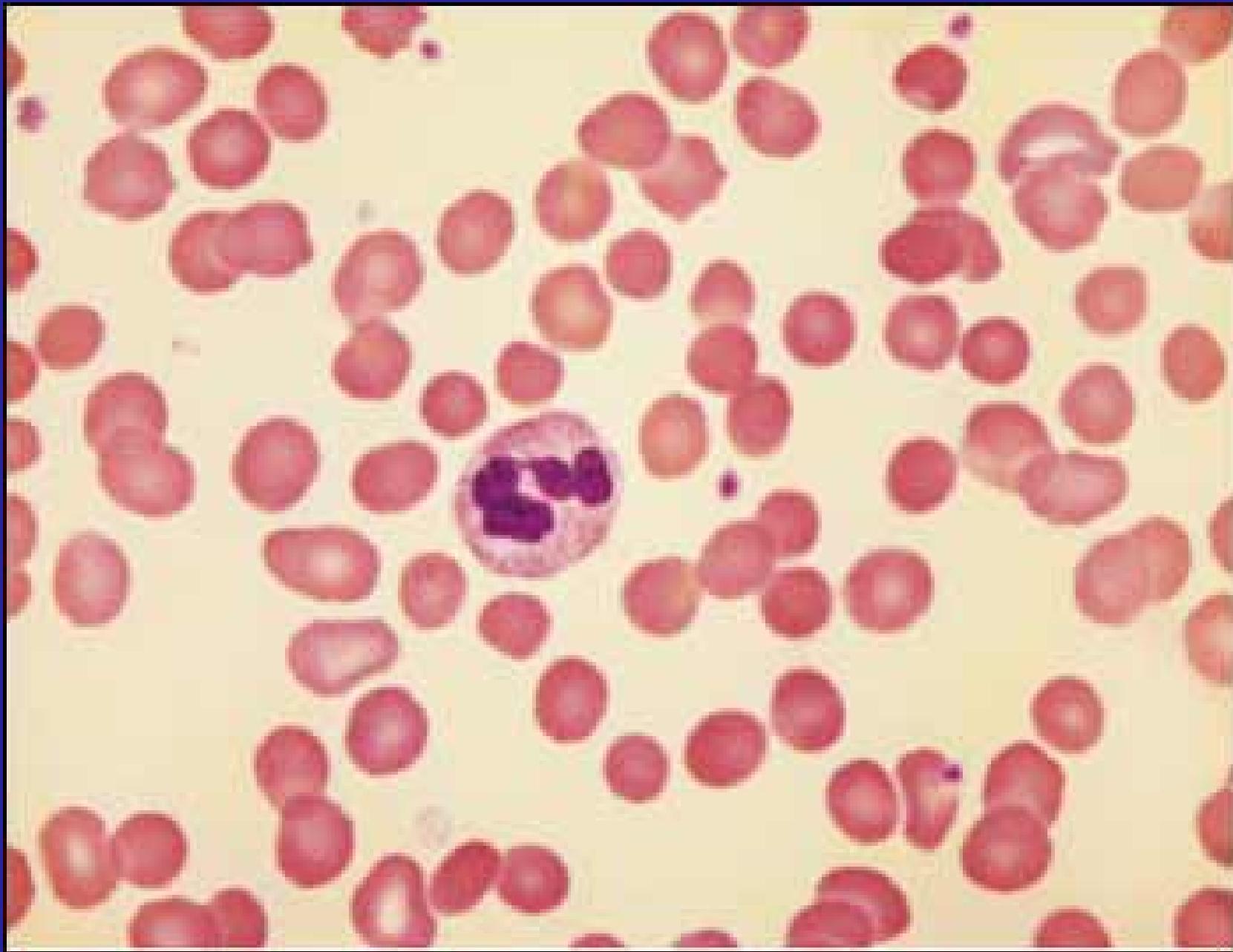
MCHC 37 g/dL (31-35 g/dL)

RDW 15 % (13-17%)

WBC 7,800 / mm³ P 50 % L 40 % M 10%

PLT 460,000 / mm³



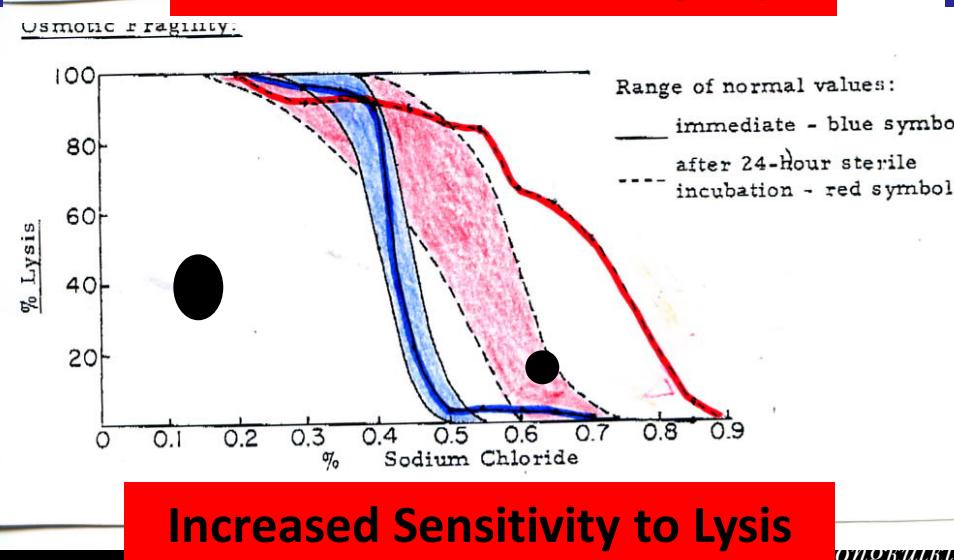
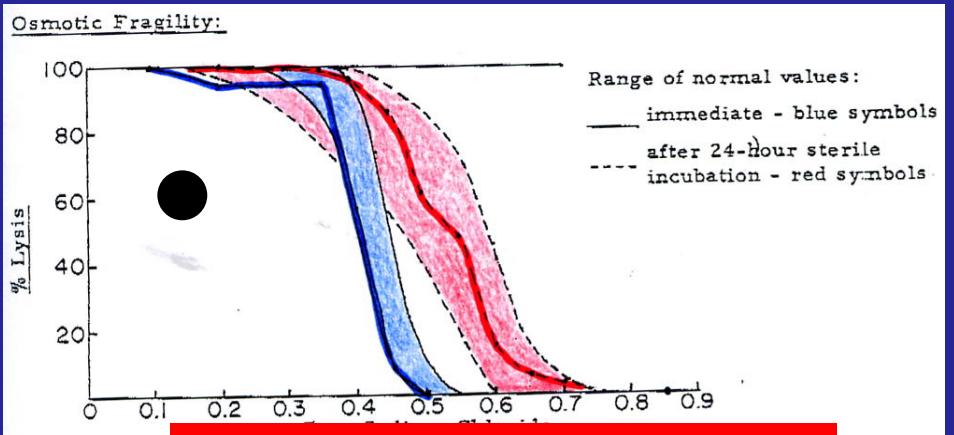


