



قسم الكيمياء الحيوية
Biochemistry Department
College of Science - King Saud University

BCH 471

Experiment (10)

Complete Blood Count (CBC)

Objectives

1. To estimate the number of RBC in blood sample
2. To estimate the number of total WBC in blood sample
3. To perform a differential count for a blood sample

Introduction

- **Complete blood count (CBC)** is a test that gives information about the cells in a patient's blood.
- A CBC test usually includes:
 - WBC count.
 - WBC differential count.
 - RBC count.
 - HCT
 - Hb
 - Platelet count
 - Mean platelet volume (MPV)
 - Red blood cell indices: There are three red blood cell indices: mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC).

- **CBC can applied by two way:**

1. Automated blood count
2. Manual blood count

- **Automated blood count:**

- CBC is performed by an automated analyzer that counts the numbers and types of different cells within the blood.
- It aspirates a very small amount of the sample through the narrow tubing. Within this tubing, there are sensors that count the number of cells going through it, and can identify the type of cell; this is called **flow-cytometry**.

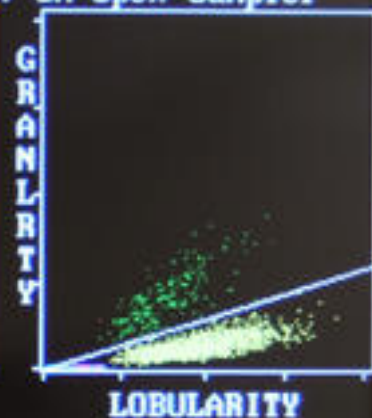
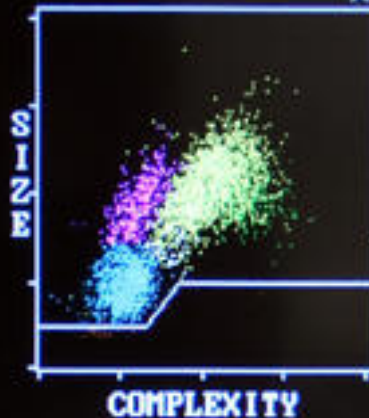


Next ID Auto
Patient -----
Sex(M/F):- DOB:--/--/--
Dr -----

RUN
Ready
Report for 00006667

23 Sep 2008 09:29
Operator ID ---
Sequence # 9289
ML: ON Open Sampler

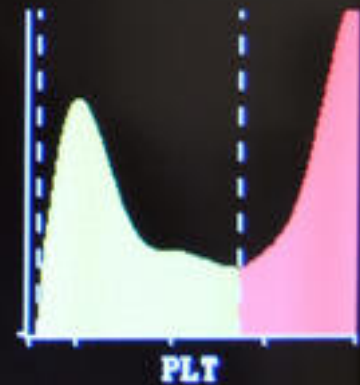
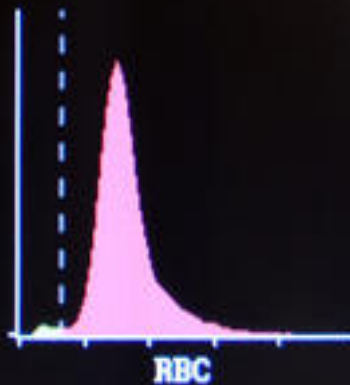
Paran: 1 Limits: 1
WBC 5.00 K/uL
NEU 2.57 51.3 %N
LYM 1.74 34.9 %L
MONO .392 7.85 %M
EOS .239 4.79 %E
BASO .056 1.12 %B



MCT:4.52

RBC 5.93 M/uL
HGB 14.1 g/dL
HCT 41.6 %
MCV 70.2 fL
MCH 23.7 pg
MCHC 33.8 g/dL
RDW 17.0 %

