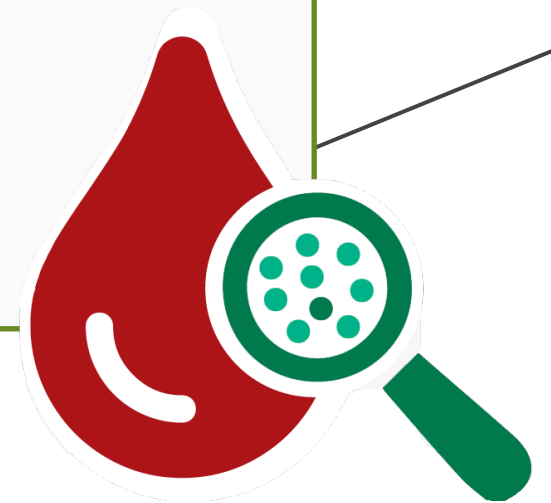
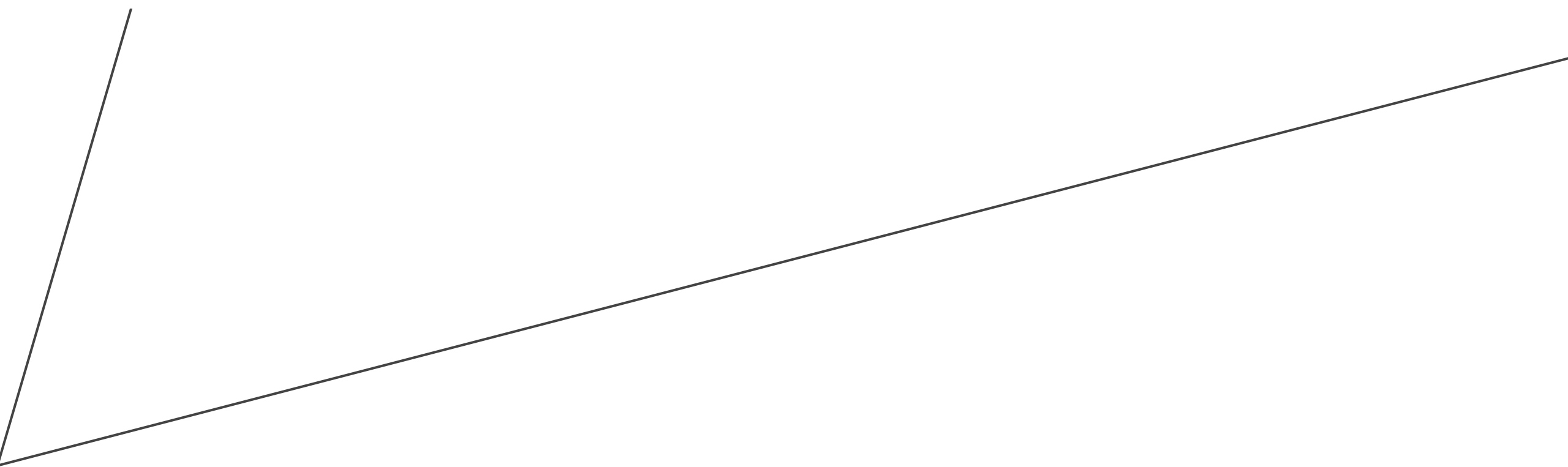


Blood Biochemistry BCH 220 [Practical]
Lab (10) Complete Blood Count (CBC)



Objectives

- To estimate the number of RBC in blood sample
 - To estimate the number of total WBC in blood sample
 - To perform a differential count for a blood sample
- 
- A decorative graphic at the bottom of the slide consists of two thin black lines. One line starts at the bottom left corner and extends diagonally upwards and to the right. The second line starts at the same point and extends diagonally upwards and to the right, but at a shallower angle than the first line, creating a wide, open triangular shape.

Introduction

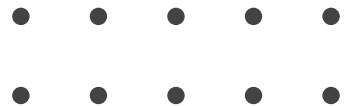
- **Complete blood count (CBC)** is a series of tests used to evaluate the composition and concentration of the cellular components of blood.