Phramongkutkla Hospital



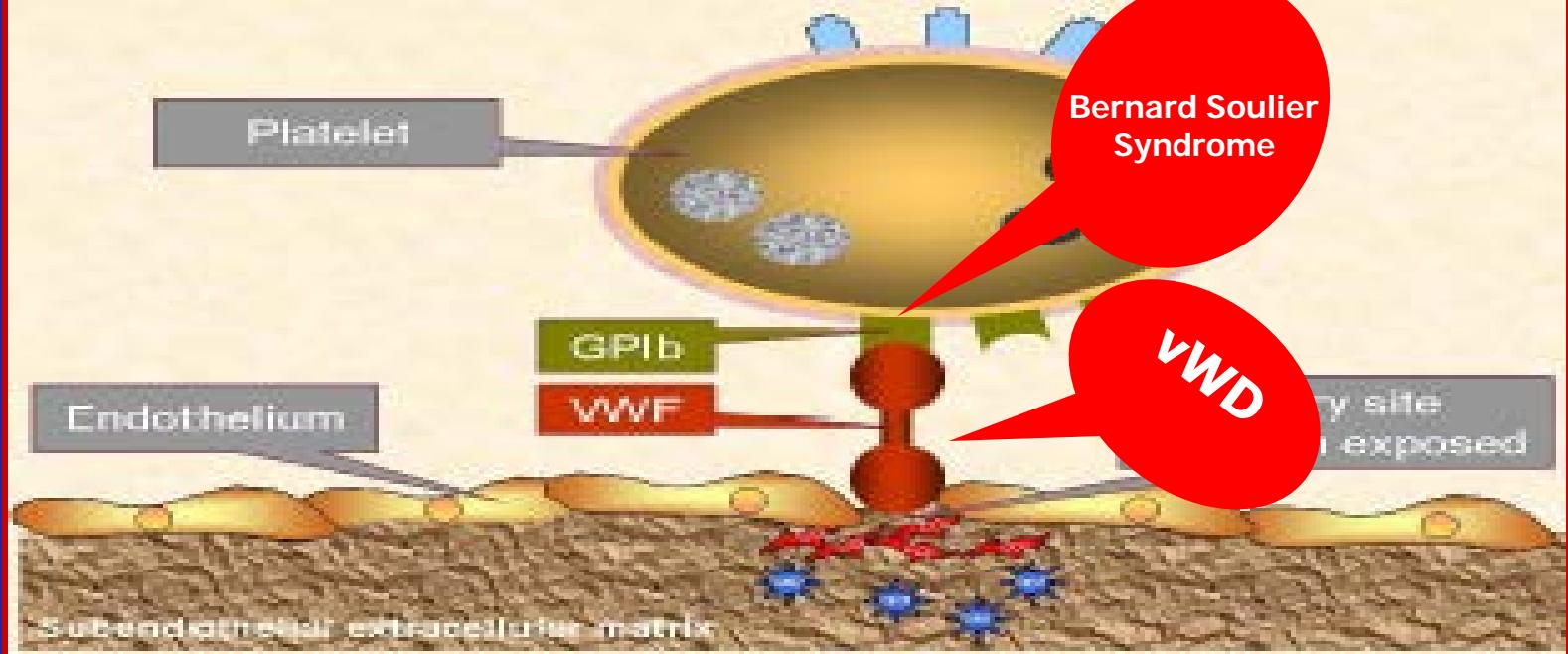


Incubated Osmotic Fragility Testing

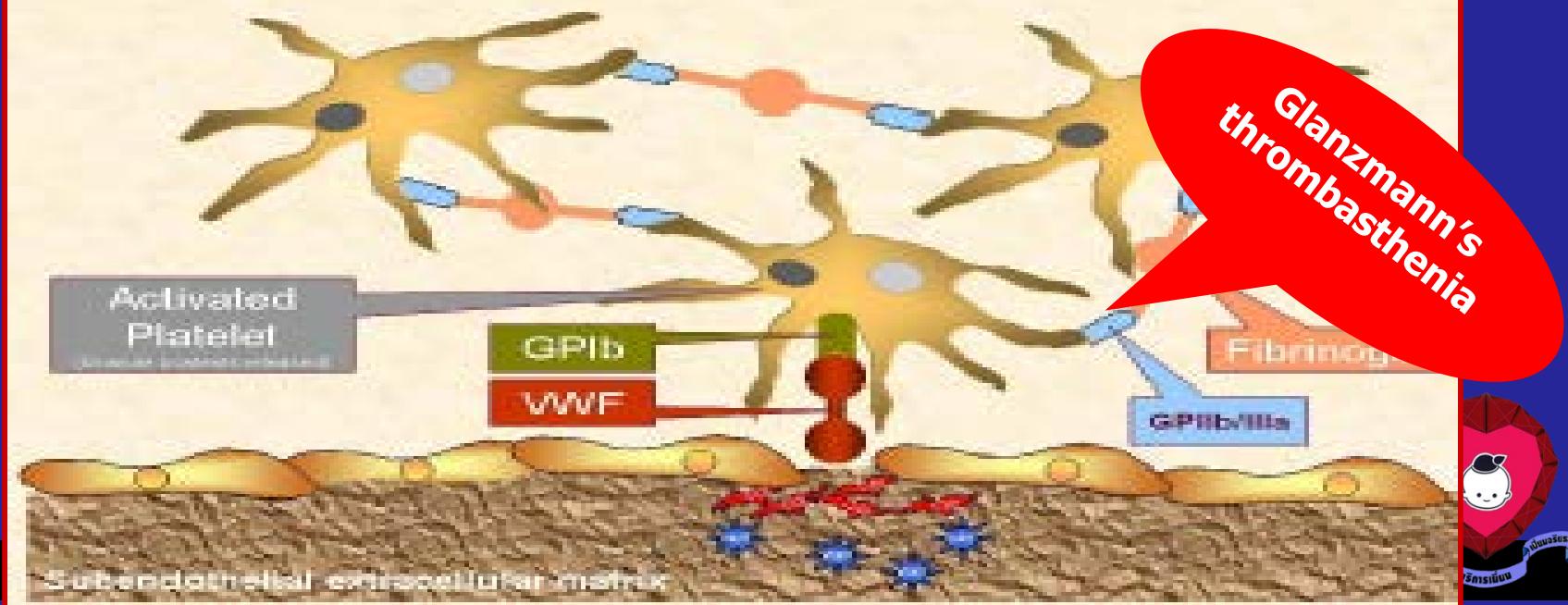


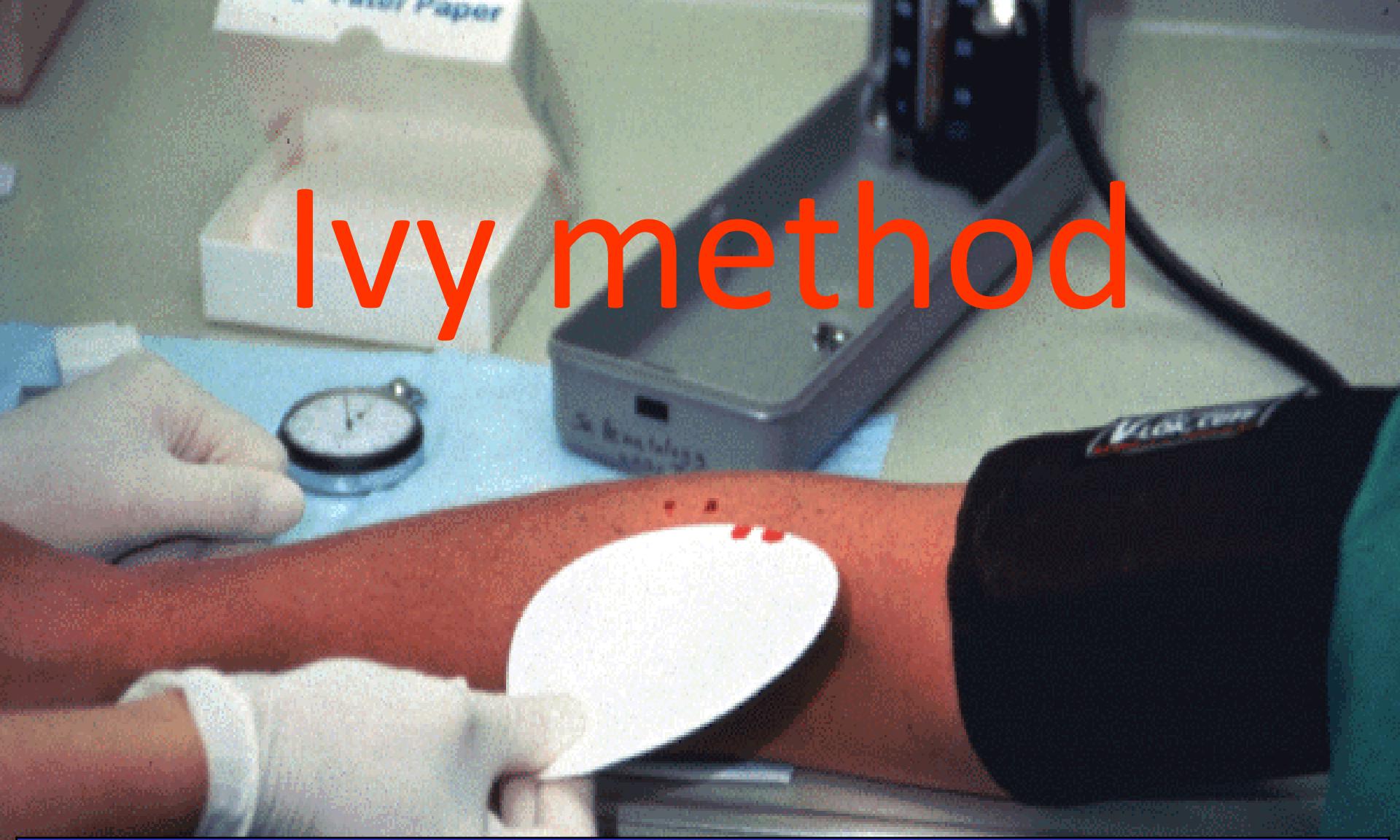
- Red cells are incubated in varying concentrations of saline (0 – 0.9%) for up to 48 hours
- As concentration of saline decreases, cells take on water and are hemolyzed
 - Normal cells around 0.5%
 - HS cells at higher NaCl concentrations
- Degree of hemolysis is detected by spectrophotometry
- Not reliable < 6-12 months of age