RBC MORPH



URI RCT:6.54

- CLEAR APERTURES
- WORK LIST
- SPECIMEN TYPE
- CUSTOMIZE REPORT
- CHANGE SAMPLER
- PRINT TICKET
- COLOR PRINT
- MAIN

LOST RIVERS MEDICAL CENTER LABORATORY

551 HIGHLAND DRIVE, ARCO, IDAHO 83213

PH (208) 527-8206 x 119

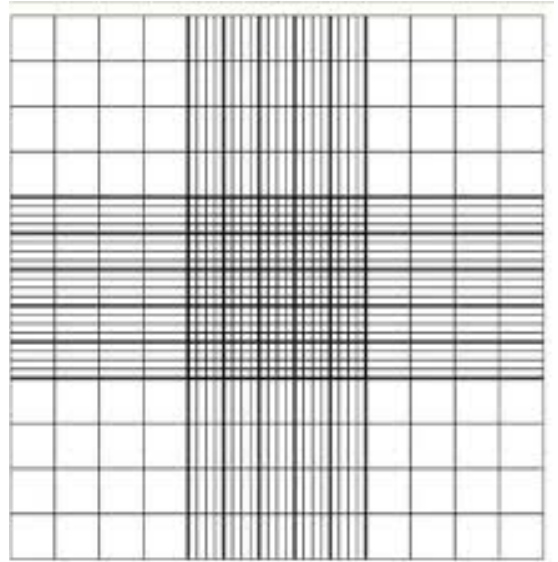
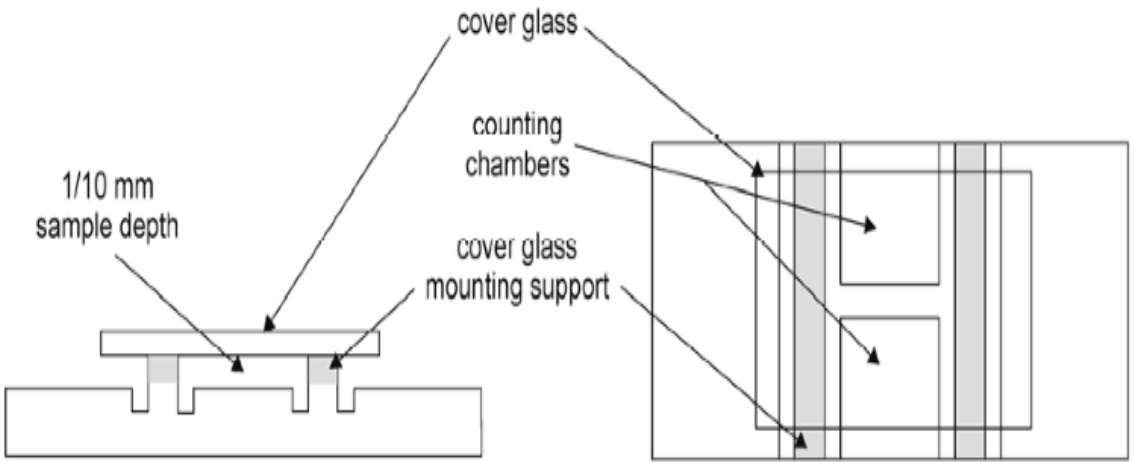
FAX (208) 527-3791

Patient: MALKIEWICZ, JUDITH A	Birth: 12/8/1950	Acc #: 55276	
Patient #: 120850JM	Age: 61 years	Fasting: UNKNOWN	
Doctor: NON-STAFF	Gender: Female	Collection Date: 2/22/2012 09:20	DZ
Home Phone: (208)588-3977		Received in Lab: 2/22/2012 09:20	DZ
		Destination	OP
		DR	SHULL/MYINT

Test Name	Result	Units	Flag	Reference Range
CBC W/ 5 PART DIFF. (X6)				<i>Run by: TB on 2/22/2012 09:33</i>
WBC	2.1	K/uL		4.0 - 11.2
RBC	4.15	M/uL		4.00 - 5.60
HGB	13.5	gm/dL		12.0 - 16.0
HCT	39.5	%VOL		35.0 - 50.0
MCV	95	fl		82 - 98
PLATELETS	172	K/uL		140 - 440
MCH	32.6	pg		26.0 - 36.0
MCHC	34.3	g/dL		27.0 - 36.0
RDW	13.2	%		9.0 - 18.0
MPV	7.3	fl		6.0 - 12.0
NEU%	55.2	%		45.0 - 65.0
LYMPH%	30.9	%		20.0 - 50.0
MONO%	9.0	%		0.0 - 11.0
EOS%	4.0	%		0.0 - 7.0
BASO%	0.9	%		0.0 - 3.0
NEUT#	1.17	K/uL		2.00 - 8.00
LYMPH#	0.65	K/uL		1.80 - 4.80
MONO#	0.19	K/uL		0.10 - 1.10
EOS#	0.08	K/dl		0.00 - 0.80
BASO#	0.02	K/dl		0.00 - 0.30

- Manual blood count

- This measurement is made with a microscope and a specially ruled chamber (**hemocytometer**) using diluted blood.