A CBC test usually includes:

- I. **WBC count**
- I. **WBC differential count**
- II. **RBC count**
- III. **HCT ?**
- IV. **Hb**
- V. **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).
- VII. **Platelet count**
- VIII. **Mean platelet volume (MPV)**
- IX. **Platelet distribution width (PDW)**

Complete Blood Count (CBC)

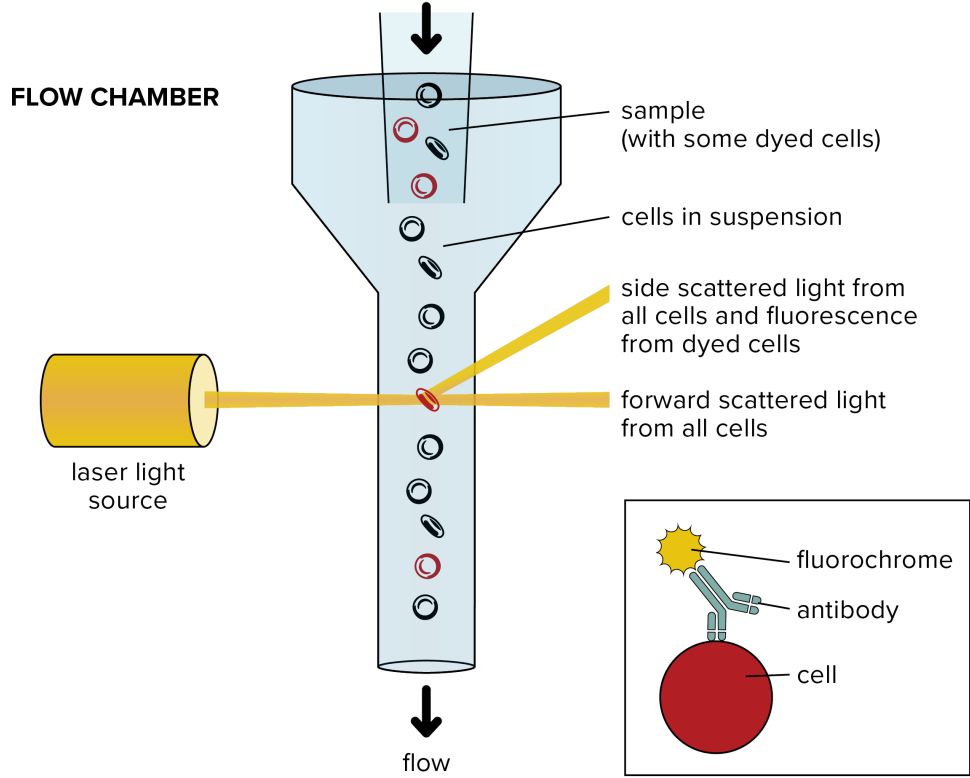
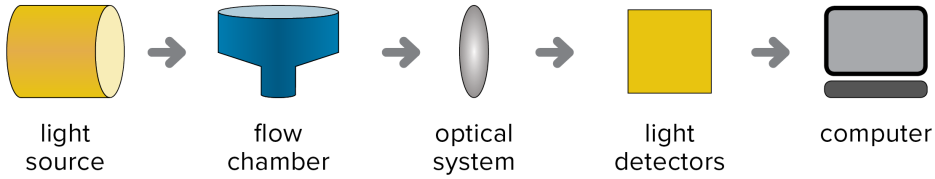
- **CBC can applied by two way:**
 1. Automated blood count
 2. Manual blood count
- **Automated blood count:**
 - CBC is performed by an automated analyzer that counts the **numbers** and **types** of different cells within the blood.
 - It aspirates a very small amount of the sample through the narrow 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*.





Flow cytometer

BASIC COMPONENTS OF A FLOW CYTOMETER



Mindray BC-2800 Auto Hematology Analyzer



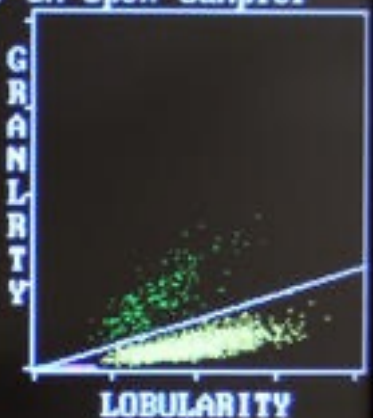
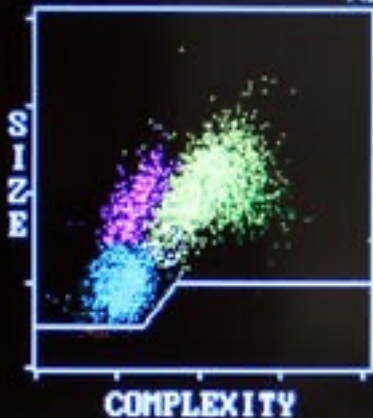
Coulter Ac•T 5diff CP (Cap Pierce) Hematology Analyzer