Platelet and Coagulation Studies



Primary Hemostasis



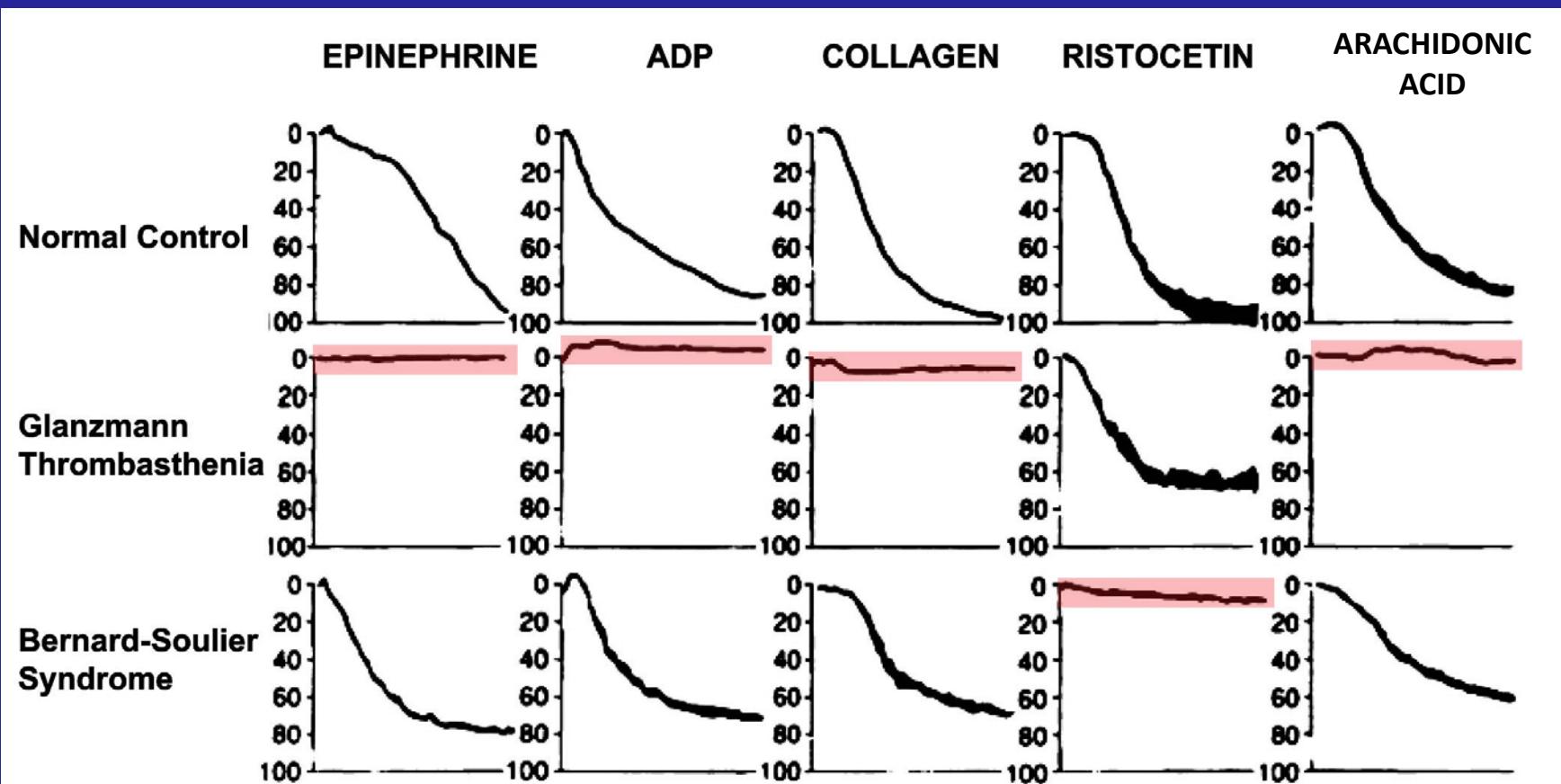


Ivy method

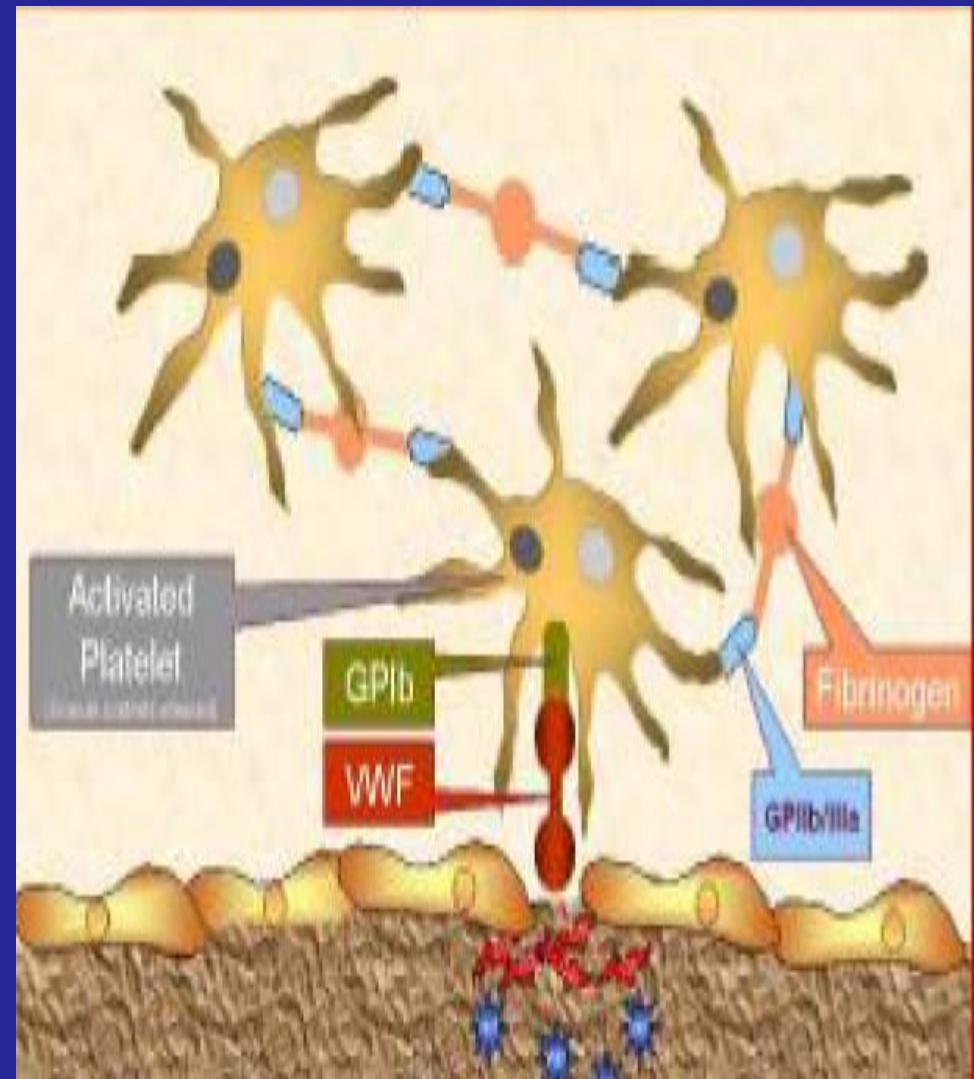
Normal range : typically 2 to 10 minutes

Accuracy if BT > 20 minutes