(A) Red blood count:

- It is test done to determination the number of RBC in a sample of blood , also it evaluate the size and shape of RBC
- It is range from 4.2 – 5.5 million RBC per cubic millimeter (mm^3)
- It is considered a very important indicator of a patient's health

Low RBC count

- ✓ Anemia
- ✓ Acute or chronic blood loss
- ✓ Malnutrition
- ✓ Chronic inflammation

High RBC count

- ✓ Polycythemia
- ✓ Congenital heart disease
- ✓ Renal problem

Normally high (RBC count)

- ✓ People who live at high altitudes
 - ✓ Smokers
- } Oxygen is low → RBC synthesis increases

Principle :

The process involves by counting cells in several squares of the grid and obtain an average number, this number is multiply by a factor that compensates the amount of dilution. The final results expresses the number of RBC /mm³ of original blood sample.

(B) WBC count :

- Total leukocytes count shows the number of WBC in a sample of blood .
- A normal WBC count is between 4,500 and 11,000 cells per cubic millimeter .
- The number of WBC is sometimes used to identify an infection or to monitor the body's response to treatment.

Low WBC count → Leukopenia

- ✓ A Condition in which the number of leukocytes is abnormally low and which is most commonly due to severe infections (such as HIV) and radiation poisoning.

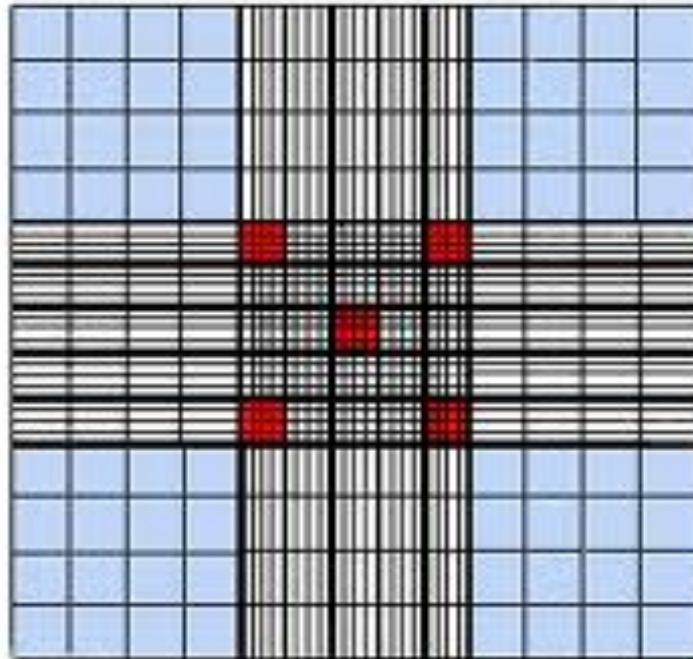
High WBC count → Leukocytosis

- ✓ A condition characterized by an elevated the number of WBC occur as a result of an infection, or cancer (Leukemia).
- ✓ It can occur normally after eating fat-rich meals .

Principle:

- It is necessary to obtain RBC free preparation of WBC from blood .
- Suspension of the red blood cell in a very hypotonic solution will lead to the destruction of RBC .