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

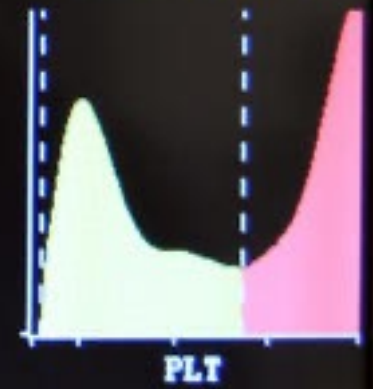
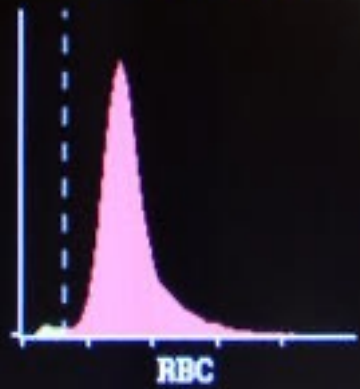
Paran: 1 Limits: 1
WBC 5.00 K/uL
NEU 2.57 51.3 %N
LYM 1.74 34.9 %L
MONO .392 7.85 %M
EOS .239 4.79 %E
BASO .056 1.12 %B



MCT:4.52

RBC 5.93 M/uL
HGB 14.1 g/dL
HCT 41.6 %
MCV 78.2 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

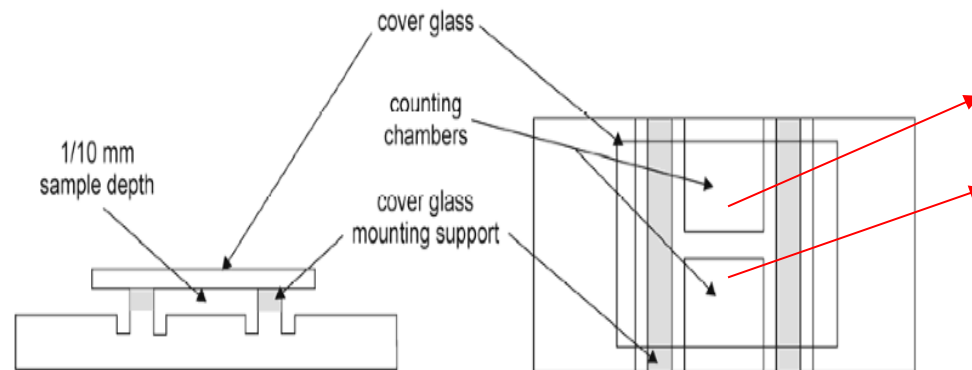
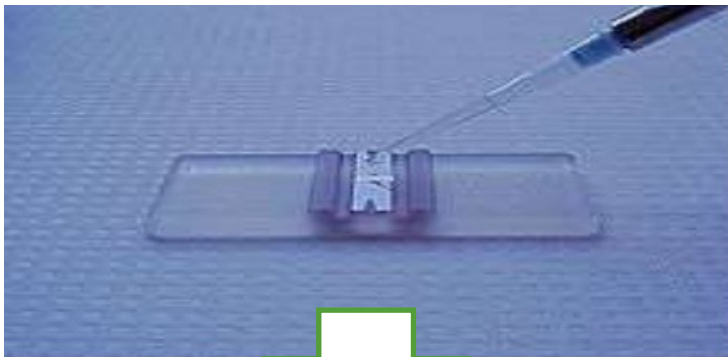
FAX (208) 527-3791