Platelet Aggregation Profiles



Clot Retraction

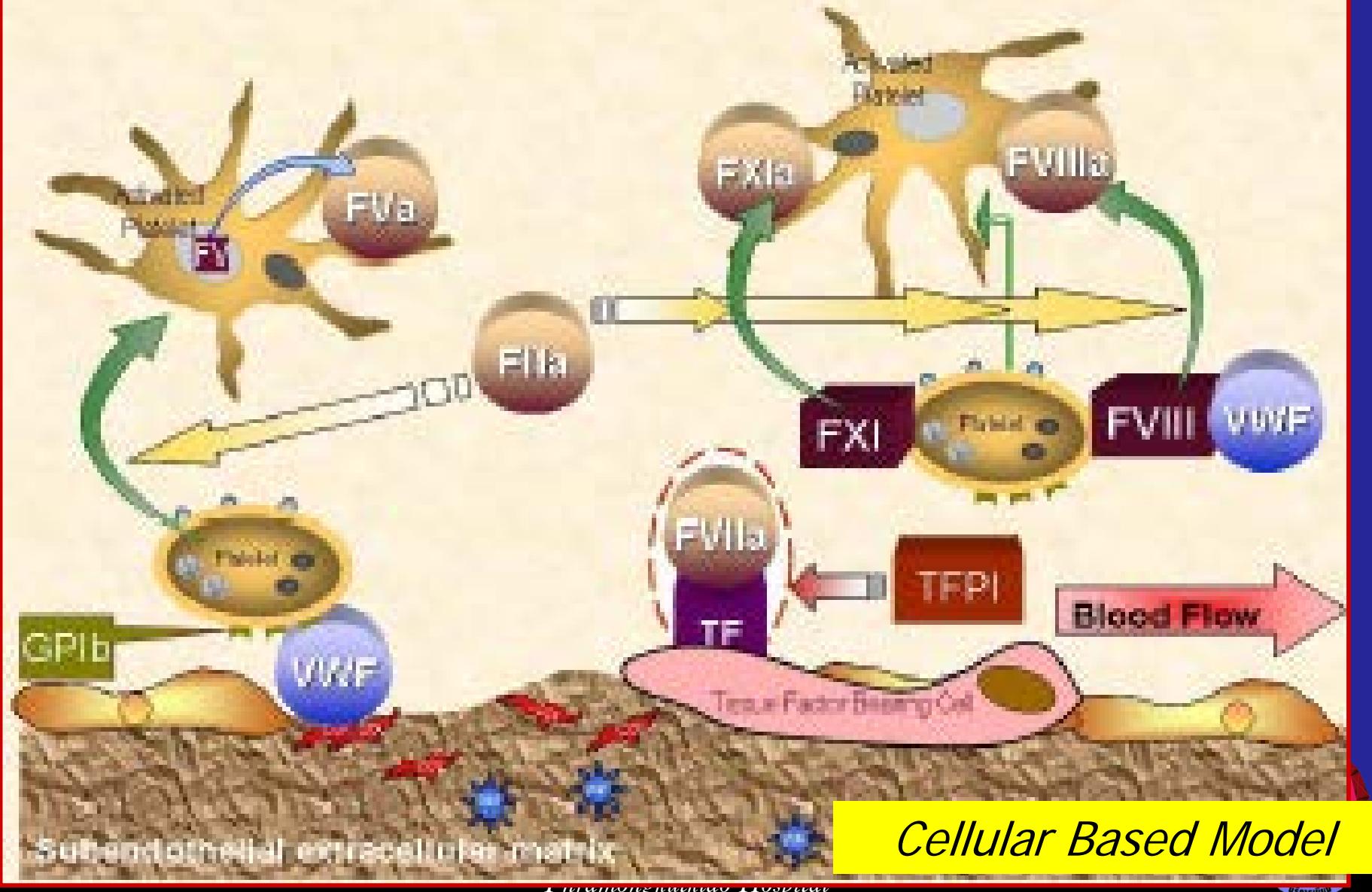


Clot retraction

- Positive in
 - GP IIb/IIIa defect (Glanzmann's thrombasthenia)
 - Afibrinogenemia/dysfibrinogenemia
- False positive
 - Thrombocytopenia