■ areas of the grid where WBC are counted



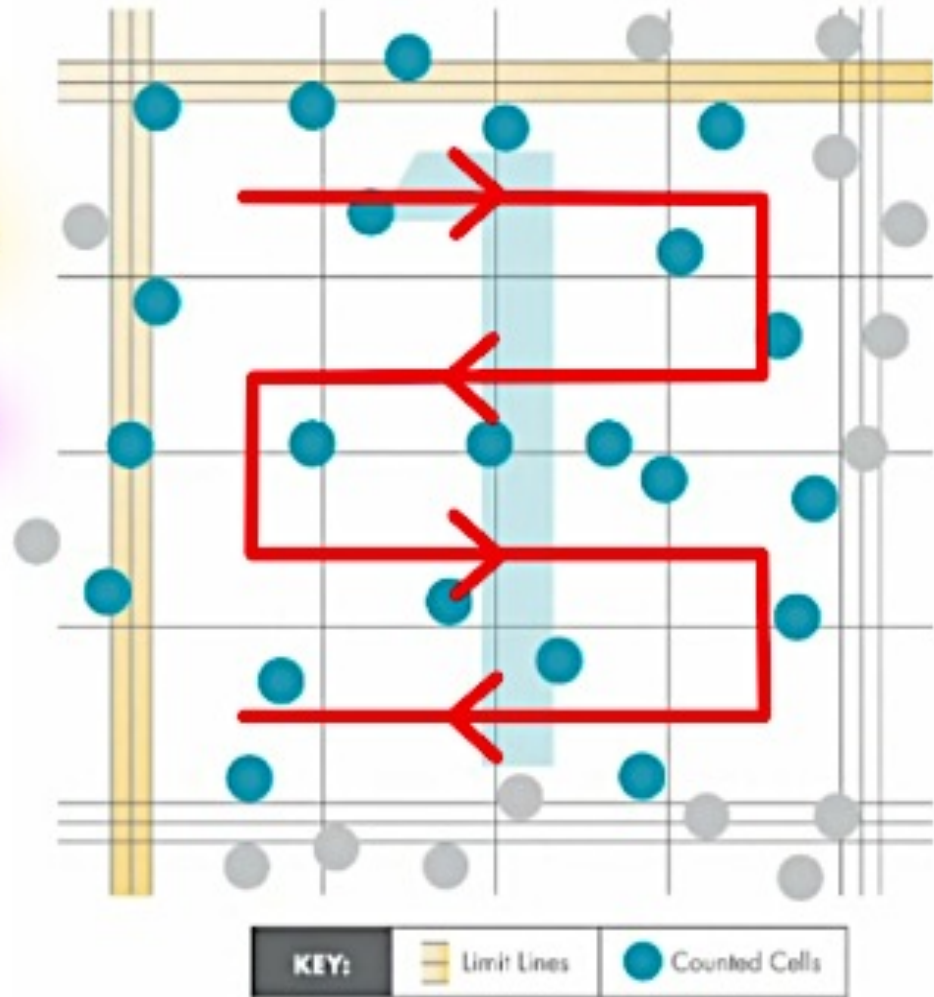
■ areas of the grid where RBC are counted

HOW TO COUNT BLOOD CELLS

Counting 2 sides in L shape
(i.e. count the Cells settle on the Top and left sides
(colored sides)
and exclude the cells on the right and the bottom sides)

Counting is Zigzag in all squares

Blue cells is counted
gray cells is not counted



Calculations:

✓ RBC blood cell count (5 squares)

- Find the sum of RBCs in 5 large squares, and divide it with 80 (5 X 16) small squares to find the average in one square, multiply it by 200 to allow for the dilution and then multiply by 4000 to obtain the number per cubic milliliter.
- The sum of RBCs in 5 large squares = $84+71+63+93+83= 394$ cells.
- The average of RBCs in one square = $394/80 = 4.9$ cells.
- RBC count = $4.9 \times 200 \times 4000 = 4$ million/mm³.

✓ Normal range = 4.2-5 million/mm³

✓ **WBC blood cell count (4 squares)**

- Find the sum of WBCs in 4 large squares, and divide it with 64 (4 X 16) small squares to find the average in one square, multiply it by 20 to allow for the dilution and then multiply by 160 to obtain the number per cubic milliliter.
 - **The sum of WBCs in 4 large squares = 16+21+17+15= 69 cells.**
 - **The average of WBCs in one square= 69/64 = 1.07 cells.**
 - **WBC count= 1 x 20 x 160 = 3200 Cells/mm³.**
- ✓ Normal range= 4500-11000 cells /mm³

(C) Differential Count

- It determines the number of each type of WBC present in the blood

Class of White Cells	White Cell Type		% of total white cell population
Granulocytes	Polymorphonuclear	Neutrophils	40 – 75
	Granulocytes	Basophiles	Approx. 1
	PMN	Eosinophils	1 – 6
	Monocytes		2 – 10
Non-granular Leucocytes	Lymphocytes		20 – 45
	Plasma cells		Ŏ

Ŏ: Rarely seen in blood, but present in the tissues.



Principle:

- Classification of polymorphonuclear granulocytes (PMN) is based on the size , shape , number and staining characteristics of their granules .
- ***Leishman's stain***
 - It is based on a mixture of methylene blue and eosin.
 - It differentiates between WBC as indicated in the following table:

	Type of Cell	Colour of the Stain
1	Neutrophils nuclei	Purple
2	Eosinophil granules	Orange – Red
3	Basophiles granules	Dark Blue
4	Lymphocytes nuclei	Dark Purple
5	Platelets granules	Violet
6	RBC	Pink