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

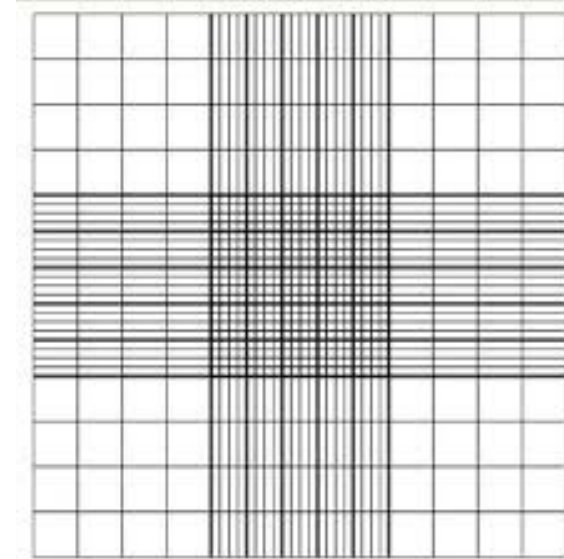
Test Name	Result	Units	Flag	Reference Range
CBC W/ 5 PART DIFF. (X6)				
<i>Run by: TB on 2/22/2012 09:33</i>				
WBC	2.1	K/uL		4.0 - 11.2
RBC	4.15	M/uL		4.00 - 5.60
HGB	13.5	gm/dL		12.0 - 16.0
HCT	39.5	%VOL		35.0 - 50.0
MCV	95	fl		82 - 98
PLATELETS	172	K/uL		140 - 440
MCH	32.6	pg		26.0 - 36.0
MCHC	34.3	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.



Counting Grid



(A) RBC Count

- It is test done to determine 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

High RBC count

- Polycythemia
- Renal problem, **Why ?**

Normally high (RBC count)

- People who live at high altitudes
- Smokers

} Oxygen is low \rightarrow RBC synthesis increases

(B) WBC Count

- Total leukocytes count shows the **number of WBC** in a sample of blood.
- A normal WBC count is between **4,500-11,000** cells per cubic millimeter (mm³).
- The number of WBC is sometimes used to identify an infection or to monitor the body's response to treatment.

Low WBC count → Leukopenia

High WBC count → Leukocytosis

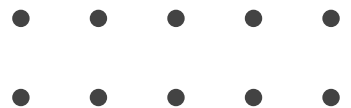
Principle

A. Regarding RBC

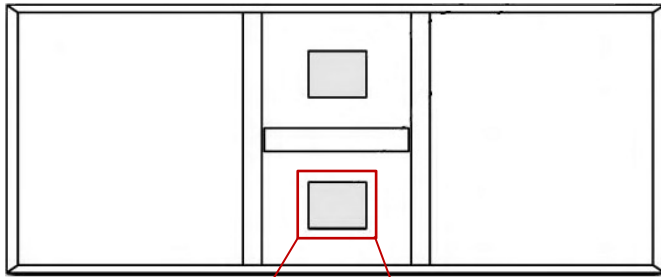
- The process involves by **counting cells** in several squares of the grid and obtain an **average number**.
- This number is **multiply** by a factor that compensates the **amount of dilution**.
- The final results expresses the number of **RBC /mm³** of original blood sample.

B. Regarding WBC

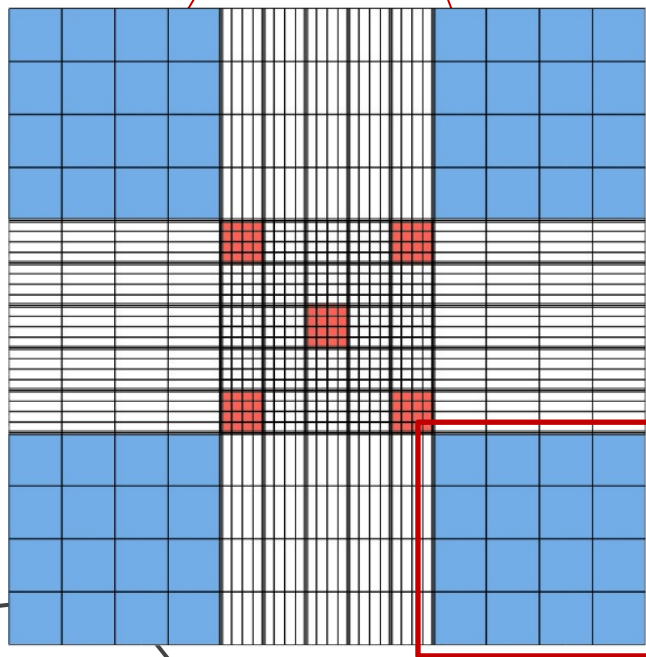
- It is necessary to obtain **RBC free** preparation of WBC from blood.
- Suspension of the red blood cell in a very **hypotonic solution** will lead to the destruction of RBC.