Secondary Hemostasis

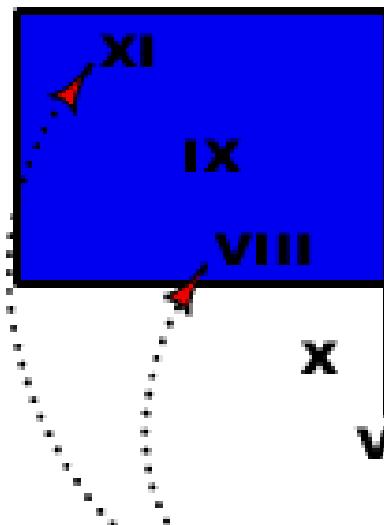


CLOTTING FACTORS AND RELATED COAGULATION TESTS

INTRINSIC SYSTEM

Water Fall Model

XII

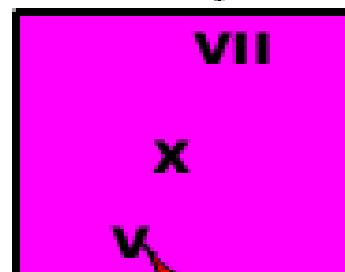


aPTT

Monitor
Heparin Therapy

EXTRINSIC SYSTEM

Tissue Factor
+



PT

Monitor
Coumadin Therapy

II → Thrombin ← II

fibrinogen

Fibrin

XIII

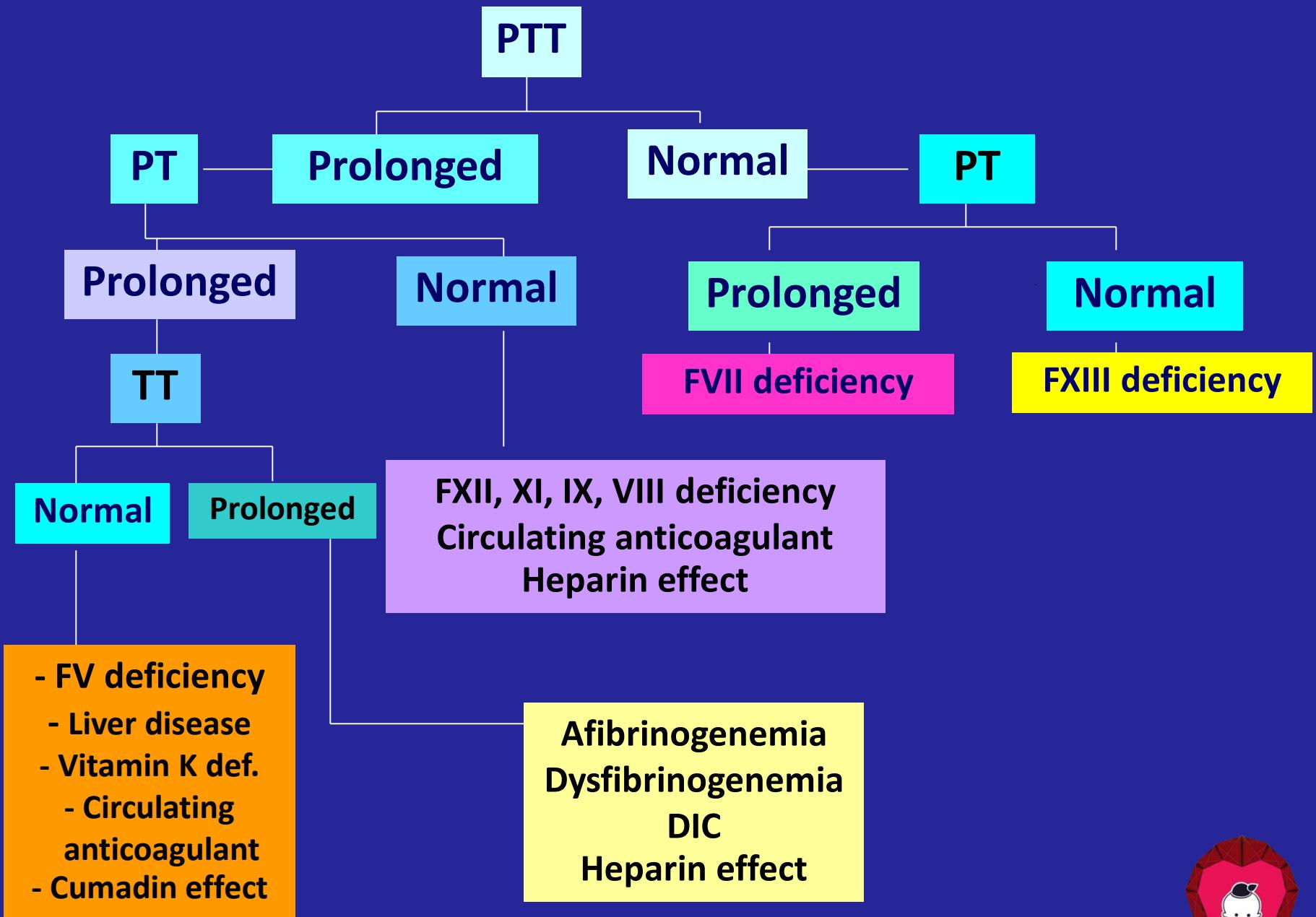
Fibrin stabilized

PT Procedure

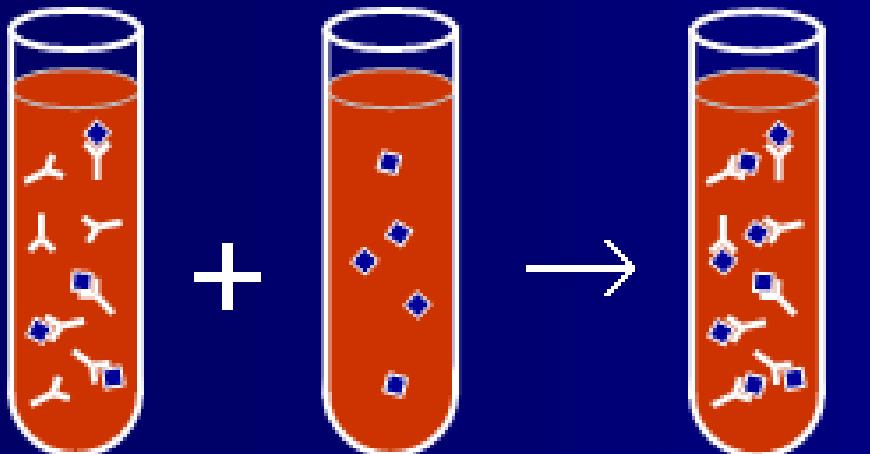
1. plasma sample
2. add Ca++
3. add thromboplastin
4. time elapsed to clot = PT

aPTT Procedure

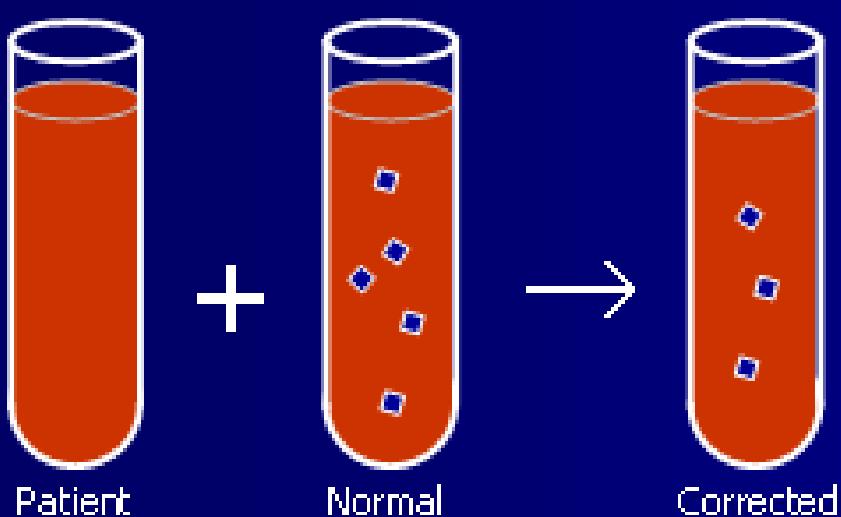
1. plasma sample
2. add Ca++
3. add thromboplastin
4. add (-) charged foreign material
5. time elapsed to clot = aPTT