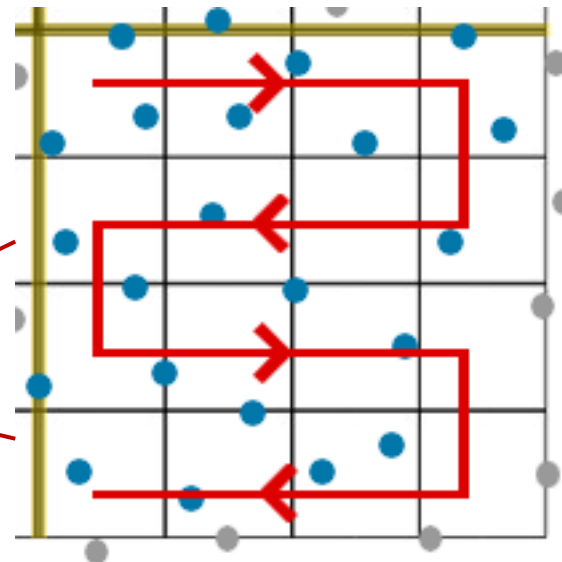
Neubauer Hemocytometer



How To Count Blood Cells ?



- Areas where **WBCs** are counted
- Areas where **RBCs** are counted



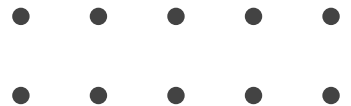
Counting **2 sides** in L shape
(i.e. count the cells settled on the top and left sides)

(Yellow sides)

And (exclude the cells on the bottom and right sides)

Counting in zigzag in all squares

Blue cells are counted
Grey cells aren't counted



Calculations

- **RBC blood cell count (5 squares)**

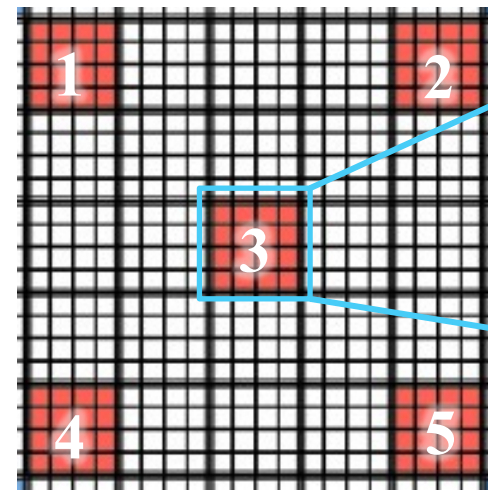
- Find the sum of RBCs in **5 large squares**, and **divide it with 80** (5×16) small squares to find the average in one square, **multiply it by 200** to allow for the dilution and then **multiply by 4000** to obtain the number per cubic milliliter.

- **The sum of RBCs in 5 large squares** = $(84) + (71) + (63) + (93) + (83) = 394$ cells.

- **The average of RBCs in one square** = $\frac{394}{5 \times 16} = \frac{394}{80} = 4.9$ cells.

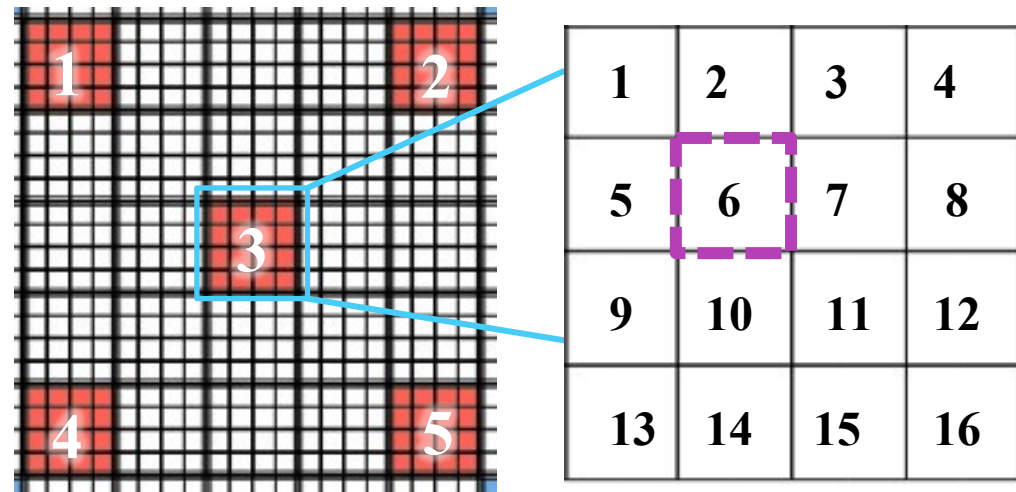
- **RBC count** = $4.9 \times 200 \times 4000 = 4$ million/mm³.