aPTT Mixing study



Clotting times
remain
prolonged =
Inhibitor

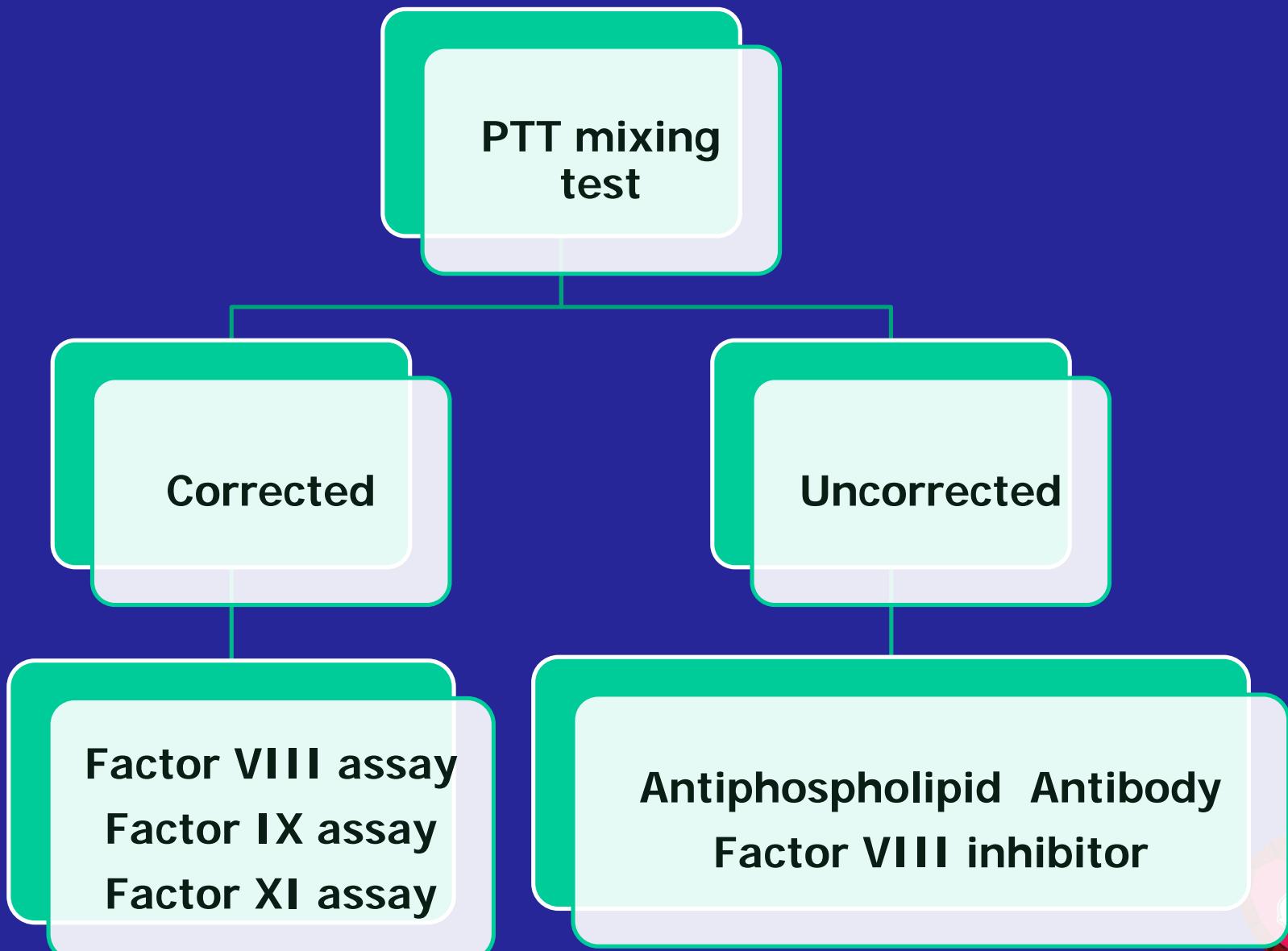


vs

Clotting times
normalize or
decrease to
near-normal =
Factor deficiency



Isolated PTT Prolonged



Mixing test

Sample	aPTT	PT
Normal control	34 sec	13 sec
Patient	54 sec	12.8 sec
Mixing study (1:1)	34.5 sec	13 sec

Diagnosis

Further study

Clotting factor deficiency

Specific factor assay e.g. FVIII:C, FIX:C

A 10 -year-old girl was planed to do elective tonsillectomy next week.

Automated CBC:

Hb 12.3 g/dL

Hct 37 %

MCV 83 fL (> 78fL)

MCH 29 pg (>25 pg)

MCHC 33 g/dL (31-35 g/dL)

RDW 14 % (13-17%)

WBC 5,800 / mm³ P 50 % L 40 % M 10 %

PLT 235,000 / mm³



Coagulogram

PT = 14 sec (12-15 sec)

aPTT = 70 sec (25-35 sec)

TT= 13 sec (10-20 sec)



Mixing test

Sample	aPTT	PT
Normal control	34 sec	13 sec
Patient	70 sec	14 sec
Mixing study (1:1)	68 sec	13 sec