- **Normal range = 4.2-5 million/mm³**



1	2	3	4
5	6	7	8
9	10	11	12
13	14	15	16

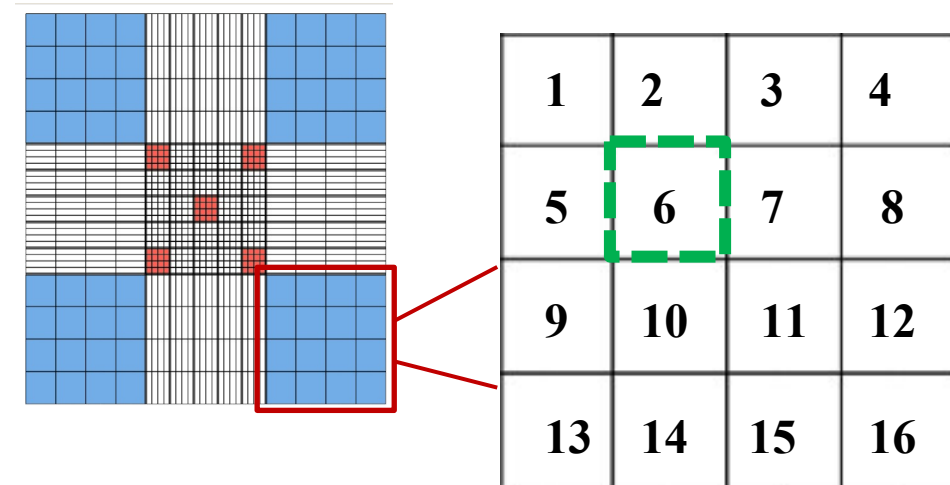
- **RBC count**= The average of RBCs in one square (1) x Dilution factor (2) x **Reciprocal of volume (3)**
- The sum of RBCs in 5 large squares = (84) + (71) + (63) + (93) + (83) = 394 cells.
- **(1) The average of RBCs in one square**= $\frac{\text{No. of cells in 5 squares}}{\text{No. of small squares}} = \frac{394}{(16 \times 5) = 80} = 4.9$ cells.
- **(2) Dilution factor**= $\frac{\text{Final volume (RBC solution)}}{\text{aliquot volume (blood sample)}} = \frac{200}{1} = 200$
- **(3) Reciprocal of volume**= $\frac{1}{\text{cubic volume}} = \frac{1}{0.00025 \text{ mm}^3} = 4000$
- **RBC count**= $4.9 \times 200 \times 4000 = 4$ million/mm³



Calculations

- **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 =** $\frac{69}{64} = 1.07$ cells.
 - **WBC count =** 1 x 20 x 160 = 3200 Cells/mm³.

- **Normal range = 4500-11000 cells /mm³**



• **WBC count**= The average of WBCs in one square (1) x Dilution factor (2) x **Reciprocal of volume (3)**

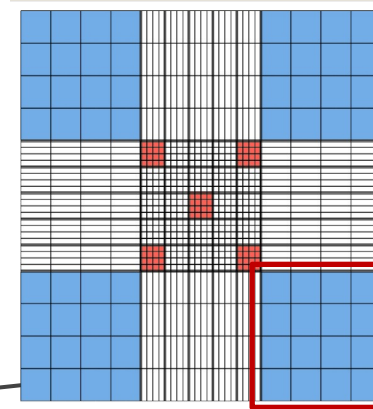
• **The sum of WBCs in 4 large squares** = (16) + (21) + (17) + (15) = 69 cells.