Diagnosis

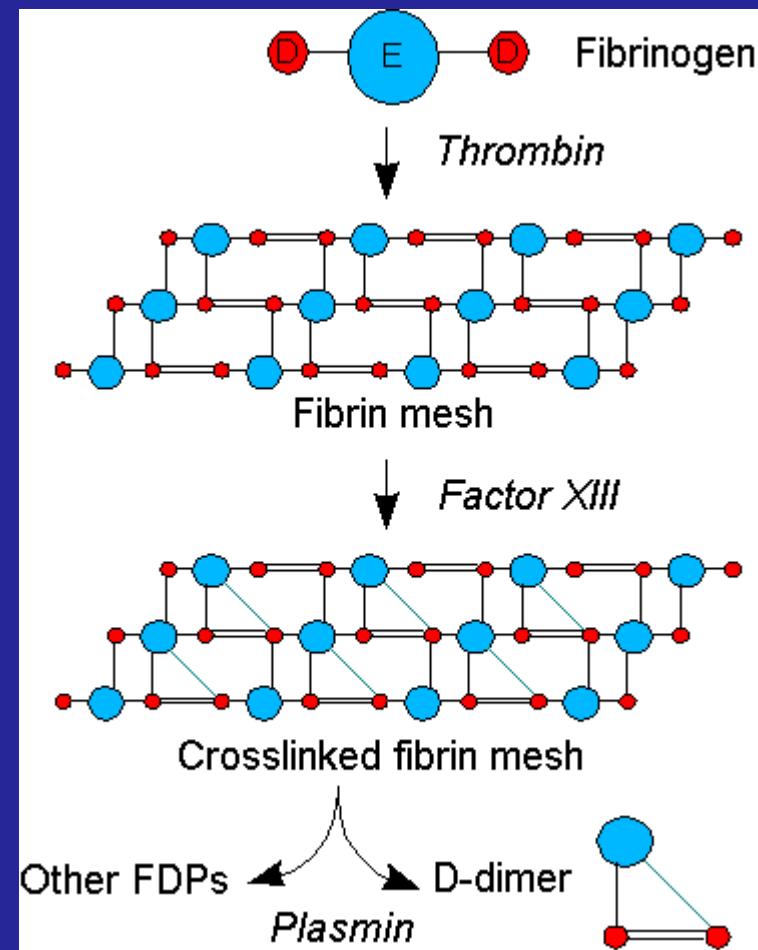
Antibodies

Further study

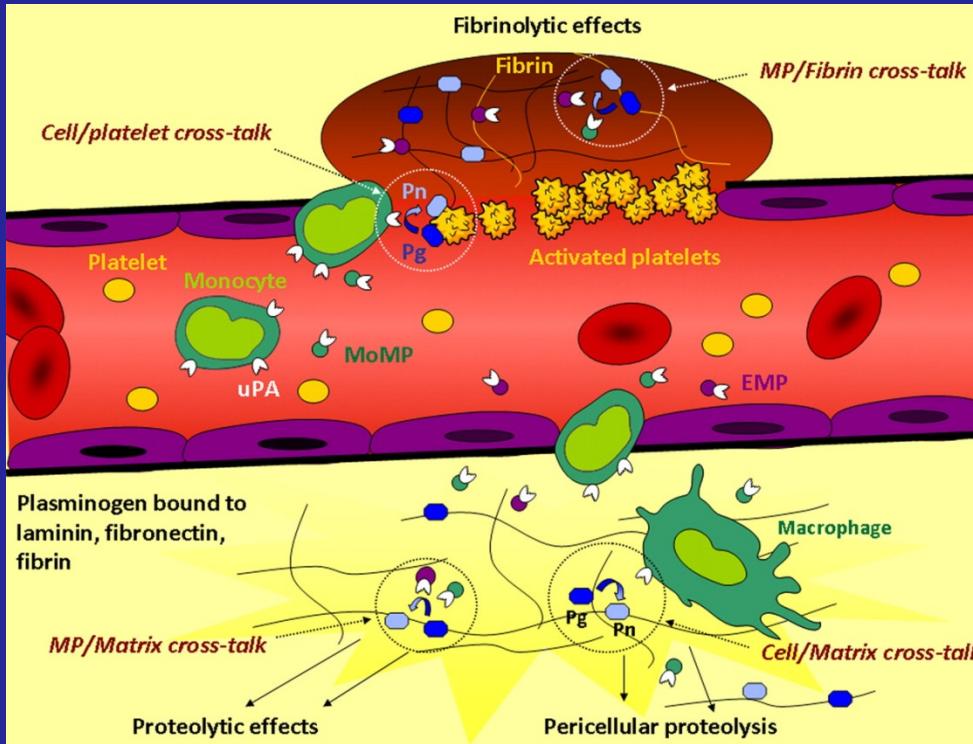
Lupus anticoagulant, β 2GPI, anticardiolipin

Clot Solubility Test

- 5M urea และ acetic acid
- Factor XIII deficiency
 - Positive : clot lysis
 - Control : normal plasma : not lysis



Euglobulin Lysis Time



Normal > 24 hrs

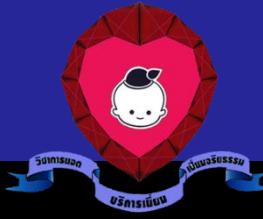
< 30 Min : Hyperfibrinolysis

> 24-48 hrs : Hypofibrinolysis

6 months old boy presented with high grade fever, jaundice for 3 weeks

- PE:BT 39°C, PR 100/min, RR 24/min,
BP 90/60 mmHg
- Moderate pale, mild jaundice, no lymphadenopathy
- Abdomen: Liver 3 cm below RCM and spleen 8 cm
below LCM
- Neuro : No stiffneck, hypotonia 2+
- Others: WNL

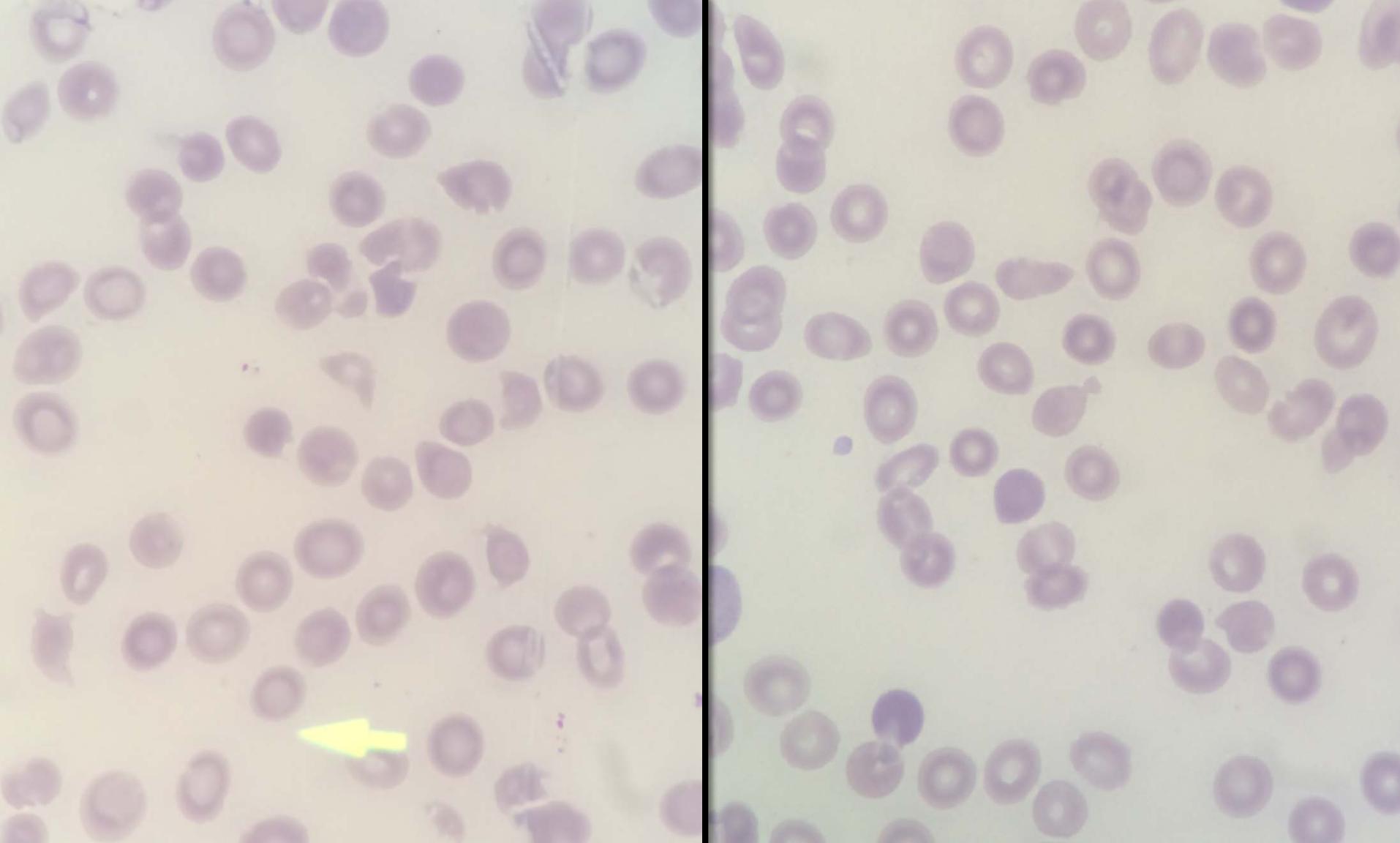




CBC	
Hb (g/dl)	5.4
Hct (%)	17
WBC	26,600
PMN (%)	39
L (%) / AL	54
M (%)	5
E (%)	0
B (%)	0
Platelet	10,000
MCV (fl)	84
MCH (pg)	26
MCHC (g/dl)	31
RDW (%)	24
Reti.	3.35 %

Blood Chemistry	
Total protein	5.1
Albumin	2.9
TB	28.8
DB	23.5
AST	394
ALT	182
ALP	243
Gamma GT	9





PBS : Normochromic, microcytic RBC, anisopoikilocytosis 1+, few schistocyte , few polychromasia, no toxic granule, no vacuolization, Platelet decrease