• **(1) The average of WBCs in one square**= $\frac{\text{No. of cells in 4 squares}}{\text{No. of small squares}} = \frac{69}{(16 \times 4) = 64} = 1.07$ cells.

• **(2) Dilution factor**= $\frac{\text{Final volume (WBC solution)}}{\text{aliquot volume (blood sample)}} = \frac{20}{1} = 20$

• **(3) Reciprocal of volume**= $\frac{1}{\text{cubic volume}} = \frac{1}{0.00625 \text{ mm}^3} = 160$

• **WBC count**= 1 x 20 x 160 = 3200 Cells/mm³.



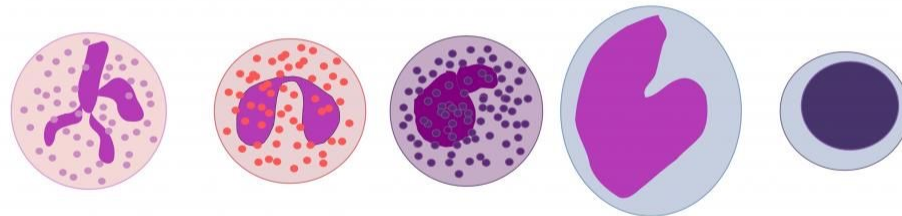
1	2	3	4
5	6	7	8
9	10	11	12
13	14	15	16

(C) Differential Count

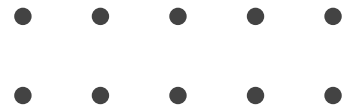
- It determines the **number** of each **type of WBC** present in the blood.

Class of white cells	White cell type		% of total white cell population
Granulocytes	Polymorphonuclear	Neutrophils	40 - 75
		Eosinophils	1 - 6
		Basophils	Approx. 1
Non-granular leukocytes	Mononuclear	Monocytes	2 - 10
		Lymphocytes	20 - 45
		Plasma Cells	*

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



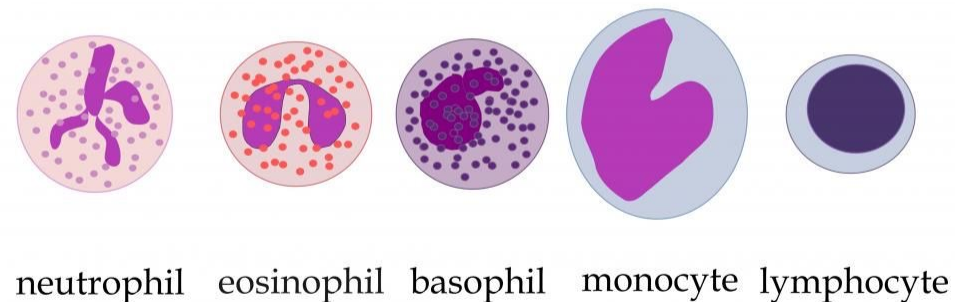
neutrophil eosinophil basophil monocyte lymphocyte

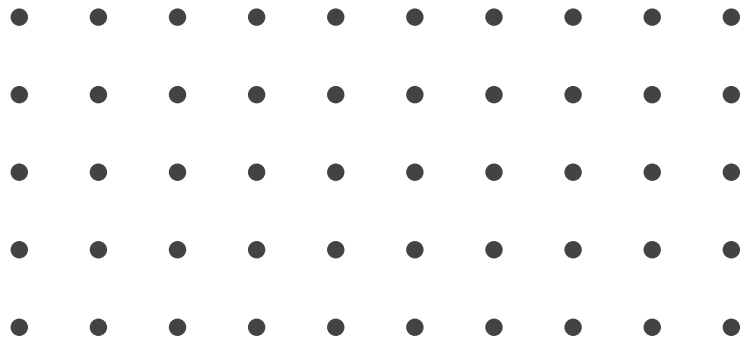


Principle

- 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	Color of the stain
Neutrophils nuclei	Purple
Eosinophils granules	Orange - Red
Basophils granules	Dark Blue
Monocytes cytoplasm	Grey blue
Lymphocytes nuclei	Dark Purple
Platelets granules	Violet
RBC	Pink





تم بحمد الله ... 