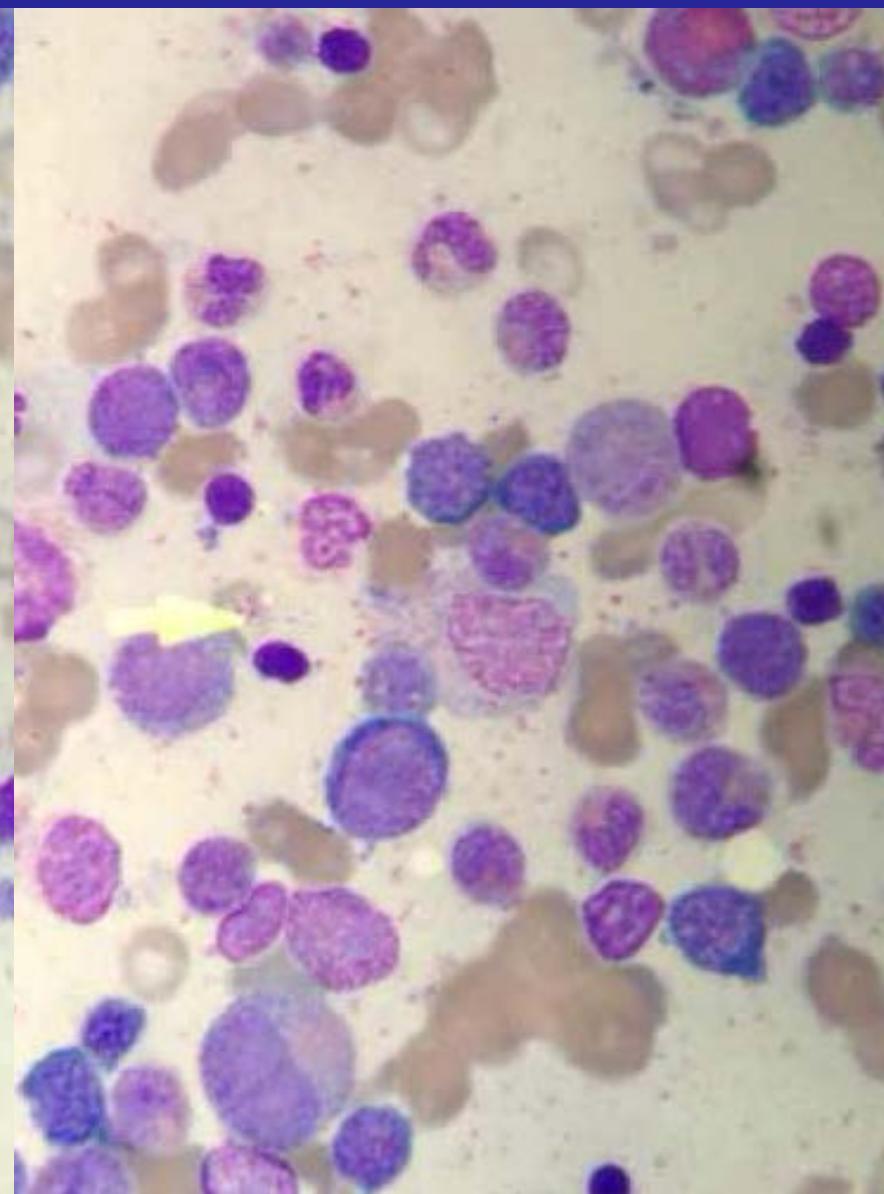
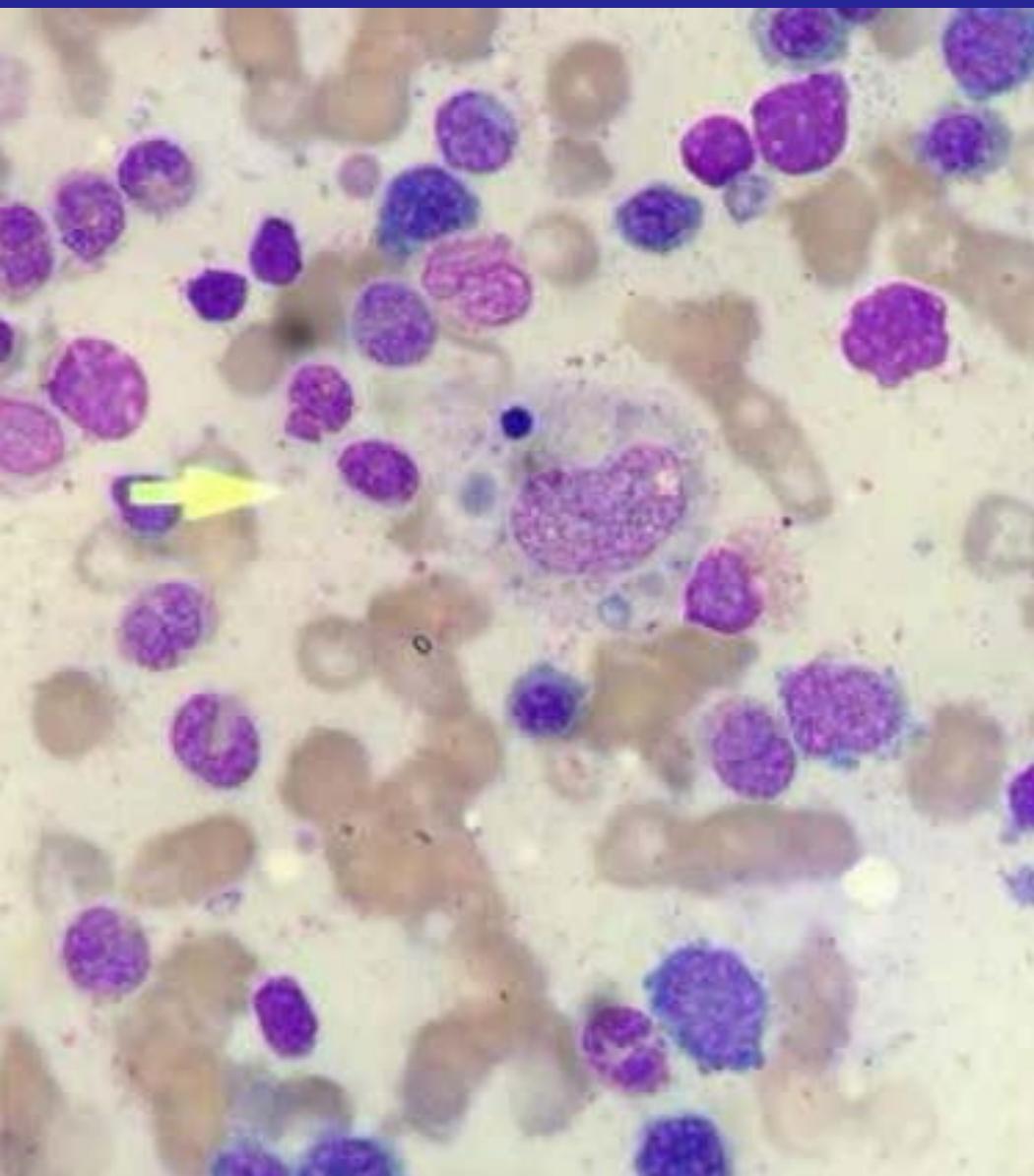
DCT : 1+ **IDCT** : negative

APTT	41.9
PT	17.8
INR	1.47
TT	17.4

Lab	
Triglyceride	206
Ferritin	1383
Fibrinogen	< 80
LDH	476
CPK	83



Investigation → Bone marrow aspiration





THANK YOU